



University Ss. Cyril and Methodius in Skopje Faculty of Agricultural Sciences and Food





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# SYMPOSIUM PROCEEDINGS

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# 2<sup>nd</sup> INTERNATIONAL SYMPOSIUM FOR AGRICULTURE AND FOOD ISAF 2015

# XXXVIII FACULTY-ECONOMY MEETING V SYMPOSIUM OF VITICULTURE AND WINE PRODUCTION VIII SYMPOSIUM FOR VEGATABLE AND FLOWER PRODUCTION X INTERNATIONAL CONFERENCE OF ASSOCIATION OF AGRICULTURAL ECONOMISTS OF REPUBLIC OF MACEDONIA VI INTERNATIONAL SYMPOSIUM OF LIVESTOCK BREEDING

Organized by

Faculty of Agricultural Sciences and Food of Ss. "Cyril and Methodius" University in Skopje, Republic of Macedonia in co-organization with Institute of Animal Sciences of Ss. "Cyril and Methodius" University in Skopje, Republic of Macedonia

# SYMPOSIUM PROCEEDINGS

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We are proud to present you with the Proceeding of Papers as an outcome of the Second International Symposium for Agriculture and Food, organized in 2015. This event represents the biggest gathering of the scientific public in the area of agriculture and food in Republic of Macedonia.

Within the framework of the general event several symposiums and conferences took place:

VIII<sup>th</sup> Symposium for Vegetable and Flower Production

V<sup>th</sup> Symposium for Viticulture and Wine Production

X<sup>th</sup> International Conference of the Association of Agricultural Economists of Republic of Macedonia, and

VI<sup>th</sup> International Symposium for Livestock Breeding.

The Faculty of Agricultural Sciences and Food, University "Ss. Cyril and Methodius" in Skopje, has organized this event for the occasion of its 68<sup>th</sup> anniversary. It also represents a faculty's commitment and responsibility for conducting and following scientific and research activities as well as for presenting novel results for the relevant stakeholders in the area of agriculture and food.

This event has confirmed the accomplishment of the faculty's vision for being a leading institution in the area of agriculture in Republic of Macedonia, recognized not only in the region but also in Europe and worldwide. The attendance of eminent authors from 21 countries, presenting more than 220 scientific papers, proofs the important role of the faculty for the agricultural and rural development. The papers were presented in 10 parallel sections covering research topics from the area of agriculture, food and environment protection.

The Symposium Proceedings includes 142 scientific papers published in two volumes. The abstracts of all submitted papers (337 in total) were published in Book of Abstracts, prior the organization of the Symposium itself. Part of the submitted papers will be published in the faculty's Journal of Agriculture and Environmental Sciences. Another part of the papers presented at this Symposium will be published in other well-known international journals, as an added value for the scientific importance of this Symposium.

We sincerely hope that with the organization of the Symposium and with the publishing of the Proceeding of Papers we significantly contribute to the science in the area of agriculture and food. We expect that the published papers will be beneficial for the scientists and the experts in their future scientific activities.

Sincerely,

Prof. Dr. Dragi Dimitrievski President of the Organizing Committee of ISAF 2015 Dean of the Faculty of Agricultural Sciences and Food CIP - Каталогизација во публикација Национална и универзитетска библиотека "Св. Климент Охридски", Скопје

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# ESTIMATION OF ACTUAL EVAPOTRANSPIRATION FOR WHEAT USING EDDY COVARIANCE METHOD IN THE THRACE REGION OF TURKEY

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#### Abstract

Along with the industrial revolution, human power has been replaced by the machine power. Accordingly, fossil fuel consumption has resulted in increased concentrations of greenhouse gases in the atmosphere. This situation has led to ongoing global warming and climate change phenomena. As the temperature rises due to global warming, it can also increase the actual evapotranspiration in the agricultural areas. For this reason, accurately determination and measurement of the evapotranspiration is extremely important. Better determination of the actual evapotranspiration allows the efficiency of water usage by irrigation. The Eddy Covariance, which is one of the micrometeorological methods, used to determine the actual evapotranspiration of vegetation, enables to measure the fluxes directly and at high frequency. In this study, actual evapotranspiration of a winter wheat cultivar (Selimiye) was indicated by the Eddy Covariance method for one growing period between 1 November 2012 and 30 July 2013, in the Kırklareli city located in the Northwest part of Turkey. At the same time, relations between the actual evapotranspiration, vegetation dynamics and meteorological variables were also analyzed.

Key words: Latent heat flux, actual evapotranspiration, winter wheat, Kırklareli/Turkey.

#### Introduction

Increasing global mean temperatures revealed the evaluation needs on greenhouse gases concentrations and the associated emission amounts. Therefore, amounts of carbon dioxide released to and captured from the atmosphere as major components of global carbon cycle have been searched by scientists. Researches especially over water, agricultural and forest surfaces allow us to understand the corresponding variations in  $CO_2$  and  $H_2O$  fluxes. By these means, the evapotranspiration is crucial to be determined but difficult to measure just because it is affected by numerous climatic factors like global radiation, wind speed, air temperature, and relative humidity (Drexler et al., 2004).

Evaluation of  $CO_2$  and  $H_2O$  fluxes over agricultural areas became more important for related studies on global carbon budget and climate change. That's why some flux networks are established to measure carbon dioxide, water vapor and energy fluxes between different terrestrial ecosystems (forests, croplands, grasslands etc.) and the

atmosphere. Concordantly, most of the related micrometeorological systems were installed firstly in forests rather than in croplands. In this way, amount of the micrometeorological tower sites is increasing in order to measure the needed mass and energy fluxes above vegetation. Studies over different plant surfaces show that continuous measurements involving whole of the vegetation-growing periods are necessary. Although many studies about greenhouse gases over different vegetation types exist since 1990's (Saito et al., 2005), the first steps have been taken yet on these issues in Turkey. But there are some efforts to increase number of research activities in order to estimate energy and gas fluxes between atmosphere and terrestrial ecosystem in Turkey. The Eddy Covariance technique has been widely used during the past 20 years to measure exchange fluxes of mass and energy between ecosystems and the atmosphere over vegetation surfaces directly (Moureaux et al., 2008). The essence of Eddy Covariance method is that vertical flux can be represented as a covariance of the vertical velocity and concentration of the entity of interest (Burba and Anderson, 2010). It is an advantageous method in terms of providing continuous long-term measurements between the atmosphere and surface over a large area. Besides, instruments of Eddy Covariance system can detect very small changes of gas concentrations at very high frequency. Micrometeorological methods like Eddy Covariance have been widely used for the estimation of energy fluxes above the canopy. Water loss by evapotranspiration can be determined by the flux of latent heat (LE) which is the amount of required energy to change the liquid phase of water into vapour (gas) phase. LE is one of the energy flux components and it can be converted to evapotranspiration dividing by latent heat (L). Hence, actual crop water consumption (evapotranspiration) can be calculated as mm.

As well known, there are three terms for evapotranspiration which are potential, reference and actual. Potential evapotranspiration based on meteorological variables under sufficient water conditions. Reference evapotranspiration also based on meteorological variables above well-watered grass or alfalfa covered surface. Actual evapotranspiration is controlled by not only meteorological variables but also actual environmental conditions such as crop type, soil water conditions, etc.

Annual global agricultural productivity varies significantly depending on meteorological conditions. Evapotranspiration affects crop growth and development. Hence, quantifying the crop water use by evapotranspiration is important to schedule and management for irrigation (Smith, 2000; Parent and Anctil, 2012). There are some direct methods like lysimeter and indirect methods like Eddy Covariance, Bowen ratio, remote sensing and scintillometer to measure actual evapotranspiration (Jung et al., 2010). In this study, as an indirect method, Eddy Covariance was chosen in order to be representative of the obtained results not only for the point that the measurements are taken but also for larger area, because direct methods give results for limited area (a few m<sup>2</sup>) or only for the point where measurement device was established. Measurements were obtained by Eddy Covariance method due to higher speed measurement (10 Hz interval) than the other methods and continuous data recording.

The purpose of this study is to determine the actual evapotranspiration of winter wheat during growing season of 2012-2013 by using Eddy Covariance method. Additionally, it is aimed to derive the crop coefficient ( $k_c$ ) of winter wheat and relationships between evapotranspiration and vegetation dynamics.  $k_c$  can be calculated by the ratio of actual evapotranspiration and reference evapotranspiration. This coefficient is used to calculate actual water consumption through .reference evapotranspiration.

Unfortunately, especially in developing countries, there are no specific coefficients for their crop types to calculate actual evapotranspiration. Thus suggested literature coefficients have to be used. For this reason, we estimated our own crop coefficients for winter wheat in the experiment field.

#### **Material and Methods**

This study was conducted at the experiment field of the Atatürk Soil Water and Agricultural Meteorology Research Station Directorate in the Kırklareli city of Turkey (41°41'53" N, 27°12'37" E). Figure 1 shows the experiment location. The Eddy Covariance measuring system and automatic agrometeorological station were established on the winter wheat field, which corresponded to an area of approx. 9.2 ha. The Eddy Covariance system includes an open-path infrared  $CO_2/H_2O$  gas analyzer (LI-7500), a 3D sonic anemometer (CSAT3, Campbell Sci.) and a data logger (CR1000, Campbell Sci). The agrometeorological station consists of the sensors for the measurement of temperature, relative humidity, wind speed and direction, soil water content, soil temperature, global solar radiation, net radiation and photosynthetical active radiation at the different levels. In the agrometeorological station, data were measured in an interval of 1 s and stored every 30 min. Calculations of this study are derived by using 30-min averages of continuous Eddy Covariance data covering 2012-2013 growing period of winter wheat (from 1 November 2012 to 30 July 2013), logged at 10 Hz.

Some of the flux and sampling errors were removed from the collected data and cross wind, coordinate, Webb, Pearman and Leuning (WPL) and rotation (double) corrections were applied to calculate water vapor and energy fluxes (Webb et al., 1980; Fuehrer and Friehe, 2002). These errors are resulted from the technical problems and weather conditions (rain, calm winds during night time and wind direction).



Fig. 1. Atatürk Soil Water and Agricultural Meteorology Research Station Directorate. The water vapor flux is expressed as latent heat flux (W  $m^{-2}$ ) which is determined as follows (Burba, 2013):

$$LE \equiv \lambda E = \lambda \frac{M_w/M_a}{\bar{P}} \overline{\rho_d} \overline{w'e'}$$
(1)

where  $\lambda$  stands for the latent heat of evaporation, which varies with the air temperature as (Aubinet et al., 2000):

$$\lambda = (3147.5 - 2.37Ta)10^3 \tag{2}$$

The latent heat flux describes the energy used in process of evaporation, transpiration, or evapotranspiration.

In order to investigate the relationship between actual and reference evapotranspiration and determine crop coefficient  $(k_c)$ , the Food and Agriculture Organization of the United Nations (FAO) Penman-Monteith equation (Allen et al., 1998) can be used:

$$ET_0 = \frac{408\Delta(R_n - G) + \gamma \frac{900}{T + 273} u_2(e_s - e_a)}{\Delta + \gamma(1 + 0.34u_2)}$$
(3)

where  $\text{ET}_0$  is the reference evapotranspiration (mm day<sup>-1</sup>);  $R_n$  is the net radiation of the crop surface (MJ m<sup>-2</sup> day<sup>-1</sup>); G is the soil heat flux density (MJ m<sup>-2</sup> day<sup>-1</sup>); T is the mean daily air temperature at 2 m height (°C);  $u_2$  is the wind speed at 2 m height (m s<sup>-1</sup>);  $e_s$ - $e_a$  represents saturation vapor pressure deficit (kPa);  $\Delta$  shows the slope of the vapor pressure curve (kPa °C<sup>-1</sup>); and  $\gamma$  is given for the psychrometric constant (kPa °C<sup>-1</sup>). For daily calculations, G may be ignored as (G~0) (Gavilian et al., 2007).

#### **Results and Discussion**

Figure 2 shows the time series of latent heat flux (LE) for winter wheat during growing season (between 1 November 2012 and 30 July 2013). Mean value of LE is calculated as  $38.54 \text{ Wm}^{-2}$ , where the maximum value is obtained as  $479.97 \text{ W/m}^2$ . As of the middle of April, the air temperature, canopy height and biomass have been increased remarkably. As a result, actual evapotranspiration (ET) is increased in that period.

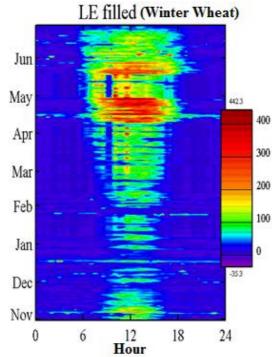


Figure 2. LE Diagram for winter wheat in 2012-2013 growing season.

Reference evapotranspiration is calculated by Penman-Monteith equation (3) whereas the actual evapotranspiration is directly measured using the Eddy Covariance method. Time series of  $ET_0$  and ET are given in Figure 3. According to this aspect, generally,  $ET_0$  is higher than ET as expected. Difference between the  $ET_0$  and ET become more prominent during the late season. Total  $ET_0$  and ET in the 2012-2013 growing season of winter wheat are 534.05 and 325.46 mm, respectively. Determination coefficient ( $R^2$ ) between  $ET_0$  and ET is calculated as 0.43.

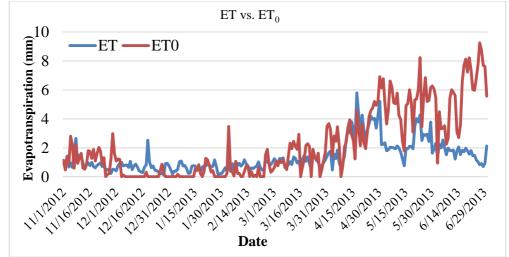


Figure 3. Time Series of Reference Evapotranspiration (ET<sub>0</sub>) and Actual Evapotranspiration (ET).

According to Figure 4, crop coefficients which were determined by FAO and calculated from the Eddy Covariance technique are established comparatively for the growing season. The average crop coefficient of winter wheat in 2012-2013 growing season was found as 0.62.

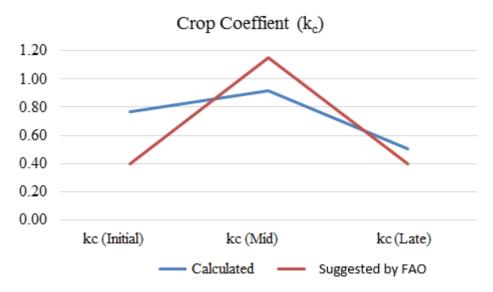


Figure 4. Calculated and suggested (by FAO) crop coefficients of winter wheat during the growing season.

#### Conclusion

In this study, one of the micrometeorological approaches (Eddy Covariance) has been used to indicate the actual evapotranspiration of winter wheat during the growing period of 2012 and 2013 in the Kırklareli city of Turkey. Additionally, crop coefficient of the winter wheat variety was calculated using actual ET measurements. Differences between  $ET_0$  and ET were revealed together with the variations in crop coefficients for the growing season in Kırklareli/Turkey.

It has been estimated daily average of  $ET_0$  and ET as 1.97 and 1.20 mm, respectively, for winter wheat during the growing period covering 271 days. Additionally, calculated  $k_c$  value for initial period of winter wheat was higher than the FAO suggested  $k_c$  whereas in the calculated  $k_c$  in the middle period was less than suggested one. The minimum difference between them has been estimated for the late growing season of winter wheat. Properly identification of these coefficients for different climates and plant varieties is extremely important when considering the importance of the crop coefficient for the calculation of actual evapotranspiration and irrigation amount. These differences can be caused by the results based on measured data of only one growing season. In order to derive accurate  $k_c$  values for winter wheat, it is necessary to use longer data set including different growing season values.

In the future, accurate measurements of evapotranspiration especially using micrometeorological approaches should be done and expanded throughout the country because it is crucial variable for sustainable agriculture; efficiency of irrigation systems;

water management of all agricultural crops and for food security. Additionally, updated  $k_c$  values would provide the opportunity of determining the actual evapotranspiration with higher precision.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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# **RECOMMENDATIONS FOR AGRICULTURAL DROUGHT ASSESSMENT UNDER LITHUANIAN CLIMATIC CONDITIONS**

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#### Abstract

Among the environmental stresses, drought stress is one of the most adverse factors that negatively impacts plant growth and productivity. Therefore, understanding and qualifying drought occurrence and its consequences on agricultural production, hydrologic cycle, and ecosystems is of particular importance. In the context of climate variability and change, water scarcity and food security, it is important to use more comprehensive data on rainfall, temperature and soils in computing drought indices. Currently for drought monitoring different indices, easily understandable, are used by end users. The Standardized Precipitation Index (SPI) was proposed by McKee et al. (1993) and it has been increasingly used during the last decades because of its solid theoretical development, robustness and versatility in drought analyses. Nevertheless for many years, the humidity of vegetation period has been described using Selyaninov's hydrothermal coefficient (HTK) in Lithuania. According to Farrago's classification, it belongs to drought indices of water balance group "supply/demand" as it uses daily values of precipitation and air temperature for the calculation of the period. Anyway drought affects water availability not only by changing regional precipitation level and temporal variability, but also by affecting water flows and soil moisture dynamics. Therefore it is essential to have soil moisture data as one of the most important factors for evaluation of drought occurrence. Finally when all meteorological data are observed and soil moisture is monitored it is very important to know what stress reaction of plants is. Results of investigation revealed a very strong negative relationship between soil moisture water content and proline content as well as between soil moisture water content and soluble sugar content in leaves. It was suggested to measure proline content. The study was done during 2013-2014 under all Lithuanian territory, covering different soils and different climatic conditions. The aim of study was to prepare agricultural drought assessment which particular could be used by extension service and crop insurance companies. Proposed recommendations under Lithuanian climatic conditions include several steps: the first is calculation of soil moisture deficit and comparison results with SPI as well with HTC; the next step is monitoring of actual soil moisture. The last step should be done when SPI, HTC and soil moisture deficit reaches critical point, and then should be taken samples of plants for measuring proline level. Key words: drought, precipitation, Watermark, plant stress

# Introduction

Drought or water deficit stress is the major environmental factor that negative impacts agricultural yield throughout the world (Selote and Khana-Chopra, 2004). Generally drought stress occurs when the available water in the soil is reduced and atmospheric conditions cause continuous loss of water by transpiration or evaporation. Under the influence of global change droughts will be more frequent and more intense in large parts of the world (Seneviratne, 2012); (IPCC, 2012); (IPCC, 2014). Drought monitoring using drought indexes often serves as an important base (Asadi Zarch et al., 2015) and can be monitored effectively using drought indices such as the Palmer Drought Severity Index (Palmer, 1965), the Standardized Precipitation Index (SPI) (McKee et al., 1995) calculated with in-situ meteorological data from weather stations or others. It is a difficult to assess droughts due to variations in temporal and spatial extends of the complex events and their severity, there is no universal drought indicator and previous studies identified significant discrepancies between the drought indices (Altman, 2012). For many years in Lithuania, the humidity of vegetation period has been described using Selyaninov's hydrothermal coefficient (HTC). According to Farago's classification, it belongs to drought indices of water balance group "supply/demand" (Farago et al., 1989). Despite the limitations of Selyaninov's hydrothermal coefficient, it is still used for the evaluation of the humidity of period in the Lithuanian climatic conditions and it is confirmed to be a drought index by the Lithuanian Government's decision.

Drought stress is characterized by reduction of water content, diminished leaf water potential and turgor loss, closure of stomata and decrease in cell enlargement and growth (Jaleel et al., 2009). During osmotic stress, plant cells accumulate solutes to prevent water loss and to re-establish cell turgor. The solutes that accumulate during the osmotic adjustment include ions such as K<sup>+</sup>, Na<sup>+</sup>, and Cl<sup>-</sup> or organic solutes that include nitrogencontaining compounds, such as proline and other amino acids, polyamines and quaternary ammonium compounds like glycine betaine (Tamura et al., 2003). The organic solutes are compatible with cellular processes and accumulate to high levels in the cytosol with increasing drought. Production of osmolytes is a general way to stabilize membranes and maintain protein conformation at low leaf water potentials (Szabados and Savoure, 2009). Proline is known to be involved in reducing the photodamage in the thylakoid membranes by scavenging and/or reducing the production of  $O_2$ . Proline accumulation in plants is caused, not only by the activation of proline biosynthesis, but also by the inactivation of proline degradation, thereby resulting in a decrease in the level of accumulated proline in rehydrated plants. It can also infer that proline acts as a free radical scavenger and may be more important in overcoming stress than in acting as a simple osmolytes (Reddy et al., 2004). Besides, proline and soluble sugars may protect membranes from damaged and stabilize the structures and activities of protein and enzymes (Lee et al., 2008; Ben Ahmed et al., 2009; Hessini et al., 2009).

Understanding and qualifying drought occurrence and its consequences on agricultural production, hydrologic cycle, and ecosystems is of particular importance. It is obvious that determining the drought hazards is quite difficult and complicated. For that, the aim of the study was to describe steps of agricultural drought assessment as recommendations which could be used by extension service and crop insurance companies.

#### Material and methods

The study was carried out under all Lithuanian territory, covering different soils and different climatic conditions (Fig. 1). Meteorological conditions were evaluated according data taken from Lithuanian Hydrometeorological service. Actual soil moisture volumetric content has been carried during 2013-2014 out in 7 different objects by thermostatic method at the depth of 20 cm in three repetitions. At the same places soil moisture content was measured by Watermark sensors (W, cbar).

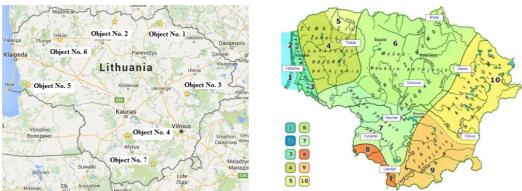


Figure 1. Objects area location

The Selyaninov hydrothermal coefficient (HTC) was calculated during 2003-2013 as follows (Selianinov, 1928):

$$HTC = \frac{10P}{\sum T_{10}},\tag{1}$$

Where P – the sum of precipitation of the calculation period, mm,

 $T_{10}$  – the sum of air temperature values higher than 10 °C of calculation period, °C.

The value of HTC < 0,3 – means very dry; 0,4-0,5 – dry; 0,6-0,7 – middle dry; 0,8-1,0 – not enough humid; 1,0-1,5 – enough humid, > 1,5 - wet (Dirse et al., 1984). Standardized Precipitation Index (SPI), accepted by the World Meteorological Organization as the reference drought index for more effective drought monitoring and climate risk management (Hayes et al., 2011) was used. SPI was calculated using the long-term precipitation record of 1982-2014 with in-situ meteorological data from weather stations using SPI SL 6.exe software available at National drought mitigation centre website (Program., 2015).

Table 1. SI I values (National Diough, 2000)			
SPI value	Interpretation	SPI value	Interpretation
$\geq$ 2.0+	extremely wet	-1.0 to -1.49	moderately dry
1.5 to 1.99	very wet	-1.5 to -1.99	severely dry
1.0 to 1.49	moderately wet	-2 and less	extremely dry
99 to .99	near normal		

Table 1. SPI values (National Drough, 2006)

In order to assess agricultural drought and plant water status it is needed to evaluate different time scales as the rainfall distribution differs a lot during separate month.

Therefore, the HTC and SPI was assess for one month period. Experimental investigation of plant response to droughts was carried out in 2013 at the laboratory of Agrobiotechnologies of Aleksandras Stulginskis University, Lithuania. Five seeds of pea cultivar "Ilgiai" were sown to each vegetation pot 0.16 m x 0.23 m (height x diameter) with peat substrate and growing in a growth chamber at 20/18°C (day/night), under illumination 50  $\mu$ mol m-2 s-1, photoperiod 16/8 h (day/night). The plants were submitted to two conditions: control and drought stress. The control plants were normally irrigated. The appropriate amount of water for the control plants was determined during the pre-experiments. Drought treatment was started three weeks after germination. Proline content was determined using a revised ninhydrin method (McClinchey, Kott, 2008) every second day.

#### **Results and discussion**

As the droughts occur depending on meteorological conditions, first of all meteorological conditions should be evaluated. As it was mentioned in introduction there is a lot different drought indexes which could be used. After analysis of existing indexes and evaluation of recommendations by WMO two main indexes were calculated and compared - HTC and SPI. The comparison of drought indices was complicated due to different methodological approaches and identified dry periods according HTC and SPI were different in 39 of the 50 cases (78 %). Excessively wet and optimally wet according HTC were 52 % and 24 % accordingly SPI. Main reasons are: interpretation of values as SPI is rated to seven levels, HTC - to six; wet period's identification according SPI is more detailed, while the HTC counts only excessively wet periods; secondly - SPI do not consider the beginning of active vegetation and if the actual vegetation starts in the middle of month SPI takes into account monthly precipitation values. Differently HTC is calculated from period when average air temperature is higher as 10 °C for three days and consider daily precipitation rate. During period of investigation the earliest beginning of active vegetation started at the end of March. This is at least 5-10 days earlier as annual average of many years. Earlier as normal, vegetation started in 2004-2005, 2009. In order to evaluate the equivalence of HTC and SPI was made correlation analysis (Fig.2). It was found out the very strong correlation (r=0.855) between HTC and SPI, thought the rating of values according HTC and SPI differed.

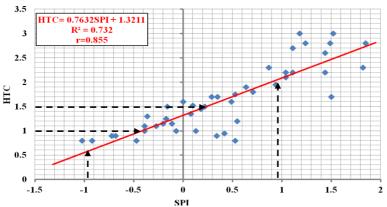


Figure 2. The relationship between Selyaninov hydrothermal coefficient (HTC) and Standardized Precipitation index (SPI)

It was found out that HTC < 0.7 equals to SPI < -0.8 and HTC < 0.5 equals to 1.0. The biggest differences determined during humid and wet periods. According to the dependence the SPI at near normal (0.2 - 0.99) are identified as wet according HTC (1 - 0.99)1.5). Still it is difficult to assess which index indicates drought better as drought is very complex. It was stated that HTC tends to overestimate as dry or wet periods, i.e. moderate humid periods are evaluated as dry or wet (Taparauskiene, 2009). Similar results confirmed by drought review done in 1961-1995 (Buitkuviene, 1998). Anyway for successful application of the standardized precipitation index in future necessary to assess the rating scale in compliance with Lithuanian Climatic conditions as well as to compare it with the actual data. As the SPI is recommended by WMO as reference drought index it is essential to integrate SPI with productive soil moisture content and to find the ways of SPI application to agricultural drought monitoring. Similar recommendations could be done regarding HTC. According research done in Lithuania, HTC does not demonstrate the actual role of meteorological conditions in plant growth (Daugeliene and Zekoniene, 2009). It would be expedient either to modify the methodology for calculation or to search for more appropriate calculation methods. HTC does not consider soil moisture, thus period of medium humidity is often evaluated as dry if there was a wet period before it or contrary (Taparauskiene, 2009). Main conclusion was done - it is not recommended to use HTC or SPI as only one index for drought evaluation. As it is evident that according HTC or SPI identification of agricultural droughts is complicated there is essential to monitor actual soil moisture. Under Lithuanian conditions after heavy rains there is accumulated sufficient amount of water in the soil. During dry periods, for operational use soil moisture can be monitored by different soil moisture indicators. In Lithuania agrometeorological service can provide observed values by Watermark sensors, still till 2013 sensors were not calibrated under existing conditions. After our study and calibration of sensors was determined critical values of soil moisture and readings of Watermark when drought should be considered (table 2).

Description of the	Object / place	Critical Watermark	Critical soil
soil	Object / place	value (cbar)	moisture (%)
Sand	No. 5 (Silute)	$\geq 80$	3.8-4.5
Coarse sand	No. 7 Varėna	$\geq 80$	3.9-7.0
Loam	Middle Lithuania (Dotnuva)	≥160	5.2-7.9
Clay loam	No.2 (Joniskis)	≥120	4.9-11.9
Silty Clay	No.1 (Birzai)	$\geq 80$	8.1-10.3
Loam Clay	No. 4 Prienai	≥160	12.0-13.0
Clay loam	No. 3. (Moletai)	≥100	7.0-10.3
Clay	No. 6 (Telsiai)	≥100	8.6-9.9

Table 2. Critical soil moisture values

Finally as it is known SPI and HTC, soil moisture content, still for crop insurance companies is essential to know what is the level of plant stress. Different plants react to water stress on different way. While some plants are adopted very well, yield reduces slightly, for others it can crucial. During investigation main attention was paid to find single parameter which reflects stress level and at the same time is easy measurable. To cope with drought stress, plant respond with physiological and biochemical changes. These changes aim at the retention of water in spite of the high external osmoticum and the maintenance of photosynthetic activity, while stomatal opening is reduced to counter water

loss (Parida et al., 2007). Various organic compatible solutes that effectively take part in plant stress tolerance include proline, glycine betaine, trehalose and several others (Ashraf, Foolad 2007; Nawaz, Ashraf, 2010; Ali, Ashraf, 2011). Normally, proline accumulation in plants is in response to drought or salinity stress occurs in the cytosol where it contributes substantially to the cytoplasmic osmotic adjustment (Chaves et al., 2003). It has been reported that proline content in leaves of many plants are enhanced by several stresses including drought stress (Abdel-Nasser, Abdel-Aal, 2002; Parida et al., 2002, Parida et al., 2007). Thus, we monitored the proline level in leaves of peas during drought period. The proline content did not change very much in control plants during the entire period of investigation, while in drought treated plants, proline content increased dramatically (Fig. 3).

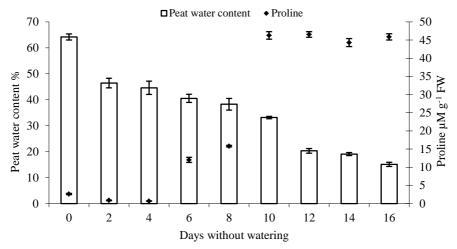


Figure 3. The effect of peat water content on proline level in the leaves of Pisum sativum

Our results of drought-induced dramatic increase in proline contents in leaves of pea agree with other research reports of accumulation of proline as a compatible osmolytes during drought exposure (Ain-Lhout et al., 2001; Nayyar and Walia, 2003; Kavi-Kishor et al., 2005; Ali et al., 2013). Significant, very strong correlation between substrate water content and proline level (R < 0.01; r = -0.87) was established. After recovery from drought, the proline content in leaves significantly decreased and was almost equal to control (data not shown). Parida et al., (2007) proposed that increased accumulation of proline in cotton under drought stress might be due to the decreased activity of proline dehydrogenase, a catabolic enzyme of proline. Our results show that the increase of proline content during water stress is an adaptive mechanism in *Pisum sativum*.

#### Conclusions

Monitoring of droughts under Lithuanian climatic conditions for extension service and crop insurance companies are following:

1. Monitoring of meteorological conditions by SPI and HTC. Drought starts when HTC is equal or less than 0.7. HTC- 0.7 equals to SPI – -0.8 and HTC – 0.5 equals to SPI – -1.0.

- 2. As HTC or SPI identification is more suitable for meteorological drought that agricultural, is essential to monitor actual soil moisture content. The critical soil moisture content, respectively to the region and soil texture varies from 3.8 till 13 %. If the watermark sensor is used the critical value of indicator varies from 80 till 160.
- 3. The proline content did not change very much under normal water content of the substrate while under drought stress proline content increased significantly. For that proline test is suggested to use as the final step for agricultural drought assessment. Test should be done when drought is monitored according HTK or SPI and soil moisture content reaches critical stage (value).
- 4. HTK, SPI and watermark values are observed by Lithuanian Hydrometeorological service and could be provided. Additionally farmers should install moisture indicators for actual soil moisture content. The proline level can be measured under laboratory and should be requested by farmer or insurance company.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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UDC: 631.445.3(497.7) Original Scientific Paper

# THE INFLUENCE OF THE SUBSTRATE ON PROPERTIES OF CHERNOZEMS IN THE REPUBLIC OF MACEDONIA

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#### Abstract

This paper presents the results of the influence of the substrate on some physical, chemical and productive properties of chernozem in the Republic of Macedonia. Chernozems form on old quaternary sediments (Ovche Pole), holocene sediments - proluvium (Stip), pliocene sediments (Veles, and in parts of Tikvesh) and the pleistocene rivers sediments (in parts of Tikvesh). This substrate is of great importance for the properties of soil: it depends on the mechanical composition and physical properties, mineralogical composition of the clay, content of  $CaCO_3$ , saturation of the adsorption complex with Ca and Mg-ions, the formation of clay-humus complexes, the release of nutrients substances in the weathering of primary minerals and etc. Analysis of variance (ANOVA) was made for all analyzed properties for samples of different size. The effect of the substrate in chernozems was established as well as its interaction with the variability of the examined properties. The significance of the differences among the mean values of analyzed properties per substrate was determined with the Tukey test, for p < 0.05 level. All statistical analyses were made with the R software package. The analysis of variance has shown that in chernozems, the parent material (substrate) has significant effect over the variability of the content of coarse, clay, physical clay (silt+clay) and physical (coarse+fine) sand. The coarse content is highest, mean (7.90%) and lowest on clay, mean (22.64%) in chernozems formed over pleistocene rivers sediments (P.R.S) and their difference from the others is statistically significant, and the content of fractions physical clay is highest, mean (51.90%) and lowest on physical sand, mean (47.44%) in chernozems formed over holocene sediments proluvium (H.S-P) and their difference from the others is statistically significant. Due to the similar bio-climatic conditions of formation there are no statistically significant differences regarding the chemical properties depending on the substrate on which the chernozemsare formed.

Key words: substrate, properties, chernozem

#### Introduction

Chernozems belong to the large group of soils - Mollisols. In the Republic of Macedonia they can be found in the central, driest region of the Vardar geotectonic zone in 4 valleys: Tikvesh, Ovche Pole, Shtip, Veles region, with a total surface area of over 30.000ha. On the pedological map of 1: 50.000 scale there are no special cartographical units for

chernozems. In these valleys they appear alongside with the regosols, rendzinas, vertisols and cinnamonic forest soils.

As a bio-climatic (zonal) soil type, chernozems can be formed on various substrates but they must have some basic common properties such as: content of carbonates, mainly loamy composition, water permeability, illite domination in the clay.

Regarding the soil genesis and properties of chernozems, the substrate has the following significance: it gives the soil a favorable mechanical composition (loams), good physical properties – good water permeability, saturation of the exchangeable complex with exchangeable Ca and Mg – as result of its richness with CaCO<sub>3</sub> and MgCO<sub>3</sub> for creation of stable microaggregates, Ca-huminohumates, and with it stable macroaggregates. The substrate contains primary minerals which, with weathering, can provide biogenic elements, and in our classification (Φилиповски, 2006), chernozems are divided into varieties based on the nature of the substrate. The mineralogical composition of the clay and the larger soil particles depend on the substrate.

#### Material and methods

The field trials were performed according to the generally accepted methodology in our country (Митрикески, et.al. 2006). After the field reconnaissance the digging locations were chosen for the basic pedological profiles (total 25) of which the following are presented in the paper 25: 9 profiles in Ovche Pole (4, 9, 10, 11, 12, 13, 16, 17, 18), 3 profiles in the Shtip region (19, 20 and 21), 5 profiles in the Veles region (23, 24, 25, 26 and 27) and 8 profiles in the Tikvesh region (28, 31, 33, 34, 35, 36, 37 and 38). The profiles are entered in a topographic map 1:50.000.

The mechanical composition of the soil is determined according to the international A – method (Resulović, et.al. 1971), and the disperzion was made with 1 M solution of Na<sub>4</sub>P<sub>2</sub>O<sub>7</sub> x 10 H<sub>2</sub>O (Thurn, et.al. 1955). The division of mechanical elements into fractions was made according to the International Classification, and the classification of the soils into texture classes was according to Scheffer and Schachtshabel (Митрикески, et.al. 2006). The carbonate content in the soil was determined volumetrically, with the Scheiblercalcimeter (Resulović, 1969); pH of the soil solution (soil reaction) was determined electrometrically, with glass electrode in water suspension and in suspension of 1N KCl (Bogdanović, et.al. 1966); the soil humus content was determined based on the total carbon, according to the Turin's method, modified by Симаков, by using 0.4 n K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>(Орлов, et.al. 1981;Симаков, 1957); the total nitrogen was determined according to the Kjeldahl method (Bogdanović, et.al. 1966) and the easily accessible potassium (K<sub>2</sub>O) and phosphorus (R<sub>2</sub>O<sub>5</sub>) were determined according to the Al method (Resulović, 1969).

Analysis of variance (ANOVA) was made for all analyzed properties for samples of different size. The effect of the substrate in chernozems was established as well as its interaction with the variability of the examined properties. The significance of the differences among the mean values of analyzed properties per substrate was determined with the Tukey test, for p<0.05 level. All statistical analyses were made with the R software package.

#### **Results and discussion**

The Vardar zone in the wider sense has a special history whose beginnings are in the Paleozoic (hircine orogenesis), Измајлов, (1958).In this geotectonic whole there is majority of clastic sediments, there are many paleogene (eocene) and neogenic (miocene and pliocene) sediments. These sediments are lithologically heterogenic, mainly carbonate. In part of them, illite is mostly present in the clay, while in the other part there is montmorillonite(smectite). Pliocene sediments are accompanied by newer lake pleistocene sediments (Филиповски, 1995).

A separate group of clastic sediments, specific only for this zone are the loess sediments. Измајлов, (1958) on the geological map of the Tikvesh coal basin R 1 : 50.000 has pointed out several regions where presence of these sediments has been determined. According to him, in the Pleistocene, after the glaciation there was arid steppe climate, when in the Tikvesh and other valleys loess sandy – clay sediments were deposited. These sediments are the second treasure for this region (Tikvesh), which according to its significance (the basis for development of viticulture was created) does not lag behind the lignite deposits present in it. These loess – clay sediments belong to medium quartery deposits (Никитин., cf. in Измајлов, 1958). On them the chernozem gets its typical properties, similarly as on loess (Филиповски, 1995). According to Spirovski, (1963), these sediments could not have been created before the withdrawal of the lakes and this substrate has also been recognized as a factor for the appearance of chernozemization by other researchers.

The substrate is important for the soil formation, but in chernozems, as stated in the introduction, its significance comes after climate and vegetation - which are so influential that they level the differences of the geological substrate. Such is the case with the typical chernozem which is formed both on sand and on clay, on granite and on loess, and in all cases it exhibits its characteristic features (CTEGYT, 1927). However, according to the same author, the most typical substrate for formation of chernozem is loess. Some chernozem researchers (Антипов-Каратаев, et.al. 1960, Гюров, et.al. 1969, Filipovski, et.al.1963etc.) have pointed out that chernozems are formed on: loess, loess sediments, redeposited loess, tertiary loams, pleistocene sandy-clay loess sediments, clay sediments, marls, alluvial and alluvial – proluvial deposits, eolian sands. Chernozems in Romania are formed mainly on loess, then on marls and alluvium (Nejgebauer, 1951). The same author has stated that chernozem can be formed on different substrates, but it is mostly (2/3) formed on loess and loess sediments, and (1/3) on various eluvial, deluvial, fluviglacial and alluvial loams, clays and sands, and even some on eluvial crushed stones from igneous rocks (Filipović, 1959). The appearance and maintenance of the calcareous chernozem are affected by the loess composition, relief and climate (loess is typically carbonate and deposited dry) Antonović, et.al. (1976). In Bulgaria, in the area of Shumen and Varna, Пенков, (1996) formation of calcareous chernozems over loess materials of old calcareousmarls has been determined.

Tertiary sediments prevail in the composition of the sediments in the valleys where chernozems are spread (Figure 1).

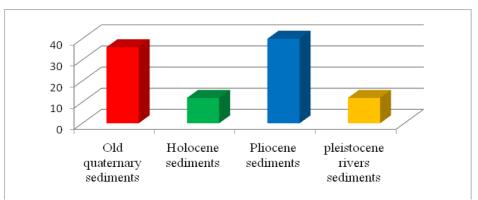


Figure 1. Percentage of parent materials in chernozems formation

According to the researches of Христов, et.al. (1973), Ракиќевиќ, et.al. (1973) and Ракиќевиќ, et.al.(1976) and according to the geological map developed by them, the studied profiles excavated in the area of the village of Erdjelija, v. of Nemanjici and v.of Mustafino, (Ovche Pole) appear over non-segregated old quaternary sediments represented with clay and sand. Most of the material which served for their formation, originates from the clay and sandy components of Neogene sediments.

Pliocene sediments found in the Tikvesh and Veles basin for the most part are represented with sands and clays. Based on the pykermyska fauna, they date back at the Pliocene Epoch. They are represented with sandy series which partially lay over Paleozoic and Mesozoic deposits, and partially over Paleogenic sediments. The sandy series have homogenous composition and they are represented with grayish-yellow sands, sandy clays and clays, rarely gravels and sandstones. It can be said that they belong to the upper Pliocene and partially, to the Mid-Pliocene.

Some of the profiles in Tikvesh (28, 31 and 38) are formed over old Pleistocene river terraces, represented with yellow and yellowish-brown clays and clay sands, poorly bound. They are considered as the oldest quaternary sediments.Филиповски, (1995) points out that the occurrence of developed automorphic soils is possible at the older dry and higher terraces (for example chernozems and other soils in Tikvesh), since the automorphic conditions last longer.

The profiles (19, 20 and 21, village of D. Trogirci – Shtip) are formed over young holocene sediments. These are young creations of the quaternary period – holocene. The deposits have heterogeneous composition and these include proluvial deposits. They are built from poorly treated and untreated pieces of Precambrian, Cambrian and Paleozoic rocks, as well as tertiary rocks bounded with clays and sands. At some places, the proluvial deposits are deep up to several meters, which indicates on intensive filling of the fields.

All cases refer to loose substrates (regolith) with finished detritation, which means that the soil genesis runs faster, and forms deep soils.

Many details on the properties (physical and chemical) of the chernozems are presented by Миткова, (1998). Here, we are going to elaborate the influence of the substrate over the properties of the chernozems. Table 1 gives the statistical mean values of the fractions of the fine earth depending on the substrate over which chernozems are formed.

When compared to the chernozems formed over eolian sediments (loess), our examined soils contain a slightly higher percentage of coarse as they are formed over water

(tertiary and quaternary) sediments. 22 out of 25 examined profiles formed over old quaternary sediments (O.Q.S), holocene sediments – proluvium (H.S-P) and pliocene sediments (P.S) have under 5% (low content of coarse), and the other 3 formed over pleistocene rivers sediments (P.R.S) up to 8%. The fine sand fraction dominates among the fractions of the fine earth, 33.59% in average for chernozems formed over O.Q.S, 31.00% for chernozems formed over H.S-P, 38.09% for chernozems formed overP.S and 32.89% for chernozems formed over P.R.S, while the fractions of silt are almost identical at the chernozems formed over O.Q.S andP.S, in average 15.98% and 16.45% silt and coarse sand at chernozems formed over O.Q.S andP.R.S, in average 23.75% and 21.36%. The values of the clay fraction are almost identical at the chernozems formed over O.Q.S, in average 26.11 %, 25.47% for the chernozems formed over H.S-Pand 25.91% for chernozems formed over P.R.S.

The analysis of the variance has shown that the parent substrate in the chernozems has an important influence over the variability of the content of coarse, clay, physical clay and physical sand. The content of coarse is the highest, in average (7.90%), while the content of clay is the lowest, in average (22.64) at chernozems formed over P.R.S and they significantly vary from the other chernozems in terms of statistics, while the content of the fractions physical clay is the highest, in average (51.90%) and the content of physical sand is the lowest, in average (47.44%) at chernozems formed over H.S-P, and they significantly vary from the other chernozems in terms of statistics.

According to the classification of Scheffer and Schachtshabel (Table 1), nine (9) of the profiles formed over O.Q.S and 10 profiles formed over P.S, represent sandy clay, three formed over H.S-P are loamy clay and three profiles formed over P.R.S aresandy clay loam.

Parent material	Profile N <sup>0</sup>	Loamy clay	Sandyclay	Sandyclayloam
old quaternary sediments (O.Q.S)	9	-	9	-
holocene sediments – proluvium (H.S-P)	3	3	-	-
pliocene sediments (P.S)	10	-	10	-
pleistocene rivers sediments (P.R.S)	3	-	-	3

Table 1. Texture classes in chernozems in relation to parent material

This speaks for the quite favorable mechanical composition, which is important for the physical properties of chernozems. The data on the mechanical composition of the examined chernozems are similar to the data of this soil type presented by other authors: Antić, et.al. (1980), Гюров, et.al. (1969), Filipović, (1959), Rode, et.al.(1972), Крупеников, (1966), Димитријевиќ, (1983) аnd Филиповски, (1996).

Table 3 demonstrates data on the chemical properties of chernozems formed over the above mentioned substrates. Due to the similar bio-climatic conditions of formation, there are no statistically significant differences with regard to these properties, depending on the substrate over which these chernozems are formed. The intensity of the accumulation of humus is poorly expressed in the examined chernozems. The statistical mean value of humus at all examined chernozems is under 1.6%. The reason for this lies in the lower quantity of organic residues (climate aridity) and the more intensive mineralization during winter, as well as the high degree of utilization of chernozems in the agricultural production (all examined chernozems are intensively used in the agricultural production). When crops are grown, the quantity of plant residues that remain in the soil gets lower as they are extracted by harvesting. When chernozems are used for growing agricultural crops (of ten monocrops), appropriate agricultural technique has not been applied (organic fertilizers are rarely used).

The Russian chernozems contain significantly more humus: (4 to 5%) in the European part of Russia, 6 to 8% in Ukraine, and 10% in the central part (Rode, et.al.1972). The percentage of humus increases from West to East. The content of humus in the chernozem of Vojvodina, being cultivated for a long period of time, amounts between 4 and 6 %, while in circumstances of natural vegetation 8.5 to 10%. According to the same author, the content of humus in the noncalcareou schernozem in Romania amounts from 3 to 5 %, as for the calcareous chernozem it amounts from 4.6 to 6.3 %. The Bulgarian chernozems usually contain 2.5 to 4.5% humus (Антипов-Каратаев, et.al. 1960). In some regions of Bulgaria, according to Донов, (1979) and Антипов-Каратаев, et.al. (1960), this percentage can be higher (4 to 8%) and lower (2%).

The total content of N is parallel to the content of humus, the statistical mean value for which is under 0.092%.

Depending on the content of  $CaCO_3$  in the substrate and its leaching intensity in the calcareous chernozems, the carbonates appear form the surface itself, while the noncalcareousappear in the transitional AC horizon. In all examined profiles, the reaction of the soil solution is not under 7.00. In all chernozems, the mean statistical value of the reaction amounts between 7.70-7.80 (slightly to medium alkaline reaction).

The studied chernozems are characterized with medium to high exchange capacity (their mean statistical value varies from 18 to 20 cmol (+) kg<sup>-1</sup>soil. Clay minerals have more important role in the exchange capacity with regard to the content of humus.

Calcareous chernozems are fully saturated with bases, while a small acidification appears at the noncalcareou schernozems in the humus-accumulative horizon A, and for some profiles in a part of the transitional AC horizon.

Depending on the parent substrate, there is no a significant difference at chernozems regarding the amount and the content of the exchangeable base cations. The base saturation percentage in the calcareous chernozems is 100%. The noncalcareouschernozems are also characterized by a high base saturation percentage (over 90%) (both of the humus horizon and a part of the AC horizon), although they do not contain carbonates.

Chernozems are characterized with relatively good productive properties (relatively deep profile, good physical and chemical properties). The examined soils are medium to poorly supplied with available  $P_2O_5$ , and rich supplied with available  $K_2O$ . Such provision of chernozems with available  $K_2O$  results from the uncontrolled fertilization with potassium mineral fertilizers, and the potassium bound between the crystal units of the illite.

# Conclusion

As a bio-climatic (zonal) soil type, chernozems can be formed on various substrates but they must have some basic common properties such as: content of carbonates, mainly loamy composition, water permeability, illite domination in the clay. Chernozems form on old quaternary sediments (Ovche Pole), holocene sediments - proluvium (Stip), pliocene sediments (Veles, and in parts of Tikvesh) and the pleistocene rivers sediments (in parts of Tikvesh).

All cases refer to loose substrates (regolith) with finished detritation, which means that the soil genesis runs faster, and forms deep soils.

The analysis of variance has shown that in chernozems, the parent material (substrate) has significant effect over the variability of the content of coarse, clay, physical clay (silt+clay)and physical (coarse+fine) sand. The coarse content is highest, mean (7.90%) and lowest on clay, mean (22.64%) in chernozems formed over pleistocene rivers sediments (P.R.S) and their difference from the others is statistically significant, and the content of fractions physical clay is highest, mean (51.90%) and lowest on physical sand, mean (47.44%) in chernozems formed over holocene sediments –proluvium(H.S-P) and their difference from the others is statistically significant.

According to the classification of Scheffer and Schachtshabel, nine (9) of the profiles formed over O.Q.S and 10 profiles formed over P.S, represent sandy clay, three formed over H.S-P are loamy clay and three profiles formed over P.R.S aresandy clay loam. This is the quite favorable mechanical composition, which is important for the physical properties of chernozems.

Due to the similar bio-climatic conditions of formation, there are no statistically significant differences with regard to chemical properties (content of humus, pH reaction, content of carbonates, exchange capacity, total exchangeable base cations and base saturation percentage) depending on the substrate over which chernozems are formed.

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# YIELD AND YIELD COMPONENTS OF CANOLA (*BRASSICA NAPUS* L.) UNDER IRRIGATION

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#### Abstract

Climate of Bulgaria is favourable for canola (Brassica napus, L.) growing but in some years is risky because of autumn and spring drought periods. Irrigation appears to be a necessary practice for obtaining high yields and high income. A two-factorial field experiment was conducted during 2011-2012 and 2012-2013 with factors: irrigation (three levels - rain-fed, full irrigation and 50% deficit irrigation) and variety (three moderateearly winter hybrids). The goal was establish the impact of irrigation on yield and yield components of canola and the importance of each yield component for seed yield accumulation. The experiment was put in a randomized complete block design as split-plot factorial arrangement. Irrigation was applied by sprinkling. Data of yield and yield components such like seed yield, seed oil content, oil yield, silique number per plant, silique length, seed mass per plant, seeds number per silique, and 1000 seed weight was obtained. Variance and regression analyses were applied to the field results. It was established that under the climatic conditions of Bulgaria the impact of irrigation on yield and yield components of canola was statistically significant. The hybrids performed up to 88% of the maximum yield under full irrigation and up to 58% of the maximum yield under 50% irrigation deficit. One of the hybrids was statistically different from the other ones by performing higher results. The increase in seed yield was due to the increase in number of branches, number of seeds in silique and 1000 seed weight. One-factor and multiple regression equations were derived.

Keywords: canola, irrigation, yield components, variance analysis, regression analyses, Bulgaria

#### Introduction

Climate of Bulgaria is moderate continental and is favourable for canola (*Brassica napus*, L.) growing. However, some of its features like great variation by time and territory of the

atmospheric moisture (55-65% for March-July rainfalls) make it risky for yield accumulation (Mitova et al., 2010; Moteva et al., 2012). Further, tendencies of warming and drought are spreading over the country (Kazandjiev et al., 2010; Moteva et al., 2011). It has been already proved that irrigation is an appropriate practice, particularly during spring vegetation, for obtaining high yield and high return form canola cultivation (Moteva and Gigova, 2013; Gigova, 2014). Experimental results from similar climates show that irrigation significantly contributes for seed and oil yield increase when the applications are properly distributed according to the water needs of the plants (Bilibio et al., 2009ab; North, 2010; Danesh-Shahraki et al., 2008; Istanbulluoglu et al., 2010). Gan et al., (2004) have established that plants, subjected to water stress during the early stages, would recover easily, while water stress during silique formation would cause considerable reduction of the yield components such like number of siliques per plant, number of seeds per silique, individual seed weight.

The goal of the paper is to establish the impact of irrigation on yield and yield components of canola and the importance of each yield component for seed yield accumulation under irrigation conditions.

## **Material and Methods**

A two-year field experiment with irrigation of three moderate-early hybrids of winter canola (*Brassica napus*, L.) was carried out in the region of Sofia, Bulgaria, during the period 2011-2013. The site is situated 42.6° N and 550 m asl. The climate is moderate continental and the region is one of the coldest and most humid in Bulgaria (Agro-ClimaticAtlas, 1982). Yearly precipitation in 1971-2000 was 636 mm that dropped to 586 mm in 1981-2010 (Moteva et al., 2014, unpublished data). The average annual temperature in 1971-2000 was 10.1°C that rose to 10.6°C in 1981-2010. The temperature total for the potential vegetation period March-October was 3546.3°C (1971-2000) and rose to 3683.5°C (1981-2001). These facts are evidence for climate warming and drought in the region. Walton et al., (1999) have established that the threshold temperature resistance of fully hardened canola is  $-15^{\circ}$ C, and for a short period (4-10 days) it withstands  $-20^{\circ}$ C. Winters in the study area are usually cold and snowy. Sometimes, air temperature in the coldest days of January drops lower than  $-15^{\circ}$ C. Frequently, there are heat waves in summer with maximumair temperatures  $>30^{\circ}$ C. Since they happen in July and August, they are not harmful to canola.

The experiment was put in a randomized complete block design as split-plot factorial arrangement in three replications. The main-plot factor was Irrigation. It consisted of three levels:  $A_1$  – rain-fed,  $A_2$  – 50% deficit irrigation,  $A_3$  – full irrigation according torefill point (*RP*) of 80% of field capacity (*FC*). The sub-plot factor was Variety:  $B_1$  – PR45D05 (Maximus),  $B_2$  - PX100CL (Clearfield) and  $B_3$  – Triangle (KWS). The area of each sub-plot was 10 m<sup>2</sup>, plant density –45-50 pl/m<sup>2</sup>, and depth of root expansion *H*=1.0 m. The soil was chromic luvisols (FAO classification) with total water content *TWC*=327 mm, total available water content *TAWC*=165 mm, and bulk density  $\alpha$ =1.5 g/cm<sup>3</sup>. Land preparation, fertilizers and weed control were applied according to the standard agricultural practices in the region. Sowing in 2011was carried out on 30 September, after a long dry period and an application for emergence, while sowing in 2012 was carried out on 25 September. The October rainfall totals in 2011 and 2012 were respectively 9.3% and 10.9% higher than month normality, which for 1973-2012 was 44.5 mm. November was

very dry in both years but soil moisture from the previous month was pretty enough to supply plants with the necessary water for the initial stage of their development.

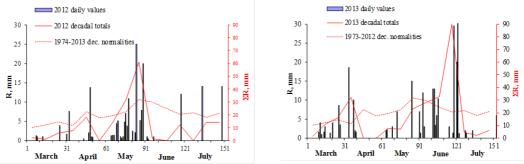


Figure 1.Distribution of daily rainfalls, decadal totals and decadal normalities in the spring period March-July of 2012 and 2013

March-July periods in 2012 and 2013 were dry and medium-dry respectively. Monthly residuals from the normalities were negative. Only in May 2012 and June 2013 they were positive with an increase of 46.2% and 98.7%. Both years had long dry periods in April-May and June (Fig.1).

The determinants for applying irrigation were daily rainfalls distribution, high air temperature and high vapor pressure deficit in May and June. Two irrigation applications were given in each year. Some irrigation water of the first application in 2012 and the second one in 2013 turned to be inefficient due to the immediate rainfalls after the applications. The application depth at *RP* was calculated as:  $m = 10H\alpha(\beta_{FC} - \beta_{RP})$ , where  $\beta$  was moisture percentage by weight (Kostyakov, 1951).

Data of yield and yield components such like seed yield (SY), seed oil content (SOC), oil yield (OY), silique number per plant (SN), silique length (SL), seed mass per plant (SM), seeds number per silique (SdN), and 1000 seed weight (ThSW) was obtained. SY was estimated at 8% standard humidity of the seeds. SOC was estimated in a Soksle apparatus. SN was counted and SL was measured at harvesting – both taken as average from 10 typical plants in 3 replications. The biometrical data such like plant height (PlH) and number of branches per plant (BN) was analogously taken at harvesting. Variance analysis was applied to establish the significance of factors' impacts on the experimental results (Shanin, 1977). Single-factor regression analysis was applied to establish the importance of yield components for seed yield accumulation. The regression analyses we proceeded in Excel environment.

#### **Results and Discussion**

The comparison between the studied hybrids showed that hybrid Triangle, which is distributed by KWS LOCHOW GMBH, was statistically different under irrigation from the other two hybrids – PR45D05 (Maximus) and PX100CL (Clearfield), which are distributed by PIONEER BULGARIA (Table 1 and Fig. 2). The differences between Triangle and the other hybrids under rain fed conditions were statistically significant only for SY, PIH, SN and ThSW in 2013 (Table 1). SY was 2.517 Mg/ha - with 0.3-0.5 Mg/ha greater than SY

of the other hybrids; Triangle plants were 142.9 cm tall - with 20-30 cm taller than those of Maximus and Clearfield; SNwas192.2 - with 50-100 more than Maximus and Clearfield; and ThSW was 3.8 g - with 0.6-1.2 g more than those of the other hybrids. There was no significant difference between the other two hybrids for any of the indices.

The greater impact of irrigation on Triangle was evident still for the deficit irrigation (Table 1). Triangle increased its SY significantly as in 2012 so in 2013 and reached 3.838 Mg/ha and 3.967 Mg/ha respectively. Analogously, OY was 1.563 Mg/ha and 1.758 Mg/ha and was significantly higher than OY of the other two hybrids. Still 50% of the needed additional water gave significant rise of Triangle PIH compared to the height of Maximus and Clearfield in both years. It was 121.7 cm against 101.7-108.5 cm in 2012 and 148.8 cm against 121.1-126.1 cm in 2013. SN also increased considerably– 245.5 against 115.6-139.5 in 2012 and 298.5 against 162.3-168.5 in 2013. SL performed divergent tendencies – keeping the same for all hybrids. SM wasn't different in 2013 but Triangle showed significantly greater SM – 7.5 g against 4.6-4.8 g in 2012. The deficit irrigation had significant impact on Triangle ThSW=4.3 g – with 1.3 g more than ThSW of Maximus. Hybrid Clearfield was also different from Maximus at 5% level.

Full irrigation had impact on all indices but the tendency of "no significant difference" between Maximus and Clearfield on one side and Triangle, on the other, kept the same (Table 1). SY of Triangle was 4.797 Mg/ha and 4.737 Mg/ha in 2012 and 2013 respectively and was statistically different from the other hybrids at 0.1% level. The difference of OY was significant at the same confidence level. OY of Triangle was 2.075 and 2.128 Mg/ha - against 1.251-1.370 Mg/ha of Maximus and Clearfield in 2012 and 1.758 and 1.887 Mg/ha - in 2013. Triangle PIH, SN and ThSW performed different from those of the other hybrids in all moisture conditions. SL and SdN tended to be less than those of the other hybrids.

Table 2 presents the impact of the main factor Irrigation on yield and structural elements of the yield. As it is seen, irrigation had significant impact on SY and OY and nearly on all yield components. Full irrigation caused considerable SY increase in all hybrids and in both years. Maximus increased its SY with 1.100-1.740 Mg/ha compared to rainfed conditions by reaching 3.346 Mg/ha and 3.793 Mg/ha in 2012 and 2013 respectively. Clearfield increased its SY with 0.95-1.88 Mg/ha by reaching 3.070 Mg/ha and 2.235 Mg/ha, and Triangle – with 1.960-2.220 Mg/ha by reaching 4.797and 4.737 Mg/ha. Analogously, OY significantly changed under the impact of irrigation. OY varied under full irrigation from 1.370 (2012) to 1.758 (2013) considering Maximus, 1.251 (2012) to 1.887 (2013) considering Clearfield and 2.075 (2012) to 2.128 (2013) considering Triangle. When 50% of the maximum irrigation depth was applied, OY also performed significant differences from the rainfed plots. It was proved that SOC significantly increased with increasing of the water amount too.

Table 1. Yields and levels of significance of sub-factor Variety							
Indices	Irrig.	A <sub>1</sub> - I	Rainfed	$A_2 - 50\%$	irr. deficit	$A_3 - Full$	irrigation
Indices	Hybr.	2012	2013	2012	2013	2012	2013
	<b>B</b> <sub>1</sub>	2.188	2.050	2.401	3.208	3.346	3.793
Seed yield (SY), Mg/ha	$\mathbf{B}_2$	2.120	2.235	2.343	3.165	3.070	4.123
	$B_3$	2.833	$2.517^{+}$	3.838++++	3.967++++	$4.797^{+++}$	4.737++++
	<b>B</b> <sub>1</sub>	39.7	43.1	40.2	42.3	40.8	43.4
Seed oil content (SOC), %	$B_2$	35.9	43.5	39.5	45.5	40.7	44.3
	$B_3$	37.9	46.3	40.6	45.8	43.4	44.9
01.11.01	<b>B</b> <sub>1</sub>	0.871	0.882	0.964	1.394	1.370	1.758
Oil Yield (OY), Mg/ha	$B_2$	0.964	0.946	0.923	1.441	1.251	$1.887^{+}$
wig/ na	$B_3$	1.370	1.092	1.563++++	$1.758^{+++}$	$2.075^{+++}$	2.128++++
	B <sub>1</sub>	97.0	118.6	108.5	121.2	121.3	127.7
Plant height (PlH), cm	$B_2$	101.6	125.1	101.7	126.1	118.0	134.4
	<b>B</b> <sub>3</sub>	105.2	142.9+++	$121.7^{+}$	148.8+++	124.3	156.4+++
	B <sub>1</sub>	5.0	7.4	5.3	8.4	6.1	9.8
Number of branches (BN)	$B_2$	4.7	7.7	5.1	8.4	4.9 <sup>00</sup>	9.5
	$B_3$	5.4	8.4	5.7	8.6	6.7	10.4
	B <sub>1</sub>	126.2	89.4	139.5	168.5	267.6	175.8
Silique number per plant (SN)	$B_2$	70.3	146.8	115.6	162.3	181.6	$254.5^{+}$
	<b>B</b> <sub>3</sub>	143.1	192.2++	245.5+++	298.5+++	489.0++	361.4+++
	<b>B</b> <sub>1</sub>	6.8	6.6	7.1	6.8	7.8	7.5
Silique length (SL), cm	$B_2$	6.5	6.6	6.9	6.8	6.8000	7.6
	<b>B</b> <sub>3</sub>	6.4	6.8	6.5 <sup>00</sup>	6.9	6.7000	7.2
	<b>B</b> <sub>1</sub>	23.6	24.2	24.9	24.8	30.0	25.2
Seeds number per silique (SdN)	$B_2$	24.4	21.0	24.4	22.0	25.9 <sup>00</sup>	23.9
	<b>B</b> <sub>3</sub>	24.1	21.0	24.1	26.3	24.8 <sup>000</sup>	26.4
	<b>B</b> <sub>1</sub>	4.7	5.8	4.8	6.3	7.3	7.8
Seed mass per plant (SM), g	$B_2$	4.2	5.4	4.6	5.8	6.2	6.8
	<b>B</b> <sub>3</sub>	5.6	6.3	7.5+++	7.8	11.3++++	9.7
	<b>B</b> <sub>1</sub>	2.6	2.6	2.8	3.0	3.0	3.2
1000 seed weight (ThSW), g	$B_2$	2.4	3.2	2.7	3.7++	3.4	3.8
	<b>B</b> <sub>3</sub>	2.5	3.8++	3.2	4.3++++	3.6+	4.4+

Table 1. Yields and levels of significance of sub-factor Variety

<sup>+</sup>significant at 5% level; <sup>++</sup> significant at 1% level; <sup>+++</sup>significant at 0.1% level

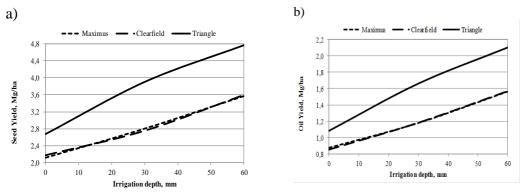


Figure 2. Seed yield increase (a) and oil yield increase (b) under the impact of irrigation

Table 2. Yields and le	evels of si						
Indices	Hybr.	B <sub>1</sub> - M	aximus	$B_2$ - Cl	earfield	B <sub>3</sub> - Ti	riangle
mulces	Var.	2012	2013	2012	2013	2012	2013
Seed yield (SY),	A <sub>1</sub>	2.188	2.050	2.120	2.235	2.833	2.517
Mg/ha	$A_2$	2.401	$3.208^{+++}$	2.343	3.165	$3.838^{+}$	$3.967^{+}$
wig/na	A <sub>3</sub>	3.346++	3.793++++	$3.070^{+}$	4.123++++	$4.797^{+++}$	4.737++
Cood all contant	A <sub>1</sub>	39.7	43.1	35.9	42.3	37.9	43.4
Seed oil content	$A_2$	40.2	43.5	39.5	45.5++	40.6	44.3
(SOC), %	A <sub>3</sub>	40.8	46.3++	$40.7^{+}$	$45.8^{++}$	$43.4^{+}$	44.9
0:1 V:-14	A <sub>1</sub>	0.871	0.882	0.763	0.946	1.075	1.092
Oil Yield	$A_2$	0.964	1.394++	0.923	$1.441^{++}$	$1.563^{+}$	$1.758^{+++}$
(OY),Mg/ha	A <sub>3</sub>	$1.370^{++}$	$1.758^{+++}$	$1.251^{+}$	$1.887^{+++}$	$2.075^{+++}$	$2.128^{+++}$
Diant haight (DILI)	A <sub>1</sub>	97.0	118.6	101.6	125.1	105.2	142.9
Plant height (PlH), cm	A <sub>2</sub>	108.5	121.2	101.7	126.1	121.7	148.8
	A <sub>3</sub>	121.3++	127.7	$118.0^{+}$	134.4	124.3	156.4++
Number of branches (BN)	A <sub>1</sub>	5.0	7.4	4.7	7.7	5.4	8.4
	A <sub>2</sub>	5.3	$8.4^{+}$	5.1	8.4	5.7	8.6
	A <sub>3</sub>	6.1++	$9.8^{+}$	4.9	$9.5^{+}$	6.7++	$10.4^{+}$
C:1:	A <sub>1</sub>	126.2	89.4	70.3	146.8	143.1	192.2
Silique number per plant (SN)	A <sub>2</sub>	139.5	$168.5^{+}$	115.6	162.3	245.5	298.5++
plant (SN)	A <sub>3</sub>	$267.6^{+}$	$175.8^{+}$	181.6	254.5++	$489.0^{++}$	361.4+++
Silique length (SL),	A <sub>1</sub>	6.8	6.6	6.5	6.6	6.4	6.8
1 0 0	$A_2$	7.1	6.8	6.9	6.8	6.5	6.9
cm	A <sub>3</sub>	7.8	7.5	6.8	$7.6^{+}$	6.7++	7.2+++
Seeds number per	A <sub>1</sub>	23.6	24.2	24.4	21.0	24.1	21.0
1	$A_2$	24.9	24.8	24.4	22.0	24.1	26.3++
silique (SdN)	A <sub>3</sub>	30.0++++	25.2	25.9	$23.9^{+}$	24.8	26.4+++
Seed mass per plant	A <sub>1</sub>	4.7	5.8	4.2	5.4	5.6	6.3
(SM), g	A <sub>2</sub>	4.8	6.3+	4.6	5.8	$7.5^{+}$	$7.8^{+}$
(JIVI), g	A <sub>3</sub>	7.3++	7.8++	$6.2^{+}$	6.8+	11.3++++	9.7+++
1000 seed weight	A <sub>1</sub>	2.6	2.6	2.4	3.2	2.5	3.8
(ThSW), g	$A_2$	2.8	3.0	2.7	3.7+	$3.2^{+}$	4.3
(1113 W), g	A <sub>3</sub>	3.0	3.2	3.4+	3.8+	3.6++	4.4

Table 2. Yields and leve	ls of significance of	f the irrigation impact

\*significant at 5% level; ++ significant at 1% level; +++ level significant at 0.1% level

Among the biometrical characteristics, PIH significantly changed under the impact of irrigation only in some plots of full irrigation, while BN - in all plots but one (Table 2). Generally, all hybrids increased their BN, compared to rainfed conditions, with around 2 per plant: by Maximus - from 5.0 (2012) and 7.4 (2013) when rainfed to 6.1 (2012) and 9.8 (2013) when fully irrigated, analogously by Clearfield - from 4.7 (2012) and 7.7 (2013) to 4.9 (2012) and 9.5 (2013) and by Triangle - from 5.4 (2012) and 8.4 (2013) to 6.7 (2012) and 10.4 (2013).

An increasing tendency in all yield components was found, except for SL (Table 2). Irrigation had significant impact on SL only for hybrid Triangle under full irrigation. It increased from 6.4 cm (2012) and 6.8 cm (2013) under rainfed conditions to 6.7 cm (2012) and 7.2 cm (2013) under full irrigation. Significant increase under full irrigation was also observed in SdN but in 2013. Triangle increased SdN too – from 21.0 (rainfed) to 26.3 (50% irr.def.) and 26.4 (full irr.). SM in all hybrids changed only under the impact of full irrigation. Maximus increased SM from 4.7 g (2012) and 5.8 g (2013) to 7.3 g (2012) and 7.8 g (2013), Clearfield - from 4.2 g (2012) and 5.4 g (2013) to 6.2 g (2012) and 6.8 g (2013), and Triangle - from 5.6 g (2012) and 6.3 g (2013) to 11.3 g (2012) and 9.7 g (2013). Irrigation had significant impact on ThSW when plants were well watered. In 2012, Clearfield increased its ThSW from 2.4 g (rainfed) to 3.4 g (full irrigation) and in 2013 – from 2.5 g (rainfed) to 3.8 g (full irr.).

Our results confirm the results obtained in different parts of the world, revised by Istanbulluoglu et al., (2010), which show that seed yield vary under rainfed and irrigation conditions in the range of 1.0–5.3 Mg/ha. As to Alberta Agriculture (1980),canola yields under rainfed conditions are good in the range 1.0-2.6 Mg/ha, while under irrigation – in the range 3.2-4.0 Mg/ha. The most advanced farmers of Australia obtained 1.8-3.6 Mg/ha seeds by irrigation while the seed yields from the experimental fields were 3.8-5.2 Mg/ha (Wright et al., 1995; Taylor et al., 1991; Taylor and Smith, 1992). Irrigation impact on canola yield in Northwest of USA contributed for 30-40% yield increase and for obtaining 3.500-4.500 lb/acre (=4.0-5.0 Mg/ha) (Kruger, 2005). Wright et al. (1988) obtained 0.8 Mg/ha canola oil without irrigation while under irrigation - 1.2 Mg/ha. Ivanova (2010) proved that seed yield from a hectare can be 2.5-3.0 Mg, while oil yield - 0.8-1.1 Mg.

SOC from our 2012-2013 experiment varied from 35.9% to 44.9%. After a thorough study of worldwide experiments, I stanbulluoglu et al., (2010) cites authors like Al Jaloud et al., (1996); Farre´ et al., (2007), Hamzei, Nasab, Khoie, Javanshir&Moghaddam (2007), Robertson & Holland( 2004) and Taylor et al., (1991) which have obtained SOC in the range of 31.0%-46.4%. In the experiments of Istanbulluoglu et al., (2010), SOC varied from 39.6% to 40.8%, OY was 1.3 Mg/ha without irrigation and 1.9 Mg/ha under irrigation. When applying irrigation, BN increased from 6.55 to 9.16, SdN - from 22.9 to 24.3, ThSW - from 3.27 g to -3.90 g, SOC - from 43.8% to 45.6%, SN - from 3425.6 m<sup>-2</sup> to 4746.8 m<sup>-2</sup> <sup>2</sup>. All changes in canola yield components of our study under the impact of irrigation demonstrated increasing tendencies. When putting their relative increase against the relative irrigation depth in a regression, the following dependencies and coefficients of determination were obtained (Table 3). It is seen that SY, BN and ThSW have very close dependence on water with high coefficients of determination- 0.79, 0.74 and 0.69 respectively. Other structural elements like PlH, SL and SM have good dependence on irrigation with coefficients of determination 0.56, 0.55 and 0.52, and SdN and SN - 0.42 and 0.41. The dependence of OY and SOC also has good relationship with irrigation water.

In similar studies, Champolivier and Merrien (1996) – cites by Moaveni et al., (2010) has established that the amount of water has greatest impact on seed number, also 1000 seed weight is sensible to the least water stress during the stage of coloring the seeds and SOC reduces with the increase of water deficit.

Considering the results in Table 3, a multiple regression analysis was applied to yield and yield components in order to establish the most important component for yield accumulation. The increases in yield components  $(\Box)$  were adopted as factors and the increase in seed yield ( $\Box$ SY) was adopted as variable, with known values from the experiment. Since canola is a non-determinant crop, we considered plant height after termination of irrigation at mid-June (to let the siliques ripen) with least impact on yield formation, while the processes of quantitative changes in seeds continued. Hence, we abandoned plant height as a factor in the multiple regression analysis. We also considered the number of branches a definite factor for yield accumulation, since they contributed for greater number of siliques per plant. The factors and the variable were standardized as relative values and are presented in the upper part of Table 4. The results of the multiple regressions are presented in the lower part of the Table and in Equation (1). The predicted by the regression equation y-values and those of the experiment have a narrow coincidence, with coefficient of determination  $R^2=0.90$ . Fisher's test showed that the experimental conditions had strong impact on the results from the experiment. Equation (1) shows that most important for SY accumulation under irrigation is the number of branches (BN) followed by seed number in silique (SdN) and 1000 seed weight (ThSW). This result is relevant to Naderikharadji et al., (2008) who have established by multiple regressions that the yield is dependent most on seed number in silique and on 100 seed weight. They also have established that silique length had the predominant effect on sees yield. As to Masoud Sinaki et al., (2007), silique length has greatest impact on yield compared to the other yield components; while Danesh-Shahraki (2008) has established that most important for the yield was the number of siliques per  $m^2$ . As to the same author, when applying water stress, the reduction of seed yield is most dependent on the reduction of the number of branches, seed mass and number of siliques.

Index	Equation	$R^2$
Seed Yield (SY)	y* = 0.7151x** - 0.6482	0.79
Seed Oil Content (SOC)	y = 0.0718x + 0.2646	0.49
Oil Yield (OY)	y = 1.4766x - 9.2672	0.43
Number of Branches (BN)	y = 0.2123x - 1.1209	0.74
1000 Seed Weight (ThSW)	y = 0.301x - 0.1924	0.69
Plant height (PlH)	y = 0,1415x - 0,5272	0.56
Silique Length (SL)	y = 0.1067x - 0.5265	0.55
Seed Mass per plant (SM)	y = 0,4564x - 2,5289	0.52
Seed Number per Silique (SdN)	y = 0,1515x - 0,5364	0.42
Silique Number per Plant (SN)	y = 0,1515x - 0,5364	0.41

Table 3. Dependence of the yield and structural elements of the yield on irrigation

\* y- relative yield increase, %; \*\*x – relative irrigation depth, %

Hybrid	Relative irrigatio n depth, %	1000 Seed Weight (ThSW)	Number of Branches (BN)	Seed Number per Silique (SdN)	Silique Number per Plant (SN)	Silique Length (SL)	Seed Mass per Plant (SM)	Seed Yield (SY)
		x <sub>1</sub>	x <sub>2</sub>	<b>X</b> <sub>3</sub>	X4	<b>X</b> 5	X <sub>6</sub>	у
B1	50	6.5	6.7	5.5	10.5	4.3	2.8	9.7
B2	50	12.5	9.5	0	64.4	4.9	10.9	10.6
B3	50	25.7	4.7	0	71.6	4	33.6	35.5
B1	50	16.5	13.5	2.3	88.4	3.5	8.6	56.5
B2	50	26.4	9.4	4.4	10.5	3.1	17.4	41.6
B3	50	13.3	2.4	25.5	55.3	1.6	23.8	57.6
B1	100	14.1	21.3	27.3	112.1	13.5	56.4	52.9
B2	100	40.7	5.7	6.3	158.2	4.8	47.8	44.8
B3	100	42.9	22.4	2.9	241.8	4.8	101.2	69.3
B1	100	24.1	31.5	3.9	96.6	13	23.8	85
B2	100	20.8	23.9	13.5	73.3	11.7	17.2	84.5
B3	100	16	21.1	26	88.1	6.4	24.4	88.2
Multiple Regression Statistics								
Coe	efficients	-0.63	-2.62	0.07	1.75	2.75	1.71	-10.85
	of the coer	ff. 0.35	1.71	0.15	0.46	0.78	0.63	13.18
$R^2$	St. err o	of y 0.90	12.68					
F	df	7.18	5.00					
SSreg	ss <sub>resic</sub>	<sub>1</sub> 6926.69	803.81					
	t	-1.81	-1.53	0.43	3.82	3.52	2.72	-0.82
	t <sub>crit</sub>	2.57						
□SY=2.75		75 SdN + 1.71	ThSW	•		•		(1)

Table 4. Data for multiple regression analysis in % and multiple regression statistics

## Conclusions

- 1. Hybrid Triangle (KWS) is statistically different from PR45D05 (Maximus) and Clearfield PX100CL (Clearfield).Under the impact of irrigation it producesgreater biomass and higher seed and oil yield.
- 2. Irrigation is an important factor for obtaining high yields from canola in moderate continental climate. It has significant effect on the yield and yield components.
- 3. Irrigation water has strongest impact on seed yield, number of branches and 1000 seed weight which was established by a single factor regression analysis.
- 4. Most important for seed yield accumulation of canola under irrigation are yield components like the number of branches, seed number in silique and 1000 seed weight, which was established by a multiple regression analysis.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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UDC: 631.67 Original scientific paper

# HAZARD PERFORMANCE INDICATORS FOR LARGE SCALE WATER SYSTEMS WITH AGRICULTURAL IRRIGATION FACILITIES

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### Abstract

Appropriate formulation of a control problem for the irrigating system is faced with the important issue: how to measure performance reliability and risk on local, subsystems' and system level. Different priorities of demand points within the system lead to different formulation of so-called acceptable system statuses, and furthermore to computation of reliability/risk performance indices. Selected examples and related discussion in this paper underline the idea of how weighting coefficient method may efficiently be used to accomplish this goal.

Keywords: system performance; hazard indicators; irrigation.

## Introduction

Surface reservoirs serve as major controllers of water storage and distribution within multipurpose water resources systems. If hazard inputs to the reservoirs occur (e.g. flooding inflows; very low inflows due to meteorological droughts), the system as a whole may experience significant water surpluses and deficits at various points within a system. Systems analysis of long term system operation is therefore of extreme importance, and pillar concern is to identify operational scenarios for avoiding hazardous situations and preserve proper dynamic performance of water system.

Being an important part of a system, agricultural water users, mainly irrigators, usually have strong interest to contract reliable water supplies during vegetation season. However, water distributor can hardly know what is a reliable water ought to be delivered on time at contracted spot(s). Probability of shortages in long term water supply is hard to determine even if very sophisticated stochastic modelling and prediction models are used for generating inputs to large scale river basin simulation models. These models are aimed at evaluating different planning, designing or management strategies and a good option is to generate sufficient number of time series of related multivariate stochastic processes related to hydrology, rainfalls, evaporation, soil moisture etc.

Simulated system performance in all instances may be of limited reliability (Biswas 2001, Hashimoto 1980, Loucks and van Beek 2005, Srdjevic and Obradovic 1995). Once output from simulation basin model is received, it is reasonable to compute so-called hazard indicators of system's dynamic performance and to evaluate the operational strategy for reservoirs implemented in the basin model. Reliability (risk),

resiliency and vulnerability are commonly used indicators as a part of rational concept applicable in evaluating performance of water resources systems on long-term basis (Hashimoto 1980, Srdjevic et al. 2007, Blagojevic et al. 2012).

Irrigating systems are differently scaled and are of different complexity. They are commonly operated on short-term basis (e.g. on-demand), related to time varying water needs of irrigated plants, soil moisture, climate regime and other relevant natural or maninfluenced conditions. The paper discusses how to define long term irrigation related hazard performance indicators, and to enable exclusive and trustful decision making about basin model outputs related to system's performance in part of agricultural irrigation.

### Materials and methods

Both hazard and regular regimes of water system operation directly depend on the irrigation method applied and system operating policy adopted for vegetation season. In most European countries, including Serbia and Macedonia, this season begins in April and lasts till September (Srdjevic 2003, Tanaskovikj et al. 2014) During the season, numerous parameters of system's behavior may be monitored and analyzed on daily, weekly or monthly basis. Balancing of waters within the system and evaluating the plant cover growing stages lead to deriving information base about system performance. At later stage, it is possible to compute values of so called performance indices (PIs) which in different ways describe dynamical behavior of the system. Based on PIs, it is an easy task to compare quality and impact of preference structures and operational policies at reservoirs, lockers and other construction elements used in simulation model of the system.

Major *hazard* PIs which are commonly used in computerized river basin simulation models are reliability, resiliency and vulnerability (Hashimoto 1980, Srdjevic and Obradovic 1995, Srdjevic 2003). Their definitions differ and often are adjusted to specific needs and purposes of the water resources system. For large scale systems with reservoirs and agricultural irrigation facilities, planners and analysts usually assess two global categories of PIs, traditional and modern. Typical traditional performance indices (TPIs) are statistical descriptors such as mean, variance, shortage index, etc. Based on the 'statistical memory' of system's performance, these descriptors bear a dozen of smoothing effect if system performance under historically recorded stresses is considered. The modern performance indices (MPIs), typically represented by reliability (or risk), resiliency and vulnerability, are conceptually different from TPIs. They put a direct focus on the 'dynamics' of the system, more precisely on behaviour of the system in various hazardous situations over long time periods.

A key issue in evaluating performance of the system is to establish criteria for distinguishing 'good' and 'bad' behaviour of the system, namely to refer to some acceptable state of the system when system and/or local demands are met with specified tolerance of shortages (Srdjevic and Obradovic 1995, Srdjevic 2007). An example situation is depicted on Fig. 1.

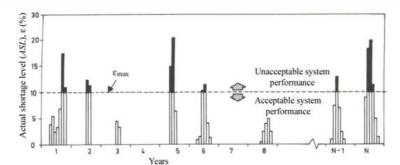


Fig. 1. System status over time based on tolerant shortage level of 10%(TSL=0.1)

<u>Reliability</u> ( $\alpha$ ) is system's ability to assure systems demands over complete time period with prescribed acceptance (tolerant shortage) level. If the planning period is N years with calendar months as discrete time frames, system operation can be simulated over 12N months and if  $n_A$  is the number of months when system was in acceptable status, then its reliability can be computed as:

$$\alpha = n_A / 12N. \tag{1}$$

Risk is opposite to reliability and is equal

 $r = 1 - \alpha = 1 - n_A / 12N = n_F / 12N$ 

where  $n_F$  is the number of months when system was in unacceptable status ('shaded' months in Fig. 1).

(2)

<u>Resiliency</u>  $(\gamma)$  defines how quickly a system is likely to recover from failure, once failure has occurred. If zero-one variable Z is introduced to relate to specific monthly states, and also zero-one variable W describes system's transition from acceptable to unacceptable state, then resiliency can be computed as:

$$\gamma = \frac{\frac{1}{12N} \sum_{i=1}^{12N} W_i}{\frac{1}{12N} \sum_{i=1}^{12N} (1 - Z_i)} = \frac{\sum_{i=1}^{12N} W_i}{12N - \sum_{i=1}^{12N} Z_i}.$$
(3)

<u>Vulnerability</u> (v) is a measure of how deep is system in unacceptable status once it went into that status. If W is an event defined as succession of k months when system behaved unacceptable, and  $w_r$  related elementary event for r-th month (during W):  $W = \{w_l, w_{l+1}, ..., w_r, ..., w_{l+k-1}\}$ , then, by definition, to each  $w_r$  corresponds some shortage in water supply,  $V(w_r)$ , say volume in m<sup>3</sup>/month, and appropriate measure may be associated to an event W:  $V(W) = V(w_l) + V(w_{l+1}) + ... + V(w_r) + ... + V(w_{l+k-1})$ . If during period of 12N months, L events of W type occur, it is possible to define a value  $V_l^{max}$ , l=1,...,L, denoting maximum monthly shortage in a sequence of k consecutive monthly shortages.

The vulnerability index may be derived in either way:

$$v = \left(\frac{1}{L}\sum_{l=1}^{L} V_{l}^{\max}\right) / D$$
(4)

or

$$v = \frac{\max_{l \in L} V_l^{\max}}{D}.$$
(5)

D is total annual demand. The first one is more risk related definition of v. The second one is more appropriate in certain cases because it contains averaged information on extreme shortages and thus is unbiased.

Irrigation supply is of interest within certain water resources systems. In southeastern European countries such as Serbia and Macedonia irrigation season starts in April and ends in September. If the planning period is N years long, with calendar months April through September as discrete time frames, the system can be simulated followed by assessment of its performance indices such as reliability (or risk as it's opposite), resiliency and vulnerability. The system operation should be simulated over n = 1, 2, ..., 12N months but assessment should include half of it (6N months), i.e. n = (4, 5, 6, 7, 8, 9), (16, 17, ...),..., ((6N-5), (6N-4), ..., 6N) months accounted for only irrigation seasons in N years.

If the state variables are defined as monthly volumes of water delivered at given control points (diversions or pumps), and if the number of control points is equal to M, then assessment of results obtained by simulation model can be undertaken to define acceptable and unacceptable states of a system. The ultimate stage should be straightforward: computation of hazard performance indices given by equations (1)-(5).

#### **Results and discussion**

The following example is selected to show how we computed actual monthly shortages in water supply within the system to determine status of the system in particular month (Cf. Fig.1). Based on this computations for all months during April-September season and during the whole period of simulation, counters  $n_A$ ,  $n_F$ , W and Z are computed later on and used for deriving values  $\alpha$ , r,  $\gamma$ , and  $\nu$  as given by Eqs. (1)-(5).

Example application assumes that there are three irrigation systems in a system with two reservoirs in cascade, Fig. 2. Let M=3 denote number of demand points and assume that irrigation systems at these points are with equivalent priorities in receiving water in all months during vegetation season and in all years. If demands  $D_1$ ,  $D_2$  and  $D_3$  in given month are equal to 70, 100 and 30 water units, respectively, the total system demand is equal to 200 units.

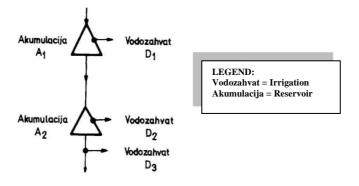


Fig. 2. System with two reservoirs and three irrigation diversions

Now, assume that simulation showed that demands  $D_1$ ,  $D_2$  and  $D_3$  are satisfied with 50, 80 and 20 units, respectively. It follows for the system that actual water shortage is

 $\varepsilon = 1 - (50 + 80 + 20)/150 = 1 - 150/200 = 0.250.$ 

Obviously, the total system demand is satisfied 75%, i.e. unsatisfied 25%. Locally, the situation is:

 $\varepsilon_1 = 1 - 50/70 = 0.286; \ \varepsilon_2 = 1 - 80/100 = 0.200; \ \varepsilon_3 = 1 - 20/30 = 0.333$ and for the whole system averaged actual shortage level (aASL)  $\varepsilon^a$  can be calculated as  $\epsilon^a$ 

$$= (\varepsilon_1 + \varepsilon_2 + \varepsilon_3)/3 = 0.273.$$

Now, suppose that demands have different priorities as shown in Table 1.

Table 1. Equal distance priorities of demands

Demand point (m)	Rank (z <sub>m</sub> )
1	3
2	1
3	2

Table 1 determines that demand  $D_1$  is of the lowest priority (z=3), compared to other two demands. If the weighting coefficients  $w_1, w_2$  and  $w_3$  are defined as reciprocal values of  $z_1$  $z_2$  and  $z_3$ , i.e.

$$w_{\rm m} = 1/z_{\rm m}, \quad m=1,...,3$$
 (6)  
weighted actual shortage level (wASL) for the system may be computed as

$$\varepsilon^{w} = \left(\sum_{m=1}^{M} w_{m}\varepsilon_{m}\right) / \left(\sum_{m=1}^{M} w_{m}\right).$$
 (7)

For above defined values the last relation finally gives

 $\varepsilon^{w} = (1/3x0.286 + 1/1x0.200 + 1/2x0.333)/(1/3 + 1/1 + 1/2) = 0.461/1.833 = 0.251.$ 

Parameter  $\varepsilon^{w}$  is more realistic than the previous one ( $\varepsilon^{a}$ =0.273) because it contains built-in priorities which recognize  $D_2$  as the highest important demand point in the system with related influence on total shortage of delivered waters. Preserving the same order of priorities, but switching to non-equal distance priority scheme, for example as shown in Table 2, gives

 $\varepsilon^{w} = (1/6x0.286 + 1/1x0.200 + 1/4x0.333)/(1/6 + 1/1 + 1/4) = 0.331/1.417 = 0.234$ which implies stronger influence of demand  $D_2$  than before because the other two points' impact is reduced twice on rank scale 1-6.

Table 2. Non-equal distance of priorities of demands Dema

and point (m)	Rank (z <sub>m</sub> )
1	6
2	1
3	4

Returning to the equal-distance scheme in Table 1, but assuming that priority matrix changes as indicated in Table 3.

Table 3. Changed equal distance priorities of demands	
Demand point (m)	Rank (z <sub>m</sub> )
1	2
2	3
3	1
it follows that new value of wASL for the system, due to	formula (7), is
$\varepsilon^{w} = (1/2x0.286+1/3x0.200+1/1x0.333)/(1/2+1/3+1/1) =$	0.543/1.833= 0.296.

This result reflects the highest influence of demand point 3 on evaluating total system shortage. Of course, local situations at demand points are not changed. Position of demand points within a system is important for both system operation and any performance evaluating process. Because spatial distribution of supplies and demands is changing from system to system, the easiest way to start analysis of system's performance is to apply equal priority strategy and deal with averages for the whole system and corresponding aASL ( $\varepsilon^{a}$ ) as reference value to distinguish acceptable from unacceptable systems states. Although reasonable and in consistence with some important practical reasons (for example, real situation to find equilibrium of different economical and technical judgments), this strategy is appropriate only if conservative approach is chosen. The other strategy, more difficult but very attractive, is to exploit recognizable relations between supplying and demanding points, to create appropriate subsystems and then, within each subsystem, apply the weighting methodology wASL ( $\varepsilon^{w}$ ) as described above. This idea may be applied from the very beginning of a planning process because it is in consistence with other major activity: defining global, regional and local goals, interests and priorities in global water resources system in which irrigation system may represent only a part.

## Conclusions

A logical framework for straightforward computation of hazard indices of water systems performance is created with focus on systems with irrigation demands. Indices are used to describe level of uncertainty and risk in supplying required waters during vegetation season to prioritized and equalized irrigation systems. By measuring also resilience and vulnerability of the system, a procedure is created to help system analysts and water planners to manipulate inputs to simulation models and recognize strategies how to protect farmers from long lasting deficits in water supply for irrigation (especially during peak demands periods), or occurrence of risky decreases of water quality in canals. The ultimate goal was to establish an adaptive global water management tool for treating hazards within large water resources systems, mainly by improving operation of lockers and pumping equipment along canals. Time frames spanned in all analyses are by assumption multiyear periods with consideration of only irrigation season months (April-September), and water quantity requirements setup at multiple points throughout hydro-systems.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

UDC: 631.67 Professional paper

# QUALITATIVE DECISION TREE FOR ASSESSING THE WATER FOOTPRINT SUSTAINABILITY INDEXIN THE CONTEXT OF WATER USE FOR IRRIGATION

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### Abstract

Water footprint is a concept receiving significant attention in recent years; not only regarding the scientific literature but also the practice (for example, ISO 14046 is the new standard for water footprinting). The sustainability of water footprint (SoWF) is a new issue and its measuring is of particular concern in ongoing related research. SoWF can be assessed from different aspects such as geographic, social, environmental, and economic), as well at different levels (locally, river basin scale, national or global level). The problems related to SoWF may be subjected to multiple criteria while evaluating attributes of sustainability, their boundaries, dynamic characteristics etc. There is no universally best set of criteria or the best water footprint sustainability assessment methodology. This paper presents the qualitative decision tree for one aspect of the problem: assessing the water footprint sustainability index, focusing on water use for irrigation only. It is based on some of the critical questions identified in the manual. Nine yes/no questions are presented as rationale setting related to water scarcity in the region, irrigation provoked environmental problems, water quality, characteristics of the source of water for irrigation, and economic justification of certain actions. Considering the problem in hand, yes/no answers to the questions could lead to four different outcomes (sustainable - index= 1; not sustainable index= 0; sustainable with environmental corrections - index=[0,1]; and sustainable with no correction regarding environment - index=[0,1]). Applicability of proposed decision tree is presented on an illustrative case study.

Key words: water footprint; sustainability index; irrigation; decision tree.

## Introduction

As water is a main resource for human activities and therefore a critical trigger for the welfare of the whole society, priority has to be given to its sustainable and effective use. In most regions of Europe, including Serbia and Italy, agricultural production systems are vulnerable to potential decrease of water availability. The impacts of climate change (increasing temperature, shifts of seasonal precipitation and decreasing summer rainfall) could cause water availability to reach critical limits. As a main water user, agriculture influences the regional water resources, which are available for key sectors. Moreover,

increasing world population and need for food, feed and bioenergy will require a more efficient agriculture and therefore irrigation will become an absolute need. However, under climate change water scarcity is likely to impose limits on irrigation enlargement in many European countries. Many attempts have been undertaken to improve the effective use of water in agricultural crop production but it appears that results are only locally valid and cannot easily be extrapolated to other regions.

In this context, the concept 'Water Footprint' (WF) was introduced by Hoekstra (2003), and later elaborated on by Chapagain and Hoekstra (2004), as an indicator that relates human consumption to global water resources. The WF of a crop is defined as the volume of water consumed for its production, where green and blue WF stand for rain and irrigation water usage, respectively. The importance of this parameter lies in the fact that international trade in commodities creates flows of what has been called 'virtual water' (Allan, 1998; Chapagain and Hoekstra, 2004; Hoekstra and Hung, 2005), by importing and exporting goods that require water. The indicator provides valuable information for a global assessment of how water resources are used. In recent years WFs and virtual water have been calculated for crops, goods, services, as well as at generic national levels (Hoekstra and Hung, 2005; Siebert and Döll, 2010).

The WF of crops forms the basis for WF estimations of crop products and derived commodities. Blue WF stands for irrigation water usage and green WF for rainwater usage. In the case of irrigation, a part of the water withdrawn from the surface or groundwater system is evaporated between the point of withdrawal and the field, another part infiltrates and returns to the water source (and can be reused), and the rest reaches the field; there is a part that turns into drainage flow, which may be available for reuse as well. The blue WF refers to the sum of evaporation from storage reservoirs, transport canals and evapotranspiration from the field, although many previous studies have focused on the physical processes that happen in the field.

The WF of a crop is defined as the ratio between the evapotranspiration (ET) and the crop yield, computed over the cropping period (WF manual, 2009). The green component in the water footprint of a crop (WF<sub>g</sub>,  $m^3$ /ton) is calculated as the green component in crop water use (CWU<sub>g</sub>,  $m^3$ /ha) divided by the crop yield (Y, ton/ha), according to WF manual (WF manual, 2009). The blue component is calculated in a similar way:

$$WF_g = \frac{CWU_g}{Y}, \qquad \qquad WF_b = \frac{CWU_b}{Y}$$
 (1)

The green and blue components in crop water use are calculated by accumulation of daily evapotranspiration (mm/day) over the total growing period:

$$CWU_{g} = 10 \cdot \sum_{d=1}^{l g p} (ET_{g})_{d}$$
,  $CWU_{b} = 10 \cdot \sum_{d=1}^{l g p} (ET_{b})_{d}$  (2)

where the factor 10 is meant to convert water depths in mm into water volumes per land surface in  $m^3/ha$ . Summation takes place from the day of planting (day 1) to the day of harvesting (*lgp* stands for length of the growing period in days).

The grey component in the process water footprint of a primary crop  $(m^3/ton)$  is defined as the volume of freshwater that is required to assimilate the load of pollutants based on existing ambient water quality standards. It is calculated by dividing the load of pollutants that enters the water system (*L*, in mass/time) by the difference between the

ambient water quality standard for that pollutant (the maximum acceptable concentration  $c_{max}$ , in mass/volume) and its natural concentration in the receiving water body ( $c_{nat}$ , in mass/volume). More information about the parameters involved can be found at WF assessment manual (WF assessment manual, 2011).

$$WF_{proc,grey} = \frac{L}{c_{\max} - c_{nat}}$$
(3)

Although the number of research activities related to the assessment of food and energy crop water footprints is steadily increasing, none of them has yet addressed the question how WFs are affected by climate change and variability and the issue of change over years has not yet received attention. Under future climatic conditions, water will likely be scarcer in many areas of Europe and related problems will represent an even more critical challenge for society. This means that the sustainability of water use for different purpose should be evaluated in present and future conditions and one of possible ways to do that is through usage of the WF concept.

#### Sustainability concept

The concept of sustainable development was born in the early 80s on the basis of considerations relating to the relationship between society and environment. Today this concept is rooted in the political dialogue in the world with increasing understanding of the interactions between social systems and environmental systems. The report of the Brundtland Commission (1987) defines sustainable development as development that meets the needs of the present without compromising the ability of future generations to meet their own needs. Economic development, therefore, is assessed not in absolute terms but in relation to the ability to contribute to the wellbeing for future generations. Since the Rio Summit in 1992, a definition of sustainable development has been introduced, which is based on the three pillars of economic efficiency, social equity and environmental sustainability. While, with the adoption of Agenda 21, signatory countries have pledged to develop quantitative information on their actions on these three pillars.

Summarizing from (Berke and Manta, 1999; Lundin, 2003; Singh et al., 2009), sustainable development indicators (SDIs) can be used to: a) anticipate and assess conditions and trends; b) provide early warning information to prevent economic, social and environmental damage; c) formulate strategies and communicate ideas; and d) support decision-making. When developing a framework and selecting SDIs, two distinctive main approaches can be identified (Lundin, 2003): a) the 'top-down' approach, which means that experts and researchers define the framework and the set of the SDIs, and b) the 'bottom-up' approach that features the participation of different stakeholders in the design of the framework and the SDIs selection process. According to Kates et al. (2001), the purpose of sustainability assessment is to provide decision makers with an evaluation of global to local integrated nature–society systems in short and long term perspectives in order to assist them to determine which actions should or should not be taken in an attempt to make society sustainable (Singh et al., 2009).

Regarding the water resources, as an important part of the environmental component, Loucks (1997) defined sustainable water resources systems as "those systems designed and managed to fully contribute to the objectives of society, now and in the future, while maintaining their ecological, environmental and hydrological integrity."

Although this concept is still valid, water management policies that promote sustainable water resource systems are becoming more difficult to identify because of nexus considerations, water scarcity, and climate change (Sandoval et al., 2011).

### **Results and discussion**

## Definition of WF sustainability index on European level

Europe is one of the major users of fresh water in the world when referenced using the indicator of water footprint for its estimation. Furthermore, it is one of the largest importers of virtual water in the world. The Southern Europe in particular is the area that mostly depends on water for its development, such as Spain, Italy, and Greece (Chapagain and Hoekstra, 2004). Therefore, the sustainability of water resources in Europe cannot be separated from an afterthought of agricultural development, which is well known coming into play in the use of water resources (Galli et al., 2012). In this paper focus is on irrigated agriculture. More in detail, when we define sustainability, considerations made must address several aspects.

### Environmental sustainability and WF

The environmental sustainability is guaranteed when the water resource characteristics (quantity, quality, etc.) are included within specific limits defined by a political convention. In this regard, when the green component of the WF is taken into consideration, the most important aspect is the availability and storage of the water into the soils. In many cases, the sustainability of green WF is compromised from land use alterations. When referring to the blue component of the WF, the most important aspect considered is the amount of water used. In the case of rivers or groundwater, their condition is considered sustainable in terms of quantity when their flows are within limits of the natural runoff. Compliance with this convention ensures users not to exceed the thresholds that could threat ecosystems and the livelihoods of people who depend on these systems. Also, the water quality and "ambient water quality standards" that people have agreed should be taken into account in the assessment of the sustainability of the WF (Richter, 2010).

## Economic sustainability and WF

Sustainability should be considered evaluating the use of water resources in terms of costeffective. In fact, the benefits of the use of water for a given product should exceed the costs associated with its water consumption (WF assessment manual, 2011).

#### Social sustainability and WF

In these terms, sustainability should be considered as the minimum amount of freshwater available to "basic human needs" (drinking, washing, cooking) and a minimal amount of water for food production to allow food security. This minimum amount of sourcing should be subtracted from a given location (e.g. basin level) only when all environmental requirements have been satisfied.

The sustainability of the WF can be assessed in different ways, depending on the criteria of analysis chosen and adopted. One of the most used systems of sustainability analysis takes into account the geographical approach. This takes into account a river basin or catchment

area. The use of this approach allows verifying, thanks to the geographical delimitation, many variables that are involved in use of water for agriculture within a specific area (Blagojevic et al., 2012: Blagojevic, 2015; Richter, 2010).

## Qualitative decision tree for assessing the WF sustainability index

Starting from the critical questions (Table 1) reported in the WF manual (2009), we have proposed a qualitative decision tree (QDR) for assessing the WF sustainability in the context of irrigated agriculture, by considering green and blue WF, while grey WF is treated indirectly (Figure 1). When the green, blue or grey WF in a catchments does not fulfil one of the criteria of environmental, social or economic sustainability, the WF cannot be considered as 'geographically sustainable'.

Table 1.Critical questions to be posed when assessing the sustainability of a water footprint	
(WF manual, 2009)	

	Environmental perspective	Social perspective	Economic perspective
Micro-	* Does the green water	* Does the water	* Is the water productivity
level:	footprint favor production at	footprint deprive	optimal?
local	the cost of valuable natural	other local water	* Can water be saved
	vegetation and biodiversity?	users?	without reducing the
	* Does the blue water footprint		production?
	violate local environmental		* Is the price of water for the
	flow requirements at any time?		user below its real economic
	* Does the grey water		cost, resulting in inefficient
	footprint		use?
	violate local ambient water		* Is water scarcity factored in
	quality standards?		into the decision to consume
			water?
Meso-	* Does the blue or green water	* Does the green,	* Is the water allocation in
level:	footprint lead to a changed	blue or grey water	time and space over different
river	runoff pattern and thus affect	footprint affect	users optimal?
basin	downstream environmental	downstream users	* Are there opportunity costs
	flow requirements?	without proper	by not consuming water for
	* Does the grey water	compensation or	another purpose providing
	footprint	benefit sharing?	more value?
	contribute to violation of		* Are there uncompensated
	ambient water quality standards		external effects on
	downstream?		downstream users?
Macro-		* In :4:4-1-1- 4	* Ano no signal ano de stien
level:	* Can the water footprint for the given purpose be sustained	* Is it equitable to have this water	* Are regional production patterns of and trade in
beyond	given the larger context of	footprint for the	water-intensive products
the river	limited freshwater availability	given purpose given	optimal (efficient) given
basin,	worldwide and other (possibly	the larger context of	the larger context of limited
global	more important) competing	limited freshwater	freshwater availability and
giobai	freshwater demands?	availability	uneven distribution
	noshwater demands:	worldwide and other	worldwide?
		(possibly more	* Are low-value water-
		important) competing	intensive products exported
		demands?	from a water scarce region?
		actinutius.	from a water searce region:

The Milestones for the basic idea of QDR are:

- As a main water user, agriculture influences the regional water resources, which are available for all key sectors. Moreover, increasing world population and the consequent increasing need for food, feed and bioenergy will require a more efficient agriculture and therefore irrigation will become an absolute need.
- Under climate change water scarcity is likely to impose limits on irrigation enlargement in many European countries.
- Blue WF stands for irrigation water usage and green WF for rainwater usage.
- QDR will be able to integrate economic, environmental and social aspects.

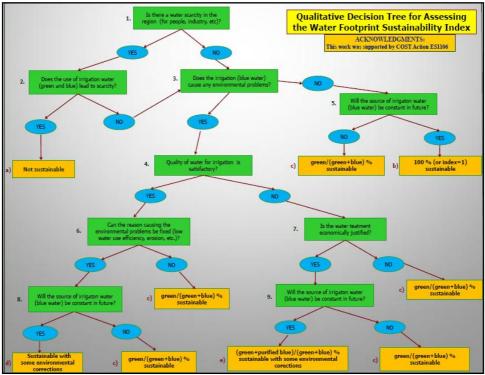


Figure. 1. Qualitative decision tree for assessing the WF sustainability

Fig. 1 presents the qualitative decision tree (QDR) for one aspect of the problem: assessing the water footprint sustainability index, focusing on water use for irrigation only. It is based on some of the critical questions identified in the WF manual (2009). Nine yes/no questions are presented as rationale setting related to water scarcity in the region, irrigation provoked environmental problems, water quality, characteristics of the source of water for irrigation, and economic justification of certain actions. Considering the problem in hand, yes/no answers to the questions could lead to four different outcomes (sustainable - index=1; not sustainable - index=0; sustainable with environmental corrections - index=[0,1]; and sustainable with no correction regarding environment - index=[0,1]).

Applicability of proposed QDR is presented on three illustrative case studies. For Vald'Orcia (Tuscany region, Italy) the result is 100% (or index=1) sustainable. For

Maremma (south of Tuscany region, Italy), considering the tomatoes, the result is green/(green+blue) 28.4% sustainable. For Stara Pazova region (Vojvodina Province, Serbia) the result is 100% sustainable for wheat, maize, sugar beet, sunflower, beans, potatoes, clover, alfalfa, meadows and apples production.

## Conclusions

Although it is very hard to define index which will be able to quantify such complex issue as sustainability of WF we made some preliminary efforts in this direction. Using some of the questions regarding sustainability from WF manual (2009), we presented qualitative decision tree (QDR) for assessing the WF sustainability index, focusing on water use for irrigation only. QDR is based on both qualitative and quantitative aspects of sustainability and it is able to integrate economic, environmental and social criteria. Developed QDR represents first phase of research and in future focus will be on the assessment of water footprint of key irrigated agricultural products, including their uncertainties, as well as on scenarios concerning WF under future climatic conditions. The use of advanced tools and updated climatic databases, climatic projections/scenarios data such as and agrometeorological models will represent the base of the future research. The use of such instruments will allow a detailed analysis of interactions between crops, climate and management that will be taken into account in the WF sustainability assessment.

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# NATURAL CONDITIONS FOR THE FORMATION OF THE SOILS UPON LIMESTONE AND DOLOMITE IN THE REPUBLIC OF MACEDONIA

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#### Abstract

In this paper are presented the results from the research of natural conditions of the soils formed upon limestones and dolomites in the Republic of Macedonia. The filed researches have been performed in the course of 2010, 2011 and 2012 during which 52 basic pedological profiles were excavated, 34 of which are calcomelanosols (WRB-Rendzic Leptosol), 13 calcocambisols (WRB-Chromic Leptic Luvisol on hard limestones) and 5 profiles of red soils terra rossa (WRB-Rhodic Leptic Luvisol on hard limestones). In addition, data were collected for natural conditions for the formation of the soils (parent material, relief, climate, vegetation, time and human impact). Surveyed soils are formed upon horn slate - hornfels facies and (massive limestone, dolomitic limestone, bituminous marbles, plate (flat) limestone, dolomitic marbles on laminated (plate) dolomite and calcite). The relief is mostly mountainous and the soils are formed at an altitude of 211-2000 m, under variety of oak and beech communities, and in subalpine belt under alpine grassland communities. Examined soils are extending into six climate-vegetation-soil regions (Submediterranean modified Mediterranean, Warm continental, Cold continental, Piedmont-continental-mountain, Mountain-continental, Subalpine mountain region). Time is a very important factor for the formation of these soils, butnone of the natural soil forming factors can't cause so intense and rapid changes in the ecosystem and soil as human factor. Moreover, its impact on the soil can be positive or negative. Key words: soil forming factors, soils upon limestones and dolomites

## Introduction

The soils formed on limestones and dolomites can be found in all mountain regions and regions which are fully or partially composed of limestones and dolomites. It can be said that these soils are present on almost all continents due to their connection with the parent material. On the basis of the soil map of R Macedonia with a 1:50 000 scale, the areas under soils formed on limestones and dolomites amount to 314.385 ha. These soils cover

12.23% of the total surface area of the Republic of Macedonia (2.571.300 ha) (Филиповски, 2003), where the calcomelanosols cover 8.61% or 221.441 ha, calcocambisols 3.61% or 92.944 ha, and terra rossa cover 0.005% or 117.7 ha. As a result of our field research, the soils formed on limestones and dolomites were found on Jablanica, Galichica, Ilinska Planina, Bistra, Suva Gora, Suva Planina at the foot of the accumulation Kozjak, Pletvar, Sivec, as well as on the higher parts in Dojran. The greatest part of the calcomelanosols and calcocambisols, due to their connection to the parent substrate, cover the abovementioned locations. In some of those regions, they have been confirmed by the authors: (Поповски, et. al., 1962); (Спировски, 1963, 1967); (Танев, 1972, 1972а); (Мицевски, еt. al., 1991); (Виларов, 1960); (Поповски, еt. al., 1969, 1969а); (Танев, 1969); (Спировски, 1964,1970,1973); (Митрикески, еt. al., 1988); (Андреевски, 1996, 2013). The terra rossa, due to its connection to the parent substrate and the climate (Mediterranean and Sub-Mediterranean), was confirmed at the foot of Galichica, right next to the border with Albania, then at the foot of Jablanica right next to Radozhda, as well as on the limestones around Dojran. The terra rossa have been soil surveyed and recorded only in two localities by the authorsФилиповски,et. al. 1972 cf. in (Филиповски, 1997) and (Поповски, 1960), and confirmed by the authors: (Виларов, 1960); (Спировски, 1963); (Spirovski, 1967); (Андреевски, et. al., 2000). On the (map N°1 and 2) can be seen location of the soils formed on limestones and dolomites in the Republic of Macedonia.

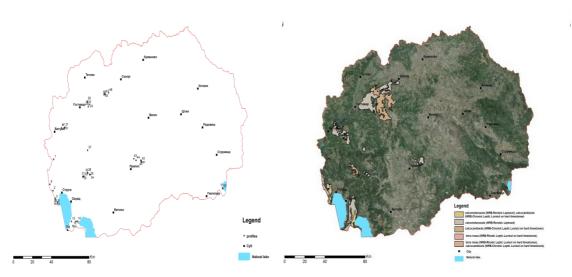


Figure 1.Map 1 and 2. Location of the soils formed on limestones and dolomites in the Republic of Macedonia.

These soils, according to their genesis and evolution differ from the soils formed on other substrates and have a line of specificities.

The opinions of the soil forming factors in the literature differ. Some authors believe that the terra rossa are lithogenic soils because they are formed similarly to the calcomelanosolson pure compact limestones and dolomites, and according to other authors they are zone soils, since they appear only in a Mediterranean and modified Mediterranean climate. Other authors combine the opinions and believe that they are lithogenicclimatogenic soils, since the substrate is not dominant factor for their formation, and the climate factor contributes to their maintenance and evolution. Similarly to the calcomelanosols, the calcocambisols can also be treated as lithogenic soils. But these soils were also not formed in some climate-vegetation zones of our limestone dolomite mountain massifs, which suggesting of the strong influence on the other soil forming factors and points to a zonal distribution (Маркоски, 2013).

## Material and methods

During 2010, 2011 and 2012 year, field researches have been carried out for the soils formed on limestones and dolomites, on various locations on the territory of the Republic of Macedonia. Following the field recognition, locations were selected for digging out of the basic pedological profiles. A total of 52 basic pedological profiles were dug out, from which 34 are calcomelanosols WRB-Molic Leptosol (WRB Classification, 2014), 13 calcocambisols (WRB-Calcic Cambisol) (WRB Classification, 2014), and 5 profiles of terra rossa WRB-Rhodic Leptic Cambisol (WRB Classification, 2014).In addition, data were collected for conditions of soil forming (parent material, relief, climate, vegetation and human impact). Since these profiles were collected soil samples for laboratory analysis. The field research was carried out in accordance with the generally accepted methodology in our country (Митрикески, et.al., 2006).

## **Results and discussion**

# Geographical position and relief

Republic of Macedonia is situated in the central part of the Balkan Peninsula, in the southern part of the temperate zone and on the borders of the subtropical zone, between  $40^{\circ}$  50' and  $42^{\circ}$  20' north latitude and  $20^{\circ}$  27' and  $23^{\circ}$  05' east longitude. Its total territory is 25.713 km<sup>2</sup>. The relief is very heterogeneous. According to (Filipovski, 1982), Macedonia is divided in 4 relief forms with varying altitudes (from 40 to 2764 m), different expositions and inclinations. We will pay special attention to the mountain relief since it is significant for the occurrence of the soils which are formed on limestone and dolomites, where these soils are mostly located.

The mountains in the Republic of Macedonia according to (Панов, 1976) belong to the Rhodope and Dinar massif. The Rhodope massif is an older one, formed in the hercin orogenic phase. According to Цвијиќ 1906 cf. in (Маркоски, 2013), the Rhodope massif is made of carbon folding, all the way to the radial movements in the neogen, it was the same land. The mountains from the Dinar group are younger and were formed in the Alpine orogenesis. In the frames of the Dinar tectonic unit according to the geological composition, the age and tectonics, the mountains belong to: the Shara group, the Vardar zone and the Pelagonia crystal massif.

According to the altitude above sea level ( $\Pi a_{HOB}$ , 1976), the mountains in the Republic of Macedonia are divided in three groups: high with an above sea level altitude over 2000 m; medium (1000 to 2000 m) and low (500 to 1000 m). All groups of mountains are significant for the genesis and the properties of the soils formed on limestones and dolomites.

The following should be mentioned from the group of high mountains: Galichica (2265 m), Jablanica (Crn Kamen 2273 m), Bistra (Mednica 2163 m), and from the group of medium mountains: Suva Gora (1784 m), Ilinska Planina (1642 m), Kozjak (1264 m) etc.The soils formed on limestones and dolomites appear in lower relief forms, like the ones formed in the Dojran region on an above sea level altitude of over 200m.

Our carbonate mountains are especially significant for the genesis of these soils, comprised of compact limestones, dolomites and marbles, where the karstification creates a specific relief. In conditions when the entire rock is covered in soil, the limestone underneath it shows a micro relief, due to which the soil has different depth on small distances. This is the cause for the occurrence of various types and subtypes of soils: organogenic, organomineral and brownised calcomelanosols, calcocambisols and terra rossa. The mountain relief in addition to the direct influence on the soil genesis of these soils affects indirectly through the rest of the soil forming factors. The soils formed on limestones and dolomites on the more inclined slopes, due to the highly expressed erosion, are usually shallower, when compared to the same soils formed on less inclined slopes, where the erosion is weaker. To gain a clearer image for the researched soils, Table 1 shows some of their external morphological properties, as well as the precise locations of the profiles. Through the above sea level altitude and exposition, the relief influences the properties of these soils. By increasing the above sea level altitude, the amount of rain is increased, the temperature drops and as a consequence of this decomposition the organic matter is slower, the content of humus in the soils increases, the processes of dealkalization and acidification are intensified, and the intensity of the chemical weathering is being slowed down. The profiles which were excavated during the terrain research are at above sea level altitude of 211 m up to 2000 m, i.e. out of a total 52 profiles, 15 are at above sea level altitude of 1500 to 2000 m (profiles 6; 11; 12; 16; 17; 18; 19; 20; 21; 22; 30; 31; 32; 33; 34), 22 profiles from 1000 to 1500 m (1; 2; 3; 4; 5; 13; 14; 15; 23; 24; 25; 26; 27; 35; 36; 37; 38; 39; 40; 41; 42; 43), 11 profiles from 600 to 1000 m (7; 8; 9; 10; 28; 29; 44; 45; 46; 47; 48) and 4 profiles from 200 to 600 m (49; 50; 51 and 52). We have confirmed that most of the profiles appear at an altitude of 1500 to 2000 m. The average above sea level altitude of the calcomelanosols is 1287 m, and according to subtypes: the organogenic are at an average above sea level altitude of 1700 m, organomineral are at 1090.67 m and brownised calcomelanosols on 1505.33 m. Calcocambisols are found at an average above sea level altitude of 1283.77 m, and terra rossa at 537.80 m above sea level altitude. The soils formed on the south and south-west expositions are dryer, warmer and often have a lower content of humus when compared to the soils formed on the north and northeast exposition. The south expositions quickly heat-up early in the spring, the snow melts quickly and conditions for a faster mineralization of the plant residues (favourable humidity and favourable temperature) due to which the soils contain a smaller content of humus.In the north and northeast expositions the snow is held longer, and the temperature is regularly lower, the organic residues are decomposition more slowly and there is a greater accumulation of humus in the soils.

#### Geological composition

The parent rocks in the soil genesis act together with the other soil forming factors. During the definition of these soils, it was pointed out that they are formed only on hard and pure carbonate rocks. Figure 2 contains an overview of the parent materials on which calcomelanosols, calcocambisols and terra rossa are formed.

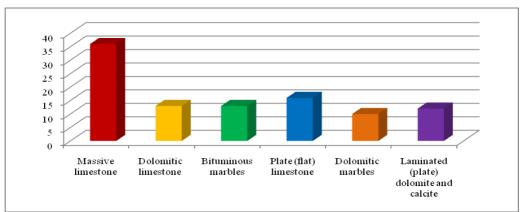


Figure 2. Percentage of parent materials in calcomelanosols, calcocambisols and terra rossa formation

It can be seen from the chart that 36% of the researched soils are formed on massive limestone (profiles 1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,49,50,51,52), 13% are formed on dolomitic limestone (16,17,18,19,20,21,22) and bituminous marbles (23,24,25,26,27,28,29,) 16% on plate (flat) limestone (30, 31, 32, 33, 34, 35, 36, 37), 10% on dolomitic marbles (38,39,40,41,42) and 12% on laminated (plate) dolomite and calcite (43,44, 45,46,47,48).On the basis of the above, it can be confirmed that the soils are formed from horn slate – hornfels facies, and their composition has an influence on the mechanical composition.

				Ceogr aphic	Geographical position				
Prof. N	Location	Soil types	Parent material	NL	EL	Altiturle (m)	Indication %	Exposition	P hytocen osi
Ħ	Jablanica	Eh	ML	41°12'17 35''	20°34'16 83''	1490	40-50	South	
t	Jablanica	41	WT	41-11 49 89	20 34 34 37	138/	0000	North-west	
t	ablanica	11		CC 00 00 14	11 04 00 07	++++		r ast	Ass. Calaminuro grandillorae-ragetum
t	Japianica	PA1	TIN	10 77 02 021	06 14 00 07	0++1	00-00	Last	
1	Jablanica	u I	ML	41 06 0/ 06	77 64 .02 07	/ 071	0,00	Last	Ass. Querco – Ustryetum carpinjoliae
†	Jablanica	u I	ML	41-14 30 22	20-32-10 30-	1902	/0-20	North	Ass. Onobrichi – Festucetum cyllenicae
1	Jablanica	it	ML	41'06 14 81'	20~37.49.94	765	40-50	North	Ass. Querco Carpinetum orientalis
1	Jablanica	Eh	MI	41'06 16,66'	20'37'46,10''	791	40-50	South	Ass. Querco Carpinetum orientalis
6	Galičica	Lf	ML	40° 54, 37' 90"	51	740	40-50	South - East	Ass. Quercetum frainetto – cerris
1	Galičica	Lf	ML	40° 54, 38' 33"	58,	740	40-50	South - East	Ass. Quercetum frainetto – cerris
	Galičica	Εh	ML	40° 57, 14' 46"		1650	50	East	
	Galičica	Еh	ML	57,	45,	1650	50	East	Ass. Seslerietum wettsteinii
	Galičica	Еh	ML	51,		1460	40-50	West	Ass. Festuco heterophillae – Fagetum
	Galičica	Lvd	ML		20°48, 32'48"	1320	30-40	West	Ass. Querco – Ostryetum carpinfoliae
	Galičica	Lvd	ML	40° 58, 06' 24"	20°48, 22' 27	1154	30-40	West	Ass. Querco – Ostbyetum carpinfoliae
	Bistra	먹크	DL	39,	20° 42, 48' 76"	1706	40-50	North	Ass. Calumintho gradiflorae – Fagetum
	Bistra	Εħ	DL	41° 38, 25' 68"	20° 41, 22' 37"	1728	40-50	North	Ass. Bruckenthalietum – Juniperetum
	Bistra	Eh	DL	41°38,25'68"	20° 41, 22' 37	1728	40-50	North	Ass. Bruckenthalietum – Juniperetum
	Bistra	Ξħ	DL	41°38,00°52"	20° 42, 44' 07"	1730	50	North	Ass. Bruckenthalietum – Juniperetum
20	Bistra	Lvd	DL	41° 38, 29' 88"	20° 41, 28' 26"	1720	50	North	Ass. Bruckenthalietum – Juniperetum
	Bistra	Eh	DL	41° 38, 22' 98"	20° 42, 25' 69	1750	40-50	North	Ass. Bruckenthalietum – Juniperetum
	Bistra	Lvđ	DL	41°38,07'51"	20°42,32'37"	1730	40-50	North	Ass. Bruckenthalietum – Juniperetum
	Suva Gora	Eh	BM	17,	,90	1350	40	South - East	Ass. Onobrichi – Festucelum cyllericae
24	Suva Gora	Eh	BM	41°48,21°62"	02,	1370	40	West	Ass. Onobrichi – Festucelum cyllericae
25	Suva Gora	Еħ	BM	05;	00, 21'	1270	20	West	
	Suva Gora	Eh	BM	49,	20° 59, 32' 07"	1060	20-30	North - West	Ass. Onobrichi – Festucelum carpinfoliae
1	Suva Gora	Lvd	BM	27;	59, 31'	1050	20-30	North	- 1
28	Suva Gora	Lvd	BM	44	20' 59, 04' 78	938	30-40	North	Ass. Querco – Ostbyetum carpinfoliae
29	Suva Gora	Lvd	BM	45,	5	830	30	North - West	
1	llinska	Lvd	PL		20 59, 07 21	1522	30-40	North - East	Ass. Calumintho gradiflorae – Fagetum
+	llinska	Lvd	LL L	1/, 30.	20 29, 11 31	1224	30-40	North - East	Ass. Calumintho gradifiorae – Fagetum
1	llinska	PA4	74	1/, 28	22, 10	0/01	00	South	Ass. Calumintho gradifiorae – ragetum
1	llinska	41	PL	17, 38'	29, 02 <sup>-</sup>	1001	30	North	Ass. Calumintho gradifiorae – Fagetum
34	llinska	41	PL	17, 39	×,	1504	30-40	North	
1	llinska	41	PL	17, 49'	8	1487	40	South	Ass. Calumintho gradifiorae – Fagetum
+	llinska	41	PL	ali	20° 58, 47' 92	1437	30-40	North - East	
37	llinska	ЧЭ	PL	17	20 58, 45' 73	1432	30-40	North - West	Ass. Calumintho gradifiorae – Fagetum
†	Pletvær	PAT	MU		40, 28.	1100	000	West	Ass. Juniperus communis intermedia
20,	Pletvar Ni	4	N C		21.40,27.57	1200	00-07	West	Ass. Juniperus communis intermedia
4 C	PI etv ar	10		ŧ[a	21 40, 2/ 22	C071	02-07	Nest	Ass. Juniperus communis intermedia
t	PI ETV ar	u u		24, 10	21 40, 50 65	11/4	40-00	North	Ass. Juniperus communis intermeata
t	Pletvar	44	Win .	24, 14	40, 41,	2001	40-00	North	лаз. липрекиз соттины плектеана
$\dagger$	PI etv ar	u u		41-24, 51 88	21 25, 54, 15	1050	00-00	North West	Ass. Juniperus communis intermedia
; ;	C Di	14			24 22, 27, 24	000	0000		Ass. Variperas communis intermedia
t	Suva Flanina	1.14		50 51	12, 02	202	09-00	Contract	Ass. Querco - Carpinetum orientatis suoass. Duxetosum
1 1 1	SUVA FLANINA	P.4		10.	21 12, 22 02	140	00-00	South	Ass. Querco - Ustryenum carpinijoliae
t	SUVA FIAMINA	44		i s	j.	1/1	09-00	SOULD - Last	Ass.Querco - Carpinetum orientatis.
0 1 0	SUVA FLANINA	4	T F F C	22, 24	21 12, 22, 30	740	40-00	L'ast	Ass. Querco - Ostryetum carpinifoliae
44	Dojran	u I	ML	n l	22 41, 35 39	CC7	40-20	East	Ass. Cocofferro – Carpinetum orientalis.
20	Dojran	Ļŕ	WI	13, 43'	22 41, 39' 22	233	40-50	South - East	Ass. Cocciferro – Carpinetum orientalis.
1	Dojran	Ħ	ML	41 13, 46' 36"	22 41, 39 98	211	40-00	North	Ass. Coociferro – Carpinetum orientalis.

## SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

This facia has a thickness of around 500-550 m, it is composed of dolomites, plate (flat) limestone with hornfels and massive limestone, and it represents a continuation in Anisian and Ladinian, and it strongly reflects on the morphology, physical and physical-mechanical properties, mineral composition and the chemical properties of the soils.

### Climate

According to (Филиповски, et. al. 1996), our country is divided in 8 climate-vegetationsoil regions, and the studied soils (calcomelanosols) are located in six climate-vegetation belts, in five (calcocambisols) and in two (terra rossa), (Table 2).

		Clin	nate-vegetatio	on-soil region.	\$		
	Sub-	Warm –	Cold-	Piedmont-	Mountain -	Subalpine	
Soil types	Mediterranean	continental	continental	continental	continental	_	
51	(modified	600-900 m	900-1100	-mountain	1300-1650	mountain	
	Mediterranean)		m	1100-1300	m	1650-	
	up to 50-500 m			m		2250 m	
					1; 2; 3;	6; 11; 12;	
Calcomelanosols	49; 52	8; 45; 47	26; 43;	5; 25; 39;	13; 23;	16; 17;	
			44; 48	40; 41; 42	24; 33;	18; 19;	
					34; 35;	21	
					36; 37		
Calcocambisols	/	29; 46	27; 28	15; 38	4; 14; 30;	20; 22	
					31; 32		
Terra rossa	50; 51	7; 9; 10	/	/	/	/	

Table 2. Climate-vegetation soil regions

The largest percentage of the profiles are located in the mountain-continental region, a total of (16) profiles, or 31%, in the subalpine-mountain region (10) profiles or 19%, then in the warm-continental (8) profiles or 15%, also (8) profiles or 15% in the piedmont-continental mountain region, (6) profiles or 12% are located in the cold-continental region, and the lowest number of profiles (4) or 8% in the continental- sub mediterranean region.

For the characteristics and the description of the climate in the regions where the soils formed on limestones and dolomites are located, we used data from the papers of (Филиповски, 1995, 1996 and 1997).

The climate of the Sub-Mediterranean modified Mediterranean region differs from the climate of the other regions in terms that the influence of the Mediterranean can be most prominently noticed. The mean annual temperature of this region is  $14.2^{\circ}$ C and the average amount of rain is between 611 up to 695 mm. According to the Lang's rain factor, the climate is semi-arid, but in four months (from VI until IX) it is arid.

The warm-continental region is dominated by the warm continental climate. The mean annual temperature in this region varies from 9.6 up to  $11,8^{\circ}$ C (mean  $10.9^{\circ}$ C), and the average amount of rain is around 700 mm.

The cold continental region covers a narrow belt of 900 to 1100 m above sea level altitude where the cold continental climate is dominant with some influence from the mountain climate, as a result of the larger above sea level altitude. The climate is colder and more humid when compared to the continental region where the mean annual temperature is  $8.6-9.6^{\circ}$ C or average of  $9^{\circ}$ C. The average amount of rain is 800-850 mm. According to the Lang's rain factor, the climate is humid.

The Piedmont-continental mountain region covers a vertical belt with an approximate above sea level altitude of 1100 up to 1300 m. The mean annual temperature is between 7.5 to  $8.6^{\circ}$ C or an average of around  $8^{\circ}$ C, and the average amount of rain is around 900 mm. The number of snowfall is greater when compared to the previous belt, and the duration of the snow cover is longer. According to the Lang's factor, the climate in this belt is humid. The same as in the cold continental region, there is not period of drought.

The mountain–continental region covers about 1300 up to 1650 m above sea level. The main difference between this and the previous region arises from the stronger influence of the mountain climate in this area and the mean annual temperature is  $1.6^{\circ}$ C lower than the piedmont-continental mountain region and on average it is  $6.8^{\circ}$ C, and the average amount of rain is 1044 mm. According to the Lang's rain factor, the climate is humid and approximates to perhumid.

The sub-alpine mountain region covers a wide belt from 1650 up to 2250 m. The climate in this region differs greatly from all the other regions due to the domination of the mountain climate which causes a significant drop in the average annual temperature, a drop of the average temperatures in all four seasons, but most prominently in the summer and spring months, causing the annual amplitude to drop. There are no hot days, and the number of days with frost increases. The amount of rainfall in this region does not differ drastically from the mountain continental region. The average temperature in this belt is only  $4.8^{\circ}$ C. The vegetation season is short, around 100 days. According to the Lang factor, the climate is perhumid.

In the described conditions where the researched soils have been formed, the climate as a factor has a different influence with the three types of soil.

For the calcomelanosols, the climate has the following meaning for soil genesis: as a result of the great daily differences of the temperatures, the long freezing and defrosting of the water, the limestone wethering physically as well (formation of crumbs, coarse soil). The great amount of rain with humidity and the perhumid nature of the climate during many months, in the soil affect so that the decedent flows are predominant and there is leaching of the formed  $Ca(HCO_3)_2$ , which is assisted by the permeability of the soil and the limestone. The long winter periods of sub-zero temperatures, as well as the dry summers do not enable more intensive microbiological processes and mineralization of the organic matter. Due to the greater period of low temperatures in these zones, the processes hydrolysis and argilisation do not take place. The clay which is released from the limestone is included in the organo-mineral complexes. The influence of the climate on the soil genesis in the calcocambiosols is very similar as well as the previous soil type.

The greatest influence and meaning of the climate can be noticed with the terra rossa. A great number of researchers agree that the genesis or the preservation of the terra rossa in that stadium is connected to the climate. The climate and the parent substrate are the main factors for the soil genesis of the terra rossa. Duchaufour, (1977) cf. in (Марковски, 2013) points out that the ferisialitic soils (and according to him the terra rossa is a member) appear in regions with sub tropic, Mediterranean and Sub-Mediterranean climate. In places where there is no influence from the Mediterranean climate, the terra rossa appears locally, and it is closely connected to a certain soil climate. These characteristics of the climate which are different in all regions have a great influence on the accumulation of organic matter and the degree of acidification which reflects of the physical, physical-mechanical, mineralogical and chemical properties of the soils.

## Vegetation

The occurrence of one or other plant communities is closely connected to the heterogenic climate, relief and soil conditions of the environment. For comments regarding the vegetation in the regions where the soils formed through limestones and dolomites are located, we used data from more authors: (Николовски, 1958), (Николовски, et. al. 1973), (Василевски, 1996), (Филиповски, et. al., 1996), (Филиповски, 1996), (Филиповски, ет. аl., 1996), (Филиповски, 1997).

The forest community of Quercus coccifera and Carpinus orientalis is spreadin the Submediteranean modified Mediterranean area, however, due to the fires and the anthropogenic impact, the plant community has changed, and these soils can be found under the plant community of Ass.Cocciferro–Carpinetumorientalis.Organomineral calcomelanosols (profiles 49 and 52) and terra rossa (profiles 50 and 51) have been excavated under this plant community. Regardless of the floristic characteristics, this community has the general specifics. This plant community is thermophilic and xerophilic with appropriate physiognomy and its own ecology. More important types of this community are the following: *Quercus pubescens; Fraxinus ornus; Cornus mas, Silene virdiflora; Cyclamen neapolitanum; Ranunculus psilostachys; Symphytum bulbosum; Carex halleriana; Tamus communis; Carpinus orientalis; Acer monospessulanum; Evonymus verrucosa; Aristella bromoides; Lithospermum purpureo coeruleum; Leontodon fasciculatum; Saxifraga buldifera; Geranium sanguieum; Iris sintenisii.* 

We have established several plant communities in the warm continental region. Terra rossa (profile 7) and calcomelanosols (profile 8) have been excavated under the plant community of *Ass. Querco Carpinetum orientalis*, while terra rossa (profiles 9 and 10) have been excavated under the plant community of *Ass Quercetum frainetto – cerris*.Profiles (29 and 46) calcocambisols and calcomelanosols (profile 45)have been excavated under the plant community of *Ass. Querco – Osttryetum carpinfoliae*, while only one organomineral calcomelanosols (profile 45) has been excavated under the plant community of *Ass. Querco – Osttryetum carpinfoliae*, while only one organomineral calcomelanosols (profile 45) has been excavated under the plant community of *Ass. Querco – Osttryetum carpinfoliae*, while only one organomineral calcomelanosols (profile 45) has been excavated under the plant community of *Ass.Querco – Osttryetum carpinfoliae*, while only one organomineral calcomelanosols (profile 45) has been excavated under the plant community of *Ass.Querco – Osttryetum carpinfoliae*, while only one organomineral calcomelanosols (profile 45) has been excavated under the plant community of *Ass.Querco – Carpinetum orientalis subass.Buxetosum*. The thermophilic types prevail in the plant communities, however, there are also moderately xerophilic types since the climate is characterized with a smaller degree or aridity. More important plant types in this community are the following: *Quercus frainetto; Pirus piraster; Malus florentina; Rosa gallica; Lathyrus inermis; Lychnis coronaria; Danaa cornubiensis; Potentilla micrantha; Helleborus odorus; Vicia barasitae; Quercus cerris; Sorbus domestica; Acer tataricum; Rubus canescens; Inula salicina; Trfolium pignatii; Asparagus tenuifolius; Stachys scadica; Luzula forsteri; Crocus moesiacus.* 

In our researches, we have also established multiple plant communities in the cold continental region. Profiles (26) calcomelanosols, and calcocambisols (27 and 28) have been excavated out under the plant community of *Ass. Onobrichi – Festucelum carpinfoliae*. Organomineral calcomelanosols (profiles 43 and 44) have been excavated under the plant community of Ass. Juniperuscommunisintermedia and one profile organogenic calcomelanosols (48) under the plant community of Ass.Querco – Ostryetumcarpinifoliae. This plant community is thermophilic and xerophilic, neutrophilic to weekly acidophilic. Since it occurs on steep, hardly accessible terrains, only small part of it is destroyed and converted into pastures and fern, and rarely into arable areas. More important types of this community are the following: *Quercus petraeae; Sorbus torminalis; Corylus avellana; Rosa arvensis; Galium pseudoaristatum; Scutellaria columnae; Luzula forsteri; Festuca hetrophylla; Fraxinus ornus; Acer campestre;* 

*Evonymus europaea; Cornus mas; Lathyrus niger; Lathyrus ventus; Brachypodium sylvaticum; Dronicum orientale.* 

The Piedmont continental- mountain region is the region in which Festuco heterophyllae – Fagetum Em extends. The beech range appears above the oak range at the altitude of 1100 to 1300 m. It covers large areas. This area contains the highest concentration of trees in our country. We have established multiple plant communities in this region. Profile (5) calcomelanosols, and one calcocambisols (profile 15), have been excavatedunderthe plant community of Ass. Querco-Osttryetum carpinfoliae and oneorgano mineral calcomelanosols (profile 25) has been excavated under the plant community of Ass. Ouobrichi-Festucelum cyllericae. Most of the calcomelanosols (profiles 39, 40, 41 and 42) and one calcocambisols (profile 38) have been excavated under the plant community of Ass. Juniperus communis intermedia. The main representatives of the floristic composition of this area are the following: Fagus sylvatica f. moesiaca; Carpinus betulus, Prunus avium; Festuca heterophylla; Galium pseudoaristatum; Lathyrus inermis; Primula acalis; Pulmonaria officinalis; Symphytum tuberosum; Lathyrus niger; Viola reichenbachiana; Corylus avellana; Quercus petraeae; Evonymus latifolia; Brachypodium sylvaticum; Potentilla micranatha; Stellaria holostea; Cyclamen neapolitanum; Scorphularia nodosa; Danaa cornubiensis; Aremonia agrimonioides; Melca uniflora.

In the Mountain continental region, calcomelanosols (profiles1, 2, 3, 33, 34, 35, 36 and 37), calcocambisols (30, 31 and 32) have been excavated under the plant community of *Ass.Calamintho* grandiflorae–Fagetum.One calcocambisols (profile 4) in this region has been excavated under the plant community of *Ass.Querno-Quercetumceries*. Organogeniccalcomelanosols (profile 13) has been excavated under the plant community of *Ass.Festucoheterophillae* – *Fagetum*, and one calcocambisols (profile 14) has been excavated under *Ass.Querco – Osttryetumcarpinfoliae*. The other calcomelanosols (profiles 23 and 24) in this climate region have been excavated under the plant community of *Ass. Onobrichi – Festucelum cyllerincae*.

Subalpine mountain The region has several plant communities. Organogeniccalcomelanosols (profile 6) has been excavated under the plant community of Ass. Onobrichi – Festucetum cyllenicae, and the other organogenical come lanosols have been excavated under the plant community of Ass. Seslerietum wettsteinii.Brownised calcomelanosols (profiles 11 and 12), and brownised calcomelanosols (profile 16) have been excavated under the plant community of Ass. Caluminthogradiflorae - Fagetum, and the other calcomelanosols (profiles 17, 18, 19 and 21), and calcocambisols (profiles 20 and 22) have been excavated under the plant community of Ass. Bruckenthalietum -Juniperetum.

The described vegetation has a great significance for the genesis of these soils. It is an important factor for the physical and chemical weathering (decarbonation) of the limestone, whereby the insoluble residue is released. The vegetation is a main source of organic matter which, combined with the conditions of the environment, contributes for the formation of various humus forms. The dense root mass of the grass vegetation is a significant factor in the formation of stable crumb structure. The vegetation, when combined with the other factors, as well as with the profile depth, contributes for evolution of the calcomelanosols into calcocambisols and terra rossa. The forest and grass plant communities are the main guardians against soil erosion.

# Time impact

The changes in the genesis and the evolution of soils, formed upon limestones and dolomites, depend both on the duration of processes (absolute age of the soil), and their intensity. The absolute age of the soil is expressed with time units, while the relative age is expressed with a number of evolution stadiums of the soils formed upon limestones and dolomites.

The time is a very important factor for the formation of these soils. Although they belong in the same class of soils as the A-C and A-R type of profiles, together with the soils formed on silicate rocks, yet when it comes to their formation, they need incomparably more time. Decarbonation and the release of silicate residue is a long-term process. We are going to compare some data related to the time needed for formation of a particular soil layer at those soils. According to Werner, cf. in (Маркоски, 2013), the formation of 1 cm soil layer of limestone with insoluble residue of 7% requires 1300-2000 years. If we calculate this, however, with a reserve of 0.14 to 0.76% residues for our limestones, we will come to a conclusion that the formation of 1 cm soil of calcomelanosols requires 15000 to 80000 years. These numbers are only approximate, however, they give us a frameork about the age of calcomelanosols, i.e. it can be said that calcomelanosols are absolutely old soils with stable residue. In this regard, but for the formation of the calcocambisols was also elaborated by (Ćirić, et al., 1981). According to this author, the formation of calcocambisols, 60 cm deep, from a limestone with 0.3% residue, requires 5.6 million years.

Most of the researchers agree that rubefication of the terra rossa is an old process that took place in the Tertiary, and also a process which still takes place in the regions with Mediterranean climate. According to these data, terra rossa would absolutely be our oldest soils, especially having in mind that the release of 1 cm layer of the limestone residue requires 8000 to 10000 years. In the regions with typical Mediterranean climate, the terra rossa are old soils, however, the contemporary bioclimatic conditions contribute for their preservation. Such contemporary bioclimatic conditions according to (Филиповски, 1997), also helped the preservation of the terra rossa soils in the Republic of Macedonia (monocyclic old soils). The deeper the autochthonous terra rossa is, the older it is.

# Human impact

None of the natural soil forming factors can cause so intense and rapid changes in the ecosystem and soil as the human factor. Moreover, its impact on the soil can be positive or negative.

In the daily struggle for survival in the hilly-mountainous areas where these soils extend, the human factor, unfortunately, causes many negative consequences with numerous activities. In order to provide new areas for producing food, meadows and pastures for the livestock, people cut out and burn the forests. By destroying the plant communities, the relationship between the soil and plant formation is deeply affected. The accumulation of organic matter and biogenic elements is reduced. At the places with more powerful solum, provided with minimum conditions for agricultural production on these soil types, the mineralization processes are intensified with the processing of the soil, and the humus content in the soil is reduced, the soil structure is worsened, which all together and with the improper tillage of the soil, contribute towards occurrence of soil erosion.

The former forest plant communities with their tree tops were protecting the soil from the direct destructive impact of the rain drops. This provides best protection against

soil erosion. Lawns also provide protection, but to a lesser extent. The erosion, especially at more inclined terrains, will affect part or the entire solum.

The impact of the human factor can be positive (planting new lawns on the erosive areas, forestation of those terrains, as well as constant protection and prevention of the above-mentioned negative anthropogenic impacts.

# Conclusion

Based on the conducted field researches on the natural conditions for formation of soils upon limestones and dolomites in the Republic of Macedonia, we can draw the following conclusions.

The soils are formed from the carbonate hornfels facies, and their composition also impacts the soil genesis. Calcomelanosols, calcocambisols and terra rossaare most frequently formed upon: massive limestone, dolomitic limestone, bituminous marbles and plate (flat) dolomites and calcite – dolomitic marble.

They are spread at the altitude of 211 m to 2000 m, i.e. out of 48 profiles, 6 can be found at the altitude of 1500 to 2000 m, 22 profiles of 1000 to 1500 m, 9 profiles can be found at the altitude of 600 to 1000 m and 4 profiles can be found at the altitude of 200 to 600 m. We have established that most of the profiles can be found at the altitude of 1500 to 2000 m.

Most of the profiles, a total of 16 or 31% extend in the Mountaincontinental region, where there are more plant communites (Ass. Calamintho grandiflorae -FagetumAss. Querno - Quercetum ceries, Ass. Festuco heterophillae – Fagetum, Ass. Querco – Osttryetum carpinfoliae Ass. Onobrichi – Festucelum cyllericae), 10 profiles or 19% in the Subalpine mountain region under plant communities (Ass. Onobrichi -Festucetum cyllenicae, Ass. Seslerietum wettsteinii Ass, Calumintho gradiflorae – Fagetum, Ass. Bruckenthalietum – Juniperetum), then,8 profiles or 15% in each of the warm continental region under plant communities (Ass. Querco Carpinetum orientalis, Ass Quercetum frainetto - cerris, Ass. Querco - Osttryetum carpinfoliae, Ass. Querco -Carpinetum orientalis subass. Buxetosum)and in the Piedmont continental mountain region (Ass. Querco-Osttryetum carpinfoliae Ass. Ouobrichi-Festucelum cyllericae Ass.Juniperus communis intermedia), 6 profiles or 12% in the cold continental region under plant communities (Ass. Ouobrichi - Festucelum carpinfoliae, Ass. Juniperus communis intermedia, Ass. Querco - Ostryetum carpinifoliae, and 4 profiles or 8% in the continental submediterranean region under the plant community Ass. Cocciferro -Carpinetum orientalis.

Although they belong to the same class of soils as A-C type of profile together with the soils formed on silicate rocks, yet, when it comes to their formation, they need incomparably more time.

The human factor has caused mainly negative changes at these soils. In order to provide new areas for producing food, meadows and pastures for the livestock, people cut and burn forests, and thus, deeply change the relationship between the soil and the plant communities. The accumulation of organic matter and biogenic elements is reduced.

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# CALCULATION OF SOIL EROSION INTENSITY IN THE SUTIVANSKA RIJEKA WATERSHED OF MONTENEGRO USING THE INTERO MODEL

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## Abstract

Soil erosion on agricultural land is a growing problem in the countries of the Balkan Peninsula being a very serious threat to soil quality. We used modelling of sediment yield and runoff in the Sutivanska Rijeka Watershed of Montenegro using the IntErO model. The studied area is characterized by the continental climate. The temperature coefficient of the region, T, is calculated on 0.99; the amount of torrential rain, hb, on 157 mm. The structure of the river basin, according to the rock permeability, is the following: f0, poor water permeability rocks, 91%; fpp, medium permeable rocks, 9%. The coefficient of the region's permeability, S1, is calculated on 0.97. The coefficient of the river basin planning, Xa, is calculated on 0.52. The coefficient of the vegetation cover, S2, is calculated on 0.74. Average river basin decline, Isr, is calculated 33.91% indicating that in the river basin prevail very steep slopes. The dominant erosion form in this area is surface erosion. Set of the geographical, climate, geology, pedology and land use inputs are included in the IntErO simulation model. The results shown that the net soil loss was calculated on 672 m<sup>3</sup> per year, specific 215 m<sup>3</sup>km<sup>-2</sup> per year. The results of this study are the determination of erosion processes; new information about the state of the runoff and soil erosion intensity, illustrating the possibility of modelling of sediment yield with such approach.

Key words: Erosion, Soil erosion assessment, watershed, Land use, IntErO model.

# Introduction

Soil erosion has long been recognized as an important physical factor in reducing soil productivity (Vaezi and Bahrami, 2014; Afshar et al., 2010; Fenton et al., 2005). It is the most widespread factor of land degradation (Erenstein, 1999).

Water erosion affects agricultural productivity in a number of ways, either directly or indirectly. It diminishes soil productivity by reducing topsoil depth (Lal et al., 2000); availability of water (Bossio et al., 2010); nutrients (Li et al., 2013) and organic matter (Fenton et al., 2005), as well as by restricting rooting depth (Stoffel, et al., 2013).

Soil erosion by water is a major environmental problem in many areas of Montenegro, threatening the sustainability of agricultural lands. Water erosion has affected 95% of the total territory of Montenegro (Kostadinov et al., 2006). Alluvial accumulation characterizes the remaining area, where the deposition of sediments is also affecting agricultural land. Erosion caused by water is dominant in terrain with high slopes due to complex physical and geographical conditions paired with reckless logging (Spalevic, 2013a).

Various researchers all over the world applied numerous approaches so far with the intention of gaining a better understanding of the erosion processes. Boardman and Poesen (2006) summarized methodologies and research. Qualitative and quantitative descriptions of hydric soil erosion have been directly performed through field observations (Bagarello et al., 2011; Della Seta et al., 2009; Porto et al., 2001; Sorriso-Valvo et al., 1995) and laboratory experiments (Leone and Sommer, 2000; Piccolo et al., 1997; Torri and Poesen, 1992). Information on sediment yield and runoff is needed for erosion risk assessment and models have proved to be good tools to understand these processes, quantifying sediment yield and runoff. For this research we used a local IntErO model (Spalevic, 2011) based on Gavrliovic method (1972).

IntErO model (Spalevic, 2011) was chosen for this study for the following reasons:

- The model has been widely used in Polimlje (2011-2015) but also wider in the South Eastern European Region;
- The model was earlier validated for simulating the erosion processes over the Polimlje Region in Montenegro (Spalevic, 2011).

With this study we received a new detailed report about the state of the runoff and sediment yield and in this format may be used further in watershed management sector of Montenegro, illustrating the possibility of modelling of sediment yield with such approach all over the country.

# Materials and methods

The study was conducted in the Sutivanska Rijeka Watershed, located in the Valley of BijeloPolje, north part of Montenegro (Figure 1).

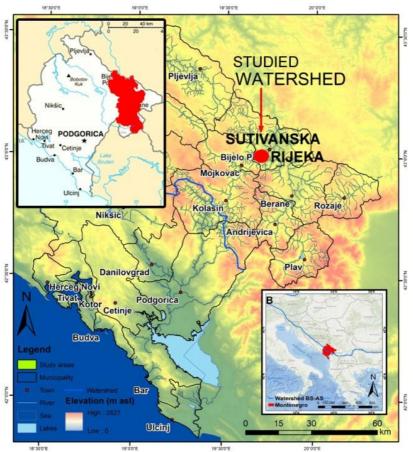


Figure 1. Study area of the Sutivanska Rijeka drainage basin



Figure 2. Details from the studied Sutivanska Rijeka drainage basin, upper part, Kurmarija (May, 2014)



Figure 3. Details from the studied Sutivanska Rijeka drainage basin, close to the inflow to the river Lim

Fieldwork was undertaken to collect detailed information on the intensity and forms of soil erosion, the status of plant cover, the type of land use, and the measures in place to reduce or alleviate the erosion processes. Morphometric methods were used to determine the slope, the specific lengths, the exposition and form of the slopes, the depth of the erosion base; the density of erosion rills.

The Sutivanska Rijeka is a one of the smallest watersheds of the Polimlje region on the territory of Montenegro and is with the surface area of 3.1 km<sup>2</sup>. The average river basin altitude is 711 m (maximum 983 m and minimum 552 m); Average elevation difference of the river basin is 159 m and Average river basin decline is 33.91%, what indicates that in the river basin prevail very steep slopes. The analysis of the geological structure and soil characteristics of the area were based on the research of the National Geological Survey led by Zivaljevic (1989) and Djuretic & Fustic (2000), who analysed all geological formations and soils of Montenegro including the Polimlje region. Climatological data were received from the Institute of Hydrometeorology and Seismology of Montenegro. We also used a Satellite images and data from the Institute of Forestry of Montenegro to prepare land use maps of the study area.

For obtaining data on forecasts of sediment yield and peak discharge from the basin we used the IntErO model (Spalevic, 2011) and the Erosion Potential Method – EPM (Gavrilovic, 1972). The model is based on the following equation:

$$W_{vr} = T \cdot H_{vr} \cdot \pi \cdot \sqrt{Z^3} \cdot F$$

where  $W_{yr}$  is the annual erosion in  $m^3yr^{-1}$ ; T, the temperature coeff.;  $H_{yr}$ , the average yearly precipitation in mm; Z, the erosion coefficient.

The erosion coefficient, Z, was calculated as follows:

$$Z = Y \cdot X \cdot (\phi + \sqrt{I})$$

where, Y is Soil erodibility coefficient; X is Soil protection coefficient;  $\phi$  is Erosion development coefficient (tables for Y, X and  $\phi$  coefficients available at Gavrilovic, 1972). F is the watershed area in km<sup>2</sup>.

The actual sediment yield was calculated as follows:

$$G_{yr} = W_{yr} \cdot R_u$$

where,  $G_{yr}$  is the sediment yield in  $m^3yr^{-1}$ ;  $W_{yr}$  is the total annual erosion in  $m^3yr^{-1}$ ;  $R_u$  is sediment delivery ratio.

The actual sediment yield was calculated as follows:

$$R_{u} = \frac{\left(\sqrt{O \cdot D}\right)}{0.2 \cdot \left(L + 10\right)}$$

where, O is perimeter of the watershed in km; D is the average difference of elevation of the watershed in km; L is length of the catchment in km.

The EPM method and the IntErO model is currently in use in the various countries by the researchers from the following countries (by alphabetical order): Albania, Austria, Belgium, Bosnia, Brazil, Bulgaria, Croatia, Czech Republic, Iran, Italy, Macedonia, Montenegro, Morocco, Serbia, Saudi Arabia (Al-Turkiet al., 2015; Barovic et al., 2015; Barovic and Spalevic, 2015; Behzadfar et al., 2015; Behzadfar et al., 2014a; Behzadfar et al., 2014b; Gazdic et al, 2015; Ristic et al., 2001; Spalevic et al., 2015a; Spalevic et al., 2015b; Spalevic et al., 2015c; Spalevic et al., 2015d; Spalevic et al., 2015e; Spalevic et al., 2015f; Spalevic et al., 2014a; Spalevic et al., 2014b; Spalevic et al., 2014c; Spalevic et al., 2014d; Spalevic et al., 2013b; Spalevic et al., 2013c; Spalevic et al., 2013d; Tazioli et al., 2015; Tazioli, 2009; Kostadinov et al, 2014; Curovic et al., 1999; Vujacic&Spalevic, 2015).

#### **Results and discussion**

*Climatic characteristics.* Analysing temperature and precipitation (1948-2014) we concluded that the studied area is characterised by rainy autumns and springs; and cold winters. The absolute maximum air temperature is 39.2 °C; a minimum of -27.6 °C. The average annual air temperature, t0, is 8.9°C. The average annual precipitation is 873 mm. Temperature coefficient for the region is calculated at 0.99. The torrential rain, hb, is calculated at 157 mm (Spalevic, 2015b).

*The geological structure and soils of the area.* Wider area consists of various types of sediment, magmatic and metamorphic rocks generated in the period from Palaeozoic to Quaternary. The broader area to which the subject river basin belongs consists of clastic and subordinate carbonate rocks from the Palaeozoic, Triassic clastites, volcanites, tuffs, limestone and dolomites, Jurassic clastic rocks with spilite and diabasic effusions and metamorphic rocks and Quaternary, mainly alluvial and deluvial sediments (Spalevic, 2015b).

We extracted specific data on the region permeability from the Geological map of Montenegro. The coefficient of the region's permeability, S1, is calculated on 0.97. A part of the river basin consisting of poor permeability rocks, fo, is calculated on 91%; of medium, fp, on 9%. Using the data of the Map of Soils of Montenegro, but also the own

results of the research, we came to the conclusion that the most common soil types in the studied river basin are Dystric Cambisols, (90%), Eutric Cambisols (5%) and Fluvisols and Colluvial Fluvisols (5%).

*Land use.* Soil erosion assessment is taking into consideration high dependencies and interactions between hydrogeomorphic processes and vegetation. Stoffel and Wilford (2012) highlighted the role of vegetation in the initiation of hydrogeomorphic processes and its impact on stream morphology.

According to the data of land use and vegetation cover, received from the Institute of Forestry of Montenegro (IoFoM), Statistical Office of Montenegro (MONSTAT), Google Maps and our own research, the Sutivanska drainage basin is mainly covered by forests, covering the area of 45% of the studied river basin. The area under grass is with 41% and arable land is 14%.

Degraded beech forests (*Fagetummontanum*) are dominating in the upper parts of the basin. On the southern exposures base forest community is *Quercetumpetraeaecerridis* Lak (forest of Sessile oak and Turkish oak). Narrow belt near river in the lower part of watershed is covered with hydrophilic forest (*Saliceteaherbacea, Alneteaglutinosae*).

Beech forests of the river basin are low forests with coppice origin as result of pretty intensive harvesting of the firewood in the past, especially anything in the area near the settlements and roads. Forest of Sessile and Turkish oak are dominated by *Quercuscerris* followed by numerous of thermophilous tree species such as: *Quercuspetreae*, *Carpinusbetulus*, *Crataegus* spp. The shrub layer is mainly composed of: *Cornus mass*, *Prunusspinosa*, *Corylusavellana*, *Rosacanina* etc.

*Soil erosion characteristics.* Water erosion is the most important erosion type in the studied river basin. It is primarily caused by precipitation, runoff and fluvial erosion in streams. The dominant erosion form in the study area is from surface runoff, but we recorded some gullies and rills also. We used the software IntErO for calculation of the soil erosion intensity and the peak discharge for the Sutivanska River Basin. Final results of the combinations of these detachment and transport processes are presented in the Table 1.

Table 1. Fait of the Intero report for the Sutivaliska River Dasin	1		
Input data	_		
River basin area	F	3.11	km <sup>2</sup>
The length of the watershed	0	9.05	km
Natural length of the main watercourse	Lv	3.05	km
The shortest distance between the fountainhead and mouth	Lm	2.81	km
Main watercourse with tributaries of I and II class	ΣL	4.69	km
River basin length measured by a series of parallel lines	Lb	3.8	km
The area of the bigger river basin part	Fv	2.01	km²
The area of the smaller river basin part	Fm	1.1	km²
Altitude of the first contour line	h0	600	m
Equidistance	Δh	50	m
The lowest river basin elevation	Hmin	552	m
The highest river basin elevation	Hmax	983	m
Very permeable products from rocks	fp	0	
Medium permeable rocks	fpp	0.09	
Poor water permeability rocks	fo	0.91	
A part of the river basin under forests	fš	0.45	
Grass, meadows, pastures and orchards	ft	0.41	
Bare land, plough-land and ground without grass vegetation	fg	0.14	
The volume of the torrent rain	hb	157.6	mm
Average annual air temperature	tO	8.9	°C
Average annual precipitation	Hgod	893.3	mm
Types of soil products and related types	Y	1.3	
River basin planning, coefficient of the river basin planning	Xa	0.52	
Numeral equivalents of visible erosion process	φ	0.27	
Results:			
Coefficient of the river basin form	А	0.58	
Coefficient of the watershed development	m	0.49	
Average river basin width	В	0.82	km
(A)symmetry of the river basin	а	0.58	
Density of the river network of the basin	G	1.51	
Coefficient of the river basin tortuousness	Κ	1.09	
Average river basin altitude	Hsr	711.17	m
Average elevation difference of the river basin	D	159.17	m
Average river basin decline	Isr	33.91	%
The height of the local erosion base of the river basin	Hleb	431	m
Coefficient of the erosion energy of the river basin's relief	Er	103.27	
Coefficient of the region's permeability	<u>S1</u>	0.97	
Coefficient of the vegetation cover	S1 S2	0.74	
Analytical presentation of the water retention in inflow	W	1.7349	m
Energetic potential of water flow during torrent rains	2gDF^1/2	98.63	m km s
Maximal outflow from the river basin	Qmax	71.1	$m^{3} s^{-1}$
Temperature coefficient of the region	T	0.99	
Coefficient of the river basin erosion	Z	0.561	
Production of erosion material in the river basin	Wgod	3658	m <sup>3</sup> yr <sup>-1</sup>
Coefficient of the deposit retention	Ru	0.184	
Real soil losses	Ggod	672.72	m <sup>3</sup> yr <sup>-1</sup>
Real soil losses per km <sup>2</sup>	Ggod/km <sup>2</sup>	215.97	$m^{3}km^{-2}yr^{-1}$
	Ugou/Kill	213.71	in Kin yi

Table 1. Part of the IntErO report for the Sutivanska River basin	

The coefficient of the river basin form, A, is calculated on 0.58. Coefficient of the watershed development, m, is 0.49 and average river basin width, B, is 0.82 km. (A)symmetry coefficient indicates that there is a possibility for large flood waves to appear in the river basin. The value of G coefficient of 1.51 indicates there is high density of the hydrographic network. The height of the local erosion base of the river basin, Hleb, is 431 m. Coefficient of the erosion energy of the river basin's relief, Er, is calculated on 103. The value of 33.91% indicates that in the river basin prevail very steep slopes.

The value of Z coefficient of 0.561 indicates that the river basin belongs to III destruction category. The strength of the erosion process is medium, and according to the erosion type, it is mixed erosion.

Using the IntErO model, the soil losses from the Sutivanska River basin were calculated on  $672 \text{ m}^3 \text{yr}^{-1}$ , specific:  $215 \text{m}^3 \text{km}^{-2} \text{yr}^{-1}$ .

# Conclusion

We studied the soil erosion processes within the Sutivanska Rijeka river basin using modelling techniques implemented in a computer-graphics environment.

The most characteristic outputs acknowledged during the field visit, laboratory analysis, studying the available literature, and finally applying the IntErO model are the following:

- In the river basin prevail very steep slopes;
- There is a possibility for large flood waves to appear in the river basin;
- There is high density of the hydrographic network;
- The dominant erosion form in this area is surface erosion;
- The river basin belongs to III destruction category (of five). The strength of the erosion process is medium, and according to the erosion type, it is mixed erosion.
- Using the IntErO model, the soil losses from the Sutivanska River basin were calculated on 672 m<sup>3</sup>yr<sup>-1</sup>, specific: 215m<sup>3</sup>km<sup>-2</sup>yr<sup>-1</sup>.

The results of this study are the determination of erosion processes; new information about the state of the runoff and soil erosion intensity, illustrating the possibility of modelling of sediment yield with such approach.

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# AN ASSESSMENT OF SURFACE WETTED PERCENTAGE AND SOIL MOISTURE FOR DRIP-IRRIGATED HORTICULTURAL CROPS UNDER FARMER CONDITIONS

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# Abstract

Drip irrigation systems normally wet only a portion of the soil area and soil volume. The wetted area in the soil for drip irrigation systems is, thus, one of the most important parameters in the design and management of the systems. This study was carried out to assess the surface wetted percentages and soil moisture for drip-irrigated horticultural crops in Eskişehir and Sakarya regions in Western of Turkey between 2008 and 2009. The drip irrigation systems of 13 different farms were selected and evaluated. The percentages of the wetted area are varied from 9.5 to 53.3 % depending on cultivated crop and lateral distance. Depth (z) and radius or horizontal width (d) of wetted area were 0.30-0.43 m and 0.25-0.38 m, respectively. The main reason of lower surface wetted percentage could be attributed to the poor system design and insufficient irrigation scheduling and management. The soil moisture content before the irrigation cycle has an undulated differences and it in the most of the fileds tested was near to the wilting point. There were no, because, any appropriate measurement based on soil moisture content or any other criteria. All these might decarease weter use efficiency for drip irrigation. In addition, wetted area and its effects on appropriate irrigation water management for drip irrigation are discussed.

Key words: drip irrigation, wetted area, soil moisture, horticultural crops

# Introduction

Drip irrigation has many advantages compared to other irrigation methods. These could be high water use efficiency, high uniformity of water distribution, elimination of surface runoff, flexibility in fertilization and fertigation, prevention of weed growth and plant disease. On the other hand, drip irrigation is one of the best techniques to use in applying water to vegetables and orchards. Recently, use of drip irrigation for vegetables, mainly tomatoes, in the study region has rapidly increased because of both increasing in yield by using drip irrigation and subsidizing of drip irrigation systems by government (Çetin and Uygan, 2008). One of the most important advantages of drip irrigation is that a grower does not need to irrigate the whole surface of the soil. Unlike surface and sprinkler

irrigation, drip irrigation only wets part of the soil root zone. This may be as, low as 30% of the volume of soil wetted by the other methods (FAO, 2014). Drip irrigation systems normally wet, thus, only a portion of the soil area and soil volume. No single correct or proper minimum value for percentage of wetted area (Pw) has been established (Keller and Bliesner, 1990; Cetin et al., 2008). A reasonable objective when plotting a design for widely spaced crops is to consider that as much as two-thirds of the potential horizontal cross-sectional area of the root system is 33% < Pw < 67%. However, when growing crops with rows and drip lines spaced <1.8m apart, Pw can approach 100% (Keller and Bliesner, 1990). Keller and Bliesner (1990) also presented and demonstrated equations for computing the wetted area as a percentage of the total crop area for a range of crop geometry and lateral layouts based on the dimensions of the spacing between emitters and lines. Since the percentage of wetted area was introduced by Keller and Karmeli (1974), it has been widely used in microirrigation systems design. The wetted pattern is one of the main design parameters, which directly relates to the initial installation cost. Excessive wetted percentage would increase the cost of drip irrigation system, lower the efficiency of irrigation, and excessive irrigation water leaches below the root zone (El-Hendawy et al., 2008). On the contrary, a small wetted percentage causes plant water stress, reducing crop yield. Every component of crop has a different sensitive level to water stress in different growing seasons, especially in a dry climate (Plaut et al., 1996).

The amount of irrigation water to apply to the root-zone by drip irrigation is dependent on the criteria chosen, for example the percentage of wetted area or canopy cover (Cetin et al., 2008). With drip irrigation, scheduling should be evapotranspirationbased; put simply, the crop evapotranspiration from the previous day is estimated and replaced the following day. A soil moisture depletion approach is difficult since the estimate of the total soil moisture available relies on knowing the soil wetting pattern, and this is difficult to accurately quantify under drip (Cristen et al., 2006). The optimal range of moisture in the soil can be maintained at all times and managed more easily, because water is applied in precise quantities on a precise schedule, according to the crop requirements. This promotes water saving, as well as enhances growth and production. In addition, the selective wetting prevents evaporation of water from areas outside the wetted zone (Anonymous, 2014). A greater surface of soil wetted throughout irrigation season did not result in more yield and IWUE. The method for calculation of the amount of irrigation water to be applied, percentage of canopy cover, was therefore more significant and appropriate for yield and IWUE (Cetin and Uygan, 2008). The percentage of wetted area can be used as a multiplier to reduce the total water applied under drip irrigation. The use of an appropriate percentage of wetted area or another scheduling approach could have considerable impact in terms of both the system design and efficient irrigation management to improve water use efficiency for drip irrigation (Cetin et al., 2008).

The objective of this study was to evaluate soil moisture, surface wetted percentage and the wetted pattern in some farms used drip irrigation systems in Western of Turkey.

# Materials and methods

*Location:* This study was carried out in the selected some farms in Sakarya and Eskişehir Provinces in Western of Turkey. The vegetable crops are almost grown in the study area and the surface drip irrigation systems are used to irrigate these crops. Eskişehir and Sakarya are located on altitude of 780 and 200 m above mean see level, respectively.

# Climate charactersitics

*Eskişehir:* The climate of area in Eskişehir is terrestrial climate and semi-arid type with an average annual rainfall of 343 mm. and average annual pan evaporation of 975 mm. July is the hottest month with mean monthly pan evaporation of 195 mm and mean monthly temperature varying between -0.2 °C to 21.4 °C.

*Sakarya:* The climate of area in Sakarya is semi-mild type with an average annual rainfall of 841 mm. July is the hottest month with mean monthly temperatures of 23.3 mm and mean monthly temperature varying between 6.0  $^{\circ}$ C to 23.3  $^{\circ}$ C.

*Soil properties:* Particle distribution and texture class are changed from sandy loam through clay. The dry bulk densities of soil were ranged from 1.20 to 1.57 g cm<sup>-3</sup>. The pH, electrical conductivity (salinity) (EC) and the content of calcium carbonate (CC) were 6.05-8.50, 0.75-6.86 dS/m and 0.07-21.1 %, respectively. However, soil pH ranged, in general, from 7.30 to 7.9. Some fields has extremly lower and higher pH value such as the field 12: 6.05 and the field 11: 8.50. Similarly, EC was 6.86 dS/m in only field 8. The content of CC in the soils were 0.07-0.08 % in field 8 and 12. The CC in the other fields ranged from 7.95 to 21.06 %.

*Dripper discharge measurements:* The manifolds were selected to measure dripper discharge, including the one closest, one middle to and most distant from the pump. The discharged flow rates of the five emitters at every control points were determined by measuring the volumes of outflow from the emitters in 15-20 minutes, using a graduated cylinder.

*Soil moisture measurements :* The soil water content was measured by gravimetric moisture before irrigation, which was determined by calculating the proportion of water loss relative to dry soil weight after oven-drying the soil samples.

*Moisture distribution pattern:* The moisture distribution pattern in the soil profile was determined by taking soil samples at different depths on the one side of the emission point after 24 hours of application. The wetting front in horizontal (d) and vertical (z) directions was measured to determine the moisture distribution pattern in the soil profile (Fig. 1). Samples were collected at 0-20, 20-40 and 40-60 cm depths for 0, 15, 30, and 45cm distances on the side of the emission point. The undisturbed soil samples were collected using a core sampler and placed in an oven for 24 hours at 104 <sup>o</sup>C. The oven dried samples were weighed and their volumes determined, finally the moisture content was determined (Mirjat et al., 2010). The percentage of wetted area was calculated as the radius of the wetted area in the field (Fig. 1, (d)) was divided to the lateral spacing (Keller and Bliesner, 1990; Yıldırım, 2003).

In this study, all the tests and collecting of the data were carried out using some field tests and some questionaires. For this, the farms used drip irrigation systems of 6 in Eskişehir and the farms of 7 in Sakarya were selected.

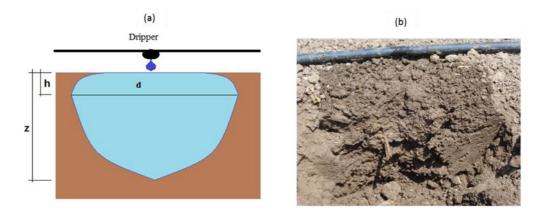


Fig. 1. Schematic pattern of wetted area measured in the field (a), real wetted area in the field (b)

#### **Results and discussion**

#### Specifications of soil and the drip irrigation systems

The texture classes of the soils in the study area were varied from SL to C. The soils of the fields in the numbers of 9, 10 and 12 were clay texture. Infiltration rate ranged from 4 to 25 mm/h. Considering soil properties such as soil texture and infiltration rate, it is understood that the farmers have not designed and constructed the drip irrigation systems according to the these specifiactions of the soils. Considering drip system design, dripper distances were usually 0.20 and/or 0.25 m. The fields in numbers of 3, 8 and 10 had dripper distance of 0.40 m. The dripper discharges were varied from 2.0 to 4.0 L/h. The measured dripper discharges are given in Table 1. Considering these values, there were significantly deviations compared to the planned dripper discharges except those of the fields numbered with 9, 10 and 11 (Fig. 2). This shows that there is a poor design of the drip irrigation systems. Thus, this might have been brought tremendous changes in discharge and the uniformity of distribution. In addition, these results shows that there has been some clogging problems. Because, the farmers has not commonly used the media filters and some chemical such as acids and chlorine to prevent precipitation and clogging. Bozkurt and Ozekici (2006) stated also that the fertlizers which included both calcium and sulfates resulted in lower system performance. They recommended acid treatment and flushing management. Thus, many factors such as physical, chemical and biological, pressure and water temperature and construction changes, have affecting on the dropper discharge and uniformity of water distribution (Samani and Nasab, 2010). Additionaly, it should be taken into account the poor quality of the systems tested area.

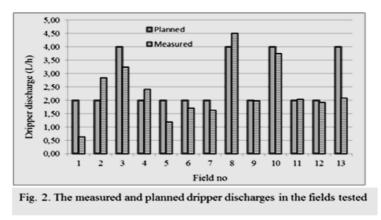
Table 1. Some properties of son and drip infiguron systems in the test area							
Farm	Infiltration	Dripper	Dripper	Dripper	Irrigation	Irrigation	Bulk
no	rate	distance	discharge	discharge	interval	duration	density
	(mm/h)	(m)	(L/h)	(measured)	(days)	(hours)	$(g/cm^3)$
				(L/h)			
1	10	0.20	2.00	0.65	7	6	1.31
2	9	0.20	2.00	2.85	2	7	1.32
3	5	0.40	4.00	3.25	4	6	1.35
4	5	0.20	2.00	2.43	3	4	1.33
5	25	0.20	2.00	1.20	7	6	1.55
6	25	0.20	2.00	1.72	3	8	1.57
7	8	0.20	2.00	1.64	7	7	1.33
8	15	0.20	4.00	4.51	4	7	1.42
9	4	0.20	2.00	1.99	2	7	1.25
10	4	0.20	4.00	3.76	5	4	1.20
11	5	0.25	2.00	2.05	2	6	1.35
12	4	0.25	2.00	1.94	7	7	1.28
13	6	0.20	4.00	2.10	10	5	1.34

Table 1. Some properties of soil and drip irrigation systems in the test area

In another data in the tested area, all of the drippeers used in the drip irrigation systems in the test area were non pressure compensating emitters. These can yield a variety of flow rates due to the variation of pressure in the laterals, usually the flow rate increases at a certain rate with the increase of pressure and decreases according to the flow pressure head characteristics of emitters (Dutta, 2008). Thus, emitters are the most important parts of a trickle irrigation system. Poor irrigation-water-application uniformity can be a cause of low crop-yields.

#### Wetted area and soil moisture

Surface wetted percentage measured in the fields are given in Table 2. The percentages of the wetted area are varied from 9.5 to 53.3 % depending on cultivated crop and lateral distance (Fig. 3). These values in the fields of 7, 8 and 12 numbers (9.5 %, 10.7 % and 12 %, respectively) were quite lower compared to appropriate values (>33 %, <67 %) (Keller and Bliesner, 1990). Surface wetted percentage in the fields of 2, 4, 9 and 13 numbers were also lower.



Farm	Cultivated	Cultivated	Planted	The interval	Radius of	Depth of	Wetted
No	crop	area	distance	distance of	wetted	wetted	area
		(ha)	(mxm)	laterals	area (d)	area (z)	(%)
				(m)	(m)	(m)	
1	Beans	0.20	0.85x0.20	0.85	0.32	0.33	37.6
2	Tomatoes	0.035	0.50x0.40	1.00	0.28	0.31	28.0
3	Tomatoes	0.80	0.80x0.40	0.80	0.38	0.38	47.5
4	Cucumber	1.00	1.50x0.40	1.50	0.31	0.30	20.7
5	Pepper	0.15	0.60x0.40	0.60	0.32	0.31	53.3
6	Tomatoes	0.035	1.10x0.30	0.50	0.35	0.32	70.0
7	Water melon	0.054	3.0x1.0	3.00	0.32	0.30	10.7
8	Kivi	0.60	4.0x4.0	4.00	0.38	0.43	9.5
9	Tomatoes	0.03	1.0x0.20	1.00	0.25	0.30	25.0
10	Rose	0.20	1.0x0.20	1.00	0.38	0.37	38.0
11	Tomatoes	0.43	0.60x0.50	0.60	0.32	0.32	53.3
12	Water melon	0.25	2.50x2.0	2.50	0.30	0.30	12.0
13	Ornamental plants	0.35	0.50x0.30	1.50	0.37	0.42	24.7

Table 2. Some data on the test areas and horizontal and vertical distances of wetted area

The vertical wetted area (z) in the fields were, in general, a little bit more than horizontal wetted area (d). Accordingly, depth (z) and radius or horizontal width (d) of wetted area were 0.30-0.43 m and 0.25-0.38 m, respectively (Table 2). Because the soil texture in the fields tested were, in general, middle texture class. The wetted area in a clay soil is much wider and deeper than with the sandy soil (Cristen et al., 2006). Thus, wetting pattern characteristics depends on emitter spacing, run time and emitter discharge and amount of irrigation water applied (Souza and Folegatti, 2010). In addition, the water advance in the soil from a point source, emitter and expansion of wet bulb depends on the other some factors such as soil texture, the period of performence and bulk density of soil (Neshat and Nasiri, 2012). On the other hand, Zhang et al. (2012) showed there was a positive linear correlation between saturated zone radius and application rate. Considering surface wetted percentages, it could be taken into account as an insufficient values for an appropriate However, the main reason could be poor system design and irrigation scheduling. insufficient applying amount of water. Because, characterization of the wetted soil volume under field conditions offer a practical means for gathering the relevant information required for system design (Keller and Bliesner, 1990). The another reason of these lower values was that the lateral distance was far from each other because of horticultural crops (fruit trees and water melon). Information about the horizontal and vertical distances by which water spreads in soils under a point source is essential for the design of cost effective and efficient drip irrigation systems. The size of the wetting pattern is influenced by many factors, including soil physical properties, soil initial conditions as well as emitter discharge rate, irrigation management, crop root characteristics and evapotranspiration (Naglic et al., 2012). Some interviews and questinaire with the farmers were carried out during the field works. Some data regarding to irrigation practices were collected. These were irrigation interval and irrigation duration for each tested system. Irrigation intervals ranged from 2 to 7 days, irrigation duration varied between 4 and 7 hours (Table 1). All these applications have been practiced by means of the farmers' experiencies. There were no any scientific approach and/or an appropriate measurement based on soil moisture

content or any other approachs. Because, the system design and management affect also soil wetting pattern (Cristen et al., 2006). In addition, the effect of irrigation scheduling techniques on the water distribution pattern could vary at different soil depths (Al-Ghobari and El Marazky, 2012).

Eventually, the wetted area in the soil for drip irrigation systems is one of the most important parameters in the designing and management of the systems. Because it affects the use of irrigation water with plant nutrition (Zhao *et al.*, 2012).

Soil moisture content of soils in the fields tested are given Fig. 4. Soil moisture content in the most of the fields are near the wilting point. It shows that the most of the drip irrigation system tested have the poor system design and insufficient irrigation scheduling and management. In addition, an appropriate amount of irrigation water could not be applied during the irrigation.

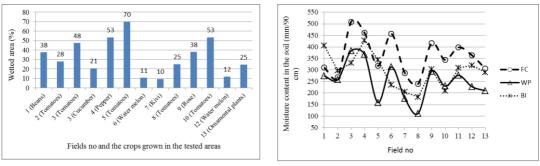


Fig. 3. The measured wetted area vs. fields and the crops grown

Fig. 4. Soil moisture content of soils in the fields tested

# Conclusions

The percentages of the wetted area in the drip irrigation systems are varied from 9.5 to 53.3 % depending on cultivated crop and lateral distance. Depth (z) and radius or horizontal width (d) of wetted area were 0.30-0.43 m and 0.25-0.38 m, respectively. The main reason of lower surface wetted percentage could be attributed to the poor system design and insufficient irrigation scheduling and management. The soil moisture content before the irrigation cycle has an undulated differences and it in the most of the fileds tested was near to the wilting point. There were no, because, any appropriate measurement based on soil moisture content or any other criteria. As a result, too large or too small surface wetted percentage might be caused yield reduction. Water distribution under drip irrigation systems is a three dimensional problem. It mainly depends on soil physical and hydraulic properties, crop evapotranspiration, drip irrigation system management and design parameters, such as emitter spacing, discharge rates, amount of water applied per irrigation. All these factors have to be considered when designing drip irrigation systems.

# Aknownledgement

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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# BIOCLIMATIC COEFFICIENT OF PLASTIC HOUSE GROWN PEPPER AS INFLUENCED BY IRRIGATION AND FERTILIZATION TECHNIQUES

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## Abstract

The field experiment was conducted with pepper (Capsicum annum L. var. Bela dolga) grown in experimental plastic house in Skopje region during the period of May to September in 2005, 2006 and 2007. The main goal of this investigation was to compare drip fertigation with furrow irrigation and conventional fertilization regarding bioclimatic coefficient of pruned pepper ("V" system). Four experimental treatments were applied in this study. Three of the treatments were drip fertigated (noted as KK1, KK2, KK3), while the last one was furrow irrigated with conventional application of fertilizer (control treatment  $Ø_B$ ). Water balance method was used for direct estimation of monthly and seasonally potential evapotranspiration (ETP) of pepper. The average seasonally ETP ranged from 4841 in treatment KK2 to 5881 m<sup>3</sup>/ha in treatment  $Ø_{\rm B}$  and it was associated with irrigation and fertilization techniques. Also, we concluded that drip fertigation frequency in every 2 or 4 days (KK1 and KK2, respectively) showed 4.5-5.5% lower evapotranspiration in comparison with KK3 (drip fertigation scheduled by tensiometers). Furthermore, the average of three-year results shows that drip fertigation treatments give lower monthly and seasonally bioclimatic coefficient as compared to the treatment of furrow irrigation and conventional fertilizer application. Generally, the lowest average seasonally bioclimatic coefficient of pepper from 1.61 has been observed in the treatment KK2, followed by treatment KK1 with 1.62 and KK3 with 1.69, and finally - the control treatment  $Ø_B$  with the highest coefficient 1.94. Also, the drip fertigation frequency pointed to differences in the bioclimatic coefficient, i.e. the treatment KK3 performed an average seasonally bioclimatic coefficient higher by 5% in comparison with KK2, i.e. by 4.3% in comparison with KK1.

**Key words:** drip fertigation, furrow irrigation, conventional application of fertilizers, pruned pepper, ETP, bioclimatic coefficient

# Introduction

Pepper is a very sensitive crop in terms of water supply. Many authors have elaborated the topic as to how much attention should be paid to irrigation practice (Doorenbos and Kassam, 1986; Jankulovski, 1997; Antony and Singandhupe, 2004; Sezen et al., 2006). The pepper water requirements during the vegetation period are quite big compared to other crops, which is a result of the poorly developed root system (Iljovski and Cukaliev, 1994; Bosland and Votava, 2000; Lazić et al., 2001) and large transpiring leaf surface (Alvino et al., 1994; Delfine et al., 2002). Water deficit during the period between flowering and fruit development reduces the final productivity of pepper (Bosland and Votava, 2000; Jaimez et al., 2000). Therefore, in order to obtain high yields, pepper needs to be provided with the adequate quantities of water throughout the vegetation period (Doorenbos and Kassam, 1986). According to Tanaskovik (2009), the proper irrigation regime is important for obtaining high yields, since the pepper is sensitive to both excessive and insufficient water.

It is generally known that one of the most important issues for the irrigation practice is proper irrigation scheduling of the agricultural crops. Depending on the parameters used as a basis for determination of the irrigation application rate and time of application, all methods may be grouped in three basic groups as follows: soil-based (soil moisture monitoring), plant-based (plant condition monitoring) and climate-based (climate characteristics monitoring) methods (Tanaskovik and Cukaliev, 2013). None of these methods sufficient on its own is for accurate determination of the irrigation application rate and time of application. The crops, the biological characteristics, the anticipated yields and the water-physical properties of soil, as well as the climate parameters have always been taken into account. It means that the ET (evapotranspiration), as well as the irrigation application rate and time of application depend on many factors, point out Tanaskovik et al. (2013). On the other hand, there is a need in the irrigation practice for a method which meets the condition requirements and is, at the same time, guick, practical and simple. Although soil moisture monitoring is the best and most accurate method when compared to the other methods, yet, the other two methods are successfully used. An example for this is Penman-Monteith method, modified by FAO, and applied by CROPWAT computer program and commonly recommended in vegetable production (Orta, 1997; Tanaskovik et al., 2013; Tanaskovikj et al., 2014). Namely, this method is an extensively applied tool for determination of water consumption based on evapotranspiration (ET) in a given structure of crops and in a particular region, and at the same time; this program can produce a complete irrigation application schedule. One of the commonly used methods for determination of the irrigation application rate and time of application in our country is the bioclimatic coefficient method (hydrophythothermic coefficient method). Until present day, bioclimatic coefficients for the following crops have been determined: hop (Iljovski, 1982), apricot (Cukaliev and Iljovski, 1994), sugar beet in Skopje region (Cukaliev, 1996), sugar beet in Pelagonia region (Jankuloski, 2000), tomato in Skopje region (Tanaskovic, 2005), a group of crops in Pelagonia region (Tanaskovik et al., 2013).

The essence of this method is that the water consumption for evapotranspiration in a given period is placed in a ratio with the sum of the average daily temperatures for the same period, which produces the bioclimatic coefficient (k). This method is based on the high correlation between ET, the temperature and the crop biology by stages. According to Tanaskovik et al., (2013), air temperature, which strongly influences ET, is most easily

measured parameter, and pretty equal during stages, decades, months, and vegetation periods. Hence, if this coefficient is determined through the field experiments, it will include the influences of the other meteorological parameters, the crop biology and conditions of the regions, the same authors reported. Therefore, the dynamics of the water consumption for all crops in the area can easily be calculated by the bioclimatic coefficient and the average daily air temperature. Rainfalls during the vegetation are included in the balancing of the water consumption.

Therefore, the main purpose of this research was to determine the bioclimatic coefficient of plastic-house grown pruned pepper as influenced by irrigation and fertilization techniques. The results from this research will have a practical application for the farmers in similar regions and conditions, first and foremost for the application of a proper irrigation application schedule and a more efficient use of water and fertilizers.

# Material and methods

The field experiment was conducted with pepper crop (Capsicum annum L. var. Bela dolga) pruned at two main shoots ("V" system) and grown in an experimental plastic house near by the Faculty of Agricultural Sciences and Food in Skopje (42° 00' N, 21° 27' E), during the period of May to September in 2005, 2006 and 2007. The soil type was coluvial (deluvial) disturbed with urban activities. The average soil pH at 0 to 60 cm depth was 7.30, while soil electrical conductivity ECe was 2.31 dS/m. The soil 0-60 cm layers contained respectively 2.80 mg/100 g available forms of N, 13.2 mg/100 g available  $P_2O_5$ and 22.5 mg/100g available K<sub>2</sub>O. According to the recommendations and literature data for the region (Jankulovski, 1997; Lazić et al., 2001), pepper planted in our conditions with yields up to 60 t/ha needs the following amount of nutrients: N 485 kg/ha, P<sub>2</sub>O<sub>5</sub> 243 kg/ha and  $K_2O$  585 kg/ha. The application of the fertilizer for the treatments was done in two portions (before transplanting and during the growing season), which is a common practice in our country. For all treatments, the first portions of the fertilizers was applied before transplanting of pepper, while the rest of the fertilizers were applied through the fertigation system for the drip fertigation treatments (Table 1) and conventional fertilization of soil for the control treatment (spread in two portions - at flowering and fruit formation). All investigated treatments received same amount of fertilizers, but by different methods and frequency of application of water and fertilizers, which was the main idea of this study. This approach enabled us to quantify the influence of the different irrigation and fertigation techniques on the bioclimatic coefficient of pepper.

Table 1. Type and amo	ount of fertilizers used	I during the vegetation	period in drip fertigation
treatments in kg/ha			
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Type of fertilizers	Amount of applied fertilizer	Period of application
15:15:15	318 kg/ha	before transplanting
0:52:34	375 kg/ha	drip fertigation during the vegetation
0:0:51+18S	802 kg/ha	drip fertigation during the vegetation
46:0:0	952 kg/ha	drip fertigation during the vegetation

\*Remark: the same amounts and quantity of fertilizers were used for furrow irrigation treatment

A drip irrigation system with integrated compensating drippers and a discharge of 4 l/h was installed. The fertigation equipment for the drip fertigation treatments was Dosatron 16, with a plastic barrel as reservoir for concentrated fertilizer. The whole amount of fertilizer

was dissolved in the barrel and the barrel was sealed to avoid evaporation of the water. The source of water was the water supply system for the city of Skopje. The irrigation of the experiment (treatment KK1, KK2 and  $Ø_B$ ) was scheduled according to long-term average daily evapotranspiration of pepper in Skopje region (Table 2). Long-term average (LTA) crop evapotranspiration was calculated by FAO software CROPWAT using crop coefficient (K<sub>c</sub>) and stage length adjusted for the local conditions by the Faculty of Agricultural Sciences and Food. Orta (1997) reported that the most suitable estimation method of evapotranspiration which could be used for irrigation scheduling of pepper is FAO modification of Penman. Some methods for determination of evapotranspiration in green pepper crop for Skopje conditions are presented in previous investigations by Tanaskovik et al., (2013). The daily evapotranspiration of drip irrigation treatments in our study was decreased for 20% (coefficient of the coverage-application of the water only on part of the total surface) similarly to Xie et al., (1999). In each experimental year, the irrigation and fertigation regime occurred from 15-20 May to 15-20 September.

Table 2. Long-term average daily and monthly crop water requirements (mm) for pepper in Skopje region calculated by FAO software CROPWAT

Months	V	VI	VII	VIII	IX	X
mm/day	1.9	3.6	5.5	5.0	3.7	1.8
mm/monthly	59	108	171	155	111	54

The irrigation scheme used in the experiment was designed according to randomized block design for experimental purposes with four treatments in three replications. Generally, the experimental treatments were set up according to the daily evapotranspiration rate. The idea was to investigate irrigation and fertilization techniques and their influence on the bioclimatic coefficient, as well as on crop potential evapotranspiration (ETP). Therefore, four experimental treatments were applied in this study:

Treatment 1 (KK1): Drip fertigation according to daily evapotranspiration with application of water and fertilizer in every two days;

Treatment 2 (KK2): Drip fertigation according to daily evapotranspiration with application of water and fertilizer in every four days;

Treatment 3 (KK3): Drip fertigation appointed by tensiometers, according to the recommendations undertaken by TEKINEL and KANBER (2002);

Treatment 4 ( $Ø_B$ ): Furrow irrigation according to daily evapotranspiration with application of water in every seven days and conventional fertilization (spreading of fertilizer on soil).

The size of each plot (replication) was 6.6  $m^2$  (25 plants in 0.75 m of row spacing and 0.35 m plant spacing in the row). Each plot (replication) was designed with five rows of crop and five plants in each row.

The plant pruning started when pepper plants were at least one foot tall (mid of May). Lateral branch shoots were pruned to form a plant structure of "V" system (two main shoots). According to Jovicich et al., (2004) and Rusevski (2005), only flower on the branch node and its adjacent leaf were left when we pruned a main shoot. Therefore, during the vegetation period, in every 10 or 15 days, all side shoots on the main shoots were removed, similarly to the recommendations of Daşgan and Abak (2003). Tie system was used to support the plants because each shoot was pruned up to 2 m high.

The bioclimatic coefficient in the research was determined as a ratio of monthly or seasonally potential evapotranspiration (ETP) and the sum of the average daily temperature for the same period (Bošnjak, 1999; Iljovski and Cukaliev 2002; Tanaskovik, 2005). The potential evapotranspiration ETP was determined monthly and seasonally by the soil-water balance method using direct measurements of soil moisture in the soil layer 0-100 cm (Cukaliev 1996; Bošnjak, 1999; Dragović, 2000; Iljovski and Cukaliev 2002; Evett 2007; Tanaskovik et al., 2013).

Collected data were subjected to statistical analysis of variance and the means were compared by using the least significant difference (LSD) at the 5% level of probability (P<0.05) test.

#### **Results and discussions**

#### The meteorological conditions during the research

Pepper needs a lot of heat during whole growing period, which is connected with its native place of origin (Jankulovski, 1997; Gvozdenović et al., 2004). If air temperature is below 15°C, there are possibilities for increasing the falling of flowers (Lazić et al., 2001; Gvozdenović et al., 2004). Also, the flowers and fruits can fall if air temperature goes above 35°C (Rylski, 1986; Đurovka et al., 2006). The optimal air temperature for growing pepper in controlled environment is 20-30°C during the day time and 18-20°C during night (Rylski and Spigelman, 1982; Đurovka et al., 2006). Bosland and Votava (2000) reported that best pepper yields can be obtained when the air temperature during day time is between 18-32°C, especially during the stage of fruit formation. According to Daşgan and Abak (2003), maximum daytime temperatures inside the greenhouse depended on the outside air temperature in the experimental plastic house during 2005, 2006 and 2007 was 22.83°C, 22.95°C and 24.10°C respectively (Table 3). During the period of the most intensive fructification (June-August), the average air temperature in all three years was in the range of the optimum values, recommended by the above mentioned authors.

Generally, pepper water requirements during the vegetation period are quite big compared to other crops, which is a result of the poorly developed root system, huge biomass exposure to strong transpiration (Iljovski and Cukaliev, 1994; Bosland and Votava, 2000; Lazić et al., 2001), large transpiring leaf surface and high stomata conductance of water vapor (Alvino et al., 1994; Delfine et al., 2002). Also, it is well known that pepper is the most sensitive to water shortage (drought) during flowering and fruit formation (Jankulovski, 1997; Bosland and Votava, 2000). Jaimez et al., (2000) revealed that water deficit during the period between flowering and fruit development reduced the final productivity. The Skopje region during the period flowering-fruit development is characterized with the highest temperatures and insolation; hence evapotranspiration has the highest rate.

		ge temperature		Average relative air humidity (%) in the			
Mantha	experimental plastic house			experimental plastic house			
Months	2005	2006	2007	2005	2006	2007	
V	20.9	20.5	21.6	72	74	73	
VI	24.1	23.6	27.1	63	71	61	
VII	28.2	27.2	31.0	60	63	53	
VIII	26.1	26.9	28.9	71	60	60	
IX	22.2	22.7	20.6	74	66	68	
Х	15.5	16.8	15.4	81	80	83	
Average	22.83	22.95	24.10	70.2	69	66.3	

Table 3. Monthly average air temperature (°C) and monthly average relative air humidity (%) in the experimental plastic house (by our measurements)

The field experiment was conducted in a controlled environment (plastic house), where precipitation does not have any influence on the crop water supply.

For normal growing of pepper and for high and quality yields, the optimal relative air humidity should range from 60 to 70%. Gvozdenović et al., (2004) reported that lower relative air humidity followed by high air temperature can affect flower and fruit falling. With the exception of October, the average relative air humidity during all three years of investigation was close to the recommended values for pepper production in plastic houses (Jankulovski, 1997) and greenhouses (Pennela et al., 2014). Data for relative air humidity during the investigation are shown in Table 3.

# The influence of irrigation and fertilization techniques on bioclimatic coefficient of plastic-house grown pepper

As was mentioned above, the bioclimatic coefficient in the research was determined as a ratio of monthly or seasonally potential evapotranspiration (ETP) and the sum of the average daily temperature for the same period. If the data for the ETP is expressed per plant stages, periods, decades or months and is compared to the average air temperature for the same period, the bioclimatic coefficient for the given period is obtained (Bošnjak, 1999; Iljovski and Cukaliev 2002; Tanaskovik, 2005). The calculation of the bioclimatic coefficient (k) and with it the determining the irrigation application rate is made according to the following formulas:

$$k = \frac{ETP}{\sum \overline{x}_{i}}$$

$$m = k \sum_{i} \overline{x}_{i} \quad \text{(for a period)}$$

$$m = k \overline{x}_{i} \quad \text{(daily)}$$

*k* - Bioclimatic coefficient for given period;

*m* - Irrigation application rate for given period in  $m^3/ha$ ;

 $\overline{x}_{t}$  - Average daily air temperature in plastic house (°C).

The potential evapotranspiration was determinated monthly and seasonally by soil water balance method by using direct measurements over the soil layer 0-100 cm (Cukaliev 1996; Bošnjak, 1999; Dragović, 2000; Iljovski and Cukaliev 2002; Evett 2007; Tanaskovik et al., 2013), and under permanent content of soil moisture and nutrients, as

well as permanent agro-technical measures. The monthly potential evapotranspiration  $(ETP_{month})$  was determinate when the active soil moisture at the end of the month  $(W_e)$  is subtracted from the soil water incomes for the same period (initial soil moisture at the beginning of each month (Wi) plus irrigation water (I) for the same period),  $ETP_{month} = (W_i+I)-W_e$ . The initial soil moisture was calculated as a difference between momentary soil moisture (MSM) and permanent wilting point (PWP) at the beginning of vegetation and each successive month. Monthly irrigation water requirements for the treatments KK1, KK2 and  $Ø_B$  were read on the volumetric meter; with periodic soil samplings for controlling of momentary soil moisture and irrigation regime. Irrigation water requirement for the treatment KK3 was obtained by tensiometers readings installed in the soil. The difference between water contents to MSM and PWP is the active soil moisture at the end of month. The sum of monthly water consumption present seasonally potential evapotranspiration or ETP=  $ETP_{month 1} + ETP_{month 2} + \ldots + ETP_{month x}$  (where ETP month 1,  $ETP_{month 2}$ ,  $ETP_{month x}$ , present each individual month included in investigation). The average 2005-2007 results for monthly and seasonally ETP are presented in Table 4.

Table 4. Average potential evapotranspiration (ETP in m<sup>3</sup>/ha) of plastic-house grown pepper for the period 2005-2007

Treatments	May	June	July	August	September	Seasonally
КК1	275.3 <sup>a</sup>	1043.8 <sup>a</sup>	1627.7 <sup>a</sup>	1512.7 <sup>a</sup>	428.5 <sup>a</sup>	4887.9 <sup>a</sup>
КК2	271.7 <sup>a</sup>	1028.9 <sup>a</sup>	1618.7 <sup>a</sup>	1495.8 <sup>a</sup>	425.0 <sup>a</sup>	4840.1 <sup>a</sup>
ККЗ	282.4 <sup>a</sup>	1088.5 <sup>b</sup>	1717.1 <sup>b</sup>	1580.3 <sup>b</sup>	438.9 <sup>a</sup>	5107.3 <sup>b</sup>
Øв	328.9 <sup>b</sup>	1245.1 <sup>c</sup>	1985.4 <sup>c</sup>	1816.9 <sup>c</sup>	505.1 <sup>b</sup>	5881.4 <sup>c</sup>

\*Values in rows followed by the same letter are not significantly different at the 0.05 probability level

The results for average monthly and seasonally ETP presented in Table 4 indicated negligible differences between the treatments KK1 and KK2, which is connected with closer irrigation interval of these two treatments. Statistically, there is no significant difference in ETP. As result of lower drip fertigation frequency, the treatment KK3 showed by 4.5 to 5.5% higher average seasonally ETP, compared to KK2 and KK1, and the differences were statistically significant. Similar pattern, except for ETP in May and June was also noted separately by months. Sezen et al., (2006) reported lower evapotranspiration in moderate frequency drip irrigation (6-10 days) and pan coefficient 0.50. In our study, the effect of drip fertigation on ETP is presented by the achieved results in treatments KK3 in comparison with  $\phi_{\rm B}$  (control treatment). Namely, in almost the same irrigation intervals, the treatment KK3 showed from 14.4 to 16.5% lower average monthly ETP and about 15% lower average seasonally ETP compared to  $Ø_B$ . The results are statistically significant at 0.05 level of probability. Higher ETP in treatment  $Ø_B$  may be associated with continuous water stress and inadequate soil moisture and nutrient content (Antony and Singandhupe 2004; Tanaskovik et al., 2013). Furthermore, the results for average seasonal ETP in our investigation were lower than those recommended by Iljovski and Cukaliev (1994), from 7000 to 8000 m<sup>3</sup>/ha and Doorenbos and Kassam (1986), from 600 to 900 mm, that is connected with proper and controlled irrigation and fertilization regime during all three years of investigation. On the other hand, Orgaz et al., (2005) reported lower ET in three stem pruned pepper (Dutch system) than ours, which was likely enough of different cultivation period and pruning system compared to our investigation. The obtained average results for the bioclimatic coefficient are given in Table 5. As can be seen, the lowest average seasonally bioclimatic coefficient of plastic house grown pepper

from 1.61 has been observed in treatment KK2, followed by treatment KK1 with 1.62 and KK3 with 1.69, and finally comes the control treatment  $Ø_B$  with the highest coefficient 1.94.

Table J. Average	Table 5. Average bioennatic coefficient (k) of plastic-house grown pepper in the period 2005-2007							
Treatments	May	June	July	August	September	Seasonally		
КК1	0.89 <sup>a</sup>	$1.60^{\rm a}$	2.11 <sup>a</sup>	2.08 <sup>a</sup>	1.43 <sup>a</sup>	1.62 <sup>a</sup>		
КК2	$0.88^{a}$	1.57 <sup>a</sup>	2.10 <sup>a</sup>	2.05 <sup>a</sup>	$1.42^{a}$	1.61 <sup>a</sup>		
ККЗ	0.92 <sup>a</sup>	1.66 <sup>b</sup>	2.23 <sup>b</sup>	2.17 <sup>b</sup>	$1.46^{a}$	1.69 <sup>b</sup>		
Øв	1.07 <sup>b</sup>	1.90 <sup>c</sup>	2.58 <sup>c</sup>	2.49 <sup>c</sup>	$1.68^{b}$	1.94 <sup>c</sup>		

Table 5. Average bioclimatic coefficient (*k*) of plastic-house grown pepper in the period 2005-2007

\*Values in rows followed by the same letter are not significantly different at the 0.05 probability level

The drip fertigation treatments (KK1, KK2 and KK3) have 14.8 to 20.5% lower average seasonally bioclimatic coefficient compared to the treatment with furrow irrigation and conventional fertilization ( $\emptyset_B$ ). The results are statistically significant at 0.05 level of probability. Such similar tendency was also noted separately by months. This can be connected with the irrigation technique and the possibility to apply nutrients through the system. According to Burt et al., (1998), maintaining moisture in soil on the necessary level by drip irrigation, despite the strong fluctuations from wet to dry soil, specific for the traditional irrigation techniques, has a positive effect on pepper production. Also, the results of our investigation correspond with those of Tanaskovic (2005), where the bioclimatic coefficient was significantly lower in drip fertigation than those in cases of tomato crop banded and furrow irrigated or banded and drip irrigated. Many other authors point out the advantages for the vegetable crops that gives fertigation compared to conventional fertilization separate from irrigation (Papadopoulos, 1996; Al-Wabel et al., 2002; Sezen et al., 2006; Cukaliev et al., 2008; Tanaskovik et al., 2011; Tanaskovikj et al., 2014; etc.). Tarantino and Onofrii (1991) reports crop coefficients of greenhouse bell pepper from 1.1 at the beginning of the cycle, up to 2.3 during the stage of maximum leaf area index, which correspond with those of the control treatment ( $\emptyset_{\rm B}$ ) in our study. Furthermore, in the present study, the drip fertigation frequency points to differences in bioclimatic coefficient, i.e. the treatment KK3 has noted an average seasonally bioclimatic coefficient higher by 5% in comparison to KK2, i.e. by 4.3% in comparison with KK1.

#### Conclusion

From the present study it can be concluded that drip fertigation at two to four days gives the lowest average monthly and seasonally potential evapotranspiration. Furthermore, a low frequency drip fertigation creates an environment for higher water consumption. The highest average monthly and seasonally ETP are obtained under furrow irrigation and conventional fertilization. The achieved results in our study, also demonstrate that irrigation and fertilization techniques have influence on the bioclimatic coefficient of plastic house pruned pepper. Thus, the lowest average monthly and seasonally bioclimatic coefficients were observed in drip fertigated treatments; while control treatment showed the highest values. Generally, the results for bioclimatic coefficients reported in this study, will enable growers in similar regions to determine the water requirements of plastic house pruned pepper and to schedule their irrigation with proper water quantity and in the right time.

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## SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

UDC: 634.63-145(4-15) Review Paper

## RECENT STUDIES ON THE SOIL CONSERVATION IN THE CONDITION OF MEDITERRANEAN REGION FOR OLIVE CULTIVATION: REVIEW

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#### Abstract

The Olive tree (Olea europae L.), a native of the Mediterranean Region basin and parts of Asia, is now widely cultivated in many other parts of the World for production olive oil and table olives. The majority of olive production in Mediterranean environments is characterized by low external inputs and is practiced in hilly areas with shallow soils. In the Mediterranean area land degradation is becoming progressively more important due to deterioration in the quality of land in terms of its capability to support land uses. Soil erosion is one of the main physical processes of land degradation, and in Mediterranean Hilly Areas is important in both its economic costs and the environmental negative effects. This review focuses comprehensively on the recent studies about alternative cultural and culture-technique measures to provide efficient soil conservation under the conditions of Mediterranean Region where the rainfed olive growing is densely practiced. **Keywords:** Olive tree, Soil conservation, Mediterranean Region

## Introduction

The cultivation of rainfed tree crops in the Mediterranean basin is associated with long processes of soil degradation. The cultivation of olive trees (Olea europaea L.), for example, dates back to ancient times. Today, olive orchards cover about 10Mha in the world, mostly under rainfed production systems (FAOSTAT, 2013). This long-lived tree is economically vital as well as socially and culturally emblematic of this environment. Over 98% of the world's olive oil is produced in Mediterranean countries (FAOSTAT, 2013), which illustrates its social economic and environmental importance. For the residents of the Mediterranean, olive oil constitutes the main source of nutritional fats and remains their most valuable export product. In particular, the cultivation of olives is concentrated on large parts of mountains and hills of Mediterranean countries where many orchards are confined to slopes or rugged land, susceptible to water erosion (Gomez et al., 2003).

Olive trees are extremely hardy. Olive groves have historically occupied hilly areas with shallow soils, where many other crops fail or produce very little. The rises in world olive oil consumption and agronomical improvements have led to intensive cultivation systems and new plantations on better soils. However, numerous olive groves in the Mediterranean area are found on soils with serious environmental limitations for agricultural use (Taguas, et al., 2015). The high soil erosion rates, loss of biodiversity, over-exploitation of water

resources and water pollution have been highlighted as the main negative environmental impacts that can be associated with this land use by several authors (Beaufoy, G., 2001;Gomez-Limon et al, 2012; Gomez et al., 2009b).

Soil erosion in olive orchards in the region has been mentioned as one of the major threats to the sustainability of this crop (Beaufoy, 2001). The on-site damage attributed to water erosion in olive orchards in Mediterranean region entails a decrease in productivity in olive yield as well as added difficulties in the access to and the movement within the orchards due to gully erosion. The major off-site effects of water erosion involve the contamination by agrochemicals (especially herbicides and fertilizers) of water bodies. This has resulted in the eutrophication of reservoirs, damages to infrastructures such as siltation of dams and rivers, and an increase in flood damages due to the large amount of sediment carried away by the flows (Gomez et al., 2014b). This poster attempts to provide a comprehensive review of most of the available experimental information.

#### Recent studies on soil conservation for olive orchard

There are basically two major approaches to soil management in olive orchards in all over the world: those which eliminate the ground cover in the area outside the projection of the olive canopy (which is achieved through plowing, the use of herbicides or a combination of both) and those which provide ground cover, usually achieved using a cover crop (seeded or spontaneous), sometimes combined with a mulch of pruning residues. In the latter case, control of the growth of the cover crop in spring is required to prevent competition for soil water with the trees, which are still mainly rainfed or, when irrigated, are under deficit irrigation. Although there are variations within each of these two main approaches (for instance, the number of times plowing is applied, whether a herbicide is or not used for bare soil management, the extension and type of the cover crop and the date of its control and the method used: either mechanical, animal or chemical), the major differences in soil management and its impact on soil properties are related to the presence or not of ground cover beyond the one provided by the olive tree canopy (Gomez et al., 2008).

In Mediterranean Region, we have found two kinds of studies measuring erosion rates in olive groves. Some are based on runoff plots and are aimed at comparing the effects of different soil managements on soil losses under the same environmental conditions (rainfall, topography, soil type) at hill slope scale. The other kind of studies are based on measurements taken on a small catchment scale (7 to 12 ha). The latter provide an integrated measurement of the balance between the different erosion processes and the deposition in the lower areas of the catchments, but are not conceived to evaluate the impact of different soil management under controlled conditions. Table 1 summarizes the results of these experiments. Hill slope-scale, plot-based experiments usually show unsustainable erosion rates when bare soil management (continuous tillage, CT, or no tillage with bare soil, NT) is used, which is in line with the observations of long-term erosion rates based on indicators such as olive mounds (Vanwalleghem, et al., 2011). Although NT and CT are different soil management methods, they result in comparable water erosion rates. It is also apparent from Table 1 that erosion rates under the same conditions and with the use of a cover crop are greatly reduced, usually below or closer to tolerable erosion rates. Within the context of erosion studies in olive orchards, tolerable erosion rates usually range from 10 to 12 t ha<sup>-1</sup> year<sup>-1</sup> (Gomez et al., 2009a) following the approach recommended for tree crops by the literature. The results shown in Table 2 on

small catchment scales also indicate relatively high erosion rates, with the exception of one experiment, where a combination of cover crops and rocky outcrops (and a notably dryer rainfall regime) within the catchment resulted in low erosion rates. These results are consistent with the concern about high erosion rates and degraded water quality due to offsite contamination (which is indirectly related to high sediment load from the cropping areas) in olive growing areas in Southern Spain (Beaufoy, 2001).

Table 1. Measured soil losses by water erosion on hill slope (plots) and catchment scale experiments in olive orchards. NT means bare soil with herbicide use and no tillage, CT bare soil by tillage, CC cover crop soil management. Sources: 1 (Gomez et al., 2009c), 2 (Gomez et al., 2010), 3 (Gomez, 2010, – Gomez et al., 2012], 4 (Francia et al., 2006), 5 (Arroyo, 2004), 6 (Gomez et al., 2014), 7 (Taguas et al, 2013), 8 (Taguas et al, 2012).

	Soil	Average Soil	Number	Slope	Steepness	Plot Area
Experiment	Management	Loss	of	Length	of	$(m^2)$
	_	t ha <sup>-1</sup> Year <sup>-1</sup>	Years	(m)	Slope (%)	
	NT	6.9	5	12	13.4	72
Conchuela I <sup>1</sup>	СТ	2.9	5	12	13.4	72
	CC	0.8	5	12	13.4	72
Benacazon <sup>2</sup>	СТ	29.6	6	60	11	480
Denacazon	CC	58.9	3	24	13.4	288
Conchuela	CT	5.6	2	24	13.4	288
$\Pi^3$	CC	5.6	2	24	13.4	288
	NT	25.6	2	24	30	480
Lanjarón I <sup>4</sup>	CT	5.7	2	24	30	480
	CC	2.1	2	24	30	480
	NT	25	2	22.5	30.5	480
Lanjarón II <sup>5</sup>	CT	8.5	2	22.5	30.5	480
	CC	6.5	2	22.5	30.5	480
	Soil	Average Soil	Number	Maximum	Average	Catchment
Experiment	Management	Loss	of	Flow	Slope (%)	Area (m <sup>2</sup> )
		t ha <sup>-1</sup> Year <sup>-1</sup>	Years	Length (m)		
Conchuela <sup>6</sup>	CT and CC	16.1	5	270	9	80,000
Puente	CC	1.8	5	150	15	61,000
Genil <sup>7</sup>						
Setenil <sup>8</sup>	NT and CT	9.3	6	340	9.5	67,000

## Conclusion

Accelerated soil erosion rates, mostly associated to water erosion are due to a combination of cultivation in sloping areas and to an inappropriate soil management, which has provided limited (or none) ground cover among the olive trees. Although the erosion rates increased with the advent of mechanization of olive cultivation in the early 1960s, this review suggests that they started to be unsustainable since intensification of olive cultivation started in the region during the early 19th century. Experimental information shows that an appropriate use of cover crops aimed to provide effective ground cover, from fall to early spring, can bring erosion down to tolerable rates, as compared to the effects of bare soil management (CT and NT). Results also indicate an improvement of soil quality when the cover crop soil management is implemented in comparison to CT of NTD (Gomez et al., 2014a).

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## SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

UDC: 416.8:546.4/.8(560) 631.67:546.4/.8(560) Original Scientific Paper

## HEAVY METAL CONTENT OF SOIL AND IRRIGATION WATER IN HEVSEL GARDENS, DIYARBAKIR

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#### Abstract

Hevsel Gardens have had an important role in the nourishment of the public since the establishment of the City of Diyarbakır until today. This study aims to determine the land structure of Hevsel Gardens, their status according to land use capability, and the heavy metal contents of the soil, irrigation water used in agricultural production and certain produces of Hevsel Gardens. The study has revealed that as of the period in which the samples have been taken, the heavy metal contents in the soil, water and plants are not at a threatening level for human health, these analyses should be repeated through the year and the quality of the irrigation water needs to be improved. It is thought that the agricultural production awareness of producers should be increased, and the production of ornamental plants, seedling production and green housing should be encouraged.

Key words: Diyarbakır, Hevsel Gardens, soil, irrigation water, heavy metal

#### Introduction

Soils an essential natural source for the survival of humans and for ensuring development and wealth of countries. Through history, development level of civilizations and life standards of people have been significantly influenced by the soil-human relationship, which has shaped the formation and development process of civilizations. One of the most important problems of our era is the production of food sources needed for the nourishment of world's ever-growing population in a safe manner and without damaging the environment.

Irrigation has a very significant role for high agricultural productivity and quality in arid and semi-arid regions. However, wrong and excess irrigation cause serious environmental problems such as elevation of ground water, saltiness, deep digging of fertilizer and pesticide residues, accumulation of trace elements in water sources, soil erosion, and illnesses and damages in the living creatures benefiting from such water and soil including the residues. Fast industrialization and ever-growing traffic increase the amount of heavy metals in the environment together with many other pollutants. This leads to many problems, especially loss of products, in plants that do not have mobility (Munzuroglu and Gur, 2000). Some heavy metals in high concentrations, negatively affect plants and the humans and animals feeding on plants. There might be toxic effects in the event that heavy metal concentrations that can be extracted in soil are above: 1 mg kg<sup>-1</sup> for cadmium (Cd), 10 mg kg<sup>-1</sup> for cobalt (Co), 0,1 mg kg<sup>-1</sup> for copper (Cu), 10 mg kg<sup>-1</sup> for

selenium (Se), 0,5-1 mg kg<sup>-1</sup> for vanadium (Va), 100 mg kg<sup>-1</sup> for nickel (Ni). It is stated that the types of heavy metal that have the most severe toxic impact are Cd, lead (Pb) and mercury (Hg) (Okcu *et al.*, 2009).

Whether it is a necessary element for plant development or not, excessive accumulation of heavy metals in tissues and organs, adversely impact the development of vegetative and generative organs of plants. Due to these toxic impacts, heavy metals cause the deterioration of may physiological incidents in plants such as transpiration, stomatal movements, water intake, photosynthesis, enzyme activity, germination, protein synthesis, membrane stability, hormonal balance (Asrı and Sonmez, 2006).

Hevsel Gardens agricultural production areas cover an area of approximately 3000 – 3500 hectares within the boundaries of Diyarbakır province, Sur Town, Alipaşa and Fatihpaşa Neighbourhoods; in 1000-1500 hectares of this land close to the river bed, poplar tree production is made and the remaining section hosts mostly vegetable production and domestic fruit production is made in a small section. Concentration of the population in cities puts pressure on agricultural production areas. Due to this pressure, agricultural production areas overlap urban structures. Nourishment habits have an important role in the memory of societies. Local markets have important functions in this regard. This study aims to carry out heavy metal analyses in the soil and agricultural irrigation water and to determine the agricultural potential of this area.

#### **Materials and Method**

The land of Hevsel Gardens are slightly inclined to the river on the banks of the Tigris (% 0-2 inclination) and have deep profile soil. This soil that is appropriate for the production of all kinds of plants has a depth of 90 cm and above. This land is a first class agricultural land which is in "Agricultural Land" class and in the deep profile soil group. Majority of the soil is clay-loam soil in terms of texture (Anonymous, 1994; Figure 1).

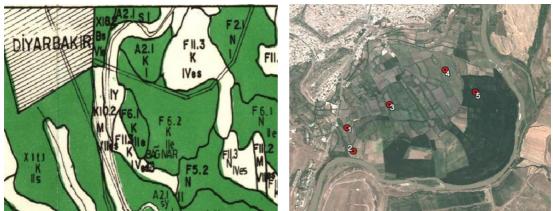


Figure 1. Location with the sampling locations of provincial land size of Hevsel Gardens

In the study carried out for the purpose of determining the general characteristics of Hevsel Gardens, visits have been paid to almost all pieces of land and soil samples have been taken from five different points representing the study area and from two different depth types (surface and underground) (Table 1).Soil samples were analysed in Diyarbakır Directorate of Provincial Food, Agriculture and Livestock, Soil Analysis Laboratory. In

Hevsel Gardens, the farmers use Hz. Suleyman spring, Benusen spring, Anzele spring, springs in gardens and Domestic Wastewater as agricultural irrigation water. Samples were taken from these water sources for the analyses. In addition to the soil and water samples, dill, rocket and stinger plants growing in Hevsel Gardens, were sampled as well and these samples were dried at 70°C for 48 hours. Heavy metal analyses of soil and water samples were made in PerkinElmer Optima 7000 DV (PerkinElmer, Inc., Shelton, CT, USA) device.

## **Results and Discussion**

## Physical and Chemical Analyses of the Soil of Hevsel Gardens

Results of the soil analyses of Hevsel Gardens are shown in Table1. According to the results of analyses, water saturation amount varies from 52% to 81%. The structure/texture of the soil samples are sand, clay, clay-loam (Table 1). The high water ratio in the samples is due to the high humidity ratio of the soil at the time of sampling (15 January 2015). The salt amount in the soil samples varies from 0.04 to 0.29. Total salt amount may vary according to the sampling date (Table 2).

Table 1. Morphological characteristics of Hevsel Gardens (LCC: Land Capability Classification)

Land No	Soil Depth	Inclination	Rockiness	Structure	LCC
1	120+	%2-6	None	Clay loam-loam	IIe
2	120+	%0-2	None	Loamy sand	Ι
3	120+	%0-2	None	Loam	Ι
4	120+	%0-2	None	Loam	Ι
5	120+	%0-2	None	Clay loam-loam	Ι

Location	Depth	Texture	Total salt (EC %)	Lime (CaCO <sub>3</sub> ) (%)	pН	Organic Material (%)	$\begin{array}{c} P_2O_5\\ (kg \ da^{-1}) \end{array}$	K <sub>2</sub> O (kg da <sup>-1</sup> )
1	0 - 30	Clay loam	0.05	13.3	7.78	2.35	17.3	208
1	30 - 60	Loam	0.04	12.54	7.84	2.44	18.7	182
2	0 - 30	Loam	0.12	5.7	7.76	1.45	20.9	76
2	30 - 60	Loamy sand	0.04	7.22	7.80	0.58	8.75	30
3	0 - 30	Clay	0.06	3.8	7.72	4.00	25.5	382
3	30 - 60	Clay	0.29	3.8	7.70	2.38	16.5	267
4	0 - 30	Clay	0.06	3.42	7.40	2.73	24.7	237
4	30 - 60	Clay	0.06	4.94	7.52	2.73	24.7	293
5	0 - 30	Clay loam	0.05	4.94	7.49	2.55	28.1	280
5	30 - 60	Loam	0.05	4.18	7.58	1.68	16.5	175

Table 2. Result of the analysis on the soil of Hevsel Gardens production site

In general, the pH of the soil that varies in texture differs from 7.40 to 7.84. There is no lime problem in the soil, the lime content (CaCO<sub>3</sub>) varies from 3.42% to 13.3%. The soil of Hevsel Gardens is in low or medium lime class in terms of lime content. The organic material of the soils varies from % 0.58 to % 4.00 and the soil is in medium-rich organic material soil class. Hevsel Gardens agricultural land is rich in phosphor and potassium and

is at a good level (Guzel *et al.* 2002). However, it is determined that the lower (30-60 cm) soil of sample 2 of the analysed soil is at a medium level in terms of potassium (K).

#### Heavy Metal Content of the Soil of Hevsel Gardens

The most important toxic heavy metals are cadmium, lead, nickel, arsenic and copper. These heavy metals reach the soil more easily due to phosphorous fertilizers and the raw materials of such fertilizers. The boundary values of some heavy metals in soil with regard to heavy metal contents are respectively  $0.2 - 1.0 \text{ mg kg}^{-1}$  for Cd, 10 mg kg<sup>-1</sup> for Co,  $70 - 10^{-1}$  for Cd, 10 mg kg<sup>-1</sup> for Co,  $70 - 10^{-1}$  for Cd, 10 mg kg<sup>-1</sup> for Co,  $70 - 10^{-1}$  for Cd, 10 mg kg<sup>-1</sup> for Cd, 10 mg kg<sup>-</sup>  $100 \text{ mg kg}^{-1}$  for Cr, 20 - 30 mg kg<sup>-1</sup> for Cu, 1000 mg kg<sup>-1</sup> for Mn, 50 mg kg<sup>-1</sup> for Ni, 10 – 30 mg kg<sup>-1</sup> for Pb, 0.5 mg kg<sup>-1</sup> for Se and 50 mg kg<sup>-1</sup> for Zn (Alloway, 1993; Guzel et al. 2002). As can be seen in Table 3, the Nickel (Ni) content of the sample soils is rather unsteady, and varies between 24.2 and 61.8 mg kg<sup>-1</sup>. The Ni content of the sample soils taken from the sampling locations 2 and 3, has been above the boundary levels (Alloway, 1993; Guzel et al. 2002). The Ni increases in the soil due to the fuel including Ni in the agricultural lands close to traffic jam locations. It is thought that this is due to the high traffic in the highway adjacent to Hevsel Gardens. The research area differs according to the depth where the Lead (Pb) content of soil samples are taken, and it is determined that the Pb content of samples taken from a depth of 30 - 60 cm is higher than that of the samples taken from the surface. This shows that Pb element moves downwards in the soil washed both with irrigation water and rain. This depth is also the depth where the plant's root system is active and may cause accumulation in the plant. The Pb content of the soil samples has been found at both depths in sample 1 and above the boundary values in samples 3 and 4. This is thought to occur with the impact of the environmental (urban) emissions in addition to the use of wastewater as agricultural irrigation water.

	1	l		2		3	4	1	4	5
	0-30	30-60	0-30	0-30	0-30	30-60	0-30	30-60	0-30	30-60
Cd	0.25±0.66	$0.25 \pm 0.26$	$0.025 \pm 3.30$	$0.10{\pm}4.47$	0.5±0.38	0.18±2.37	$0.12{\pm}1.34$	$0.2\pm0.97$	$0.06 \pm 2.98$	0.12±2.71
Co	7.0±0.42	6.5±0.46	16.8±0.72	$11.8 \pm 1.06$	9.5±0.22	8.4±0.15	7.1±0.67	11.4±0.23	6.8±0.29	8.4±0.40
Cr	22.0±0.68	18.3±0.49	37.8±0.49	35.3±1.43	4.3±0.26	26.9±0.36	$14.2 \pm 0.88$	23.8±0.45	16.1±0.89	18.4±1.65
Cu	39.3±0.01	43.8±1.61	74.0±1.08	50.3±1.08	17.8±1.00	40.6±0.51	49.6±1.28	72.6±0.60	34.4±0.50	44.8±0.84
Mn	$236.3 \pm 0.64$	214±0.19	356.3±0.42	572.3±0.23	666.0±0.31	$308.0 \pm 0.26$	$400.5 \pm 0.27$	315.6±0.11	145.8±0.23	168.4±0.61
Ni	30.5±1.47	27.0±0.60	61.8±0.77	$56.0 \pm 0.68$	$52.8\pm0.80$	50.1±0.78	$24.2\pm0.42$	38.4±1.00	26.7±0.10	30.8±0.71
Pb	35.3±0.26	41.5±0.51	7.0±0.33	$5.75 \pm 0.78$	13.3±0.35	39.2±0.89	28.7±0.16	42.6±0.51	15.3±0.51	19.8±0.29
Se	Tr	Tr	Tr	Tr	Tr	Tr	Tr	Tr	Tr	Tr
Zn	36.5±0.24	33.0±0.56	37.8±0.51	$24.0\pm0.38$	42.8±0.23	38.2±0.40	27.5±0.51	43.4±0.92	19.2±0.67	20.8±0.92
Fe	$13162 \pm 0.42$	$13297 \pm 0.08$	12945±0.20	19102±0.57	512.3±0.14	$19635 \pm 0.02$	9585±0.07	8376±0.37	11250±0.18	12486±0.24

Table 3. Heavy metal content of soil samples taken from different locations and depth in Hevsel Gardens (ppm (mg kg<sup>-1</sup>)  $\pm$ % RSD)

Cadmium is widely used in the manufacture of dye, glass, textile, electricity, batteries, fungicide, insecticide, metal alloys and synthetic polymers. Cd of more than 3 mg kg<sup>-1</sup> in soil causes toxic effects. When the Cd contents of the soil samples of Hevsel Gardens are analysed, it is seen that the total Cd concentration in the soil is within acceptable limits for agricultural soil (Table 4). As Cd has the highest water dissolution characteristic among heavy metals, it also has a high emission rate in nature. Copper is fixed strongly in the environment by oxides and clay minerals and especially organic materials. The heavy metal concentration in the soils has been determined as 39 mg kg<sup>-1</sup> for copper, and in cases where it is above this value, toxic effects can be seen (Yaylali and Tuysuz, 2012). When Hevsel Gardens soil is assessed from this point of view, is can be stated that there is a

significant level of copper pollution. Intensive agricultural activity in these regions and extensive use of chemical pesticides including Cu are thought to be a factor increasing the Cu content. In addition, human related, other agricultural pollutants, wastewater, etc. sources can cause copper pollution in soil. Chromium boundary values of 10-100 mg kg<sup>-1</sup> in soil are considered normal. There is no chromium pollution in the agricultural soil of Hevsel Gardens, and the highest chromium value has been found in sample 2 (37.8 mg kg<sup>-</sup> <sup>1</sup>). This result shows us that there is no contamination and the existing chromium can be due to natural material. Cobalt accumulation is within the notified boundary values in samples other than sample 2 (0 - 30 and 30 - 60 cm) and 4 (30 - 60 cm) in the survey area. Soil sample no. 2 has high Co content (16 mg kg<sup>-1</sup>) and it is thought that there can be Co accumulation here. Especially the high Co content in the 2<sup>nd</sup> station shows that there is accumulation in the soil due to usage of phosphoric and nitrogenous fertilizers. Important sources for the Co in the soil are: direct cobalt salts or fertilizers including Co such as the phosphate fertilizers applied on the topsoil in the implementations for missing elements (Adriano, 1986). It is seriously discussed that lack of selenium is determinant in cancer and cardiovascular diseases. Selenium is not an essentially necessary plant nutritive element; however, it is one of the most significant antioxidants in the world (Irmak and Semercioglu, 2012). Hevsel Gardens soil samples include trace amount of selenium, and there is no pollution. There is no Zn pollution in the survey area, the highest Zn value has been found in soil sample, no. 4 at a depth of 30 - 60 cm (43.4 mg kg<sup>-1</sup>). In soil, iron content of 0.02 mg kg<sup>-1</sup> is defined as low, 2.0 - 4.5 mg kg<sup>-1</sup> is defined as medium and more than 4 - 50 mg kg<sup>-1</sup> is defined as sufficient (Lindsay and Norvell, 1969). There is negative correlation between useful iron and the lime content of soil and a positive correlation between the amount of clay and organic material. In 40% of the soil in GAP (South-eastern Anatolia Project) region, the amount of iron is less than 4.5 mg kg<sup>-1</sup>.

#### Heavy Metal Content of the Agricultural Irrigation Water Used in Hevsel Gardens

There are old records indicating that the wastewater of the city also reached the gardens in the irrigation of Hevsel Gardens. Documents dated 1893, show the complaints and actions filed by the producers who thought that prevention of the wastewater for health protection was hindrance of production in the gardens. Today, the irrigation water of agricultural production areas of Hevsel Gardens are provided from five different sources. These sources are; Hz. Suleyman Spring, Benusen spring, Anzele spring, natural spring water and domestic wastewater (Figure 2).



Figure 2. Irrigation water sources Hevsel Gardens (left: spring water, right: deep well) In the analysed irrigation waters, the lowest Ni amount was observed in Anzele spring  $(0.0002 \text{ mg kg}^{-1})$ , and the highest amount was observed in natural spring water (0.00315 mg kg<sup>-1</sup>) (Table 4). Considering zinc content, it is determined that the lowest Zn content is in domestic wastewater as 0.00305 mg kg<sup>-1</sup>, and highest Zn content is in natural spring water as 0.00897 mg kg<sup>-1</sup>. The Zn content in Hz. Suleyman, Benusen-Spring water and Anzele spring is 0.00886 mg kg<sup>-1</sup>, 0.00572 mg kg<sup>-1</sup> and 0.00323 mg kg<sup>-1</sup> respectively (Table 4). According to Water Pollution Control Regulation Directive for Technical Procedures (Anonymous, 1991), the Zn content allowed for irrigation waters varies between 2.0 and 10 mg kg<sup>-1</sup> depending on climate, soil and irrigation conditions.

Location				Para	meters (p	opm)			
Location	Ni	Zn	Se	Pb	Cu	Fe	Cr	Cd	Mn
Hz. Suleyman spring	0.0015	0.0089	Tr	0.0045	0.0052	0.0134	0.05	0.0087	0.0392
Benusen	0.0009	0.0057	Tr	0.0006	Tr	0.0002	0.0379	0.0003	0.0002
Domestic Wastewater	0.001	0.0031	0.0001	0.0089	0.0079	0.0028	0.012	0.0494	0.0537
Natural spring water	0.0032	0.009	0.0034	0.0032	0.0109	0.0049	0.0166	0.0615	0.0042
Anzele water	0.0002	0.0032	Tr	0.0109	0.0058	0.0046	0.0137	0.0054	0.0068

Table 4.Trace element and heavy metal contents of agricultural irrigation waters in Hevsel Gardens

The selenium content in irrigation waters has been found at trace amounts in Hz. Suleyman, Benusen and Anzele spring waters. Selenium content is the highest in Domestic Wastewater and spring waters as 0.07 mg kg<sup>-1</sup> and 0.00344 mg kg<sup>-1</sup> respectively. However, according to Water Pollution Control Regulation Directive for Technical Procedures (Anonymous, 1991), the maximum allowed Selenium content in irrigation waters is 0.02 mg kg<sup>-1</sup>. Pb content in irrigation waters vary from 0.00063 to 0.01089 mg kg<sup>-1</sup>. While the lowest Pb amount is seen in Benusen spring, the highest Pb level is in Anzele spring with 0.01089 mg kg<sup>-1</sup> (Table 4). According to Anonymous (1991), there is no restriction for irrigation in terms of Ni, Zn, Se and Pb contents in the sources providing water for Hevsel Gardens. Cu contents differ between 0.00522 and 0.0109 mg kg<sup>-1</sup> in the water samples apart from that received from Benusen spring. According to Anonymous (1991), the allowable Cu content in irrigation waters is between 0.2 and 5.0 mg kg<sup>-1</sup>. Therefore, when the Cu contents in waters received from Hz. Suleyman, Anzele, domestic wastewater and natural spring water and used in irrigation, are analysed separately, it can be said that it does not exceed the maximum total Cu amount of 190 kg ha<sup>-1</sup>, which does not cause accumulation neither in the soil or in plants; therefore, will not have adverse effects for human health. Fe content has been found as 0.01337, 0.00015, 0.00275, 0.00486 and 0.00462 mg kg<sup>-1</sup> in Hz. Suleyman, Benusen, Domestic Wastewater, Natural Spring Water and Anzele spring respectively (Table 4). According to Water Pollution Control Regulation Directive for Technical Procedures (Anonymous, 1991), the allowable Fe content in irrigation waters is between 5.0 and 20.0 mg kg<sup>-1</sup>. Cr contents of irrigation waters have been found as 0.05, 0.03791, 0.01198, 0.01659 and 0.01373 mg kg<sup>-1</sup> in Hz. Suleyman, Benusen, Domestic Wastewater, Natural Spring Water and Anzele spring respectively (Table 4). Maximum allowable Cr amounts vary from 0.1 to 1.0 mg kg<sup>-1</sup>. the manganese (Mn) amounts that should be between 0.2 and 10.0 mg kg<sup>-1</sup> according to Water Pollution Control Regulation Directive for Technical Procedures (Anonymous, 1991), have been found as 0.03924, 0.00016, 0.05374, 0.0042 and 0.00678 mg kg<sup>-1</sup> in Hz. Suleyman, Benusen, Domestic Wastewater, Natural Spring Water and Anzele spring respectively. The results show that there is no restrictive condition in the irrigation water in terms of Fe, Cr and Mn amounts. When the Cd values whose existence is an important problem, are reviewed, the Cd content is found as 0.00872, 0.00028, 0.04936, 0.06146 and 0.00535 mg kg<sup>-1</sup> in Hz. Suleyman, Benusen, Domestic Wastewater, Natural Spring Water and Anzele spring respectively. The allowable maximum Cd amounts vary between 0.01 and 0.05 mg kg<sup>-1</sup> (Anonymous, 1991). Among the irrigation waters, the Cd content is found just above the boundary value only in natural spring water.

## Conclusions

Vegetables with fruit and green leaves, consumed fresh without being processed, and largely preferred by consumers as they are easily found and cheap are more prone to water related pollution than other types of food, depending on the quality of irrigation water used in the land they are grown. Occurrence of foodborne diseases as a result of consuming these types of food is high. Hevsel Gardens have high risk in this regard. As is seen in certain examples that can occur in developed countries as well, there have been death cases due to outbreak of Escherichia coli in plants whose leaves are consumed, such as lettuce and spinach. The basic reason for this is irrigation with surface waters with which Domestic Wastewater is mixed up. The level of use of ancestral seeds by producers is rather low. The seeds used are materials sold in the market by firms. Use of seeds with local properties should be encouraged. Such as Lice tomato, etc. Construction should not be allowed in the agricultural production areas of Hevsel Gardens, and expansion of residents in this area should be prevented. In this way, the first class agricultural land can be protected against domestic waste and construction. The laboratory in the water treatment unit of Diyarbakır Waterworks Authority (DİSKİ) has sufficient infrastructure and hardware. Therefore, routine analyses should be made to detect risk causing elements in irrigation water, soil and plants and the relevant public institutions should be warned to take the necessary measures. In addition, below are certain measures to be taken in Hevsel Gardens for soil, irrigation water and food safety.

## Measures to be taken for the sustainability of agricultural soil

- Hevsel Gardens does not have a serious saltiness problem. Most of the soil is oligohaline soil class. This can be periodical, which may be observed due to the period of analysis. Therefore, the analysis should be repeated in various periods through the year.
- Even though Hevsel Gardens agricultural production areas have gentle slope, there is the threat of erosion. In order to prevent this, soil cultivation and irrigation methods should be applied with the appropriate soil cultivation devices and at the right time.
- Highly nitrated fertilizers are used in order to increase productivity. This causes accumulation of nitrate in the soil and plants and adversely affects human health. Regular analyses should be made from product growing and to marketing stages to determine this and there should be supervision.

## Recommendations for the water used in the irrigation of agricultural production areas

- Formation of complex structures due to the accumulation of heavy metals existing in the irrigation water through time, adversely affects human, animal and plant health.
- The quality of the agricultural irrigation water should be improved. For this purpose, the usefulness of the existing waters should be determined and the water should be brought to the farms with a closed system.

- The quality of the irrigation water may affect the saltiness of the soil itself. Because salts are brought to the soil mainly through irrigation water.
- The Ministry of Agriculture should subsidise the farmers for transforming their farm irrigation systems into closed and pressured systems.
- It should be ensured that domestic wastewater reaches the Tigris River with a closed circuit system.

## Points to Consider for the Food Safety of the Grown Produces

- Therefore, regular analyses should be made to determine the nitrate content and nitrate limit excess in every plant in Hevsel Gardens in the active growing period before the produces are marketed.
- The highway adjacent to Hevsel Gardens is a busy road. There is emission of Cd, Pb, S and other heavy metals from the exhaust pipes of the vehicles on the highway. These toxic elements cause a risk in the consumption of especially the plants whose leaves are consumed. For this purpose, measures should be taken to decrease the traffic on the highway.
- A mechanism should be established to regularly control the microbial pollution in the vegetables whose leaves are consumed, that may arise from irrigation water and soil.

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## USE OF REMOTE SENSING TECHNIQUES IN DETERMINATION OF PADDY AREAS

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#### Abstract:

Nowadays, agricultural enterprises and related government organizations should consider economic, social, cultural, technological and environmental factors in decision making processes like whole sectors. Classic methods are used for obtaining this type of information may cause major load in terms of monetary and time. Also, safeties of obtained information are disputable. The information obtained by satellite technologies could be provided much shorter time, more economic and high accuracy by assistance of qualified staff and processing of methods which are accepted by international institutions. As known, Turkey applied to be member of European Union and they should obey different rules of EU like agriculture policies in regard to this application. In this case, cultivated area has to be determined accurately in order to implementation of right agriculture policy in European Union. In this study, the main purpose is to determine paddy production areas which are extend along the Meric River in the year of 2011. Paddy areas were determined using high resolution Rapideye satellite image by supervised classification techniques. In the result of study, paddy production area was found as 41021.18 ha.

Key words: Rapideye, remote sensing, paddy, classification

#### Introduction

The studies are about investigation and development of natural resources which are limited for meeting human requirement should be more effective due to rapidly increasing of world population and technologic development. Wealth of natural resources has ground of economic development and it steps up the development.

The total changes of planted areas and obtained yield which have been determined by survey studies have been reported to farmers for many years. But, this ordinary method takes up more time and it isn't always reliable. Therefore, the usage of new techniques is absolute required for planning of effective land use. Such a database covering the agricultural areas can create opportunity to get information about other natural resources (Tugac and Torunlar, 2000).

The developments of remote sensing and geographic information systems have great importance in the studies about determination of land cover or land use which are most dynamic element in the earth, effective usage of natural resources and rural planning. Geographic information systems and remote sensing techniques are most widely used methods because of some reasons like high accuracy, short working time and low cost in long term (Anonymous, 2002). Obtaining data is a generally expensive process. Especially, spatial and temporal validity of the data which are variable based on time are restricted. Land management shows rapidly changes due to the activities and production of land user. For this reason, large volume data should be obtained, so it is required to be spent great effort. Besides that spatial information are continually required in land management studies. As it is considered from this point of view, the satellite images which supply data from large areas persistently are very effective way to discover the features of land use (Rosenfeld and Chutirattanapan, 1994).

It is necessary to detect amounts of production and planting areas for crop production during our participation process in the European Union (Eraktan, 2002). For this reason consumption demand of agricultural products and our current production amounts are should be put forward. The crop pattern in the agricultural areas can be determined by many personnel who have to work in field during long period by the reason of dynamic structure in agricultural production and their activities are performed in wide areas. This situation causes both to be achieved data with very large costs and obtained inaccurate results based on the information are getting from declaration of farmers. Marketing prices could be determined realistically through the amount of production are detected properly and this case will provide that preventing price fluctuations when farmers sell their products and leading fulfillment of social expectations. The right agricultural policies can be only provided by obtaining information with accurately, quickly and economically. Remote Sensing (RS) and Geographical Information Systems (GIS) have been used in agriculture sector depend on occurring developments in technology (Chuvieco, 1993). Many studies have shown the utility of satellite remote sensing for Agricultural monitoring (Gallo and Flesh, 1989), mapping of cultivated areas (Fang et al., 1998) and observing the stages of crop growth (Reed et al., 1994) from local, regional to global scale (Maselli and Rembold, 2001).

Paddy (Orvza sativa L.) is one of the most important staple food crops, feeding over half of the world's population. In 2010, the global paddy production was approximately 672 million tons from a cultivation area of around 154 million ha (Zhao et al., 2015). Turkey is contributing to paddy production approximately in 99,400 ha and 900,000 ton in 2011. Paddyproduction areas have been planned to improvement within the scope of "Paddy production development project" by the Ministry of Agriculture. In addition, it has been planned with this project that paddy purchase can be taken under control by public institutions and private sector so unregistered economy can be prevent. As to paddy remote sensing, most of studies were focused on field area mapping production estimation (Bailey et al., 2001; Ribbes and Le Toan, 1999; Fang et al., 1998; Kurosu et al., 1997; Xiao et al., 2002; Sun et al., 2009; Peng et al., 2011; Jeong et al., 2012; Nuarsa et al., 2012; Kuenzer and Knauer, 2013; Gumma et al., 2014; Mosleh and Hassan, 2014; Gumma et al., 2015). In this study, it was attempted to determination of paddy cultivation areas in Edirne Province where is the most paddies producing place in both Thrace region and our country. Therefore, crop classification process was carried out by using of Rapideye satellite image belonged to 2011. Supervised classification and maximum likelihood algorithm are used in the classification process. In the result of accuracy analysis, overall accuracy was determined as 85,56%. Finally, it was seen that enough accuracy was cached in this study.

## Material and methods

The study area has been conducted Edirne city. The study area ranges from  $40^{\circ} 41^{\circ}$  E to  $41^{\circ}$  29 E and from  $26^{\circ} 00^{\circ}$  N to  $26^{\circ} 45^{\circ}$  N west Thrace in Turkey. Annual precipitation is 600 mm and annual mean air temperature is  $13,5^{\circ}$ C.

The study area is under influence of Black Sea and Mediterranean Sea climate along coastline is neighbor of Marmara Sea. Interior part of the study area has continental climate. While the winter seasons are cold and rainy, the summer seasons are hot and little rainy. Meric and Ergene rivers are used as main water resource. Mericriver is a much fresher water resource than Ergeneriver. As for that Ergene River passes through large industrial zones like Corlu and Cerkezköy and their wastewater has arise great pollution in Ergeneriver. The paddy cultivation areas in this study are comprised of the regions which are close to river bed and bottom flat land.

Rapideye, which was launched in August 2008, is a constellation mission consisting of five identical small satellites. It provides high-resolution multispectral imagery in five optical bands corresponding to the blue, green, red, red-edge, and near-infrared (NIR) parts of the electromagnetic spectrum (440–850 nm). The satellites are placed in a single sun-synchronous orbit at an altitude of 630 km. At nadir, the revisit period is 5.5 days, and the ground sampling distance is 6.5 m (Hyun and Jong, 2015).

Rapideye satellite image was selected to use for classification. 3A Rapideye satellite image was acquired in August 2011. Crop classification with satellite images was basically carried out of processes consisting of pre-processing, image enhancement, classification and accuracy analysis. In the pre-processing stage, radiometric and geometric errors which were occurred in satellite imagery were eliminated. In the image enhancement processing, contrast enhancement and visual enrichment using different filters were achieved to increase the visual interpretation power.

In this study, classes are determined by the way of supervised classification. Sufficient numbers of samples were obtained to define surface properties of study area in supervised classification. Ancillary information on crop calendar and crop condition was also obtained from State Hydraulic Works (DSI) records and through informal interviews with farmers. Field sampling were systematically conducted in the paddy field during May and September in 2011. Ground data collection was organized to collect training area for various theme classes of paddy, forest, water, agriculture, pasture and roads. Afterwards, classification process was conducted by using of spectral values in selected samples. Maximum likelihood decision rule was implemented in classification process. Each pixel in image data was added to land cover category which was most similar type for pixel. After, all of data were categorized; results were taken in the form of maps and statistical tables (Lillesand and Kiefer, 2000).

Accuracy analysis was conducted to investigate suitability between assigned class for a pixel and actual class of it. The compatibility of actual class belonged pixels are obtained from existing maps and GPS measurements with classified pixels are controlled (Göksel, 1996). The samples belonged actual classes were randomly selected. As the pixel values of the sampling points which are basis of the classification may cause deviation for evaluating the results, land cover map can be useful (Erdas Field Guide, 2002).

After the process of comparison between selected pixel and reference information, accuracy of assigned classes was acquired by error matrix of classification. The error

matrix was formed to describe analyzable degree of accuracy statistically with kappa coefficient. The success of classification was determined by accuracy of producer and user.

#### **Results and discussion**

In the study area, six main classes were composed for type of land cover. These were; paddy, forest, water, reeds, roads and agriculture. Classified image of study area was obtained by using of Rapideye satellite image was given in the Figure 1.

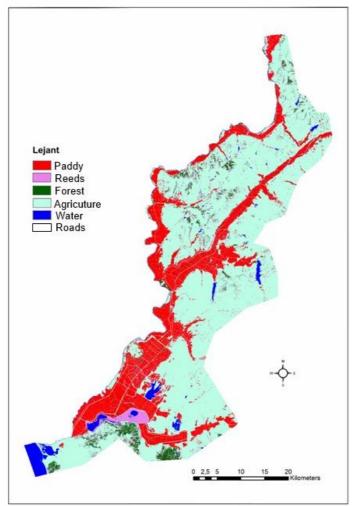


Figure1. Land use and vegetation cover distribution in 2011

As it is seen in Table 1, After classification process belonged to year of 2011, Agriculture, Paddy, Forest, Water, Reeds and Roads were determined as 131475.10 ha, 41021.18 ha, 6855.11 ha, 5618.82 ha and 1878.44 ha, 2300.10 ha respectively. Accordingly, it was seen that paddy area covered 21.7 % of study area. Accuracy assessment was derived from random selected point in order to reveal accuracy percentage of supervised classification technique. The results of accuracy assessment were given in Table 2. Overall accuracy was determines as 85.56 % and kappa statistics was 0.8232. Hyun and Jong, (2015) indicated that generally, higher spatial resolution differentiates similar crop species and

growth processes better than lower resolution in relatively small, paddy-dominated landscape areas.

8											
LUVC	Area (ha)	Percentage (%)									
Paddy	41021,18	21,7									
Agriculture	131475,10	69,51									
Forest	6855,45	3,6									
Reeds	1878,44	1,0									
Water	5618,82	3,0									
Roads	2300,10	1,21									
Total	189150	100									

Table1. Land use and vegetation cover distribution in 2011

Table 2. Accuracy assessment of land use and land cover classification

Class Name	Reference	Classified	Number	Producers	Users
	Totals	Totals	Correct	Accuracy	Accuracy
Paddy	17	16	15	88,24	93,75
Pasture	9	11	8	88,89	72,73
Water	11	11	11	100,00	100,00
Road	25	21	18	72,00	85,71
Forest	11	10	9	81,82	90,00
Agriculture	17	21	16	94,12	76,19
Totals	90	90	77		

Overall classification accuracy is 85,56 %. Overall Kappa statistics is 0,8232

#### Conclusions

In this study, it was tried that paddy cultivation areas were determined by using of highresolution Rapideye satellite image. Paddy can be detected easier on the grounds of growing under water during long period and covering land very well in the following periods. Paddy areas were determined as 41021.8 ha in the study area.

According to accuracy assessment, it was seen that Rapideye satellite image could be successfully used for this type of study. A single image was used in this study from Rapideye has led to a problem with the separation of the spectral of the mixed crop pattern due to seasonal changes. In particular spectral values of the bare agricultural fields and roads caused to confusion during classification.

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## SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

UDC: 631.8(497.11) Original Scientific Paper

# THE POSSIBILITY OF DIFFERENT CULTIVATION FIELD CROPS ON TECHNOGENIC SOIL

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#### Abstract

On the surface coal mines are the biggest surfaces degraded land. In order to preserve the land fund, it is necessary on such surfaces to take measures recultivation in order to increase soil fertility. Soil fertility is closely linked with the quality of the land and makes the ability of the soil to provide a satisfactory crop production. On the open pit coal Kolubara, which is of strategic importance for Electric Power Industry of Serbia, research in the four years was conducted in order to establish the concept of field crops production (plant species and diet in plants) in order to revitalize degraded land after the completion of exploitation coal. In the experiment were grown soybeans, corn, sorghum and sunflower, where in order to plant nutrition was used fertilizer, manure, celuflora (by-product in the production of paper) and microbiological preparations (with symbiotic and associative diazotrophs). Based on the results at the end of the four-year period, it was found that the basic agrochemical soil properties increased with the introduction of manure and celuflore, and that are smaller when worn to microbiological preparations and mineral fertilizers. The highest value of agro-chemical properties of the soil were found in the plots where are grown soybeans. In the third year of the survey were measured yields of green mass of plants in three growth stages. On the yield of green mass selection of plant species has had an impact on the level of p < 1%, a selection of nutrients is not statistical significantly affected.

Key words: cultivated field, fertilizers, recultivation, soil, yield green mass

## Introduction

In recent decades at the global level becomes more acute struggle for food production and preservation of the basic agricultural resource, land. Large areas of agricultural land have been excluded from the production after the opening of mining places. Owner and user of mined coal has a legal obligation to introduce land in the process of biological cultivation, as should agricultural land back to the original function of after the exploitation. So far, the recultivation was moving in the direction of afforestation field. The economic effect of recultivation would be much higher if on these surfaces was possible the cultivation of crops used in human and / or animal nutrition. In this regard it is important that the practices are changing negative physical and chemical properties of degraded land and be an effective

and quick. These procedures should create favorable conditions in the soil for the growth of cultivated crops, and the materials used must not pollute the already vulnerable area. Preserving the land fund for agricultural production should have primary importance for any country (Sekulić et al., 2010). Recultivation is complex mining, engineering and agricultural measures, which are used for restoration, improvement, biological productivity and the value of agricultural terrain disturbed by surface mining out (Pavlović, 2000). Recultivation creating natural habitats of plants and animals, which have the technical, environmental, economic and aesthetic function in our environment (Bauer, 2006).

In this regard have been set and the main objectives of this study to investigate the effect of different materials for various food crops, grown in tailings at landfills surface mine Kolubara.

## **Materials and Methods**

4-year (2008-2011) investigation was carried out at recultivated land RB "Kolubara". The total area with tracks was  $7185.2m^2$  (253 x 28.4 m).

(factor A): The following crops were used: Corn-(Zea mays L.) – planted in the density of 57,000 plants ha<sup>-1</sup>, Sorghum - (Sorghum bicolor L.) - planted in the density of 246.000 plants ha<sup>-1</sup>, Soybean - (Glycine hispida) - planted in the density of plants 460.000 plants ha<sup>-1</sup>, Sunflower - (Helianthus annuus L.) – planted in the density of plants 57.000 plants ha<sup>-1</sup>. Cultivars of domestic origin and seed with high germination produced at the Institute of Field and Vegetable Crops in Novi Sad were used.

(factor B): The different types of fertilizer applied on the experiment were: 1. Celuflora in quantities of 40 t ha<sup>-1</sup>. Celuflora is by-product from the pulp production industry. Chemical analysis determined the reaction alkaline pH 8.02 in KCl with a lot of high content of  $P_2O_5$  (69.2 mg.kg<sup>-1</sup>) and K<sub>2</sub>O (84.4 mg.kg<sup>-1</sup>). 2. Manure - in quantities of 40 t ha<sup>-1</sup>. 4. Microbiological - Immediately before planting were inoculated seeds of all plant species specific bacteria that are able to fix atmospheric nitrogen: symbiotic nitrogen fixers-*Bradyrhizobim japonicum* for soybean; and associative nitrogen fixers-*Azotobacter chroococcum, Azotobacter vineland* for maize, sorghum and sunflower. 5. Phosphorus, potassium, and nitrogen were applied on the whole area (600 kg.ha<sup>-1</sup> of 15-15-15 fertilizer NPK) + nitrogen fertilizer AN (34.5% pure nitrogen) equivalent to 200 kg.ha<sup>-1</sup>.

(factor C): During the development of the plants was measured green mass and  $C_1$  before flowering,  $C_2$  in the process of formation of fruits and grains and  $C_3$  at the stage of wax ripeness of maize and sorghum, forming 2/3 of pods on the plant of soybean and the formation seeds in head of sunflower. Basic agrochemical properties of the soil, according to the determination methods: the content of humus - Tjurin method of oxidation of organic matter, the content of total N - automatic method - CHNS analyzer, determination of mineral N - method Wehrmann and Scharpf, spectrophotometric determination of P<sub>2</sub>O, flame photometric determination of K<sub>2</sub>O, determination of Ca and Mg - Kappen method.

#### **Results and discussion**

In Table 1 are presented at the beginning of this barren soil characteristics of the study period. The soil had slightly acid to slightly alkaline reaction. The level of the basic parameters of productive capacity, - humus and total nitrogen, were very low. Also small amounts of nitrogen forms ( $NH_4$ ,  $NO^3N$ ) were detected. The levels of phosphorus and

potassium in the soil were different. The levels of 5.41 mg  $P_2O_5/100$  are very low and indicate that the soil requires a high amount of fertilizer, even 130-150% higher than that established based on the yield of crops. The soil was well supplied with potassium in Serbia.

1 4010 1.1	Tuble 1. Agroenement properties of the son ready for recultivation											
pF	ł	Humus	Total N	$NH_4^+-N$	NO <sub>3</sub> -N	Ca	Mg	$P_2O_5$	$K_2O$			
H <sub>2</sub> O	KC1	%	%	mg.k	mg.kg <sup>-1</sup>		kg <sup>-1</sup>	mg.kg <sup>-1</sup>				
7.56	6.70	1.04	0.14	12.6	10.2	4800	76.2	5.41	22.2			

Table 1. Agrochemical properties of the soil ready for recultivation

According to Molnar and Stevanović (1986) quantitative and qualitative changes of humus in anthropogenic soils are very slow and can be determined longtime plowed organic fertilizers, as well as plants that produce a large amount of vegetative matter that is plowed. In this way, increase the intake of plant remains, which slowly decompose, which affects microbial activity and education humus. Raising the level of soil fertility after recultivation, is a slow process and is based on the regular application of organic fertilizers. Soil incorporation of organic matter and its interactions with mineral fertilizers results in high yields. According Cvijanović et al., (2007) may be said that to achieve stable yields the most reasonable combination of organic and mineral fertilizers, whose implementation must be balanced with measures to protect the environment, which correlates with researching Kostina, N. V et al. (1995).

Using manure can be influenced to improve the physical properties of the soil, as well as increasing its biogeny. The basic characteristics of manure is high content of organic matter (usually 20-40% C on dry matter basis) low concentration of nutrients and their small accessibility. Due to the high amount of compound C was found the highest percentage of humus (1.58%) at the end of the test period. (Table 2).

plants	р	Н	Humus	Ν	C/N	NH <sub>4</sub> - N	NO <sub>3</sub> -N	Ca	Mg	$P_2O_5$	K <sub>2</sub> O
	H <sub>2</sub> O	KC1	%	%	-	mg.l	kg <sup>-1</sup>	mg.	kg <sup>-1</sup>	mg.1	00g <sup>-1</sup>
Soybean	7,57	6,44	1,89	0,21	5,22	9,50	6,76	5.562	686	6,97	38,0
Corn	7,47	6,37	1,48	0,25	5,92	12,9	6,38	5.777	637	2,99	32,7
Sorghum	7,05	5,78	1,70	0,19	5,19	22,4	10,7	5.408	782	2,97	24,5
Sunflower	7,19	5,93	1,24	0,17	4,23	26,2	3,57	5.316	708	2,77	23,5
Average	7,32	6,18	1,58	0,21	5,14	17,7	6,85	5.516	703	3,92	29.7

Table 2. Effect of fertilization manure on the basic agrochemical soil properties

Application celuflore with 28% humus, had significantly affected the level of humus in the soil (1.33%). (Table 3).

	pН		Humus	Ν	C/N	NH <sub>4</sub> -N	NO <sub>3</sub> -N	Са	Mg	$P_2O_5$	K <sub>2</sub> O
	$H_2O$	KCl	%	%	-	mg/	′kg	mg/	kg	mg/1	00g
Soybean	7,52	6,35	1,72	0,21	4,75	16,8	6,00	5.900	582	3,98	28,1
Corn	7,61	6,35	0,91	0,22	2,40	5,75	5,54	6.146	668	2,72	20,0
Sorghum	7,30	5,82	1,33	0,18	4,28	8,25	5,87	5.480	717	4,22	26,3
Sunflower	7,02	5,82	1,36	0,18	4,38	32,5	6,16	4.763	630	5,22	24,5
Average	7,36	6,08	1,33	0,20	3,95	15,82	5,90	5.572	649	4,03	24,7

Table 3. Effect of fertilization celuflorom the basic agrochemical soil properties

The use of fertilizer has a 50% impact on yields. Due to its rapid solubility plants are available and directly influence the microbial processes and yield of plants. However, due to the lack of organic matter, constant use will lead to reduced soil fertility (Vasin, 2008). In the Table 4 showed the lowest percentage of humus at the end of the test period (0.99%). This points to the fact that the necessary application of a combination of organic and mineral fertilizers.

Table 4. Effect of fertilization with mineral fertilizers on agrochemical soil properties

Plants	р	Н	Humus	Ν	C/N	NH <sub>4</sub> -N	NO <sub>3</sub> -N	Ca	Mg	$P_2O_5$	K <sub>2</sub> O
Fiants	H <sub>2</sub> O	KCl	%	%	-	mg.ł	دg <sup>-1</sup>	mg.	kg <sup>-1</sup>	mg.1	.00g <sup>-1</sup>
Soybean	7,19	5,75	0,99	0,20	2,87	12,2	3,95	4.364	650	4,81	26,6
Corn	7,52	6,33	1,01	0,17	3,45	19,8	4,23	6.761	722	3,48	28,1
Sorghum	8,03	6,60	1,04	0,15	4,02	11,0	6,72	6.423	624	2,54	24,2
Sunflower	7,75	6,37	0,95	0,17	3,24	14,4	5,59	5.962	735	2,66	22,8
Average	7,62	6,26	0,99	0,17	3,39	14,3	5,12	5.583	607	3,37	25,42

Also, it is advisable to enter a special group of microorganisms that its activities make available basic nutrients (NPK). In addition, many groups of microorganisms affect the improvement of soil structure which increases water-air properties of soil and the soil ecotoxicity (Milosević et al., 2010). Since the plant through its root system allocates certain metabolites in the land that affect the abundance and diversity of microorganisms, their number varies depending on the plant species and weight of the root system. After lysis of cells in their land remains a large amount of organic matter. When using certain groups of microorganisms likely to influence the increasing of the indigenous population, which increased biomass, as defined by the level of humus of 1.16% (Table 5).

	pН		Humus	Ν	C/N	NH <sub>4</sub> -N	NO <sub>3</sub> -N	Ca	Mg	$P_2O_5$	K <sub>2</sub> O
	$H_2O$	KCl	%	%		mg.l	kg <sup>-1</sup>	mg	.kg <sup>-1</sup>	mg.1	00g <sup>-1</sup>
Soybean	7.78	6.68	3.83	0.27	8.22	11.9	10.6	6085	562	14.9	28.8
Corn	7.53	6.33	1.74	0.19	5.31	10.8	6.31	5777	695	5.13	27.4
Sorghum	7.78	6.53	1.61	0.19	4.92	11.1	10.6	6453	600	4.33	30.8
Sunflower	7.56	6.16	1.28	0.17	4.36	11.2	4.21	5009	630	3.40	23.1
Average	7.66	6.42	1.16	0.20	5.70	11.25	7.53	5831	622	6.94	27.25

Table 5. Effects of fertilization on microbial fertilizer agrochemical soil properties

Yields of green mass (t.ha<sup>-1</sup>) depend on is types of crops and utilization phase (Table 6). Statistically significant differences at the level p < 1% were found with plants. The lowest yield was in soybeans in the first stage of exploitation and the largest was a sunflower in the third stage. Influence of fertilizers on yields is not statistically significant. The

interaction between the factors AC (types of crops and development phases) was statistically significant at p < 1%. Similar results have come Licina et al. (2004) in studies of growing lettuce on various types of tailings and different sources hraniva.primena liquid organic fertilizers and combining them with other materials increased significantly lettuce yield. Way of nutrition of plants with the addition of flavoring land, even in problematic tailings can increase their productive capacity.

A-plants		B- types of fertilizers			C- dev	AB			
					C <sub>1</sub>	C <sub>2</sub>	C <sub>3</sub>		
		B <sub>1</sub> .NH <sub>4</sub> NO <sub>3</sub>				3.67	6.67	13.10	7.81
A		B <sub>2</sub> -celuflora				2.78	9.67	15.20	9.22
Soybe	ean	B <sub>3</sub> manure				8.83	12.44	16.10	12.46
		B <sub>4</sub> - microbiological				6.83	14.00	17.80	12.88
A	1		AC			5.52	10.67	15.55	10.59
		B <sub>1</sub> .NH <sub>4</sub> NO <sub>3</sub>				17.89	21.00	29.00	22,63
A <sub>2</sub>	2	B <sub>2</sub> .celuflora				15.67	18.44	25.56	19.89
Cor	'n	B <sub>3</sub> _manure	15.67	17.89	22.89	18.82			
		B <sub>4</sub> - microbiologica	17.44	23.11	25.33	21.96			
A <sub>2</sub>	2		AC			16.66	20.11	25.69	20.82
		B <sub>1-</sub> NH <sub>4</sub> NO <sub>3</sub>				13.00	27.20	31.10	23.77
A	3	B <sub>2</sub> -celuflora				10.20	16.11	26.00	17.44
Sunflo	ower	B <sub>3</sub> _manure				19.67	25.00	30.10	24.92
		B <sub>4</sub> - microbiological				11.78	27.89	33.20	24.29
A <sub>3</sub>	3	AC				13.66	24.05	30.10	22.60
		B <sub>1-</sub> NH <sub>4</sub> NO <sub>3</sub>				11.67	21.56	29.89	21.04
A <sub>4</sub>	Ļ	B <sub>2</sub> -celuflora				17.00	20.89	22.32	20.07
Sorgh	um	B <sub>3</sub> _manure				14.11	21.36	24.99	20.15
		B <sub>4</sub> - microbiological				16.56	20.55	22.65	19.92
A <sub>4</sub>	Ļ		AC			14.83	21.09	24.96	20.29
		B <sub>1</sub>				11.56	19.11	25.77	18.81
D	r	B <sub>2</sub>				11.41	16.28	22.27	16.65
BC	- -	B <sub>3</sub>				14.57	19.17	23.52	19.09
		B <sub>4</sub>				13.15	21.39	24.75	19.76
С						13.15	18.98	24.07	18.57
Level		А	В	С	AB	А	C	BC	ABC
F-test		**	ns	**	ns	*	*	ns	ns
LSD	5%	4.1	3.9	1.9	9.1			.2 4.8	
LSD	1%	5.4	5.2 2.6 13.1			5	5.9		25.9

Table 6. Impact of crops, fertilizers and stages of development on green mass yield (t.ha<sup>-1</sup>)

#### Conclusions

Based on the results, the production of field crops can be organized on land that is recultivated. On the height of the yields of green mass very significantly affected by the type of forage crops and exploitation phase and their interactions. The choice of measures in recultivation repairs substrate mainly linked to a type of culture that will be on this land to cultivate. Considering the fact that the research was at the worst soil properties, such as chemical and physical in relation to other developed surfaces in RB "Kolubara" it can be concluded that with intensive measures of production, a combination of organic material and mineral fertilizers, it is possible to positively influence the process of humification in the soil.

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## PHYSICAL PROPERTIES OF CALCIC CHERNOZEM INFLUENCED BY IRRIGATION

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#### Abstract

The complementary character of irrigation over the precipitation, in Vojvodina province northern part of Serbia, requires a correct on-site irrigation management depending on plant needs, precipitation and soil properties. In these conditions the soil is exposed to higher water influence. Despite its positive impact irrigated agriculture can also have negative influence on natural resources, especially soil. Inappropriate watering regime can lead to deterioration in physical properties of soil. In order to investigate how long term irrigation affects the carbonate chernozem in Vojvodina region, a field survey was conducted at the Experimental field Rimski Sancevi, Department of Vegetable Crops, Institute of Field and Vegetable Crops, near Novi Sad. According to World reference base for soil resources, the examined soil is classified as calcic chernozem. The analyzed soil samples were taken from the irrigated as well as the rainfed plots, from the soil profile, by horizons. The quality of water used for irrigation was found to belong to C3S1 (U.S. Salinity Laboratory). Basic physical properties of soil were analyzed (mechanical composition, bulk density, specific weight, porosity, soil permeability (k-Darcy) and soil water retention at -33 kPa, -625 kPa and -1500 kPa). The investigated soil is characterized by uniform mechanical composition. Somewhat smaller percent of particles <0,002mm were found in irrigated soil. Long term irrigation had no significant influence on bulk density and soils porosity in the surface layer, as well as on specific weight and k-Darcy coefficient in the whole soil profile. Although established higher values of water retention on non irrigated soil, they are within the limits characteristic for the investigated soil type. Based on the obtained results it can be concluded that irrigation had no influence on physical properties of investigated soil. However, an application of basic principles of rational irrigation, as well constant monitoring of irrigation water quality is obligatory. Key words: irrigation, soil, physical properties

#### Introduction

Irrigation is an important factor in the development of agriculture. It is a prerequisite for plant production with the basic aimof increasingyield thatare, according to Bruinsma, (2003) generally higher in irrigated agriculture than inp roduction without irrigation, in order to reduce the effect of drought. And beyond, irrigated agriculture is closely related to the food security, as well as poverty reduction in developing countries (Rosengrant et al., 2009). Despite the positive economic and social impacts, negative effects of irrigated agriculture on natural resources, especially over-exploitation, pollution and degradation of

water and land are inevitable (Özerol et al., 2012). Although it is a powerful factor of changes in ecological conditions, incompetently managed irrigation can cause negative processes in the soil and its deterioration (Issaeva and Parfenova, 2004).

In irrigated conditions it is necessary to synchronize a large number of factors in order to avoid adverse effects to soil. The specificity of production under irrigation requires a corresponding complex of technological process, which is different from the measures applied in the cultivation of crops in rainfed conditions (Kresović and Tolimir, 2009). Minashina (2009) stated that long term irrigation of steppe soils in the South of Russia caused deterioration of soil productivity, by the improper design of irrigation systems, on the one hand, and by the low tolerance of chernozems toward increased moistening upon irrigation, on the other hand. The authors analyzed the factors and regimes of soil formation under irrigated conditions and concluded that irrigation induced changes in the soil hydrology also lead to changes in the soil physicochemical and other properties. The deterioration, of irrigated chernozems was related to the absence of adequate experience in irrigation of steppe soils, unskilled personnel, improper regime of irrigation, and excessively high rates of watering. To improve the situation, the training of personnel is necessary, as well as improved irrigation regime. Korolev, (2007) came to similar findings, stating that irrigation, which is widespread in the central region of Russia Plain, often causes undesirable changes in the physical properties of chernozem. They have found that irrigated and non irrigated soil differ in density, porosity, and conclude that irrigation has caused the changes of the basic properties of the soil. According to Khitrov (2008), irrigation leads to the change of texture, especially in the upper layers of the chernozem. Gajic (1999) found a significant deterioration of the physical and mechanical properties of irrigated chernozem compared to rain fed, due to a significant decrease in calcium carbonate and humus. The same authors found that arable horizon of irrigated soil was susceptible to deformities due to processing in the wet state and has a greater tendency to form crust due to the instability of structural aggregates, and concluded that irrigated chernozem has a much lower carrying capacity.

However, it is not necessary to agricultural practices always have a negative impact on soil quality and productivity. It is possible through management practices of soil and water quality and nutrient to improve or maintain good soil quality and sustain productivity at the same time (Sahrawat et al., 2010). According to Burcea et al., (2013) the correct application of irrigation water of good quality, can have an indirect positive effect on the physical properties of soil by stimulating the natural processes of wetting and drying.

Irrigation in Vojvodina province has a complementary character (Pejić et al., 2012) over the precipitation, and requires a correct on-site management depending on plant needs, precipitation and soil properties. In view of the opposite stand point, the investigation was performed on the chernozem soil which is according to Nešić (2011) predominant soil type in the region and the highest quality soil. The aim of this paper was to determine the influence of long-term irrigation on physical properties of chernozem in Vojvodina province.

## Material and methods

Research on the effect of irrigation on the physical properties of chernozem, was carried out during 2011, at the Experimental Field of the Institute of Field and Vegetable Crops, Novi Sad, in Rimski Šančevi, Vojvodina province. A field survey was performed, specific sites where chosen and soil pits were dug. Pedological profiles were opened to the depth of 2 m, and the external and internal morphology was described. The soil was classified as calcic chernozem according to WRB (FAO, 2006). Soil samples were collected from the soils irrigated (Irr) by sprinkling from 1946 and also from locations that were not irrigated, rain fed (R). Intact and disturbed soil samples were taken from the six profiles, three from irrigated and three from rainfed soil plots, from the middle of established genetic horizons. The samples were submitted to laboratory analyses.

The following parameters were analyzed: mechanical composition according to pipette method with preparation in sodium pyrophosphate, texture class according to Tommerup, bulk density with Kopecki cylinders of 100 cm<sup>3</sup> of volume, specific weight by Albert-Bogs method with xylol, total porosity was calculated based on the values of bulk density and specific weight, soil permeability (k-Darcy) was done with constant pressure and soil water retention at -33 kPa using the Porous plate and at -625 kPa and -1500 kPa using the Pressure membrane apparatus (calculated in mass%).

Statistical analysis of experimental data was accomplished by standard analysis of variance (ANOVA) and Duncan's LSD test using the Statistica 12.0 software package. The data presented in the tables and graphs represent the average values retrieved from three replicates.

## **Results and discussion**

The quality of irrigation water plays an important role in crop production and has a great influence on physics and chemical properties of soil (Jalal and Merrikhpour, 2008). The quality of water used for irrigation at the investigated locations belongs to C3S1, according to U.S. Salinity Laboratory. Although the water is high-salinity, it can be used on soil with adequate drainage, but salinity control may be required.

Based on the distribution of mechanical fractions the investigated chernozem is characterized by heterogeneous mechanical composition. The fraction of total clay (clay and dust) occurs with the significantly highest percentage throughout the soil profile, except in the C horizon in the irrigated variant. The fraction of coarse sand is the least present throughout the soil profile (figure 1. and 2.). In the arable horizon fine sand is the dominant fraction in irrigated variant and with somewhat the same share as clay fraction in rain fed variant. This horizon is classified as loamy clay. The transitional AC horizon of the irrigated chernozem has a smaller share of clay and dust than the rain fed variant, and belongs to clay loam and loamy clay respectively. Dust is the dominant fraction in C horizon in rain fed variant (in both C1ca and C2G), and in the Cca horizon in the irrigated variant, while in the C horizon the percentage of fine sand is the highest. The C horizon is classified as clay loam and fine sandy loam in irrigated and as dusty clay in rain fed horizon. Significantly smaller percentage of clay was found in the surface layer of irrigated soil, but no detrimental effect of irrigation was found. These results are in accordance to results of Belić et al., (2003), who found no significant changes of chernozem properties in irrigated conditions, i.e. no significant difference in distribution of mechanical fractions throughout the soil profile. The same author stated that reduced content of clay fractions in the surface layer may be a consequence of irrigation water containing soda, which causes clay minerals to exceed the dispersed state, causing their migration.

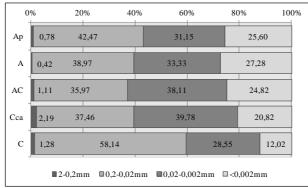


Figure 1. Average values of mechanical composition of irrigated soil

0	% 20	20%		60	60%		%	100%
Ap	0,23 32,83			32,23		34,71		
A	0,15 32,00			34,93		:	32,94	
AC	0,49 31,26			39,70			28,55	
C1ca	0,88 20,26		5	1,24			27,63	
C2G	0,20 22,56			51,97			25,19	
	∎2-0,2mn	n ■0,2-0	0,02mm	■0,02-0,	002mm	□<0,	002mm	

Figure 2. Average values of mechanical composition of rainfed soil

The determined bulk density values (Table 1) were in average for the soil profiles 1,49 and 1,57 g/cm<sup>3</sup>, and specific weight 2,62 and 2,61 g/cm<sup>3</sup>, on irrigated and rain fed soil, respectively. The bulk density of the investigated soil samples in arable soil layer show an increased compaction, on both irrigated and rain fed soil, and increases with the soil depth. No statistical significance was found between bulk density values in both Ap and A horizon of irrigated and rain fed soil. These values were significantly higher in deeper soil layers of rain fed variant. Irrigation did not influence the values of specific weight throughout the soil profile. The average values of total porosity are very low; the arable horizon is poorly porous. These values correspond to higher values of bulk density. Irrigation had no effect on total porosity, except in C horizon, but these differences were not assigned to irrigation, but to the different properties of substrate.

Horizon	Bulk density (g/cm <sup>3</sup> )		Specific we	ight (g/cm <sup>3</sup> )	Total porosity (%)		
	Irr	R	Irr	R	Irr	R	
Ар	1,48	1,45	2,54	2,48	39,18	38,17	
А	1,46	1,51	2,55	2,56	40,53	38,77	
AC	1,41	1,47	2,64	2,63	46,52	44,03	
Cca/C1ca	1,55	1,69	2,65	2,67	41,52	36,75	
C/C2G	1,54	1,73	2,73	2,73	43,34	36,37	
average	1,49	1,57	2,62	2,61	42,22	38,82	

Table 1. Pysical properties

The results of investigation are compliant with the results of Burcea et al., (2013), who reported that after long-term irrigation of maize soil compaction vary in a wide range of 1,23 to1,45g/cm<sup>3</sup> for the layer of soil up to40 cm, while the irrigated land ranges from 1.21 to 1.41g/cm<sup>3</sup>. The authors found no effect of irrigation for investigated soil layer, except at a depth of 10-20 cm, but point out the indirect effects of irrigation, that is, tillage performed at slightly higher soil moisture as well as reducing the stability of structural aggregates, which caused a greater susceptibility to soil compaction. The values of total porosity correlated with the values of density and ranged from 49 to 55%. The higher compression of the chernozem and reduced total porosity under the influence of irrigation, especially in arable layer, but also due to intensive impacts of agricultural machinery on the land during the growing season was found by many authors (Challa, 1987; Zamfir et al., 2003; Bezuglova and Yudina, 2006; Korolev2007; Walker and Lina, 2008; Kalinitchenko et al., 2011). According to Buoraci Bašić, quoted by Gajić et al., (2004) soil compaction is one of the major problems of modern intensive land use in agricultural production due to the increased use of heavy machinery as well as additional processing and transportation of the soil. Studying the impact of longterm use of agricultural machines ina gricultural production Gajic et al., (2004) concluded that there was a significant increase of compaction of meadow black soils in the layer up to 30cm.

The investigated soil was medium to very permeable, in both variants. Also no significant differences were found, somewhat higher values of percolation rate (Figure 3) were established in rain fed soil (up to  $3,34\cdot10^{-3}$ cm/s) in arable horizon compared to the irrigated soil ( $3,94\cdot10^{-4}$ cm/s). Values of percolation rate mainly depend on soil mechanical composition and porosity (Bošnjak, 1999) and are in compliance with the results of Vučić (1964), who determined the rate from 5,8  $10^{-3}$  to 1,9  $10^{-4}$ cm/sfor carbonate chernozem of loess terrace. These results are in consistence to those obtained by Belić et al, (2005), who found intermediate values of water permeability in the order of  $10^{-3}$ , in both irrigated and rain fed chernozem, similar in all soil layers except of topsoil were it was slightly smaller.

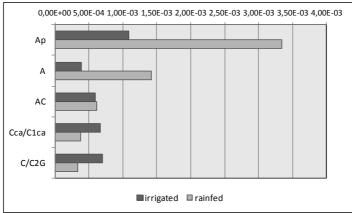


Figure 3. Average values of soil permeability (k-Darcy)

Obtained soils water retention values (Table 2) at 33 kPa correspond to water retention capacity, at -625 kPa to lento capillary moisture and at -1500 kPa correspond to permanent wilting point. The highest statistically significant values of water retention were measured on all horizons on rain fed variant, which can be attributed to somewhat higher percentage of clay in investigated soil layers. The highest values were obtained in arable layer in both

variants, which decreases with the increase in soil depth. The results of the soils water retention are typical for the investigated soil and are in compliance with the data obtained by Vučić (1964), who stated that the water holding capacity in chernozem of the loess terrace amounts to 25-27%, lentocapillary moisture amounts to 16% and permanent wilting moisture to 11-13% for A and AC horizon.

Horizon	-33 kPa (%)		-625 k	Pa (%)	-1500 l	kPa(%)
	Irr	R	Irr	R	Irr	R
Ар	24,88	29,70	14,29	17,54	12,54	15,58
А	27,11	30,45	15,63	18,68	13,55	16,45
AC	26,67	28,94	14,58	16,40	11,06	13,96
Cca/C1ca	24,68	27,52	11,12	14,81	9,25	12,21
C/C2G	15,66	26,70	5,90	13,71	5,06	11,22
average	23,80	28,66	12,30	16,23	10,29	13,88

Table 2. Soil water retention

Water retention is correlated with the content of the clay, that is the value of retention increases where the clay content increases (Challa, 1987). According to Markoski and Mitkova (2012) differences in retention of water depends on the concentration of clay and clay minerals as well as humus content. Al-Rumikhani (2002) studied the changes in water capacity of sandy loam irrigated by center pivot, for a long term period and found that the volume of water decreased with increasing soil depth. A similar tendency was found in permanent wilting point. The differences in the values of permanent wilting point was attributed to the greater volume of micropores in samples within irrigated soil, compared with a smaller volume of micropores outside of irrigated soil. Reduction of these values with soil depth is mainly due to a decrease in the clay in the deeper layers of the soil, thus reducing the water content in the layers that have less micropores.

#### Conclusions

Based on the results obtained in this research as well as the results obtained by other authors the following conclusions can be drawn:

Long term irrigation did not influence the mechanical composition, that is, the distribution of mechanical fractions throughout the investigated soil profiles. However, smaller percentage of clay was found in the surface layer of irrigated soil.

The bulk density of the investigated soil shows substantially increased compaction, on both irrigated and rain fed soil, and increases with the soil depth. No statistical significance was found between bulk density values in arable horizon of both irrigated and rain fed soil, which points to the great influence of agricultural machinery on investigated soil. Irrigation did not influence the values of specific weight throughout the soil profile. The values of total porosity correspond to values of increased bulk density. The investigated soil is poorly porous, especially the arable horizon. The investigated soil was medium to very permeable, in both variants, with somewhat higher values of percolation rate in rain fed soil. Obtained soils water retention values are typical for the investigated soil. Higher values of water retention were determined on rain fed soil, which can be attributed to somewhat higher percentage of clay. The results of the investigation imply that irrigation had no influence on physical properties of investigated chernozem soil.

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# SOIL PROPERTIES IN TOBACCO PRODUCTION IN CROATIAN PODRAVINA AND POŽEGA VALLEY

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#### Abstract

In Podravina and Pozega Valley in Croatia, tobacco is an agricultural culture of particular interest and importance, with very long tradition. However, tobacco production takes place on soils with very different physical and chemical properties, which directly affects the yield and quality of tobacco. For the purpose of better knowledge of soil properties that are used in regular tobacco production in Croatia and the needs to remove present restrictions, it was organized during three-year (2006-2008) field and laboratory soil research. Soil properties of 72 locations were analyzed in total and it has been found that the production of tobacco during the three-year period took place mainly on 9 soil types, with the 17 lower pedosystematics units. A soil type luvisol was identified on most locations (58%), with the most represented pedosystematic unit of luvisol on loess typical. Confirmed data show that the soils on which tobacco is grown in the studied areas of Croatia are extremely heterogeneous in their physical and chemical properties, and that the majority of them have limited suitability for growing tobacco. Among other things, a larger part of soil is partly unsuitable for growing tobacco due to unfavorable soil reaction (pH), unfavorable humus content and unfavorable soil texture. Summing up the above mentioned data and the results of chemical and physical properties of soil on family farms in the area of flue-cured tobacco cultivation, it could be concluded that it is necessary to calcify most of the soils, to correct humus content with agrotechnical measure and avoid heavy soil texture wherever possible, especially on soils which are occasionally over-moistened. Key words: Tobacco, soil types, soil properties, Croatia

#### Introduction

In Podravina and Pozega Valley in Croatia, tobacco is an agricultural culture of particular interest and importance, with very long tradition. However, tobacco production takes place on soils with very different physical and chemical properties, which directly affects the yield and quality of tobacco. Therefore, systematic field and laboratory studies of soil in tobacco production were carried out in Croatia during the period from 2006-2008, in order to come to a better knowledge of the soil properties used in regular production of tobacco, and the needs to remove the present restrictions. (Husnjak *et al.*, 2006-2008). The aim of this paper is to show the soil properties in tobacco production in Croatia and point to the need for agroglomeration measures for soils arrangement for the purpose of more intensive production of tobacco.

## Materials and methods

Sampling and determination of the soil types was carried out during the regular production of tobacco in 48 representative family farms in the Podravina and 24 family farms in Pozega valley, figure 1.

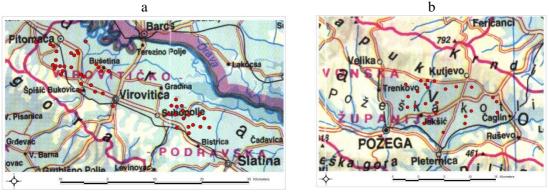


Figure 1: Locations of soil sampling on Podravina (a) and Pozega Valley (b)

At each location (area  $6 \times 6$  m), a half-pedological profile was dug, and soil samples were taken for determination of soil physical and chemical properties from plough and sub-plough layer. Analysis of particle size of distribution and chemical properties of soil (soil reaction (pH), humus, carbonate and total nitrogen content, as well as plant available phosphorus and potassium) were carried out under standard ISO laboratory methods:

- Treatment of soil samples for analysis (ISO 11464 : 1998),
- Determination of particle size distribution- method by sieving and sedimentation, modified with Na-pyrophosphate (ISO 11 277:1998)
- Determination of soil reaction (ISO 10390 : 2005)
- Determination of soil carbonate- Gas-volumetric method (ISO 10693: 1995)
- Total N in soil- modified Kjeldahl method (ISO 11261: 2004)
- Humus content in the soil-dichromate method by Tjurin (Tjurin, 1937)
- Plant available phosphorus and potassium- Ammonium lactate method, according to Egner- Riehm Domingo (Egner et al., 1960)

Bright Virginia type of tobacco was growing on all farms.

#### **Results and discussion**

Analyzing the soil classification in some locations, it has been found that the production of tobacco took place mainly on 9 soil types, with the 17 lower pedosystematics units, table1. Luvisol types were determined on most locations (49 or 58%) where tobacco is grown, wherein the most prevalent pedosystemic unit is luvisol on loess typical, next are luvisol on loams typical and luvisol on sandy loess. According to the representation of the soil units, more prevalent distric cambisol occurs in seven locations (about 10%), and eutric cambisol has appeared at four locations. Other soil types are equally represented as they occur at the 3 locations.

Table 1. Number	of location according to soil types presented in to						
Soil type	Lower systematical soil units	]	Number	of locati	ons		
Son type	Lower systematical son units	2006	2007	2008	Total		
Rendzinas	on calcareous sand	1	1	1	3		
Ranker	on sand			3	3		
Entrio	on loess, luvic	2			2		
Eutric Cambisols	on loam, luvic			1	1		
Cambisois	on loam, typical		1		1		
Distric	on silicateous sand, typical	1		1	2		
Cambisols	on siliceous calcareous deposits, typical	2		2	4		
Cambisois	on siliceous loamy deposits, luvic			1	1		
	on loess, typical	8	5	4	17		
	on loess, typical with sand beds	1	2	3	6		
Luvisol	on loams, typical		7	5	12		
LUVISOI	on loess, pseudogleyic		1	1	2		
	on sandy loess, typical	4	6	1	11		
	on gravel silicate deposit	1			1		
Pseudogley-gley	typical	2	1		3		
Fuglava	hypogleyic	1		1	2		
Eugleys	amphigleyic, vertic	1			1		

Table 1. Number of location according to soil types presented in tobacco production (2006-2008)

The soil properties in tobacco production in Croatia are shown on the base of a detailed analysis of the number the locations and important physical and chemical properties of the soil, which have a crucial influence on the yield and quality of tobacco, table 2.

Analyzing data related to soil reaction, it is possible to conclude that tobacco production in Croatia mainly takes place in very acid soils. Specifically, the total number of locations, among them 70% present a very acid soil, with about 20% of locations are acid soils, at 4% of the locations are slightly acidic soils, and at about 7% of the locations are neutral and less alkaline soils. As far as contents of humus, tobacco is predominantly (46% of locations) grown on very poorly humic soils (0.5-1.5% humus), then the poorly humic soils (1.5-2.5% humus) that are represented at 44% of locations, then the medium to good humic soils (2.5-3.5% humus) which are represented on 5% of locations, and at least on an extremely poorly humic soils (<0.5% humus), which are represented on only 4% of the locations.

1 4010 2	. I mary	on nui	noer the	locations	in order	to son properties in prough son layer					
Soil re	action	Hur	nus	Nitrogen		Soil te	xture	Phosp	horus	Potas	sium
		con	tent								
pН	No.	%	No.	%	No	Class*	No	mg/	No.	mg/	No.
KCl	locat.		locat.		locat		locat.	100 g	locat.	100 g	locat.
<4,5	50	<0,5	3	<0,06	9	S	1	<5	7	<5	1
4,5-5,0	7	0,5-1	12	0,06-0,1	41	LS	2	5-10	18	5-10	4
5,0-5,5	7	1-1,5	21	0,1-0,2	19	SL	16	10-15	21	10-15	18
5,5-6,0	2	1,5-2	24	0,2-0,3	2	Si,L,SiL	47	15-20	12	15-20	28
6,0-6,5	1	2-2,5	8	>0,3	1	SiCL	4	20-25	5	20-25	14
>6,5	5	2,5-3,5	4			SiC	2	>25	9	>25	7

Table 2: Analysis of number the locations in order to soil properties in plough soil layer

\*Legend of texture class: S-sand;LS-loamy sand; SL-sandy loam; Si-silt; SiL-silty loam; L-loam, SiCL-silty clayey loam; SiC- silty clay

From the aspect of the nitrogen content, tobacco is predominantly grown on 57% of locations of moderately poor soils (from 0.06 to 0.1% nitrogen), then on a well-supplied soils with nitrogen (0.1-0.2% nitrogen) which are represented on 26% of locations, then on poor soils (% nitrogen <0.06), which are represented at 12% of locations, and at least in rich soils supplied with nitrogen (nitrogen% > 0.2), which are represented on only 4% of the locations. Analyzing data related to the texture of the soil, it is possible to conclude that tobacco production in Croatia mainly takes place on soils with silty loam texture. Specifically, total number of locations, among them 65% present the silty, silty loam and loamy texture. Furthermore, in 22% of locations, texture of the soil is sandy loam, in 4% is loamy sand or sand, and in about 8 percent of the texture is heavier, clayey and silty loam clay. Considering the content of nutrients phosphorus and potassium, it has been found that the tobacco supply with these nutrients is mostly moderate (10-20 mg / 100 g), followed by low phosphorus and rich in potassium. The minimum number of location refers to the soil is very poor supply and the soil richer supply. Virginia tobacco is grown on different soil types, or soils with different suitability. According to the literature (Collins 1993; Cristanini 2005; Tursic and Kozumplik 1984; Tursic 1989; Cosic 1998) this type of tobacco grows best in sandy loamy soils with low humus content, ie. organic matter (0.7 to 1.3%), slightly acid reaction (pH in KCl 5.5 - 6.5), very persistent looseness, good drainage, and the stability of structural aggregates. On soils with higer ratio of sand, the yield of tobacco is lower, and its quality is often lower because of the difficulty of maintaining the necessary soil moisture and nutrient supply. On the hard clayey, silty clayey and silty clay soils, the yield is much higher, but due to the increased supply of soil nitrogen quality of tobacco is significantly reduced. Confirmed data show that the soils on which tobacco is grown in the studied areas of Croatian, are extremely heterogeneous in their physical and chemical properties, and that the majority of them have limited suitability for growing tobacco. Among other things, a larger part of soil is partly unsuitable for growing tobacco due to unfavorable soil reaction (pH), unfavorable humus content and unfavorable soil texture. Summing up the above mentioned data and the results of chemical and physical properties of soil on family farms in the area of flue-cured tobacco cultivation, it could be concluded that it is necessary to calcify most of the soils, to correct humus content with agrotechnical measure and avoid heavy soil texture wherever possible, especially soils which are occasionally over-moistened.

# Conclusion

Soil type Luvisol was determined on most locations (49 or 58%) on which tobacco is grown, and the most representative pedosystemic units were luvisol on loess, typical. After that follows Distric Cambisol (7 locations or 10%), with the most common soil type Distric Cambisol on siliceous loamy deposits.

Tobacco production is carried out mainly on soils with silty-loam texture, very acidic and very poorly humic and on soils moderately poor with nitrogen and moderately supplied with phosphorus and potassium.

Analyzing data show that the soils on which tobacco is grown in the studied areas of Croatia are extremely heterogeneous in their physical and chemical characteristics and that the majority of them are of limited suitability for growing tobacco. Therefore, for the intensive cultivation of tobacco, it is necessary to calcify most of the soils, to correct humus content with agrotechnical measure and avoid heavy soil texture wherever possible, especially soils which are occasionally over-moistened.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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UDC: 633.18-18(560) Original Scientific Paper

# EFFECT OF FOLIAR SILICON APPLICATION ON RICE PLANTS IN IPSALA PLAIN

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#### Abstract

Silicon (Si), uptaken in high quantity by rice plants, is one of the main nutritional elements. Effects of foliar applications of 0,1000,1500,2000 and 2500 cc/ha doses of silicic acid (Si(OH)4) with the concentration rate 28g/L on Edirne and Osmancik-97 rice varieties were investigated in this study. Applications were performed twice after tillering and florescence. Trial was set up according to a split plots (100 m<sup>2</sup> (5x20m)) in a randomized complete blocks design (RCBD) with four replications in Ipsala plain of Edirne province in 2014. As a basic fertilizer 400 kg/ha 15:15:15 NPK was firstly used in the plots. As top dressing 250 kg/ha ammonium sulfate (AS) 21% and 200 kg/ha AS were applied at onset of tillering and at panicle stage of rice plants, respectively. Silicon application was found as significant (P<0.01) on the whole properties investigated. Decline of the properties (plant height, panicle length, grain number per panicle, neck blast and lodging) and enhance of the properties (number of panicle per square meter, yield, 1000 kernel weight, milling rate) were determined with increasing of silicon doses. Differences between the varieties tested were significant for the properties (plant height, number of panicle per square meter, neck blast, yield, output of 1000 kernel weight and lodging rate). The study suggested that optimum silicon dose for rice plants was 2000 cc/ha for Ipsala condition. Keywords: Neck blast, quality, rice, silicon, yield

#### Introduction

Rice is one of the most important foods for nutrition, providing 21% of energy and15% of protein for human(Zibaee, 2013). Global rice production in 2012 was 738.1 million tonnes with an average yield of 4.5 tonnes per ha in a 162.3 million ha area. (Anonymous, 2014). However, Turkey has 98,000 ha of paddy rice plantation, half of which isin the Thrace and Marmara regions. The most productive region is Thrace, which covers 10-15 percent of Turkey's total rice plantation area. Ipsala district in the Edirne region alone produces 20,000 ha of paddy rice. The most preferred varieties of rice are the Calrose, Baldo and Osmancık in Turkey (Karabina, 2015). Plant nutrition is an important factor affecting yield and quality. In the last few years, rice yield has been diminishing, one of the major reasons of which is nutritional imbalance. Although required in a minute quantity, micronutrients are important as much as macronutrients (Khuang et al., 2008; Esfahani et al. 2014). Among all the micronutrients assimilated by plants, silicon (Si) alone is consistently

present at concentrations similar to macronutrients. Its concentrations in different plants range from 0.1% (similar to P and S) to more than 10% of dry matter of whole plant (Epstein, 1999). Hydrated amorphous silicon compounds are likely to be deposited in different cellular parts such as cell lumens, cell walls and intercellular spaces. Silicon is an important micronutrient for healthy and competitive growth of all cereals including rice (Brunings et al., 2009). Silicon is absorbed by plants in the form of silicic acid in the same magnitude as other micronutrients. It is also the only beneficial element that does not cause toxicity or serious injury to plants under excessive amounts (Synder et al., 2007). Effects of Silicon on yield are related to the deposition of the element under the leaf epidermis which triggers a physical mechanism of defense and accordingly production of phenols, which stimulates phytoalexin production, reduces lodging, decreases transpiration losses and increases photosynthesis capacity and decreases transpiration losses of crop plants. Plant tissue analysis revealed that the optimum amount of silicon is necessary for plant development (Korndorfer et al., 2004; Datnoff and Rodriguez, 2005; Liang et al., 2006; Ning et al., 2014). Thus, the present study was conducted to explore the effect of foliar application of Si on yield and yield components of Osmancık and Edirne (Baldo)rice cultivarscommonly grown in Turkey.

# Material and methods

*Ecological Properties of the research area*: Soil samples taken from the experiment field were analysed at soil laboratory in Keşan, Edirne. Concentrations of macro and micro nutrients of the samples were N (0.07%), P (4.59 ppm), K (101.78 ppm), Ca (4.772 ppm), Mg (447.04 ppm), Na (36.27 ppm), Cu (1.57 ppm), Fe (36.53 ppm), Mn (69.99 ppm), Zn (0,34 ppm). Soil structure was clay-loam (53) and other properties were pH (7.05), organic matter (1.34%), total salt (0.14%), total lime (4.43%).Meteorological data belonging to the period of growing season of 2014 were given in Table 1.

Variable	April	May	June	July	August	September
Minimum temp. (C <sup>o</sup> )	3.6	4.8	10.9	14.3	13.6	6.8
Maximum temp.(C <sup>o</sup> )	25.0	31.1	33.6	34.6	36.5	31.1
Average temp.(C <sup>o</sup> )	14.1	18.6	22.3	25.3	25.6	19.6
Precipitation (mm)	48.7	89.0	88.5	97.8	12.7	105.3
Relative Humidity (%)	74.6	68.7	67.2	61.9	61.0	71.4

Table 1.Monthly temperature, precipitation and relative humidity of the experimental area\*

\*Regional Meteorology Station, Edirne climate station

*Application and Experimental design*: Foliar applications of 0,1000,1500,2000 and 2500 cc/ha doses of silicic acid (Si(OH)4) with the concentration rate 28g/L on *Edirne* and *Osmancik-97* rice varieties were performed twice after tillering and florescence by spraying on leaves. Trial was established according to a split plots (100 m<sup>2</sup> (5x20m)) in a randomized complete blocks design (RCBD) with four replications in Ipsala district of Edirne province in 2014. Rice seeds were sown on May 14th in the experiment field and harvested on October 1th. As a basic fertilizer 400 kg/ha 15:15:15 NPK was firstly applied to the plots. In addition, As top dressing 250 kg/ha ammonium sulfate (AS) 21% and 200 kg/ha AS were used at the beginning of the tillering and at the panicle stage of rice plants, respectively.

Assessment of the plants: The same cultural treatments such as ploughing, cultivation seed rate, fertilizers and weed control were implemented for all the plots. With maturing of the plants, the traits (plant height, panicle number per square meter, panicle length, seed number per panicle, neck blast rate and lodging rate) were measured. After harvest yield, 1000 kernel weight, total milling yield, net miling yield were also determined.

*Statistical analysis*: The values obtained from the trial were subjected to analysis of variance using JMP statistical software and differences among means were compared using LSD at 5% probability level.

## **Result and discussion**

Variance analysis table of investigated characteristics was given in Table 2. Statistical analysis showed that the effect of Si application on whole properties were significant at 1% level of probability. In the present study, with increasing silicon doses, plant height, panicle length, seed number per panicle, neck blast and logging rate decreased at the rates of 11-14%, 7-26%, 18-31%, 19-50%, 25-64%, respectively. Whereas increasing doses of silicon reduced the panicle number (5-31%), yield (0,4-24%), 1000 kernel weight (0,8-4,9%), total milling yield (0,4-2,5%), net milling yield (0,8-5%). Due to the increase in the number of panicles per area, leaf application of Si enhanced yield,1000 kernel weight and milling rates. These results corroborated the findings of Datnoff and Rodriguez (2005), Crusciol et al.(2013), Horuz et al. (2013) and Ghasemi et al. (2013). In the present study, increasing doses of foliar applications of Si were found to be efficient to decrease the neck blast rate and logging rate. The authors (Korndorfer, 2004; Datnoff and Rodriguez, 2005; Liang et al., 2006; and Ning et al., 2014) stated that effects of silicon on rice are in regard to a physical mechanism of defense. In the study conducted by Jinab et al. (2008) in silicon accumulating crops, it was found that the silicon in plant health and growth had an effective role. Findings of some authors (Ma and Takahashi, 2002; Synder et al., 2007) revealed that adequate uptake of silicon can increase the tolerance of agronomic crops in particular rice to abiotic and biotic stresses. In this regard, Low silicon uptake increases the susceptibility of rice to diseases such as rice blast, leaf blight, brown spot, stem rot and grain discoloration and makes them more susceptible to insect feeding and abiotic stresses that adversely affect crop yield and quality (1000 kernels weight, panicle per area, milling rate, biological yield, kernel yield, protein content and starch content) (Kobayashi et al., 2001; Rodrigues et al., 2001; Kim, 2002; Akhtar et al., 2003; Datnoff and Rodriguez, 2005; Massey and Hartley, 2006; Santos et al., 2011; Ahmad et al., 2012; Crusciol et al., 2013; Ghasemi et al., 2013).

In our study, there were also significant difference (P<0,01) between varieties (*Osmancık-97* and *Edirne*) for the plant heigh (107 and 117cm), panicle number per area (106 and 102 number), neck blast rate (2,83 and 3,83%), yield (5743 and 5410 kg/ha), 1000 kernel weight (26,7 and 30,5g), total milling yield (70,35 and 64,72%), net milling yield (60,7 and 55,8%) and logging rate (15,5 and 39,8%), respectively. In this context, Horuz and Korkmaz (2014) reported that optimum Si dose for rice grown in saline soils was 200 mg kg-1 and silicon fertilization could be a practical way of reducing the deleterious effect of soil salinity and alkalinity in rice cultivation. However, Çatak et al. (2010) stated that about increasing concentrations of Si did not cause any significant change in germinating rates of seeds of cv. Kıral, *Osmancık-97* and *Baldo* genotypes incubated under photoperiodical induction and dark conditions. The authors also

underscored that Si applications led to statistically significant differences for *Osmancik*-97 root fresh and dry weights, Baldo hypocotyl and root fresh and dry weights and sheath dry weights under photoperiodic induction. All these changes occurred in the morphological and quantitative development parameters were observed to have significant effects at genotypes level.

In addition, in the present study, there were no significant differences between variety x silicon interaction and plant height, panicle number, panicle length, neck blast, yield, 1000 kernel weight, total milling yield, net milling yield except for logging rate and seed number per panicle. Seed number per panicle was the higher on *Osmancık-97* at control application and logging rate was the higher in *Edirne* variety plots at control application.In another study carried out by Horuz et al. (2013), rice grain yield (1.56 - 45.85%) significantly increased (P<0.01) by silicon fertilization in 83% of the paddy soils. In addition, It was found that paddy soils require different doses of Si (50-200 mg kg-1) and averagely 87 mg kg-1 of optimum Si dose for the Bafra region soils.

## Conclusion

In the study, silicon application significantly increased panicle number, yield, 1000 kernel weight, milling rates and decreased plant height, panicle length, seed number per panicle, neck blast and logging rate. The results of the study showed that the most suitable and economic silicon dose for rice is 2000 cc/ha. In addition, the present study suggested that silicon micronutrient is essential for rice cultivation to gain higher yield and quality crop in Ipsala plain.

Table 2. Theettec	Table 2. The effects of toliars ill conaplication on some characters of $Edime$ and $Osmancik$ incevane thes at Ipsalacon dition in 2014.	cationon somec	haracters of <i>Ed</i>	<i>irne</i> and <i>Osm</i>	<i>lancik</i> ncevaneties	at Ipsalacon	dition in 2	014.			
	Silicon	Plant	Panicle	Panicle	Seed	Neck	blo:V	1000	Total	Net	Logging
Variety	Doses	height	number	Length	Number	blast	(herba)	ΚW	Munung viald	Milling	Rate
	(cc/ha)	(cm)	$(\text{per } m^2)$	(cm)	Per Panicle	(per m <sup>2</sup> )	(bilga)	(g)	yıcıu (%)	yield (%)	(%)
	0	119,4	91,7	19,53	134,4a	4,1	5214	26,18	69,57	59,4	28,5e
	1000	105,6	96,5	18,02	108,9b	3,2	5200	26,39	69,82	59,89	19f
Communit 07	1500	105,6	106,95	16,84	97,5bc	2,95	5590	26,77	71,12	61,52	15g
Osmancik-9/	2000	103,9	115,2	15,87	80,35c	2,1	6204	27,47	71,27	62,52	9,75h
	2500	101,8	119,2	14,02	76,25c	1,8	6509	26,75	96'69	60,01	Si
	Mean	107,26b <sup>®</sup>	105,91a	16,86	99,48	2,83b	5743a	26,71b	70,35a	60,67a	15,45b
	0	129,5	87,7	19,95	87,35b	5,05	4905	29,9	64	54,67	57,5a
	1000	115,6	92,5	18,68	108,9b	4,25	4885	30,16	64,23	55,1	45b
Edirne	1500	115,7	103,3	17,55	97,5bc	3,93	5298	30,6	65,43	56,59	37,25c
(Baldo)	2000	114	111,1	16,67	80,35c	3,13	5914	31,4	65,57	57,52	33d
	2500	111,6	115,9	15,22	76,25c	2,78	6046	30,58	64,36	55,21	26
	Mean	117,28a	102,1b	17,61	90°07b	3,83a	5410b	30,53a	64,72b	55,82b	39,75a
	0	124,5a	89,7c	19,74a	110,88a	4,58a	5060b	28,05c	66,78e	57,04d	43 <b>a</b>
	1000	110,7b	94,51c	18,35b	108,9a	3,73b	5040b	28,28bc	67,02d	57,49c	32b
Maan	1500	110,6b	105,11b	17,19c	97,5a	3,44b	5444b	28,68b	68,28b	59,05b	26,13c
INTCALL	2000	109b	113,13a	16,27d	80,35b	2,61c	6059a	29,43a	68,42a	60,02a	21,38d
	2500	106,7c	117,53a	14,62e	76,25b	2,29c	6277a	28,6b	67,16c	57,61c	15,5e
	Mean	112,3	103,99	17,23	94,78	3,33	5576	28,62	67,53	58,24	27,60
CV (%)		1,56	6,88	3,23	15,5	19	9,5	1,45	0,18	0,42	8,9
	Variety	0,28**	0,93**	SN	NS	3,04**	14,84**	0,05**	0,01**	0,01**	3,33**
LSD (0,05)	Si Aplication	1,81**	7,39**	0,814**	15,24**	0,7**	54,75**	0,43**	0,12**	0,25**	2,54**
	Var. X Si Appl.	ISI	SN	SN	21,56*	SN	SN	SN	SN	SN	3,59*
<sup>©</sup> Differentlettersind	<sup>®</sup> Differentlettersindicatesizuificant differences (P<0.05) the according to LSD test. * and ** Significant at the 0.05 & 0.01 levels . respectively. <sup>1</sup> Not significant	ices (P<0.05) thea	accordingto LSD	test, *and **	Significant at the 0,05	&0.01 levels	, respective	elv. <sup>1</sup> Not sig	mificant		

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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UDC: 635.21-186(497.715) Original Scientific Paper

# THE INFLUENCE OF FOLIAR FERTILIZING WITH ORGANIC FERTILIZERS ON POTATO (Solanum tuberosum) YIELD IN THE GEVGELIJA AREA

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#### Abstract

The basic goal of this research is to determine the influence of foliar fertilization with organic fertilizers over the potato yield in the Gevgelija region. The experiment was set according to random block-system on fluvisol soil with high concentration of available forms of nitrogen, phosphorus and potassium. It was performed during the vegetation period of 2013, on the territory of the village of Negorci, near Gevegelija, with four variants and three repetitions in twelve rows. Each variant involves 90 plants in total. The experiment involves the following variants: 1. Control (Non-fertilized); 2. Humustim; 3. Ingrasamant foliar. and 4. Bioflor. During the vegetation period, total of three treatments have been performed by foliar feeding with 0.5% solution of the above given fertilizers. Following the potato harvest and the measurements of the potato yield, it was concluded that the foliar fertilizing and the high concentration of available forms of nitrogen, phosphorus and potassium have positive effects over the quantity of the potato yield in all variants. The highest yield of 38.71 t/ha was achieved in the variant no. 3. Ingrasamant foliar.

Key words: potato, organic fertilizers, foliar fertilizing, fluvisol soil

#### Introduction

Agricultural production, which relies on the use of organic fertilizers in plants' food, as well as the non-use of chemicals for protection of plants falls into the group of organic agricultural vegetable production. It includes all techniques of agricultural production that apply methods for obtaining healthy food, and preserving the environment at the same time. Being a key to successful production, these techniques are supported by the natural fertility of the soil. Respecting the biological cycles and the establishment of relative biological balance in vegetable production, the final product is profitable returns characterized by good quality, while maintaining ecologically clean environment. The yield and quality of plants depend on the biological properties of different plant species, climate and soil conditions, method and rate of irrigation, as well as the correct and controlled plants' food. The plants' food will produce complete and positive results if fertilizers are used in optimal quantities and at a certain time period of vegetation. Moreover, in order to determine the exact quantities of fertilizers, the properties of the soil need to be known. The implementation of the soil fertility control system is a preventive measure for healthy food (Trpeski, 1992).

Potato (Solanum tuberosum) is among the most important and the most intensive horticultural crops in the Republic of Macedonia. Due to the rich and refined chemical composition, the potato is mainly used for human nutrition.

Potato can be successfully cultivated in all soil types with the exception of light and sandy soils. Moderately hard soils with good air and water regime, and enough nutrients are the most favourable for cultivating potato (Lazić et al., 2007).

Foliar nutrition represents application of water soluble fertilizers, directly through the leaf. Foliar fertilizers are quickly absorbed in plants and most frequently are used as additional feeding for plants and alternative feeding in conditions when plants indicate the greatest need for nutrients or in cases of deficient soil fertility (Kovačević, 2012). The use of foliar fertilizers in agriculture is increasingly spreading and such fertilizers are environmentally friendly and target-oriented, since compared to soil fertilizers, they are absorbed directly into the organism in limited quantities. The effectiveness of foliar fertilizers is assessed on the basis on absorption and availability of elements, reduction of phytotoxicity, deficit, impact of physiological processes over the yield and the quality of culture (Petrov, 2014).

#### Material and methods

The scientific-research experiment was performed on the territory of the village Negorci, near Gevgelija (Republic of Macedonia), in an area which has been used for vegetable production for years, and was intensively fertilized with mineral and organic fertilizers. The size of the testing lot is  $64.8 \text{ m}^2$ . The research included potato (Solanum tuberosum) – Volumnia. The experiment was conducted during the vegetation period in 2013, following generally accepted norms and methods for setting up field experiments according to (Filiposki, 2004). The experiment was set according to random block-system, with four variants and three repetitions in twelve rows. The distance of planting is 0.6 m between rows and 0.3 m within the row. Tubers are planting in open furrows at a depth of 8-10 cm. The foliar application of fertilizers is performed with 0.5% aqueous solution of fertilizer, by using a backpack sprayer on every 15-20 days, four times during the vegetation. The first application of fertilizers is performed manually on 21.06.2013. During the harvest period, the yield was measured separately, per variations and per repetitions. The experiment involves the following variants:

- 1. Control (variant without application of agro-technical measure fertilizing);
- 2. Variant with application of foliar fertilizer –Humustim;
- 3. Variant with application of foliar fertilizer Ingrasamant foliar;
- 4. Variant with application of foliar fertilizer Bioflor.

The fertilizer of the second variant Humustim, is categorized in the group of organic fertilizers and is characterized by the following chemical properties: total organic matter = 58.63%, total dry matter = 12.38% humic acids = 20.40%, fulvic acids = 2.15%, N = 3.00%. P<sub>2</sub>O<sub>5</sub> = 1.02%, K<sub>2</sub>O=7.92%, Ca=3.70%, Mg=1.03%.

Bioflor, is categorized in the group of organic fertilizers, and according to the manufacturer declaration. it is characterized by the following chemical properties: pH =

7.93, organic matter = 45.00%, dry matter = 27.00%. N = 1.80%,  $P_2O_5 = 0.42$  %,  $K_2O = 0.33$ %. Ca = 0.10%. Mg = 0.03%. Fe = 0.002%, Cu = 0.002%, Mn = 0.0008%, Zn = 0.0001%.

The third variant is performed by application of the foliar fertilizer Ingrasamant foliar, from the group of organic and mineral fertilizers, and according to the manufacturer declaration, it is characterized by the following chemical properties: N=0 g/l,  $P_2O_5$  =130 g/l.  $K_2O$  =130 g/l. ME in chelated form, plant extracts 0.005 g/l.

The fertilizer of the fourth variant, Bioflor, is categorized in the group of organic fertilizers, and according to the manufacturer declaration, it is characterized by the following chemical properties: pH = 7.93, organic matter = 45.00%, dry matter = 27.00%, N = 1.80%, P<sub>2</sub>O<sub>5</sub> = 0. 42 %, K<sub>2</sub>O = 0.33%, Ca = 0.10%, Mg = 0.03%, Fe = 0.002%, Cu = 0.002%, Mn = 0.0008%, Zn = 0.0001%.

Prior to the setting of the experiment, average samples were taken at a depth of 0-20 cm and 20-40 cm, for the purpose of determining the chemical properties of the soil and the analysis included:

- soil solution pH-reaction (in H<sub>2</sub>O and N KCl), determined with potentiometric titration using pH meter, with combined glass and calomel electrode (Bogdanovic, 1966);
- carbonates content determined by using Scheiblar calcimeter, and 10% HCl (Mitrikeski and Mitkova, 2013);
- content of organic carbon and humus determined according to the Kotzmann method (Bogdanovic, 1966);
- total nitrogen content, determined according to the Tjurin method (Bogdanovic, 1966);
- content of physiologically available forms of nitrogen, determined by Tjurin and Kononova method (Bogdanovic, 1966);
- content of physiologically available forms of phosphorus, determined according to the AL-method and reading of spectrophotometer (Bogdanovic, 1966);
- content of physiologically available forms of potassium, determined according to the AL-method and reading of flame photometer (Bogdanovic, 1966);
- determination of hydrolytic acidity and amount of base cations in absorbed carbonate-free soils according to the Kappen method (Mitrikeski and Mitkova, 2013);
- calculation of the cation absorption capacity and the degree of saturation of the soil with base cations (Mitrikeski and Mitkova, 2013).

#### **Results and discussion**

Best potato yields are achieved at fertile, moldy and deep soils, rich in organic matter, with good water-air properties. As regards the mechanical composition of the soil, potato requires soils with easy to medium heavy mechanical composition.

Potato development and yield are frequently conditioned by pH reaction to the soil. For normal growth and development and fertile yields, the reaction to the soil should be neutral to slightly acidic. As extreme reaction the pH of the soil is considered less pH 4.5 and pH greater than 7.5 (Egumenovski, 1994). Our research concerning the reaction of the soil solution indicated that the soil solution has a neutral reaction.

		-	1									
Depth	<b>p</b> ]	H		%			lable fori g/100 g s		eq.	.mmol/1	00g	%
in (cm)	$H_2O$	N KCl	CaCO <sub>3</sub>	Humus	Total N	Ν	$P_2O_5$	K <sub>2</sub> O	$H^*$	$\mathbf{S}^{*}$	$T^*$	$\mathbf{V}^{*}$
0-20	7.35	6.71	0.00	3.30	0.20	13.61	126.00	65.35	1.39	41.64	43.03	96.77
20-40	7.40	6.73	0.00	2.61	0.16	12.44	122.33	64.28	2.45	57.01	59.44	95.91
Average	7.38	6.72	0.00	2.96	0.18	13.03	124.17	64.82	1.92	49.33	51.24	96.34

Table 1. Chemical properties of the soil

The soil was characterized with neutral pH reaction and is moderately of humus origin. The soil is characterized by high capacity absorption (> 30 eq.mmol/100g) absolutely dry soil and high saturation of the soil with base ions (Ca and Mg more than 90%).

The values obtained for the nutritional elements note immense imbalance of nutritional regime in the soil, i.e. imbalance of the content of physiologically available nutritional elements. The content of the three main macro-biogenic elements is relatively high, and the situation with the phosphorus is particularly alarming. According (Egumenovski, 1994) for the production of potato mercantile of NPK ratio should be 1:1.5-2: 2.4 to 3.

The potato crop as well responds to fertilization. Rich and quality yields are achieved by applying combined fertilization with organic and mineral fertilizers. To obtain a yield of 30 t / ha required to provide 150 kg of nitrogen, phosphorus 180 kg, 240 kg potassium and 90 kg calcium, (Egumenovski, 1994). In our case, at extremely high concentrations of nutritive elements in the soil, application of soil fertilizers would have a negative effect and would further deteriorate the current soil fertility, including negative influence over other components of the environment. Having in mind the ultimate goal in vegetative agricultural production (obtaining higher yields characterized by better quality), only application of foliar fertilizers and cultivation of cultures for which other soil properties would be primarily fitting would satisfy these needs while maintaining a clean environment (Petrov, 2014).

	Repetition		Total crop	Average	Yield	Index	
Variant	т	П	Ш	variation	per plant	(t/ha)	%
	1	11	111	(kg)	(kg)		
1	15.41	16.33	16.26	48.00	0.53	29.48	100.00
2	19.43	20.46	20.66	60.55	0.67	37.19	126.15
3	20.02	21.33	21.68	63.03	0.70	38.71	131.31
4	16.61	17.20	16.54	50.35	0.56	30.93	104.92
LSD 0.05= 1.1890		LSI	0.01 = 1	.7299			

Table 2. Yield of potato kg, t/ha and index indicator

Table 2 above presents the average values of potato yield obtained in the experiment. These values indicate the following:

The highest yield was obtained in the variant number 3, applying the foliar fertilizer Ingrasamant foliar, i.e. 38.71 t/ha which is by 9.23 t higher than the control variant with no agro-technical measure fertilization.

The amount of the yield in other variants is not negligible. The yields for variant 2 was 37.19 t/ha, and slightly lower yield 30.93 t / ha was obtained in variant number 4 Bioflor.

If the yield mass is translated into index indicators compared to the control variant, an increase of 4.92% in the yield is noted in variant 4, increase of 26.15% in variant 2, while the highest increase in the yield compared to the control variant was observed in variant 3 i.e. 31.31%;

Based on the results obtained from the LSD test, statistically significant difference was noted in all variants for the two levels of testing (0.05 and 0.01);

The increase in the yield in variants 2 and 3 is characterized by high economic and statistical justification, while variant 4 is marked by significantly lower feasibility.

Very high yield of potato was obtained on fluvisol, even without application of fertilization. i.e. 29.48 t/ha, which is of great importance in practice and confirms the importance of this soil type for the agricultural production;

Data interpreted from Table 2 on potato yields in all variants are relatively high compared to data indicated in the scientific records in books on yields of potato in the Republic of Macedonia. For example, (Egumenovski, 1998) emphasizes that the average potato yield in the Republic of Macedonia is about 16.2 t/ha but the same author points out that organized production, yields would have reached up to 50 t/ha.

## Conclusion

Based on data obtained from the research, the following conclusions can be inferred:

In high soil fertility conditions with previously conducted soil analysis, the application of ecological foliar fertilizers consisted by macro and micro biogenic elements and plant extracts without soil fertilization enables in average 35.61 tones per hectare yields. This is very important especially if we want to protect the environment from additional fertilization in high fertility soil, as well as to reduce production costs of potato crop.

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# SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

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UDC: 635.615:551.573(560) Original Scientific Paper

# DETERMINATION OF ACTUAL EVAPOTRANSPIRATION OF WATERMELON USING A MICROMETEOROLOGICAL METHOD

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#### Abstract

Agricultural areas play an important role within the global water consumption. Hence, planning the water sources become vital in semi-arid countries especially by means of current global climate change. As a developing country, also Turkey assigns the major water consumption to agricultural activities. Concordantly, irrigation scheduling is an important application affecting both the production quantity and quality. In order to obtain optimum scheduling and irrigation system design, crop water consumption should be determined reasonably. In most of the developing countries, however; no continuous measurements exist for the measurement of evapotranspiration, which is also a component among greenhouse gases. In these countries, the actual evapotranspiration is mainly calculated using appropriate equations and crop coefficients, which were developed abroad. In fact, the actual evapotranspiration should be measured and determined by data collected within shorter time intervals and higher precision instead of using conventional methodology, equations and technologies. Yet the recent studies consider only globally major crops such as wheat, maize, sunflower, etc. even in the developed countries; whereas Turkey is the second biggest watermelon producer throughout the world. Therefore, the goal of this study was determined as taking a first step to fulfill the needs of both of the above mentioned subjects. The study was an applied research during which the actual evapotranspiration of watermelon had been measured using a globally accepted micrometeorological method, Eddy Covariance, (EC). Related measurements were conducted within the growing period of watermelon (from May to September 2012) in the Kırklareli city in NW of Turkey. It has been estimated that cumulative actual evapotranspiration of watermelon was 422 mm with a daily average of 3.27 mm. In conclusion, it can be said that the actual ET of watermelon was measured for the first time in the world by using the EC Method.

Key words: Eddy covariance, drought, water management, irrigation, Kırklareli/Turkey.

# Introduction

Precipitation regimes have been changed in most areas throughout the world because of global climate change. In many regions, however; the intense increase periods cause decreases because most of precipitation become run off although total amount precipitation

amount remains the same. This situation affects also the agriculture negatively, which is vulnerable especially in rainfed agriculture. Therefore, the importance of irrigation schedule in order to obtain optimum water usage is increasing. Water requirement of crops varies to crop types and climate so it is essential to determine the plant water consumption (actual evapotranspiration, ET<sub>a</sub>) for different phenological stages and climates. There are a lot of common methods for determining  $ET_a$  in the world. These techniques can be classified as applied and computational approximations. The most well-known and used approaches like Lysimeter, Bowen Ratio Energy Balance Method and EC are difficult to conduct because of the strains in field studies, high cost and data analysis problems. On the other hand, there are concerns about the consistency of computational methods which are even more executable than applied methods. To evaluate this consistency, comparative relationships between applied methods and ET<sub>a</sub> values obtained from empirical formulas were handled in lots of studies. For example, ET<sub>a</sub> was determined by the EC method over peach orchards in Portugal for two growing periods and these values were then compared with these of the FAO 56 approach (Allen et al., 1998). Finally, new crop coefficients were determined with ET<sub>a</sub> obtained from EC approach (Paco et al., 2006). In a study conducted over the northern forest (Canada), ET values of surfaces with different characteristics were determined by EC. Moreover, daily ET rates of a region consisting of mature trees were compared with daily ET values of fire exposed and cut forest regions (Amiro et al., 2006). Parent et al (2012) determined the ET of potato by EC method during one growing period in 2012 in Canada. Here, the variation of  $ET_a$  was obtained to determine the optimum irrigation schedule with the purpose of decreasing negative effects of meteorological variables on yield. Beside these studies, lots of researchers measured ET of various crops and compared the reliability of corresponding ET calculation methods (Cho et al., 2013; Pereira, 2004; Moeletsi et al., 2013; Raziei et al., 2013). The above-mentioned studies are at local scale however, so they are not representative for different climate types. Represented by 20% of the total global production, Turkey is the second biggest watermelon producer after China (Figure 1). In this connection, ET<sub>a</sub> of watermelon crop was measured and analyzed by EC method during the growing period of 2012 on the experiment field of Atatürk Soil Water and Agricultural Meteorology Research Institute Directorate, in the Kırklareli City of the Thrace Region in Turkey for the first time.

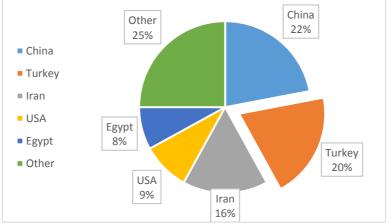


Figure 1: Rate of global watermelon production (FAO, 2014)

## **Materials and Methods**

#### Study Area

The field study was conducted in the Kırklareli city (41.70 N, 27.20 E) located in the Thrace Region of northwest Turkey for a growing period between 14.05.2012 and 19.09.2012. Figure 2 shows the location of experiment field, represented by an area of approximately 2 ha. The research territory belongs to a temperate, continental and Mediterranean climate, with a long term annual mean temperature of 13.2 °C (from 1950 to 2014). Long term annual mean total precipitation is 578 mm (TSMS, 2015). Monthly distribution of precipitation can be seen in Table 1. During experiment period the weather conditions were generally at climate means.

Table 1: Long term (from 1950 to 2014) average total precipitation, rain	y days and temperature
values for experiment site (TSMS, 2015).	

	January	February	March	April	May	June	July	August	September	October	November	December
Total precipitation (mm)	61.8	47.3	48.4	43.4	50.0	50.6	26.8	20.8	34.5	50.0	63.8	70.8
Rainy Days (Days)	11.0	9.1	9.2	10.4	10.0	8.5	4.8	3.7	4.8	7.1	8.7	11.6
Temperature (°C)	2.9	4.0	6.9	12.0	17.3	21.6	23.9	23.3	19.1	13.9	9.0	5.0



Figure 2: Location of study area

#### Measurements

EC measurements were conducted using two types of sensors; namely a 3D sonic anemometer (CSAT3, Campbell Scientific) and an infrared gas analyzer (LI-7500, LI\_COR Biosciences) in a temporal resolution of 10 Hz. To examine the EC measurements simultaneously with meteorological variables like maximum, minimum and mean temperatures, relative humidity (Hygrometer MP100A, Rotronic Instrument Crop), wind speed and direction at 0.5, 1, 2, 5, 10 m level (NRG #40C Anemometer and NRG #200P Wind Direction Wane, NRG Systems), global solar radiation (CMP3, Kipp&Zonen), net radiation (NR LITE, Kipp&Zonen), soil temperature at 2, 5, 10 and 20 cm level, soil water content at 0-30, 30-60 and 60-90 cm (CS 616 TDR type, Campbell Scientific) were also measured during the experiment period. All meteorological measurement data were collected and recorded by a data logger (CR1000, Campbell Scientific Inc.).

During the growing period crop reached up to 5 m horizontally and 30 cm vertically. Harvest has been applied gradually between 31.07.2012 and 19.09.2012. EC

variables were measured at 10 Hz resolution and covariances of 30 minutes data were calculated. All variables were measured in 1 second resolution by the agricultural meteorology station and recorded in 10 and 30 minutes averages except precipitation, which was recorded as totals.

#### Methods

After technological developments in 90's the Eddy Covariance (EC) technique has become the mostly used micrometeorological method for direct measurement of  $ET_a$ . These developments allow the measurements to be conducted in higher resolutions such as 10 Hertz. EC basically needs the measurement of measure two variables; 3D wind speed and concentration of the material (gas) of interest. Covariance between these two parameters stands for the explanation of the vertical fluxes in consideration such as  $CO_2$ ,  $H_2O$ ,  $CH_4$ etc. Equation 1 shows how latent heat flux (LE) is calculated according to the EC (Launiainen, 2005):

$$LE = L\rho \overline{w'q'} \tag{1}$$

where LE, represents latent heat flux (Wm<sup>-2</sup>);  $\rho$  (kgm<sup>-3</sup>) is the air density; L shows the latent heat of vaporization; w' (ms<sup>-1</sup>) is the deviation of vertical wind speed; q' stands for the deviation of water vapor (kgm<sup>-3</sup>), respectively. Over bar represents the mean.

This is not a mathematically complex method, but it requires a lot of caution for setting up and data processing. Moreover, necessary instruments for the application of this method are costly. Nevertheless, the EC is accepted as one of the most accurate direct approximations and is widely used in the world (Aslan, 2015).

#### **Results and discussion**

Measured daily mean volumetric soil water content (SWC) time series can be seen in Figure 3. SWC is always above wilting point which are 0.12 (0-30 cm), 0.18 (30-60 cm) and 0.177 (60-90 cm)  $m^3/m^3$  but after the last irrigation (July 3<sup>rd</sup>) SWC (0-30 cm) decreased up to the harvest (July 31<sup>st</sup>) and causes to the water stress for watermelon. In contrast, SWC in deeper layers of 30 – 60 cm and 60 – 90 cm show that there was no water stress during the growing period. So crop did not expose any water scarcity. Pick values have been observed after irrigation (24 May; 17 June; 3 July) and effective precipitation.

Results of measured meteorological variables during the water melon growing period are shown in Figure 4. During growing period only in May, 71.4 mm precipitation occurred while its total corresponded to 88.2 mm (0.1 mm resolution). Global and net radiation values together with relative humidity were also lower in May than the rest of the growing period. As expected, temperature variations were getting higher values during summer months but lower in May caused by below climate averages because of rainy days.

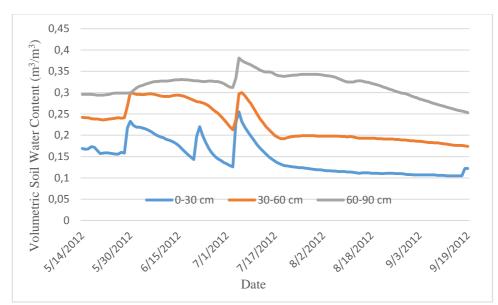


Figure 3: Variations of daily mean volumetric soil water content (SWC) during the studied period in watermelon growing season (14.05.2012/19.09.2012.)

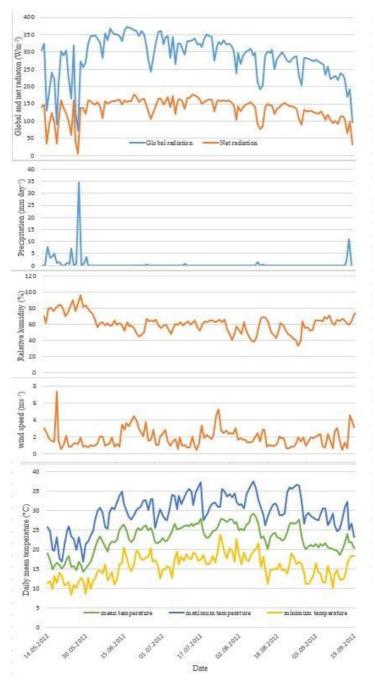


Figure 4: Variations of measured meteorological variables during the growing period in 2012

It is obviously seen that experiment period has lower precipitation (droughty) and nearly same atmospheric temperature values compared to the climate averages (Table 2).

	May	June	July	August	September
Monthly total precipitation (mm) (long term – TSMS 2015)	50.0	50.6	26.8	20.8	34.5
Monthly total precipitation (mm) (2012)	68	3.9	1.1	1.9	12.8
Average temperature (°C) (long term – TSMS 2015)	17.3	21.6	23.9	23.3	19.1
Average temperature (°C) (2012)	15.5	22.8	25.1	23.8	20.2

$T_{11} 2 C_{11} 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1$	term averages and measurements	C 0010
I aple 2. Comparisons of long	term averages and measurements	of 2012 growing period
ruble 2. comparisons of long	term averages and measurements	of 2012 growing period.

Latent heat flux (LE) values have been divided by latent heat (L) and estimated  $ET_a$  was derived as W m<sup>-2</sup>. Then,  $ET_a$  values were converted into mm. Within the growing period, daily mean  $ET_a$  was calculated as 3.27 mm day<sup>-1</sup> while daily maximum and minimum  $ET_a$  values were recorded as 7.26 mm day<sup>-1</sup> on 13.07.2012 and 1.01 mm day<sup>-1</sup> on 14.05.2012, respectively. In this period, the total water consumption (total  $ET_a$ ) of the crop was calculated as 422 mm. During June and July, the crop was in the middle of its growing stage, so  $ET_a$  was getting higher values. Because of decelerated growth rate and application of the first harvest (31<sup>st</sup> July)  $ET_a$  values were decreasing during August and the rest of the growing period. In addition to daily variation, 24 hour-variation of LE as W m<sup>-2</sup> is also shown in Figure 5. As the global radiation was in increase,  $ET_a$  also increased and get its highest values nearly at noon while also  $R_g$  was highest. Lack of radiation throughout cloudy days like in May cause the  $ET_a$  to get lower values.

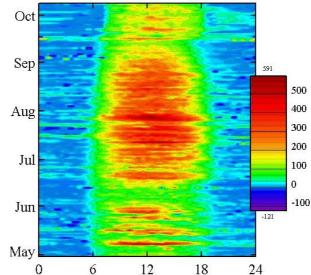


Figure 5: Hourly and daily variations of LE (Wm<sup>-2</sup>) during the growing period of 2012

# Conclusion

In this study,  $ET_a$  of watermelon was determined using the EC method for the first time in Turkey and probably the World. Although EC is one of the most used and reliable methods to measure  $ET_a$ , it should be compared with other methods such as Bowen Ratio Energy Balance and lysimeter. Also periodic measurements of multiple growing seasons at different climatic conditions are important to better understand the ecosystem behavior.

Further studies could serve this aim by covering successive growing periods with different agrometeorological conditions.

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# HYDROCARBONS POLLUTION ENVIRONMENTAL IMPACTS ON FLORA OF FIER DISTRICT

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#### Abstract

Oil industry activities have influenced environmental pollution in general showing direct impacts on ecosystems and living creatures. The fauna and flora situation analysis of the hydrocarbon contaminated area in Fier region indicates a reduction on quantity and quality of kinds. The study of Patos-Marinza area of the vegetation environment around Seman and Gjanica rivers shows a small number of kinds and difficulties in their occurrence. Hydrocarbon contaminated areas are characterised by a large extension including: field habitats with natural vegetation, field habitats with agricultural cultures, Mediterranean shrubs habitats, forest habitats, vegetation around riverbeds streams canals or ponds habitats, etc. The present study aims "to identify and assess the environmental situation in Patos-Marinza industrial area as well as the environmental risk related to environment users. The study basic hypothesis is: "Patos-Marinza oil industry environment is polluted by hydrocarbons with impacts on the flora, fauna and man health". Based on the bio indicators types, we assess that: This situation is related to the contamination of oil and gas processing industry, technological wastewater discharges, oil films and jellies observed in river waters, the high level of H<sub>2</sub>S, etc. The objectives of this study are: the identification of environmental aspects of oil pollution on flora and fauna of the area under study". With reference to the observation and analysis, it results that the fauna and flora composition of this area, including mammal types and domestic birds, mammals used in agriculture, forest fauna, river and pond freshwaters fauna which are significantly damaged due to contamination as of decades in Fier area.

Key words: Environmental contamination, hydrocarbons, flora, rehabilitation

# Introduction

Industrial oil area of Fier is one of the earliest in Albania. The oilfield of Patos was discovered in 1928, that of Marinza, in 1957. During its 70 years of history, the oilfield of Patos-Marinza has already produced over 120 million oil barrels. Environmental issues are evident in the area under study. Pollution of soils, waters and air has caused adverse effects related to soil fertility, reduction of plant productivity, contamination of food chain up to disease spread on people. Activities of oil industry are distinguished for environmental issues caused during extraction, accumulation, storage, transport and product processing.

This area is relatively rich in natural resources and biodiversity (flora and fauna). It is populated by autochthonous communities. In a summarized way, for determining the level of environmental sensitivity, the estimation is based on the following factors:

- this area is inhabited
- land is fertile
- developed agriculture
- rich in surface waters
- phreatic waters are near the surface
- rich in biodiversity
- long-term industrial activity

Referring to different authors, oil pollution has high dangerousness on environment and people's health. It is estimated that oil contains around 10 000 components, a part of which is classified of high dangerousness on wildlife and people's health, especially benzene and its components. Changes in the distribution of spontaneous flora (family, gender and species) are a useful indicator for identification of pollution pressure on wildlife.

The presence and identification of spontaneous species is an indicator for the pollution level in a certain area. Species might show different sensibilities or might be tolerant to the presence of hydrocarbons in environment. Pollution from hydrocarbons affects the distribution of species in the affected area. Some species might be more sensitive than others toward oil pollution. Other organisms might be more tolerant to oil and may be able to colonize and dominate the contaminated areas (Peterson et al., 2003).

Ecosystems are complicated systems composed of various types of organisms linked among them hierarchically in a complexity of interactions and in a continuous change dynamics. Environmental changes can be originated from anthropogenic and natural sources. The acute situation of pollution can threat the ecosystems (Eggen et al., 2004).

Albanian flora is distinguished for its high diversity as result of geographic position of Albania, topography and diversified climate. In the study area several autochthonous species are identified. The region under study is crossed by a large number of rivers, often with very wide beds and covered by special hydro-hygrophilous vegetation. The free flow of Seman and Gjanica river crossing the area through a mosaic of habitats created as a result of various aspects of waters flow (calm waters, pockets, rattling waters, rocky or gravel places) favors the biodiversity or provides environments for a wider scale of organisms. Surface water pollution from oil causes disorders on soil and aquatic ecosystems, (Linden 1976; Feder et al., 1990; Cram et al., 2006; Jackson et al., 1989; Peterson 2003). A small part of oil can be dissolved in water. This is the most bio-available part, and for this reason it is the part of the highest potential or the highest risk causing damages to wildlife. Agricultural lands flora is poor and monotonous in types. In such habitats, many spaces of plain and hilly area are included, which are exposed to sun radiation or shaded by cultivated plants. The number of species here is low and they are directly threatened by human activity. From the cryptogramic flora in these habitats grows about 1% of the total number of species. The phanerogamic families with the highest distribution scale and highest number of types are: Asteraceae, Poaceae, Fabaceae, Cruciferae, Papaveraceae, Charyophyllaceae, Polygonaceae, Convolvulaceae and Primulaceae. In about 60% of surveys, the species with the highest frequency and in highest amount are: Dittrichia viscosa, Papaver rhoeas or poppy crop and then we might mention: Avena sterilis, Lactuca seriola, Alopecurus myosuroides, Sinapis arvensis, Capsella bursa-pastoris, Sonchus vulgaris, Anagallis arvensis, Medicago lupulina etc.



Picture. 1 Invasive type Dittrichia viscosa (Marinza) Pictu

Picture. 2 Dominated by Rubus ulmifolius (Marinza)

Natural and semi-natural communities in Fier region, nowadays have a fragmentary range. Climax formation or ever green Mediterranean forests dominated by evergreen oak (*Quercus ilex*), are almost disappeared. It is preserved only in some limited and fragmented micro-habitats and far away from oil extraction and processing industry as well as inhabited centers. There an open forest is formed with distanced trees thus with a well-developed bush and herbaceous floor. The most characteristic bushes are: *Pistacia lentiscus*, *P. terebinthus*, *Rosa sempervirens*, *Cercis siliquastrum*, *Rhamnus alaternus*, *Hedera helix*, *Arbutus unedo*, *Phyllirea angustifolia*, *Ph. latifolia*, *Viburnum tinus*, *Lonicera etrusca*, *L. implexa*, Cistus incanus, *Daphne gnidium*, *Myrtus communis*, *Coronilla emerus* etc.

In the herbaceous floor are mostly distinguished *Euphorbia characias, Teucrium chamaedrys, Stachys officinalis, Rubia peregrina, Asparagus acutifolius, Ruscus aculeatus, Asplenium adianthum-nigrum, Symphytum tuberosum, Dorycnium hirsutum, Cerinthe major etc.* 

Vegetation of these environments is often represented by various degradation phases of former primitive forests dominated by oak (Quercus ilex). More often, in these environments are seen bush formations dominated by the kermes oak (*Quercus coccifera*). Along with the kermes oak there is another degradation phase dominated by Spanish broom (Spartium junceum) representing the dominant species of these environments. Along with this, its flora is represented by species such as Phlomis fruticosus, Ruscus aculeatus, Asparagus acutifolius, Teucrium polium, Cistus incanus, Cistus salviafolius, Bellis perennis, Smilax aspera, Dactylis glomerata, Arum italicum, Anemone hortensis, Cynodon dactylon, Anthoxanthum odoratum, Briza maxima, Chrysopogon gryllus, Poa bulbosa, Micromeria juliana, Cynosurus echinatus and by bushes such as Pistacia lentiscus, Myrtus communis, Phillyrea angustifolia, Paliurus spina-christi, Pyrus amygdaliformis, Juniperus phoenicea etc (Demiri, 1979). A further degradation it is noticed on this vegetation, not only from the hydrocarbon pollution, but also from the occasional burnings of its inhabitants aiming at "improvement of pastures, herb quality and intensive pasturing". The main indicators of this degradation are the wide distribution of species such as: Galactites tomentosa (thorny species), invasive species Dittrichia viscosa and anthropogenic species such as *Rubus ulmifolius*, *Pteridium aqualinum*, *Asphodelus aestivus*, *Andropogon ischaemum*, etc.

Frequently, the serious damage of forest formations of *Quercus frainetto* e *Quercus cerris* has lead to their replacement with Macedonian oak (*Quercus trojana*) an indicative oak type of very eroded and dry rocky soils with basic lime rock often in surface. Along with the above accompaniments, an important place in the rivers shores of the area is taken by the dominant accompaniments of galingale (*Cyperus longus*). The dominant type is accompanied by *Carex* sp. div., *Lysimachia nummularia, Veronica scutellata, Eleocharis palustris, Alisma plantago - aquatica* etc and saw-sedge (*Cladium mariscus*).

## Material and Method

This study is realized during 2012-2015, in Fier district. The method used is based on comparisons of the flora of the area contaminated by hydrocarbons, with that of another area not contaminated (with the same geographical and climatic conditions). The study of Patos-Marinza area (Fier district) of the vegetation environment around Seman and Gjanica rivers shows a small number of kinds and difficulties in their occurrence. To judge the environmental aspects, changes in environmental components and environmental effects, information is gathered and observations and measurements are made in the area under study. Based on the bio indicators types, we assess that: This situation is related to the contamination of oil and gas processing industry, technological wastewater discharges, oil films and jellies observed in river waters, the high level of  $H_2S$ , etc.

#### **Results and Conclusions**

- Observations show that the abandoned terrains by human activity, in the study area, particularly in the district of Ballsh, evolve toward the primitive forest with dominant species of evergreen oak.
- Compared to the flora of cultivated areas, as a result of hydrocarbons pollution, the most dominant plants of the abandoned terrains are: *Calystegia sepium, Daucus carota, Erigeron epiroticus, Conyza canadensis, Lactuca seriola.*
- The spontaneous vegetation of abandoned terrains left uncultivated is a source of infection for cultivated terrains nearby.
- Oil industry activities have influenced environmental pollution in general showing direct impacts on ecosystems and living creatures (flora and man health).
- In terms of flora and fauna damage from hydrocarbons, there is an indispensable need for artificial and safe afforestation which would go towards a natural forest formation in harmony with physiognomy of sustainable landscape. Sustainable landscape and its evolution should be followed step by step, because the opposite process may occur. Also, abandoned lands could be invaded by intensive erosion and therefore degraded to complete desertification.
- Effects on reproductive capabilities may lead to modification of natural cover, modification of habitats and ecosystems in oil industry areas or in other accidentally polluted areas or polluted by gas emissions or byproducts of fossil fuels.
- The effects of oil byproducts on chlorophyll are a significant finding confirming the fact that the productive capacity of species is directly affected. The effects may occur on genetic and reproductive activity of plants (Malallah, G., 1997).

#### SECTION 8. NATURAL RESOURCES MANAGEMENT AND ENVIRONMENT PROTECTION

- Oil extraction and processing activity in the region, besides the anthropogenic activity, has caused a high level of pollution and significant loss of biodiversity. Two are the most visible indicators of this activity:
- 1. Growth if infection rate from different diseases
- 2. Higher infection rates from pests



Picture. 3 The effects of pollution (necrosis) in the leaves of Phragmites australis and Rubus ulmifolius



Picture. 4. The effects of pollution (necrosis) in the leaves of Populus canadensis and Calystegia sepium as a result of oil pollution from hydrocarbons

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# STUDIES ON CYCLAMEN COUM MILL. IN SINITE KAMANI NATURAL PARK, BULGARIA

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#### Abstract

Sinite kamani Natural Park is located on the southern slopes of the Eastern Balkan range. Over 1060 species of vascular plants have been established for its territory until now, 42 species are protected under the Biological Diversity Act. The object of the present study is one of them – Cyclamen coum Mill. This work aims to make an assessment of the status of Cyclamen coum Mill. populations after undertaking in-situ measures for their preservation and for soil fertility of the territories inhabited by them. The study was conducted during the vegetation period of 2013 - 2015. The populations were observed during each of the phenophases of their development. Soil samples of 0-20 cm layer were collected from every studied area. The samples were analyzed for: pH; mineral nitrogen, available potassium and available phosphorus content. As a result of the studies could be concluded that in Sinite kamani Natural Park Cyclamen coum Mill. forms populations on Eutric Luvisols and Regosols. It grows successfully both in acidic soil reaction, and in slightly acidic to neutral reaction. The soils are with low mineral nitrogen content, well stocked with available forms of potassium and phosphorus. The undertaken during 2013 - 2014 insitu measures for preservation of Cyclamen coum Mill., including assistance in its natural reproduction, control of Pteridium aquiline (L.) Kuhn, erosion control mesh, have led to an increase from 93.8% to 142.8% in the number of the populations in the natural park. Key words: Sinite kamani Natural Park, Cyclamen coum, soil fertility

## Introduction

Sinite kamani Natural Park is situated on the southern slopes of Eastern Balkan range above the town of Sliven on an area of 11380,8 ha. Except as a natural park the territory was declared a NATURA ZONE under Directive 92/43 EEC for conservation of habitats (BG0000164), an important place for plants (BGIPA101) and an area of European importance for the protection of natural values (Korine site №000006400).

The climate in the park is moderate continental and the relief is heavily indented with many rock formations, ravines, caves and waterfalls. Its territory is traversed by many rivers and deepravines, the largest river is Ablanovska river. The varied topography and climatic conditions, the relatively great displacement in altitude, the variety of soils and underground rocks, the long geological history and dynamic historical past and present of the Sliven region have contributed to the formation of exceptional biodiversity on the territory of the park. The currently known vascular plants are over 1060 species (Grozeva

*et al.*, 2004; Petrova, 2004; Petrova *et al.*, 2009; Georgieva & Petrova, 2009; Grozeva *et al.*, 2014) and represent just over 25% of those known in Bulgarian flora, which according to Petrova & Vladimirov (2007) are about 4000 species. Bulgarian endemic species are 9, Balkan ones - more than 40 species, and those protected by the Biological Diversity Act (2002) - 42 species (Grozeva *et al.*, 2004, Petrova *et al.*, 2009, Georgieva & Petrova, 2009). Object of this study is one of the protected species registered on the territory of the park - *Cyclamen coum* Mill. (Picture 1). The species is included in Appendix 2 of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (1992). In Bulgaria the species is found in oak forests and bushes on the Black Sea coast, North-Eastern Bulgaria, Eastern and Central Balkan Range, Thracian plain, Tundzha hilly country and Strandza region (Peev, 1982; Assyov & Petrova, 2012). The species is reported for the Eastern Balkan Range from the Chukata area near shady wooded areas by Urumov (1906). In Bulgarian scientific herbaria (SOM, SOA, SO) there are no herbarium materials from the Eastern Balkan Range. Grozeva *et al.* (2014) reported 3 populations of the species on the territory of the park.



Picture 1. Cyclamen coum Mill.

The aim of the study is to assess the status of the populations of *Cyclamen coum* Mill. in Sinite kamani Natural Park after applying *in*-situ measures to stabilize them and to identify their requirements to the soil reaction and soil fertility.

## Materials and Methods

The study was conducted during the vegetation period of 2013-2015. A total of 3 populations of *Cyclamen coum* Mill. on the territory of Sinite kamani Natural park were studied (Table 1). The registered populations were visited during phenophase reaching normal size of leaves, beginning of flowering, flowering, fruit formation, fruit bearing, falling of mature seeds and period after falling of leaves. To assess the status of each population Methodology for monitoring vascular plants in Bulgaria has been used. The voucher specimens are deposited in the herbarium of the Institute for Biodiversity and ecosystem research, Bulgarian Academy of Sciences (SOM).

Locality	Ecological conditions	Population area
Ablanovo area - the plants are placed into four groups: N 42° 42.599` E 026° 17.192`; N 42° 42.599` E 026° 17.192`; N 42° 42.598` E 026° 17.193`; N42° 42.596` E 026° 17.194`, 533-537 m a.s.l.	The terrain is slightly sloping (9°), non-eroded, facing west. The parent material is marl, the soil type - Eutric Luvisols. An asectatorial species in the herbaceous tier of a mixed deciduous forest incorporating <i>Quercus</i> <i>cerris</i> L., <i>Quercus robur</i> L. and <i>Carpinus orientalis</i> Mill. In the herbaceous tier are registered <i>Ranunculus</i> <i>constantinopolitanus</i> d'Urv, <i>Viola alba</i> Besser, <i>V.</i> <i>odorata</i> L., <i>Scila bifolia</i> L., etc.	The population has an area of 27 m <sup>2</sup> .
Chukata area - the plants are in 3 groups: N 42° 44.207` E 26° 24.508`; N 42° 44.206` E 26° 25.509`; N 42° 44.203` E 26° 25.510`, 699-729 m a.s.l.	The terrain has a slope of 12°, slightly eroded, facing south. The parent material is limestone, the soil type – Regosols. The soil in that studied area was eroded with weak acid soil reaction. An asectatorial species in the herbaceous tier of a mixed deciduous forest incorporating <i>Acer pseudoplatanus</i> L., <i>Acer campestre</i> L., <i>Quercus robur</i> and <i>Carpinus orientalis</i> . There are single shrubs <i>Rosa canina</i> L., <i>Crataegus monogyna</i> . In the herbaceous tier are registered <i>Corydalis solida</i> (L.) Clairv., <i>C. marschaliana</i> Pers., <i>Viola odorata</i> .	The population has an area of 43 m <sup>2</sup> .
Enyova bulka area - the plants are in 3 groups: N $42^{\circ} 42.914^{\circ}$ E $26^{\circ} 24.180^{\circ}$ ; N $42^{\circ} 42.919^{\circ}$ E $26^{\circ} 24.179^{\circ}$ ; N $42^{\circ} 44.918^{\circ}$ E $26^{\circ} 24.177^{\circ}$ , 473-484 m a.s.l.	The terrain has a slope of 26°, heavily eroded, facing southwest. The parent material is limestone, the soil type - Eutric Luvisols. The soil samples were characterized with acid to neutral reaction. An asectatorial species in the herbaceous tier of a mixed deciduous forest incorporating <i>Quercus robur</i> and <i>Carpinus betulus</i> L. There are single shrubs of <i>Rosa</i> <i>canina</i> . In the herbaceous tier <i>Corydalis solida, Viola</i> <i>odorata, Scilla bifolia</i> L. have been registered.	The population has an area of 1475 m <sup>2</sup> .

Table 1. Studied population of Cyclamen coum Mill.

Soil samples from of 0-20 cm layer were collected from every studied area. The samples were air-dried, and plant residues and stones were removed, after which the samples were crushed and sieved with particle size less than 2 mm. All samples were analyzed for: pH with 1:2,5 soil:water ratio; mineral nitrogen content using spectrophotometer JENWAY 6705 UV/VIS; available potassium and available phosphorus content by the Egner-Riem method. The concentration of available potassium was determined by AAS using Analyst 800 Atomic Absorption Spectrometer, Perkin Elmer. Visual soil assessment of soil texture and soil structure in the field was performed (Houškova, 2005).

## **Results and discussion**

Status of the *Cyclamen coum* Mill. populations on the territory of Sinite kamani Natural Park. During the field studies in growing season 2013 a total of 3 populations of the

species were found in the park territory (Table 1). New populations were not registered in 2014 and 2015. The assessment of their condition in the 3-year period of the study showed that the main threats to all populations are two: anthropogenic impact due to their proximity to an asphalt and/or forest road and difficult seedling regeneration due to the abundant dead forest cover during the fruiting period. In the population from Ablanovo area traces of horses were registered in 2014 probably due to tree cutting nearby and in the population from Chukata area in 2014 - scattered branches and household waste, and in 2015 - entry of Pteridium aquilinum (L.) Kuhn along the southeastern border of the population. Due to the proven ornamental value of the species, collection of bouquets during flowering and trampling of specimens during the other phenophases of their development could jeopardize the development and even the existence of populations. Since the natural park is also a place for relaxation, restriction of anthropogenic impact is hard to achieve. Therefore, efforts during the vegetation periods of 2013-2015 were aimed at assisting the seed propagation in order to increase the number of the population. Each year in the late flowering phenophase and early fruiting phenophase of the flowering plants in each population isolation bags were placed for collecting mature seeds (Picture 2). The collected seeds were replanted in pre-selected areas of each population. The sown areas were rolled and mulched.



Picure 2. Plants with placed isolation bags.

During the vegetation period of 2014 the terrain of the population in Chukata area was cleaned of the fallen branches and scattered household waste, and in 2015 at the beginning of April mechanical control of *Pteridium aquilinum* (L.) Kuhn along the border areas of the population started. It will be held four times during vegetation and in August a one-time revealing of its roots is planned.

As a result of systematically applied *in*-situ measures, the number of all populations for the period 2013-2015 has increased (Table 2, Figure 1). In 2014, the most significant is the increase for the population in Chukata area - by 56%, followed by that in Ablanovo area – by 40%, and during the vegetation period of 2015 the highest increase of the number was found in the population of Ablanovo area - by 71.4%, followed by Enyova bulka area - by 24.4%. On average for the period 2013-2015 the number of the population in Enyova bulka area and Ablanovo area increased approximately one and half times, and in Chukata area - with 93.8% (Table 2). The more rapid increase of the populations in

Enyova bulka and Ablanovo area, according to our observations, is due to the remoteness of the populations from asphalt roads, while the population in Chukata area is in close proximity to an asphalt road time and anthropogenic impact is enhanced. The slowest is the rate of increase of the population in Enyova bulka area, which according to the analyses is due to the smaller number of flowering specimens as well as the considerable slope of the terrain, which in heavy rain and stronger winds creates a prerequisite for destruction both of the mature seed and a large part of the young plants.

The population in Ablanovo area was registered for the first time in 2010 and then the numbers of its fruiting specimens was 12. Juvenile specimens were not been described, since the population was registered in a later stage of development of the plants. During the vegetation period of 2011 it was found that the number of the population comprised 25 specimens, including 14 flowering, in 2012 - 28 specimens, including 15 flowering and in 2013-30 specimens, including 12 flowering ones, i.e. for the 3-year period of 2012-2013 the number increased by 20%, while with the implementation of in-situ measures the increase in the number is seven times higher. The other two populations were registered for the first time during the vegetation period of 2013 and a similar comparison cannot be made. As is evident from the data about the population in Ablanovo area, the assistance of natural seed propagation has increased significantly the number and contributed to stabilizing the status of the population. However, having in mind that the number of mature seeds in each fruit box of Cyclamen coum Mill. is from 12 to 22, and the flowering plants in each population are between 6 and 41 in different growing seasons, the achieved increase in number by implementation of *in*-situ measures is far from the theoretically possible. Our observations revealed that the germination of seeds is good (over 70%), but the plants that emerge from them are very fragile and very vulnerable. A threat to them is both possible trampling by passing people and/or animals, and heavier rain, strong winds, sudden drought. With more prolonged waterlogging observed during the vegetation period of 2014, annual plants rotted very quickly. It is hard for them to live through lack of moisture in the soil. Given the good germination rate of seeds, faster increase and full stabilization of all populations could be achieved by implementing *ex*-situ measures as well - growing collected mature seeds in laboratory conditions and returning the grown annual plants in the natural populations of the species.

Area	2013	2014	2015	Increase of the number for the
Alca				period 2013 – 2015, %
Ablano vo area	30 specimens, incl. 12 flowering and 18 juvenile ones.	42 specimens, incl. 27 flowering and 15 juvenile ones.	72 specimens, incl. 41 flowering and 31 juvenile ones.	140.0
Enyova bulka area	21 specimens, incl. 10 flowering and 11 juvenile ones.	41 specimens, incl. 19 flowering and 22 juvenile ones.	51 specimens, incl. 24 flowering and 27 juvenile ones.	142.8
Chukat a area	16specimens, incl. 6 flowering and 110 juvenile ones.	25specimens, incl. 13 flowering and 12 juvenile ones.	31specimens, incl. 16 flowering and 15 juvenile ones.	93.8

Table 2. Number of the Cyclamen coum Mill. populations for the period 2013-2015.

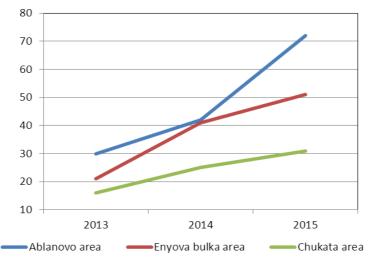


Figure 1. Number of the population by years.

Characteristics of soils in the registered population of *Cyclamen coum* Mill.

In the studied areas two soil types are found: Cinnamon forest soil in Chukata area and near Enyova bulka area and Leached cinnamon forest soil in Ablanovo area. Both types are referred to Luvisols WRBSR (2006). Due to the high slope gradient over 12° in Chukata area soils are highly eroded, which give us reason the soils in the area to be assigned to intrazonal ones - Regosols (Regosols, WRBSR, 2006). Soil structure in Ablanovo area and Chukata area is crumb-like-powdered and in Enyova bulka area it is high in skeletal elements. The mechanical composition of soils in the three areas is characterized by slightly sandy clay mechanical structure on the top surface horizon. The light mechanical composition of all studied soils determines favourable air mode, good water permeability and low moisture capacity. According to pH values in aqueous extract the reaction of the studied soils varies from highly acidic to neutral (Figure 2).

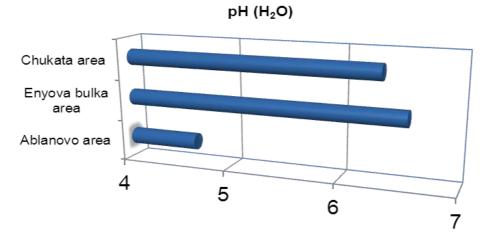


Figure 2. Reaction of soil samples in the studied areas.

The values of pH in aqueous extract show that cyclamate develops successfully both in strongly acidic reaction -  $pH_{(H2O)}$  - 4.66 in Ablanovo area and weakly acidic to neutral with  $pH_{(H2O)}$  values from 6.35 to 6.60. Carbonate forming materials in the cinnamon forest soil in Chukata area and Enyova bulka area cause the slightly acid to neutral reaction of these soils, while the leaching process under the influence of forest vegetation in Ablanovo area causes acidific ation of soil in the studied area. Regarding supply of humus in the soil, the studied soils are characterized by very low to medium humus content (Figure 3). The lowest is the humus content of soils in Enyova bulka area - under 1%, and it is over 2% in Chukata and Ablanovo areas. Cinnamon forest soils have low humus content below 4%, and the total stock of humus in the one-meter layer varies quite widely from 10 to 25 t per decare, 50% of it being in the surface 20-30 cm.

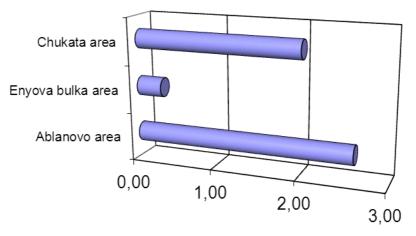
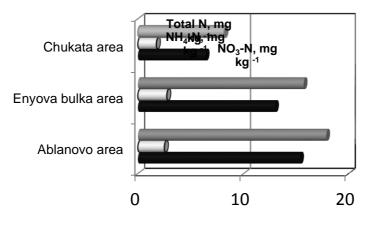


Figure 3. Humus content, % in the studied soils.

All soil samples are poorly stocked with mineral nitrogen. The data about content of ammonium, nitrate and total mineral nitrogen, mg/kg are presented in Figure 4. The content of the total mineral nitrogen is much less than 40 mg/kg, which determines low stock level. According to literature data (Gyurov & Artinova, 2001; Koynov *et al.*, 1998) the contents of digestible nitrogen in the cinnamon forest soils and leached cinnamon forest soils is insufficient, because the conditions for nitrification are not very favourable, therefore the low content mineral nitrogen in the studied soils is not surprising. The results from the chemical analyses for content of digestible forms of phosphorus show moderate stock level in the samples from Enyova bulka area and Chukata area and low level in the leached cinnamon forest soils is most of the leached cinnamon forest soils is medium to low, varying quite widely, but always with a pronounced tendency of decrease from slightly to strongly leached cinnamon soils. Soil samples are characterized by relatively good stock level of content of digestible forms of potassium over 13 mg/100 g soil. Therefore, there is a favourable potassium mode in all studied soils.



Mineral N, mg kg<sup>-1</sup>

Figure 4. Mineral nitrogen stock in soil from *Cyclamen coum* Mill. habitats in Sinite kamani Natural Park.

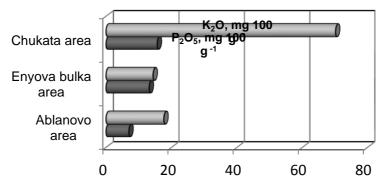


Figure 5. Stock of digestible forms of phosphorus and potassium in soil from *Cyclamen coum* Mill. habitats in Sinite kamani Natural Park.

## Conclusion

The protected species of *Cyclamen coum* is indigenous to the territory of Sinite kamani Natural Park. It forms populations in Ablanovo, Chukata and Enyova bulka areas in rare deciduous forests; at an altitude of 473 to 729 m on cinnamon forest soils. The species is not demanding with regard to soil reaction. It grows on soil with a soil pH from acidic to neutral, poorly stocked with digestible forms of nitrogen and well stocked with digestible phosphorus and potassium compounds. Favourable environment for its development are the light in mechanical composition soils with good permeability and the presence of powdery structure or the presence of skeletal elements have no significant influence. Main threats to the populations in the Natural park are anthropogenic impact, especially during flowering and the hindered seedling regeneration due to the abundant dead forest mat. The implementation of *in*-situ measures aimed primarily at assisting the seedling regeneration of populations helped to increase their number for a period of three years from 93.8 to 142.8%. However, having in mind that flowering specimens are highly vulnerable in unauthorized collection of bunches and that yearlings that had evolved from seedsare highly sensitive to sudden climate changes, it is imperative observations and the implementation of *in*-situ conservation measures of the three registered populations to continue and if possible to include *ex*-situ conservation measures.

## Acknowledgement

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## VARIATION OF ALBEDO DURING THE GROWING PERIOD OF DIFFERENT WINTER WHEAT CULTIVARS

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### Abstract

The greenhouse gases emitting to the atmosphere, have been affecting both people's life and the climate of the regions substantially especially in recent years. Thus, many studies have been focused on the factors that have impacts on climate change. Climate change influences people's daily lives and also many business sectors. One and important of these sectors is agriculture. Increase in evaporation due to global warming requires the application of correct irrigation methods to provide the food safety. Actual evapotranspiration amounts of the plants can be determined by the meteorological parameters. The most significant parameter to determine the actual evapotranspiration is net radiation. This parameter is calculated by using the values developed for different plants by the international foundations because it is not measured in meteorological observation stations. This situation may cause the errors. The most important mechanism controlling the net radiation is albedo. Albedo is expressed as the ratio of reflected radiation and incoming radiation and it varies according to the land surface properties. This study is on the determination of the albedo values for five different winter wheat varieties commonly grown in the Thrace region located in the northwest of Turkey. Identifying the actual evapotranspiration accurately by using correct albedo values, allows the sustainable use of water for irrigation.

**Key words:** Winter wheat, albedo, actual evapotranspiration, sustainable water, Kırklareli/Turkey.

## Introduction

Land surface albedo is an important parameter, which controls the energy budget between land-atmosphere interactions. Albedo is defined as the ratio of reflected solar radiation and global radiation. It mostly depends on land colour, soil moisture, and sky conditions. Changing in albedo is affect the micro and regional climate (Zhang et al., 2013). Doughty et al. (2011) investigated whether increasing agricultural albedo can cause a 0.25 °C cooling regional climate in a small changes (0.01) of albedo. This cause a difference in order to calculate actual evapotranspiration ( $E_a$ ), net radiation ( $R_n$ ), which controlls the evapotranspiration mechanism, of over interested area must be properly determined. Crop albedo diurnal, seasonally or annually varies relation to earth-atmosphere energy balance. Crop albedo also has an important role in actual evapotranspiration with related to  $R_n$ . In order to determine the crop albedo is a complex process by atmospheric and surface conditions: diurnal solar position, cloudiness, soil type and soil water content (Giambelluca et al, 1999; Iziomon and Mayer, 2002).

In developping countries, actual evapotranspiration is calculated by using net radiation, which is calculated by using global radiation values from meteorological station which is located in nercitie sunless agricultural areas. Therefore, it is an important wellestimated net radiation which has component of surface albedo and global radiation.

The purpose of this research, it is aimed to how the five winter wheat albedo variation of 2014-2015 growing period changes, as a function of time. In addition, differences estimated and measured net radiation and actual evapotranspiration of winter wheat were investigated.

### Materials and methods

In Figure 1 the winter wheat field is located in Kırklareli, Northwestern part of Turkey, at 41°42'N and 41°42'E. Meteorological data was collected in Atatürk Soil Water and Agricultural Meteorology Research Station Directorate during 2014-2015 growing season from 2014 November 14 to 2015 June 26.

In this period, phenological observation and leaf area index (LAI) values were measured (LAI-2200C; LiCor, Nebraska, USA) periodically. Short wave and reflected radiation were measured by (Kipp&Zonen, Delft) CMP6 and CNR4 continuously with five different parcels.

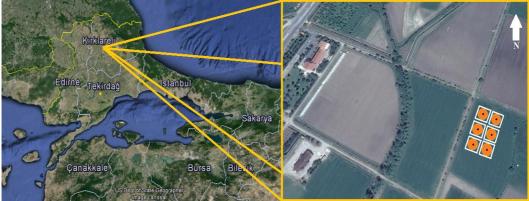


Figure 1. Atatürk Soil Water and Agricultural Meteorology Research Station Directorate

In order to calculate net radiation, global radiation measurement and albedo values are required (Eq. 1):

$$R_{ns} = (1 - \alpha)R_s \tag{1}$$

where  $R_{ns}$ , net short wave radiation (MJ m<sup>-2</sup> day<sup>-1</sup>);  $\alpha$ , surface (winter wheat) albedo;  $R_s$ , global radiation measurement (MJ m<sup>-2</sup> day<sup>-1</sup>).

Net long wave radiation( $R_{nl}$ ) and net radiation( $R_n$ ) can be calculed using equation 2 and 3:

$$R_{nl} = \sigma \left[ \frac{T_{max,K}^{4} + T_{min,K}^{4}}{2} \right] \left( 0.34 - 0.14\sqrt{e_a} \right) \left( 1.35 \frac{R_s}{R_{so}} - 0.35 \right)$$
(2)  
$$R_n = R_{ns} + R_{nl}$$
(3)

Reference evapotranspiration is calculated FAO Penman-Monteith equation:

$$ET_{0} = \frac{0.408\Delta(R_{n} - G) + \gamma \frac{900}{T + 273}u_{2}(e_{s} - e_{a})}{\Delta + \gamma(1 + 0.34u_{2})}$$
(4)

where ET<sub>0</sub>, reference evapotranspiration (mm day<sup>-1</sup>);  $\Delta$ , slop evapour pressure curve (kPa °C<sup>-1</sup>); Rn, net radiation (MJ m<sup>-2</sup> day<sup>-1</sup>); G, soil heat flux(MJ m<sup>-2</sup> day<sup>-1</sup>);  $\gamma$ , psychrometric constant (kPa °C<sup>-1</sup>); T, air temperature (°C); e<sub>s</sub>, saturated vapour pressure (kPa); e<sub>a</sub>, averaged vapour pressure (kPa); u<sub>2</sub>, averaged wind speed at 2 m (m s<sup>-1</sup>).

Being one of themostcrucial parameter for irrigation, good estimates of actual evapotranspiration are needed. Moreover, proper use of water is an important subject for agriculture with related to global warming and climate change.

#### **Results and discussion**

Figure 2 shows the time series of daily mean surface albedo and phenological stages of winter wheat. After sowing, the surface remained bare until germination (Table 1). Surface albedo varied between  $0.134\pm0.013$  and 0.160 for bare soil conditions and first germination phases of winter wheat, respectively. Analysis on soil texture classification resulted as a sand fraction of 66.67%, while 20.83% and 12.50% were obtained for silt and clay ratios, consequtively.

For the 5 different cultivars, mean albedo rates of the 2014-2015 growing period were 0.191 (Gelibolu), 0.208 (Selimiye), 0.191 (Bereket), 0.173 (Pehlivan), and 0.179 (Kate1A), respectively. Daily mean albedo of all winter wheat varieties increased slightly until the earing stage. With in the flowering and grain filling stages, daily mean albedo decreased gradually. During maturity, albedo of winter wheat was in increase.

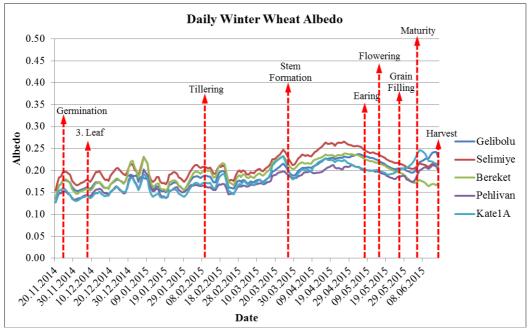


Figure 2. Time series of daily mean albedo of winter wheat

Generally, seasonal average albedo values for crops are using for the calculation of the components of net radiation. But albedo value of crop is associated with surface future ssuch as growing stages, development and climatic conditions etc. Table 1 presents the variation of albedo values for different values according to the phenological stages.

Phenological Stages	Date	Gelibolu	Selimiye	Bereket	Pehlivan	Kate1A
Sowing	11.11.2014	$0.098 \pm 0.002$	$0.113 \pm 0.004$	$0.095 \pm 0.003$	$0.090 \pm 0.005$	$0.090 \pm 0.002$
Germination	19.11.2014	0.136±0.037	0.152±0.037	0.134±0.037	0.121±0.031	0.125±0.033
3. Leaf	08.12.2014	$0.164 \pm 0.012$	$0.181 \pm 0.015$	$0.162 \pm 0.015$	$0.144 \pm 0.010$	$0.144 \pm 0.012$
Tillering	10.02.2015	0.176±0.027	$0.194 \pm 0.027$	$0.180 \pm 0.032$	$0.160 \pm 0.025$	0.157±0.024
Stem Formation	27.03.2015	0.184±0.021	0.206±0.025	0.198±0.025	0.170±0.018	0.182±0.030
Earing	08.05.2015	0.219±0.016	$0.247 \pm 0.018$	0.227±0.013	$0.200 \pm 0.011$	0.210±0.016
Flowering	18.05.2015	$0.223 \pm 0.008$	$0.236 \pm 0.006$	$0.217 \pm 0.005$	$0.196 \pm 0.005$	$0.198 \pm 0.005$
GrainFilling	25.05.2015	$0.204 \pm 0.004$	0.219±0.003	$0.200 \pm 0.005$	$0.182 \pm 0.004$	$0.195 \pm 0.005$
Maturity	04.06.2015	$0.199 \pm 0.007$	$0.207 \pm 0.007$	$0.181 \pm 0.011$	$0.181 \pm 0.010$	0.212±0.010
Harvest	26.06.2015	0.231±0.014	0.213±0.010	0.171±0.011	$0.207 \pm 0.012$	0.231±0.019

Table 1: Variation of albedo values of five wheat varieties according to the phenological stages.

Figure 3 indicates that the diurnal profile of winter wheat's albedo has an asymmetrical distribution; as mentioned (Ogundunte and Giesen, 2004; Bsaibes et al., 2009). Among all of the evaluated cultivars, "Selimiye" variety had the maximum diurnal albedo, while "Pehlivan" variety of winter wheat showed minimum diurnal albedo values. Each of these five varieties had different albedo values, as expected.

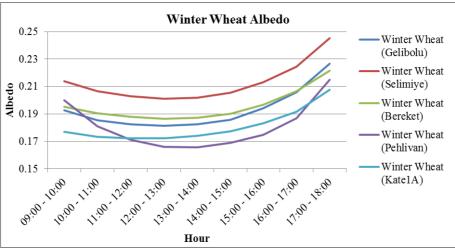


Figure 3. Diurnal profile of albedo for 2014-2015 growing season

As mentioned earlier, the Leaf Area Index (LAI) was also measured periodically during the whole growing period of winter wheat (Figure 4). After the tillering phenological stage, LAI had been increased until flowering, whereas it decreased slightly once the maturity stage was reached. The maximum LAI was measured as 4.778 for the variety of Pehlivan.In the senesence period, the highest LAI value was observed for "Kate1A"

variety. For his reason, in that period albedo of Kate 1A was higher than the values of other varieties.

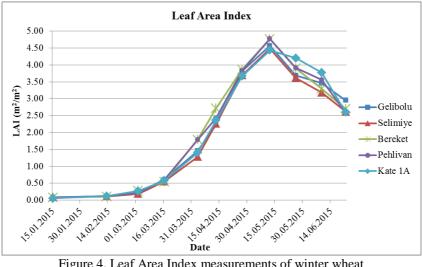


Figure 4. Leaf Area Index measurements of winter wheat

In Figure 5, time series of net radiation data calculated using suggested (reference; default) and measured albedo for five different winter wheat varieties are given. There are differences between the R<sub>n</sub>data. This is because in the application albedo value is using as a reference value, which is a constant value through out the growing period. Altough albedo varies during the growing period, is accepted a constant value for the calculation of net radiation. In 2014-2015 growing season, the daily average net radiation was a maximum for "Kate1A" variety as 80.71 Wm<sup>-2</sup>, where as the day time average  $R_n$  was 76.71  $\text{Wm}^{-2}$  for the same variety. In contrast, the day time average  $R_n$  was mimimum for"Pehlivan" variety as 65.77 Wm<sup>-2</sup>.

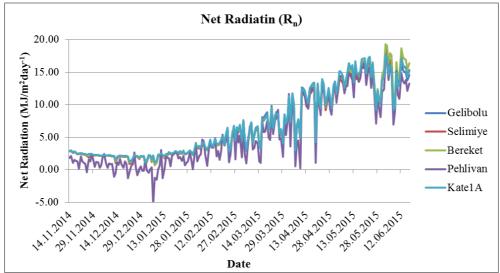


Figure 5. Time series of net radiation for different wheat varieties

# Conclusion

This study is the first through out analysis of crop albedo in Turkey. Results of this research may provide reliable albedo values that are needed for the determination of actual evapotranspiration of winter wheat as well as calibration of remote sensing data. This research was a part of a extensive project, which included also another rotational crop, namely sunflower in the Thrace. Additionly, daily mean albedo values were used for calculation of net radiation values. We can recomendate the usage of albedo values for different phenological stages like crop coefficients instead of a single albedo value for whole growing period.

## Acknowledgements

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UDC: 712.25(498) Original Scientific Paper

# STUDY ON ASSESSMENT OF THE GREENERY AREA IN THE CENTRAL PARK FROM TIMISOARA

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## Abstract

The existence of a green cadastre in a large city is required in the sense of an optimal, European level approach regarding essential ecologic, economic and social relationships between the city's peer and its inhabitants. The carrying out of such studies represents the only way to acquire knowledge about the real situation of green spaces belonging to a city's patrimony, including not only parks, but the entire street greenery. This paper is designed as an inventory of the vegetation fund, as well as an organizational project, based on ecologic and landscaping criteria. The right care is essential for the maintenance and improvement of existent trees and for planning the future economic growth of urban green spaces and requires a professional and competent approach, which can only be accomplished by experts. It is also the only way to manage this important city component from a technical and economic point of view, simultaneously succeeding in optimally estimating the public green spaces conditions, their quality, and also in finding the best solution to intervene in these green spaces with the purpose of carrying out a better management (Ciupa et al., 2005). The Central Park is situated on the right shore of the Bega channel, between the Cathedral and Republic Avenue, belonging to the park bouquet of this shore. The park's surface is of 7.91 ha.

Key words: management, green cadastre, landscape

# Introduction

The purpose behind setting up a green cadastre was based on the technical-economic principles of a better city management. The achievement of a city green cadastre is deemed as indispensible when approaching the issue of optimal relations of social and ecologic nature between a city peer and its inhabitants, from a modern angle. It is the only way to get to know the reality of green spaces in a city, including parks as well as the entire street greenery. The current paper receives thus the character of a vegetation fund inventory, as well as that of a ecologic and landscaping criteria organizational project.

# Material and methods

For each woody vegetation element identified in the field according to its position in the plan, after the topographic determinations, the following characteristics have been collected and registered in code as follows (Timisoara City Hall, 2001):

1. Identification number corresponding to the one in the plan –NRI.

2. Scientific name – SPE.

3. Age – VÎR: 1– 10 years – code 11; 11– 20 years – code 12; 21– 40 years – code 20; 41– 60 years – code 30; 61– 80 years – code 40; 81–100 years – code 50; over 100 years – code 60.

4. Height classes – INA: 1 - 1 - 5 m; 2 - 6 - 10 m; 3 - 11 - 15 m; 4 - 16 - 20 m; 5 - 21 - 25 m; 6 - 26 - 30 m; 7 - 31 - 35 m.

5. Crown diameter – DCO.

6. Crown structured (standard) volume – VRE.

7. Ecologic value – VAE. Tree and shrub ecologic value is determined by their influence on the surrounding areas' physical-climatic factors. This influence depends directly especially on the tree and shrub crown, respectively on its volume and branch and leaf density. The effects of the tree and shrub crown reflect on the following climatic elements: solar energy absorption; atmospheric turbulence – wind intensity reduction;  $CO_2$ absorption; oxygen emission; filter effect on the solid particles from the atmosphere; negative ion emission; phytoncide emission; phonic isolation. In order to determine this relative structured volume, we can use the following formula: Ecologic value = crown volume x crown density indices As a maximum crown density index, with the value 1, one may take the de sycamore, linden, spruce, fir crown one. For all other species, this index is established by estimation, its minimum possible value being 0.5. For the efficiency in the ecologic value expression, absolute values are grouped as follows: standard crown value  $\rightarrow 2 \text{ mc} - \text{cod } 1$ ; 2,1 - 10 mc - cod 2; 10,1 - 20 mc - cod 3; 20,1 - 40 mc - cod 4; 40,1 - 80 mc - cod 5; over 80,1 mc - cod 6.

8. Landscaping value –VPE. The landscaping value is an element of high importance in characterizing the park's woody vegetation. The park's recreational and educational function is fully correlated with this value. This value generally depends on three characteristics: general species physiognomy in singular port; the specimen's height, crown and trunk, which depend especially on the age; trunk anomalies such as: tree forks, twisting etc. Regarding the general species physiognomy, the following are basic elements: general crown shape, leafage and structure, leaf colour, including its variation in time, blooming, remnant fructification. Another basic characteristic is the specimen's size which impresses through its grandeur and, implicitly, through the specimen's age estimation. Anomalies impress because of their rarity and singularity. From these characteristics, the last two have a permanent character, while the first presents a conjunctural, dynamic character and thus of a variable impressive value. Resineferous trees are the exception, for whom this characteristic is permanent, thus increasing their value. The landscaping estimation stages used in the project are: Very low – code 1; Low – code 2; Medium – code 3; High – code 4; Very high – code 5; Exceptional – code 6.

9. Global value – VGL. It is established by the computer using the formula: VGL = VAE x VIT x VPE This value may vary between 0 when the vitality is 0 – dried tree and 108, at maximum values (VAE = 6; VIT = 3; VPE = 6). The maximum value is encountered very rarely, as in the example of a monumental sycamore forest. This global value bears a special significance for the establishment of a park's importance, or for the establishment of penalties in the case of some element destruction.

10. Proposed works. Several works are foreseen: intact maintenance — code 1; toileting, pruning — code 2; dry extraction — code 3; biologically inadequate extraction — code 4; landscaping inadequate extraction — code 5.

### **Results and discussion**

The total number of species, 85, is reduced when considering park size, position, history and prestige. However, when considering the proportion of the spontaneous species tree and shrubbery number, characteristic to phytoclimatic areas, of 45% (maple, ash, tilia and elm tree species - 37% and sambucus, prunus cerassifera, cornus sanguinea - 8%), one can draw the same conclusion regarding the more natural and less processed park. Resineferous trees participate only in a 12% degree. Still, one may observe the existence of a valuable vegetal fund, made up by older species with an obvious ornamental character, such as: platanus, pinus, aesculus, wisteria, juniperus virginiana, taxus, which represent 14%. Overall, the vegetal fund is thus valuable enough to allow for a prospective park restructuring according to current requirements.



Picture 1. Central Park - Google Earth view



Picture 2. Central Park – current situation plan

The 68% proportion of specimens below 40 years of age proves the fact that the park shows an essentially young component in its structure. Older, over 80 years old, specimens, left over from prior landscapings, which constitute the golden park fund, especially with regards to the species aspect, represent 4%, which is quite high, given their size, ecologic and landscaping value. The average crown coverage degree is of 49%, with variations between 4% on main alleys, and 80% within lot 3. The high coverage degree confirms the enclosed massive character of the largest park part.

Due to lack of space, in Table 1 I presented trees and shrubs description inventory only for 40 specimens out of a total of 2182.

N o	Species denomination	Age	Height	Crown diam.	Struct. volume	Ecol. value	Landsc. value	Proposed works
1	Prunus avium	12	5	3,0	9.54	2	2	1
2	Fraxinus excelsior	11	5	3.0	11.28	3	2	1
3	Tilia Platyphyllos	30	22	8,0	281.33	6	4	1
4	Tilia Platyphyllos	30	22	7,0	107.73	6	4	1
5	Sophora japonica	40	23	10,0	384.65	6	4	1
6	Fraxinus excelsior	40	24	6,0	158.24	6	4	1
7	Juglans regia	40	23	6,0	79.10	5	4	1
8	Sophora japonica	40	23	8,0	180.84	6	4	1
9	Acer platanoides	30	13	5,0	41.23	5	2	1
10	Populus alba	40	16	3,0	12.72	3	4	4
11	Acer pseudoplatanus	20	16	6,0	158.24	6	3	1
12	Acer platanoides	20	15	6,0	118.72	6	3	1
13	Acer pseudoplatanus	20	16	5,0	82.46	6	3	1
14	Acer pseudoplatanus	12	12	2,0	13.16	3	2	1
15	Fraxinus excelsior	40	25	6,0	158.27	6	4	1
16	Populus alba	40	10	2,0	3.15	2	3	4
17	Acer pseudoplatanus	11	5	2,0	4.41	2	2	1
18	Celtis occidentalis	40	16	14,0	430.78	6	6	2
19	Acer pseudoplatanus	40	18	6,0	90.40	6	4	1
19	Fraxinus excelsior	40	15	8,0	105.49	6	4	1
20	Acer platanoides	20	10	4,0	26.39	4	3	1
21	Fraxinus excelsior	30	16	5,0	82.46	6	3	1
22	Acer platanoides	20	6	3,0	14.84	3	2	1
23	Morus alba	30	15	7,0	161.56	6	4	1
24	Fraxinus americana	20	4	3,0	5.68	2	2	1
25	Sophora japonica	30	18	8,0	210.98	6	4	1
26	Pinus nigra	30	15	3,0	4.26	2	4	2
27	Acer negundo	20	9	4,0	20.08	4	2	1
28	Tilia Platyphyllos	20	9	4,0	20.08	4	2	1
29	Tilia Platyphyllos	40	22	7,0	123.12	6	4	1
30	Tilia Platyphyllos	20	12	4,0	20.08	4	3	1
31	Tilia Platyphyllos	20	11	4,0	30.16	4	2	1
32	Sophora japonica	40	23	7,0	107.73	6	4	1
33	Morus nigra	40	17	8,0	241.12	6	4	1
34	Sophora japonica	40	18	8,0	140.70	6	4	1
35	Sambucus nigra	20	4	2,0	2.17	2	2	1
36	Sambucus nigra	20	4	2,0	2.17	2	2	1
37	Acer pseudoplatanus	30	14	5,0	31.44	4	3	1
38	Tilia cordata	30	15	6,0	113.04	6	4	1
39	Sophora japonica	40	20	7,0	134.61	6	4	1
40	Sophora japonica	40	16	5,0	35.34	4	4	1

Table 1. Trees and shrubs description inventory

1 a0	le 2. Centralizing species sit	uation – specime	en no. and crowi	n surta	ace		
No		Crown surface		No	Species	Crown	Specim
	Species denomination	(m <sup>2</sup> )	Specimen no. (buc)		denomination	surface	en no.
1	A his 11	× ,	. ,			$(m^2)$	(buc)
1	Abies alba	76.145	4	47	Pinus mugho	10.205 793.63	2
2	Abies concolor	18.840	3	48	Pinus nigra	793.63 5	73
3	Acer campestre	693.155	33	49	Pinus ponderosa	2.551	2
4	Acer monspessulanum	292.805	16	50	Pinus strobus	178.19 5	17
5	Acer negundo	1744.270	73	51	Pinus sylvestris	76.930	11
6	Acer platanoides	2934.391	183	52	Platanus acerifolia	2437.4 25	33
7	Acer pseudoplatanus	2057.485	92	53	Populus alba	361.88 5	15
8	Acer tataricum	38.465	1	54	Populus euroamericana	28.260	1
9	Aesculus hippocastanum	2891.322	116	55	Populus nigra v. italica	33.755	3
10	Ailanthus altissima	200.175	24	56	Prunus avium	46.315	5
11	Buxus sempervirens	37.680	12	57	Prunus cerasifera	256.10 6	39
12	Carpinus betulus	124.030	3	58	Prunus padus	7.065	1
13	Catalpa bignonioides	54.950	3	59	Prunus pissardi	492.19 5	30
14	Celtis occidentalis	201.745	3	60	Pseudotsuga taxifolia	64.566	10
15	Chamaecyparis lawsoniana	16.681	11	61	Pseudotsuga taxifolia glauca	3.140	1
16	Chamaecyparis pisifera sq.boulevard	0.196	1	62	Quercus borealis	38.465	1
17	Cornus mas	105.975	4	63	Quercus imbricaria	66.725	2
18	Cornus sanguinea	65.940	15	64	Quercus robur	1123.3 35	15
19	Corylus avelana	7.850	2	65	Rhamnus frangula	3.140	1
20	Crategus monogyna	17.466	3	66	Robinia pseuoaccacia	1514.2 65	102
21	Deutzia scabra	4.710	3	67	Salix matsudana	155.43 0	4
22	Forsythia intermedia	32.751	12	68	Sambucus nigra	472.57 0	119
23	Fraxinus americana	650.962	144	69	Sophora japonica	1953.8 96	44
24	Fraxinus excelsior	6071.915	256	70	Spiraea vanhouttei	25.120	15
25	Ginkgo biloba	130.310	3	71	Symphorycarp hus albus	20.410	11
26	Gleditsia triacanthos	12.560	1	72	Syringa vulgaris	170.12 6	49
27	Gymnocladus dioicus	268.470	3	73	Taxus baccata	244.91 9	24
28	Hibiscus syriacus	11.775	9	74	Thuja occidentalis	0.196	1
29	Juglans nigra	1075.450	16	75	Thuja occidentalis wagneri	1.366	4
30	Juglans regia	73.790	3	76	Thuja occidentalis aureo- variegata	0.196	1
31	Juniperus chinensis	161.121	23	77	Thuja	11.586	19
51	Jumperus chinensis	101.121	23	//	occidentalis	11.300	19

Table 2. Centralizing species situation – specimen no. and crown surface

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					fastigiata		
32	Juniperus chinensis aureo- variegata	2.355	3	78	Thuja orientalis	24.248	12
33	Juniperus horizontalis	3.140	1	79	Til;ia cordata	1685.0 74	48
34	Juniperus virginiana	261.601	30	80	Tilia platyphyllos	3935.8 47	152
35	Koelreuteria paniculata	25.120	2	81	Tilia tomentosa	12.560	1
36	Lonicera fragantissima	142.870	35	82	Ulmus campestris	932.77 6	48
37	Lonicera xylosteum	47.296	20	83	Wisteria sinensis	26.690	8
38	Magnolia kobus	24.335	4	47	Pinus mugho	10.205	2
39	Malus floribunda	104.405	5	48	Pinus nigra	793.63 5	73
40	Morus alba	206.455	6	49	Pinus ponderosa	2.551	2
41	Morus nigra	441.170	18	50	Pinus strobus	178.19 5	17
42	Paulownia imperialis	7.065	1	51	Pinus sylvestris	76.930	11
43	Phioladelphus coronarius	85.204	45	52	Platanus acerifolia	2437.4 25	33
44	Picea abies	28.173	6	53	Populus alba	361.88 5	15
45	Picea orientalis	12.560	4	54	Populus euroamericana	28.260	1
46	Picea pungens argentea	4.710	3	55	Populus nigra v. italica	33.755	3
					TOTAL	38681.0 07	2182.000

## Proposal for the re-landscaping of the Central Park from Timisoara

The following objectives were proposed for the improvement of the Central Park aspect: Urban furniture rehabilitation and modernization in order to increase the life quality of the Timisoara city inhabitants, re-landscaping the main interests points, as well as the creation of a new area which should give the visiting public one more reason to return to this park, all these objectives creating an impact on the overall city development. The urban furniture types for this area are as follows: benches, garbage bins, lamps.In addition, the creation of new spaces was proposed, such as a free speech stage and a small hearing hall for school shows. Also, it was proposed that the old swimming pool should be transformed in a recreation base with a bicycle lane which should run across the park, a terrace, an open air sports space, and instead of the old pools we proposed to set up Japanese gardens. Also, the spring fountain refurnishing, found on the main park alleys, is desired, the reorganization of the space surrounding the monument, the creation of a new "wheel" is proposed, similar to the one in London (London Eye), as well as of an exotic plant species green house, and the furnishing of a skateboard and parkour space too. The vegetation influence is directly or indirectly reflected in the people's health. Air purity, lower day or season temperature amplitudes or the tree shadow exercise a direct physical-sanitary action on the organism, while the line, shape and colour harmony, the aesthetic tree, shrub and flower grouping enchant the eye, creating a positive state of mind which, in its turn, positively influences the general state of mind.



Picture 3. Top view proposal

The suggested solution respects, as accurately as possible, the current park alleys and structure, and establishes new links to existing and proposed areas. The new park landscaping is the object of a study proposing the park rehabilitation, completion and adaptation for the creation of a harmonious, modern ensemble fit for a European city.



Picture 4. 3D perspective (Japanese garden representation)



Picture 5. 3D perspective (skateboard and parkour court)

Green spaces, as surfaces covered with vegetation, are defined by their capacity to improve the microclimate, the phonic regime, through the utilitarian equipment and ornamental degree of the resting, recreation and sport areas and through the global aesthetic value (Timisoara City Hall, 2001).



Picture 6. 3D perspective (chess space)

Picture 7.3D perspective (spring fountain furnishing)

The importance of green spaces resides in their ecologic and social-economic functionality (Patrascoiu N, 1972), in their capacity to fulfil certain natural and environmental protection functions with the common purpose to accomplish several functions useful to society. Urban green spaces are an integrant part of community life, contributing to the social image, and the degree of landscape architecture representation in a settlement's urbanism reflects its social and economic development status (Florincescu, 1999).



Picture 8. 3D perspective (theatre stage)

Picture 9. 3D perspective (free speech stage)

The "Timisoara Central Park Rehabilitation" project is situated in the context of local, regional, national and European initiatives and policies which target the elimination of problems generated by the urban public infrastructure degradation, at the level of urban communities, for an increase in life quality and the insurance of a balanced regional development. The project constitutes a viable proposition since it answers the needs identified at a local, regional and national level regarding the urban public infrastructure rehabilitation and the improvement of urban public services, an increase in green space per capita, pollution reduction and city landscape value increase. The objectives proposed by the project will contribute to the Timisoara city identity consolidation and to rendering the initial values of a central space, in the context of overall city development. In the present proposition, the vegetation does not undergo major transformations, no ecologically valuable trees will be cleared away, about 20 shrubs will be transplanted and a diversified range of ornamental shrubs will be proposed for plantation among flowers in access and intersection areas, so as to render them more colourful. Species proposed for the landscaping: shrubs (Potentilla Fruticosa, Weigela florida, Euonymus Japonicus, Berberis varieties, Philadelphus coronarius, Prunus serrulata, Buxus sempervirens, Thuja occ. Smaragd, Juniperus Varieties, etc).

## Conclusions

The following works are foreseen: toileting and pruning, dried branch cutting, extraction of the dried specimens, as well as of the inadequate ones, from a biological and landscaping point of view, the execution of a minimum of 4-5 mowings in summer, with immediate grass collecting. Park infrastructure rehabilitation works will effect in creating an environment optimal for recreation and walking activities, thus improving the area's functionality, its proper use as a recreational area respectively. By properly exploiting the Central Park, a perfect harmony and an elegant balance is created between the green area and the historic centre. Also, the current work will increase the number of ornamental

species, thus adding spaces maintaining colour for a longer period during the season, and, from a spatial point of view, the park will regain its former role, as a Central Park, of a special recreational importance.

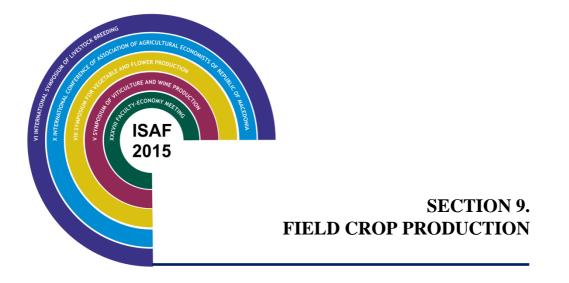
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## THE ROLE OF CULTIVAR IN PRODUCTION OF WHEAT CERTIFIED SEED MATERIAL

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#### Abstract

In the recent years, the production of wheat has been reduced to the level of national consumption in R. of Macedonia, but it is still subject of quantitative and qualitative changes. The main requirement to achieve high and stable yields is to choose and use certified seeds, by applying appropriate technology of cultivation. The main objective of this research was to determine the genetic potential ofsome wheat cultivars for production of quality seed material, thereby indirectly affecting the provision and use of sufficient quantities of quality and cheap seed. The basic indicators of the seed quality (purity, moisture content, germination energy, total germinability and mass of 1000 grains), were analyzed during three years, on three cultivars of wheat. The obtained results were within the prescribed legal values, indicating excellent seed quality. The average value of the purity of the finished seed was 98.9%, moisture content 10.2%, germination energy 93.6%, totalgerminability 96.9% andmass of 1000 grains 42.7g.

Key words: wheat, certified seed, indicators of quality.

## Introduction

For exploitation of the economic potential of wheat, it is necessary to produce high quality seed. Certified seed is a kind of guarantee for the quality of seed or a precondition for high yields. This means that if the production is aimed at high yields, it is necessary use of high quality seeds (Ujevic, 1988). Seed quality is an essential, critical and cheapest input (Rana, 1997), and dynamic instrument that would improve the production potential of crops. The use of agro-technical measures (fertilization, irrigation, plant protection) will not give good results and economic benefits without the use of quality seeds (Milosevic *et al.*, 2010). Seed quality can be estimated on the basis of a number of features and depends on many factors such as viability, the percentage of germination, the germination energy, moisture content, storage conditions, viability and health (Akbari *et al.*, 2004).

The basic precondition for achieving high and stable yields in wheat is the use of quality seed for sowing, combined with the application of appropriate technology of cultivation. It should be noted that certain varieties are genetically predisposed with greater potential in relation to the other, or with the use of these wheat varieties, can be produced better seeds from lower categories. For this reason, and starting from the fact that still use the wheat seed is relatively low, the aim of this research was to explore the potential of some wheat varieties in terms of production of quality seed material, which can indirectly affect the production of a larger quantity seed material of this crop.

### Material and methods

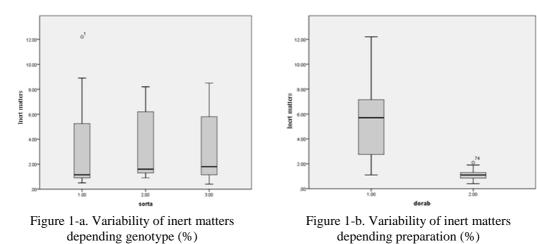
Samples of wheat seed produced in the period 2008 - 2011 were used as testing material. The studies for analyze and evaluation, included data of natural and prepared seed from three varieties: Pobeda, Emese and Altana.

The tests were performed on average seed samples of category C1. The seed is produced in ZK "Pelagonija" - Bitola, and the natural seed is completed in its preparation center. The results of the test were statistically processed by ANOVA method of analysis of variance and compared with Tukey's test.

#### **Results and discussion**

#### Purity of the seed

The quantity of pure seed and the total inert matters, primarily have major implications on the economic effect on the competitors. Because of the economic impact, often happens to neglect the quality of preparing, thus to get a lot smaller percentage of inert material in order to get clean seed. The differences that arise in terms of pure seed and inert material for individual wheat varieties presume that they are result of the characteristics that possess the varieties (Mladenovski and Nikolovski, 2000).



The results for the purity of seed, we can see that the percentage of purity in prepared seed is increased compared with the natural. The percentage of prepared seed in relation with the tested year, shows that the average values were lowest in 2008 (95.4%) and highest in 2011 (98.6%). From the results obtained in our research, (Table 1, Figures 1-a, and 1-b) we can see that the average value of inert substances in the natural seed of the four year

research is 5.3%, while the prepared seed 1.1%. The results show that, after preparation of the seed, the content of inert substances is statistically significantly decreased.

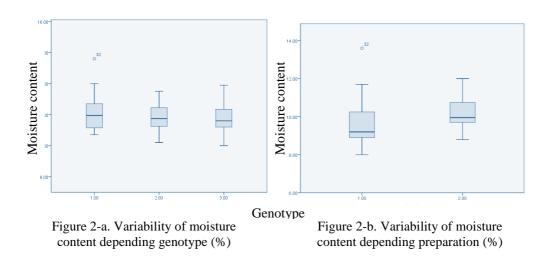
Preparation	Natural seed				Prepared seed		
Seed purity	94.7			98.9			
Inert matters	5.	34b			1.11a		
Moisture content	9.	55a		10.20b			
Germination energy	91	.43a			93	3.58b	
Full germination	95	.98a			96	5.87b	
Absolute mass	42	2.33			4	2.74	
Year	2008		2009	2010	)	2011	
Seed purity	95.4		96.5	96.6		98.6	
Inert matters	4.55c		3.52b		)	1.43a	
Moisture content	9.69b		9.33a	11.14c		9.33a	
Germination energy	91.80a	9	93.70b	93.63b		90.87a	
Full germination	96.53		96.30	96.73		96.13	
Absolute mass	41.27a	2	40.72a	42.81	b	45.33c	
Genotype	Pobeda		Eme	se		Altana	
Seed purity	96.9		96.	7		96.8	
Inert matters	3.10		3.33			3.24	
Moisture content	10.06b	10.06b 9.		9.83a		9.73a	
Germination energy	82.63	82.63 92.		92.83		92.05	
Full germination	96.58			96.33		96.38	
Absolute mass	42.17b		45.4	47c 39.96a		39.96a	

Table 1. Basic elements of certified seed (%)

## Moisture content

The moisture content of the seed is one of the main components that characterize the quality of the seed. The moisture content of the seed depends on its maturity, the conditions during the harvest and the conditions in which the seed is stored after harvesting and preparing (Ilieva *et al.*, 2011).

The survey results for this property are shown in Table 1, Figures 2-a, and 2-b. According to the results, the average moisture content of the test period was 9.6% for the natural seed, or 10.2% of prepared seed. In research, the highest average seed moisture in 2010 was (11.1%), and this value is statistically significantly higher than other years. Identically same average moisture was obtained in 2009 and 2011 (9.3%) and in 2008, the content was 9.7%. In terms of varieties, the average moisture to Pobeda is the highest with 10.1%, followed by Emese (9.8%) and Altana (9.7%). The divergence in the variety Pobeda, has statistically significant level of 95%, compared with other tested varieties.



### Germination energy and full germination

The germination energy and full germination of seed indicate an ability to develop a normal plant, and the main factors that determine the germination are moisture, air and heat (TeKrony *et al.*, 1987).

In these studies (Table 1, Figures 3-a, and 3-b), the energy of germination in natural seeds by an average of four years was 91.4% and prepared seed - 93.6%. The results for energy have statistical significance in terms of unfinished seed, which clearly indicates the need for seed developing. When analyzing the values of the varieties, the lowest average energy of germination was shown in variety Altana (92%), Pobeda had 92.6% and 92.8% in Emese variety. In relation with this characteristic, in none of the varieties were obtained statistically significant differences, but the results show that the energy of germination is much higher than the minimum prescribed legal value.

According to surveys of Statkic *et al.* (2008), micro-agro-cultural conditions have an influence to the overall germination during the vegetation period, and physiological properties of the tested varieties.

Based on the results obtained in our research for the overall germination, which are shown in Table 1 and Figures 4-a, and 4-b, generally, this property is a high value, regardless of variety, year and finishing. The total germination before preparing was 96% and 97% in prepared seed. These values are statistically significantly different at the level of 95% statistical confidence, indicating that no matter how high is the total germination in natural seeds, preparing shows a significant positive effect. In terms of varieties, the variety Pobeda showed the highest germination 97% and varieties Emese and Altana, the average germination was not statistically significant and was 96.4%.

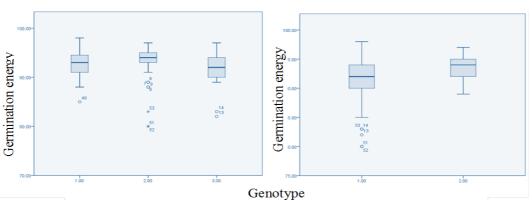


Figure 3-a. Variability of germination energy depending genotype (%)

Figure 3-b Variability of germination energy depending preparation (%)

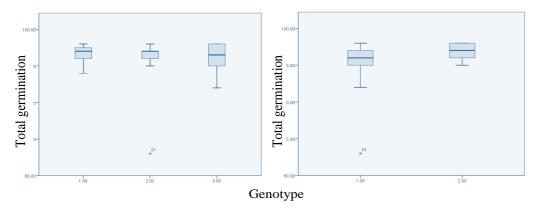
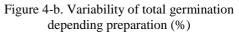


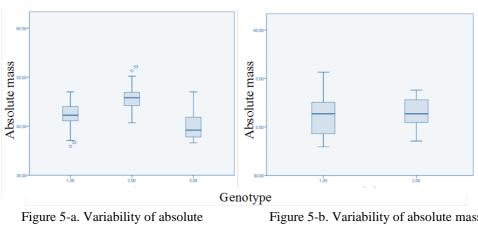
Figure 4-a. Variability of total germination depending genotype (%)



### Absolute mass

The absolute mass (mass of 1000 grains) depends on the development of the plant and can vary in different years. Plants in the initial stage of development, during the germination and growing receive food from the endosperm where are the reserves of nutrients. Seeds with greater mass generally have better developed embryo and give more developed and more resistant plants in the early stages of development (Miric *et al.*, 2007).

The results (Table 1, Figures 5-a, and 5-b) shows that all tested varieties have relatively high absolute mass of seed. The average mass of 1000 grains in natural seed was 42.3 g, and in the prepared seed 42.7 g. The results showed that the variety Altana has the lowest absolute mass (39.9 g). The absolute mass of variety Pobeda was 42.2 g and it was significantly higher than the variety Altana, while the variety Emese obtained value (45,5g) that positively statistically deviate from Altana and Pobeda.



mass depending genotype (%)

Figure 5-b. Variability of absolute mass on depending preparation (%)

## Conclusions

Based on the received results for the quality characteristics of the seed of three wheat genotypes (Pobeda, Emese and Altana), can be concluded that the purity of the seed and the presence of inert matters directly depend on the conditions of cultivation, agroecological conditions (the year), as well as the quality of preparation.

The moisture content of the seed depends on the genetic characteristics of the genotype and conditions in the collection phase. As refers to the energy of germination, there were not obtained results which indicate the influence of the variety on this characteristic. The energy mainly depends on the conditions of cultivation and can be improved with good quality of preparation of the seed. The full germination also is influenced by genotype, but can be increased with preparing the seed and removing all impurities. The value of this property for the test period, in the natural seed accounted for 96% and 97% in the prepared seed. The absolute mass is genotype characteristic, but is strongly influenced by the conditions of cultivation (agro-technics, nutrients, water). The mass of 1000 grains in tested genotypes varied in the range of 40 g in variety Altana, to 45.5 g in Emese. The seed preparation does not have a significant impact on improving absolute mass, if the natural seed has been already produced with high absolute mass.

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# ASSESMENT OF YIELD AND YIELD COMPONENTS OF SOME MAIZE HYBRIDS GROWN IN VELES AND NEGOTINO REGION

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## Abstract

The research has analyzed the yield and yield components of 6 maize hybrids (ZP341, ZP360, ZP434, OS499, ZP677 and ZP704) in Veles and Negotino reions (R. of Macedonia). The experiment was performed during 2011 and 2012, in randomized block design in three replications. Most of the hybrids, shown one cob per plant, excluding ZP434 with 1.14 in Negotino and 1.06 in Veles. The length of the cob varied and it depends on the year, location and genotype. The largest cob was determined on ZP434 (18.11 cm), and the shortest on ZP677 (15.94 cm). The number of rows per cob is a genotype characteristic, but it also depends on the growing conditions too. The biggest number of rows for both locations, was obtained in hybrid ZP704 (14.87), and the smallest in ZP360 (13.19). The Grain weight in Negotino region varied from 183.3 g (OS499) to 131.1 g (ZP677) and from 148.8 g (ZP704) to 79.7g (ZP360) in Veles region. The most important analyzed property was grain yield. The hybrid OS499 showed general highest average yield with 8.05 t ha<sup>-1</sup>. The highest average yield per location, varied from 9.64 t ha<sup>-1</sup> <sup>1</sup> in Negotino to 6.94 t ha<sup>-1</sup> in Veles. During the examination, the highest average value for hectoliter weight was obtained in hybrid OS499. Regarding statistical reliability, genotype and year showed strongly significant impact on all analyzed properties, except the number of cobs per plant. The interaction between location and genotype, showed the greatest impact on the expression of all analyzed properties, with a statistical confidence level of 99%.

Key words: maize, yield, yield components.

# Introduction

The geographic position of the Republic of Macedonia enables it to be a part of the countries where is corn grown. According to the Annual Statistics, out of all arable agricultural area (509,000 ha in total), corn was sown on 28,644 ha (http://www.stat.gov.mk/PrikaziPoslednaPublikacija.aspx?id=69). Many years ago, the average yield is very low, between 4 and 4.5 t ha<sup>-1</sup>. The reasons for that low average yield of corn by surface unit are due to several reasons: the use of non-hybrid seed, inappropriate agricultural-technique, as well as the absence of irrigation systems and inappropriate

choice of hybrids in particular regions (Boshev, 2002). The appropriate choice of hybrids could be the most significant factor for yield improving, which can affect the maximum use of genetic potential of different genotypes in different regions (Bavec and Bavec, 2002). From this point of view, the aim of this research was monitoring the morphological, productive and qualitative traitsof the corn hybrids of various FAO maturity groups in order to choose the most appropriate hybrids for the agro-ecological conditions of Veles and Negotino regions.

# Material and methods

The research was conducted in 2010 and 2011 at two locations in the regions of Veles and Negotino. The following commercial genotypes (hybrids) of maize were used: ZP341, ZP360, ZP434, OS499, ZP677 and ZP704 selected in the Maize Research Institute Zemun Polje - Serbia and the Agricultural Institute of Osijek - Croatia. These hybrids were grown by applying standard agricultural and technical measures which are used for this crop. Examination samples were set in randomly picked block system, in 3 replications, with 4 rows in a plot. Only data from the 2 inner rows of the plots were used for analysis and planned measurements, by using twenty plants per plot. The results were statistically analyzed by ANOVA method of variance analysis, whereas the analysis of internal differences of researched genotypes was done with M stat C statistical package.

## **Results and discussion**

### Soil and climatic conditions

Examination samples were set on cinnamon forest soil (chromic cambisole), characterized by small amount of humus, fine water permeability, loose structure and possibility for easy access in the root system in the deeper layers (Filipovski, 1993).

Months	Monthly average		Average	maximum	Average minimum		
	2010	2011	2010	2011	2010	2011	
V	17,6	16,5	23,6	22,5	11,6	11,2	
VI	21,2	21,7	27,8	28,4	15,1	15,6	
VII	24,0	24,4	30,5	32,5	17,6	17,0	
VIII	25,2	25,3	33,7	33,3	18,1	17,7	
IX	18,7	22,5	25,7	29,9	12,4	15,7	
Average annual	21,3	22,1	28,3	29,3	14,9	15,4	
∑t <sup>°</sup> (V-IX)	3268	3378					

Table 1. Average, maximum and minimum monthly temperature during vegetation in Veles region (°C)

Considering the necessary temperature during corn vegetation period, it can be concluded that the warmth conditions for all researched hybrids were satisfied in both years of research (Tables 1 and 2). Comparison of the average monthly temperatures between the two regions shows that in the region of Negotino, the temperatures for all months during vegetation are somewhat higher from region of Veles, both in year 2010 and 2011.

Months	Monthly average		Average 1	naximum	Average minimum		
	2010	2011	2010	2011	2010	2011	
V	19,3	17,6	25,9	24,3	11,6	10,8	
VI	23,0	23,3	30,2	30,2	15,8	15,2	
VII	25,7	26,3	33,7	34,6	17,8	17,0	
VIII	27,2	26,6	35,7	35,2	18,9	17,5	
IX	20,4	23,3	28,0	31,5	13,2	15,2	
Average annual	23,1	23,4	30,7	31,2	15,5	15,1	
$\sum t^{\circ}$ (V-IX)	3540	3583					

Table 2. Average, maximum and minimum monthly temperature during vegetation in Negotino region (°C)

Table 3. Percipitation in Veles and Negotino region (mm)

-	Veles		Negotino	
Years	2010	2011	2010	2011
Months				
Ι	24,1	22,2	22,5	45,1
II	94,5	31,3	82,8	31,0
III	52,7	18,6	71,4	27,8
IV	81,1	26,2	32,2	22,9
V	51,4	54,7	34,8	51,8
VI	32,1	8,3	40,2	4,8
VII	27,0	36,4	30,8	27,0
VIII	0,8	11,0	14,7	12,6
IX	49,5	17,7	32,4	61,5
Х	162,9	24,5	194,2	29,0
XI	80,4	2,0	29,6	7,1
XII	55,2	28,6	68,4	71,2
∑ I-XII	711,7	281,5	654,0	391,8
$\sum$ V-IX	160,8	128,1	152,9	157,7

The amount of rainfall per month, as well as, total precipitation for for two regions, is presented in Table 3. In Veles region, 2010 was significantly more rainy than 2011. Total amount of rainfall in this region in the first year of research is more than twice higher from the second year. Similarly as in Veles, in Negotino region, in 2010 there was more rain than in year 2011, but not in the amount of Veles region. On the other hand, in the period of vegetation, the sums of rain in both years are almost the same.

# Number of cobs per plant

The maize yield amongst other factors depends on the number of cobs by plant, but only in conditions of sufficient nutrition (Jevtic and Jevtic, 1996). In different cases, most cobs could be counterproductive, i.e. to decrease size and nutrition of the base cob, resulting in even lower yield by surface unit, although there are larger number of cobs by plant (Maddoni *et. al.*, 1998; Boshev, 2002).

		ZP341	ZP360	ZP434	OS499	ZP677	ZP704	Average
Negotino	2010	1,05	1,00	1,14	1,00	1,00	1,00	1,03
	2011	1,00	1,00	1,00	1,00	1,00	1,00	1,00
	Average	1,02	1,00	1,07	1,00	1,00	1,00	
Veles	2010	1,00	1,00	1,06	1,00	1,00	1,00	1,01
	2011	1,00	1,00	1,00	1,00	1,00	1,00	1,00
	Average	1,00	1,02	1,03	1,00	1,00	1,00	
	AVERAGE	1,01	1,01	1,05	1,00	1,00	1,00	
	0.066	J LOD	0 100	т.		1	LCD	0.004

Table 4. Number of cobs per plant

Genotype  $LSD_{0.05}=0,066$  Location  $LSD_{0.05}=0,189$  Interaction genotype x location  $LSD_{0.05}=0,094$ 

In this research, most hybrids had only one cob by plant, and part of them had formed a second cob. At the location of Negotino, the hybrid ZP434 showed the highest value which is 1.07 cobs by plant for both years in average, although at the location of Veles the average value of this hybrid was 1.03 (Table 4). In average, the hybrids OS499, ZP677 and ZP704 have formed only one cob per plant in both locations for both years. Hybrids ZP341 and ZP360 formed 1.01, while hybrid ZP434 showed the highest value of 1.05 cobs by plant. In both cases the differences between genotypes are not statistically significant in both localities, and there is no significant difference at the interaction genotype by locality.

### Cob length

It is evident from the results of the cob length (Table 5) that at Negotino locality the smallest cob length has the hybrid ZP677 (16.22 cm) and it is statistically different from all other genotypes at this locality.

The longest cob length at this locality was noted with the hybrid ZP341 (18.81 cm), whereas significant differences are evident only with hybrids ZP434, OS499 and ZP677. At Veles locality the shortest length was measured with the hybrid ZP360 (14.64 cm) whereas the longest with ZP434 (18.58 cm). The highest value at this location with the hybrid ZP434 has significant differences in comparison with values of all other genotypes, whereas the smallest value of ZP360 is statistically significant in comparison with all other hybrids, except for ZP341.

		ZP341	ZP360	ZP434	OS499	ZP677	ZP704	Average
Negotino	2010	20,28	19,38	19,24	18,92	17,35	20,12	19,21
	2011	17,33	16,70	16,04	15,77	15,09	16,09	16,17
	Average	18,81	18,04	17,64	17,35	16,22	18,10	
Veles	2010	16,77	15,72	20,11	17,94	16,41	18,06	17,50
	2011	14,09	13,55	17,04	13,80	14,92	17,20	15,10
	Average	15,43	14,64	18,58	15,87	15,67	17,63	
	AVERAGE	17,12	16,34	18,11	16,61	15,94	17,87	

Table 5.	Coh	longth	(am)
Table 5.	COD	lengui	(CIII)

Genotype LSD<sub>0.05</sub>=0,9015 Location LSD<sub>0.05</sub>=2,573 Interaction genotype x location LSD<sub>0.05</sub>=1,275

## Number of rows per cob

The row number per cob is also one of the genetic traitsof a given hybrid. In the research of Hallauer *et al.* (1988) there have been confirmed variations in the row numbers of cob, primarily as a genetic difference of hybrids, whereas Bocanski *et al.* (2001) determined connection of number of rows with mass of 1000 grains and the contribution. In our research, the row number by cob was measured between 13.19 (ZP360) to 14.87 (ZP704). At Negotino locality, the lowest annual contribution was measured at OS499 (13.27), while the biggest number at ZP677 (14.67). The hybrid ZP677 in this region showed significant differences ZP677 and ZP341. In Veles region, the smallest number of rows was measured with ZP341 (12.65), whereas the largest number with the hybrid ZP704 (15.51) which is statistically different from all other genotypes (Table 6).

		ZP341	ZP360	ZP434	OS499	ZP677	ZP704	Average
Negotino	2010	14,86	14,63	14,67	14,48	15,70	15,81	15,02
	2011	12,70	12,62	12,22	12,06	13,65	12,65	12,65
	Average	13,78	13,62	13,44	13,27	14,67	14,23	
Veles	2010	13,75	13,71	14,38	15,40	15,38	15,89	14,75
	2011	11,55	11,82	12,19	11,84	13,98	15,13	12,75
	Average	12,65	12,77	13,28	13,62	14,68	15,51	
	AVERAGE	13,21	13,19	13,36	13,44	14,68	14,87	

Table 6. Number of rows per cob

Genotype LSD<sub>0,05</sub>=0,516 Location LSD<sub>0,05</sub>=1,473 Interaction genotype x location LSD<sub>0,05</sub>=0,730

## Grain mass per cob

Grain mass per cob is a parameter which has direct influence on contribution per surface unit, i.e. a greater grain mass at a cob leads to a bigger contribution. This traitin other conditions is a genotype feature, but largely depends on growth conditions (Severini *et al.*, 2011). Favorable climate conditions, appropriate agro technics and appropriate choice of hybrids are the basic conditions for getting bigger grain mass by cob. The calculated results for this traitare shown in Table 7. According to these results, on average for both localities during the two years of research, the lowest value of grain mass was noted with hybrid ZP677 (113.98 g) and highest with hybrids OS499 (161.04 g) and ZP704 (160.06 g). At the analysis of interactive influence on factors genotype by locality for the two years of research, hybrids with largest grain mass per cob (OS499 and ZP704) showed significant differences only in comparison with ZP677.

		ZP341	ZP360	ZP434	OS499	ZP677	ZP704	Average
Negotino	2010	195,57	206,97	188,14	199,96	140,24	190,33	186,87
	2011	167,15	178,42	156,78	166,64	121,94	152,27	157,20
	Average	181,36	192,69	172,46	183,30	131,09	171,30	
Veles	2010	104,07	85,47	149,90	156,88	101,49	152,45	125,04
	2011	87,45	73,68	127,04	120,68	92,26	145,19	107,72
	Average	95,76	79,58	138,47	138,78	96,88	148,81	
	AVERAGE	138,56	136,14	155,46	161,04	113,98	160,06	

Table 7. Grain mass per cob (g)

Genotype  $LSD_{0,05}=19,44$  Location  $LSD_{0,05}=55,49$ 

Interaction genotype x location  $LSD_{0,05} = 27,5$ 

# Grain yield

The corn grain is the main product because of which this cereal is grown, although at some parcels it is grown for silage, where the whole plant is used. The yield is the final parameter at cultivating corn and all agro measures strive towards producing a higher yield per surface unit. Since corn is robust plant with a wide leaf mass and realizes its vegetation in our condition during the summer period, it is evident that water is the first and basic factor for producing tall and stable contributions of corn.

All authors that have dealt with this subject have concluded that as a result of changes with elements which determine yield, as well as morphological changes of plants due to drought, the yield can be decreased from 50% to 100% (Dragovic *et al.*, 1997, Tomov *et al.*, 1997; Djevic and Miodragovic, 1998). However, in conditions of sufficient water access, nutrients as well as genetic potential of the hybrid are the factors for getting high and stable yields.

According to the results of this trait(Table 8), in Negotino locality the lowest contribution was noted with the hybrid ZP677 (6.56 t ha<sup>-1</sup>), whereas the highest contribution was got with the hybrid ZP360 (9.64 t ha<sup>-1</sup>). Apart from the hybrid ZP360, a higher contribution of over 9 t ha<sup>-1</sup> was marked with ZP341 (9.07 t ha<sup>-1</sup>) i.e. OS499 (9.16 t ha<sup>-1</sup>). On the other hand, at Veles locality, the lowest contribution was noted with ZP360 (3.98 t ha<sup>-1</sup>), which had the best results in Negotino locality. The lowest average for both years of research at both localities was with hybrid ZP677 (5.70 t ha<sup>-1</sup>), while the highest average was noted with the hybrid OS499 (8.05 t ha<sup>-1</sup>).

Tuble 6. Oful	<u> </u>							
		ZP341	ZP360	ZP434	OS499	ZP677	ZP704	Average
Negotino	2010	9,78	10,35	9,41	10,00	7,01	9,52	9,34
	2011	8,36	8,92	7,84	8,33	6,10	7,61	7,86
	Average	9,07	9,64	8,63	9,16	6,56	8,56	
Veles	2010	5,20	4,27	7,50	7,84	5,08	7,62	6,25
	2011	4,37	3,68	6,35	6,03	4,61	7,26	5,38
	Average	4,79	3,99	6,92	6,94	4,84	7,44	
	AVERAGE	6,93	6,82	7,78	8,05	5,70	8,00	

Table 8. Grain yield (t ha<sup>-1</sup>)

Genotype LSD<sub>0.05</sub>=0,973 Location LSD<sub>0.05</sub>=2,776 Interaction genotype x location LSD<sub>0.05</sub>=1,376

### Analysis of variance

Apart from the statistical analysis for every single traitby using LSD test for internal comparison of hybrids, as well as the comparison of the two localities with the aim of determining the effect of factors (locality, genotype and year), there has been a three-factor analysis of variance - ANOVA upon analyzed characteristics. With this analysis one can determine the separate influence of factors upon the examined features, as well as interactive influence of factors upon features.

Source of variation	Replication	Location	Genotype	Year				
Df	2	1	5	1				
Number of cobs per plant	0,002	0,001	0,005	0,011				
Cob length	3,900**	34,806**	8,898**	133,498**				
Number of rows per cob	0,851*	0,129	7,055**	86,002**				
Grain mass per cob	2382,135**	55752,398**	4012,242**	9938,620**				
Grain yield	5,940**	139,529**	10,013**	24,840**				

Table 9. Influence of factors on yield components and grain yield

Based on the determined results of the influence of factors upon examined traits (Table 9), it is evident that genotype and year have a significantly strong influence upon all analyzed features, except on the number of rows per cob. Apart from separate analysis, there was conducted analysis of interactive influence on examined factors upon examined traits (Table 10).

 Table 10. Interaction between factors

	Replication	Location	Location	Genotype	Location	Error
		* Genotype	* Year	* Year	* Genotype	
					* Year	
Df	2	5	1	5	5	46
Number of cobs per	0.002	0.002	0,001	0,005	0,002	0,004
plant	0,002	0,002	0,001	0,005	0,002	0,004
Cob length	3,900**	8,977**	1,856	1,150	1,416	0,738
Number of rows per cob	0,851*	2,262**	0,640	0,583	1,011**	0,242
Grain mass per cob	2382,135**	3811,047**	685,240	148,546	92,667	343,279
Grain yield	5,940**	9,528**	1,708	0,371	0,232	0,859

# Conclusions

Based on the determined results, several conclusions can be drawn. Considering the number of cobs per plant, most hybrids have one cob. The cob length, varied by year, region and genotype. On average, the hybrid ZP434 had the longest cob (18.11 cm), whereas ZP677 the shortest (15.94). The largest average number of rows for both localities was noted with ZP704 (14.87) and the smallest with ZP360 (13.19). On average for both years on both localities, the lowest value of grain mass per cob was noted with the hybrid ZP677 (113.9 g), whereas the highest with OS499 (160 g). The grain yield was the most important element in this research. In Negotino, the highest yield was found with ZP360 (9.64 t ha<sup>-1</sup>), whereas in Veles with OS499 (6.94 t ha<sup>-1</sup>). On average for the two localities, the best yield was noted with OS499 (8.05 t  $ha^{-1}$ ). The genotype and the year have a significantly strong influence on all analyzed characteristics, except on the number of cobs. The interaction locality x genotype showed the biggest influence on expression of all analyzed characteristics with statistical certainty at the level of 99%. As most stable of all examined hybrids, depending on year and locality, were hybrids from FAO 400, i.e. OS499 and ZP434. Considering the fact that both examined regions have similar climate conditions, these hybrids can be recommended as most dependable for cultivating in the regions of Veles and Negotino.

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### UDC: 633.11-116.424(497.7) Original scientific paper

# A LONG-TERM VARIABILITY OF THE WHEAT YIELD AND QUALITY IN REPUBLIC OF MACEDONIA

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## Abstract

The most important cereal and a strategic crop in Republic of Macedonia is the wheat. Domestic and foreign varieties that have different yield and quality are grown. Seed yield is varying from year to year, and beside from the genotypes, it depends on the climatic conditions on different locations and years. In order to assess the variability of yield and quality of wheat varieties grown in the R. of Macedonia, 44 wheat cultivars from three locations were analyzed in the period from 2002 to 2009. The varieties were assessed on different locations. The average seed yield ranged 3.90 t ha<sup>-1</sup> in 2003 to 8.10 t ha<sup>-1</sup> in 2009. The lowest average yield was detected in location Prilep  $(4.21 \text{ t ha}^{-1})$  and the highest in Skopje (7.90 t ha<sup>-1</sup>). On all three locations in the same period, higher relative yield compared to the control Radika, had the cultivars Prima, OSK-314/00, Tina, Mambo, IJZK-13/95 and SK-17/98-1/4, while lowere relative yield had Preslav, Milena, Mizaja, Bajka, Anastasija, Stamena, Sonata, Verbun Kos, Kodman, Makedonska rana, Balkanija and Lucija. Based on the relative yield the most stable were OSK-314/00, Tina and Mambo. The best results in location Skopje were achieved by the cultivar OSK-314/00, in Prilep by Vizija and in Strumica by Prima. Skopje represented ideal location for variety evaluation and assessment of wheat cultivars included in this study. Key words: wheat, yield, quality, variability.

# Introduction

Wheat represents a strategic culture in the Republic of Macedonia which is extensivly grown on its territory. The present assortment of varieties is caracterized with yields and quality that changes from year to year, and depends on the conditions of cultivation as well as the year and location. To provide increased yield and better quality is a complex challenge that requires an integrated approach, which among other things, includes an explanation of the interaction between the properties and the environment in different genotypes in a certain period of time. Although newly created varieties are characterized by high yields, there is a need of a constant creation of new varieties that will be better adaptable to the conditions of the external environment.

Considering the needs of creating more yielding varieties, the main objective of these researches was the assessment of the variability of yields for wheat varieties that

grown in Republic of Macedonia in a long term. By analyzing the long-term data from a wider assortment, we can get informations upon which the future strategies could be made and the directions of wheat selection could be projected.

# Material and methods

The data from the field trials of wheat, which were the subject of the research of the National Variety Commission, are used for the purpose of these studies. Different varieties were tested in three locations through the Republic of Macedonia: Strumica, Skopje and Prilep between 2002 and 2009. The experiments were set according to the method of randomized block design in five replications. The data on the yield of each location, in each year of cultivation, were analyzed by the method of analysis of variance, and average yields of analyzed varieties are mutually compared with LSD-test. To determine the locations of the achieved highest yields, average yields of wheat (an average value of all tested varieties in one year) in the appropriate localities are compared with LSD-test.

All varieties tested in the year of testing, are standardized in terms of the variety Radika. The obtained data were used to compare the relative contribution of all varieties grown in all locations from 2002 to 2009. The stability and adaptability of the tested varieties, as well as representativeness of locations for testing and cultivating wheat are determined by the GGE Biplot technique (Yan *et al.*, 2000). All statistical analyzes were made by using the statistical package R 3.0.3.

## **Results and discussion**

### Variability of wheat yield by location

In the period 2002 - 2009, the wheat varieties were tested in three locations: Strumica, Skopje and Prilep (Table 1). The lowest yields of the wheat have been achieved in 2003, an average of  $3.90 \text{ t} \text{ ha}^{-1}$  of three locations, the yield that is significantly statistically lower than other years of testing. This low yield primarily dues to the extremely low yield which was noted in Strumica and Prilep this year. The yield in Skopje this year does not deviate significantly from the annual average over this locality. The lowest yield in Strumica and Prilep is noted in 2003 (2.56 and 1.78 t ha<sup>-1</sup>, respectively). The highest average yield from three locations was received in 2004 (8.10 t ha<sup>-1</sup>), when also, the highest average yield was achieved in the locality of Skopje (9.77 t ha<sup>-1</sup>). Locality of Prilep is characterized by statistically significant lower average yield of wheat in the period from 2002 to 2009 (4.20 t ha<sup>-1</sup>), compared with the average yields in Strumica (6.74 t ha<sup>-1</sup>) and Skopje (7.89 t ha<sup>-1</sup>). Although the average yield in Skopje is the highest in comparison from other localities, it is not statistically significantly different from the average yield, achieved in Strumica. In all other years of testing the yield of wheat compared to the long-term annual average does not deviate significantly.

The interaction between genotype and environment is the biggest problem for breeders who produce varieties for specific regions (Khan *et al.*, 2014; Sabaghnia *et al.*, 2013). To be successful, a genotype should give good results in different growing conditions, or have a high average yield and consistent characteristics at different locations during more years (Gauch *et al.*, 2008). The breeders, often examined a series of genotypes

in more locations, before registration of commercial varieties and start of cultivation (Naghavi *et al.*, 2010). Because of that, the identification of these genotypes is very important (Annicchiarico, 2009). The studies that deal with the examination of the interaction between genotype and the environment, enables some external basis for selection of genotypes that will be used for mass cultivation or for a specific area with defined climatic conditions (Khan *et al.*, 2007; Yan and Rajcan, 2002).

V		Location	,	Maan adala
Year	Strumica	Skopje	Prilep	Mean yield
2002	5,16	7,49	6,14	6,26c
2003	2,56	7,36	1,78	3,90d
2004	9,13	9,77	5,41	8,10a
2005	7,66	8,16	4,10	6,64bc
2006	7,49	9,70	4,35	7,18b
2007	5,59	7,20	4,31	5,70c
2008	8,62	5,93	3,31	5,95c
2009	7,7	7,56	4,18	6,48bc
Mean yield	6,74b	7,90b	4,21a	6,28
LSD <sub>0,05</sub>	For location	1,544	For your	0,851
LSD <sub>0,01</sub>	For location	3,565	For year	1,257

Table 1. Average and mean yield per location and year (t ha<sup>-1</sup>)

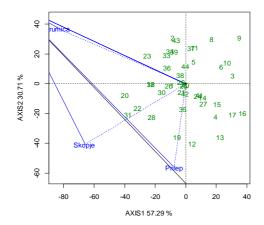
### Variability of wheat varieties and representativeness of location

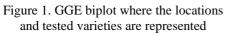
In order to compare all tested wheat varieties in period between 2002 and 2009, a relative contribution of each variety was calculated in terms of the variety Radika (code 1) in the corresponding year of the examination. Higher relative contribution in terms of the variety Radika (100 %) on all three localities showed only varieties 20, 31, 22, 28, 18 and 32 (Table 2). A large number of the tested varieties to all three localities had lower relative contribution in terms of the variety 1 (4, 5, 6, 8, 9, 10, 11, 24, 27, 37, 38 and 42).

Besides the yield amount it was necessary to assess its variability, in order to identify stable genotypes that can be grown in different climate areas. By determining the stability of individual varieties, the best varieties for specific regions included in the survey will be defined.

						cu to the standar	(, . )		
Code	Cultivar	Strumica	Skopje	Prilep	Code	Cultivar	Strumica	Skopje	Prilep
1	Radika	100	100	100	23	Liberta	113,91	107,1	86,14
2	Aglika	115,97	79,65	96,66	24	Verbunkos	82,12	99,11	95,51
3	Preslav	76,74	73,45	98,18	25	Suveges	88,96	104,4	89,89
4	Progres	71,88	94,91	104,26	26	Suba	96,25	108,6	91,76
5	Milena	95,49	86,5	92,4	27	Kodman	78,37	98,44	98,13
6	Mizaja	78,13	83,63	86,02	28	Mambo	105,3	110,6	114,9
7	Slaveja	96,88	94,47	102,13	29	Golubica	93,6	100,2	93,63
8	Bajka	91,67	75,66	84,19	30	Klara	98,23	112,4	94,01
9	Anastasija	80,56	63,94	84,5	31	OSK-314/00	111,7	125,3	107,1
10	Stamena	78,13	77,65	88,15	32	SK-17/98-1/4	110,38	106,6	101,3
11	Sonata	99,31	80,97	90,58	33	OSK-266/3	107,73	96,44	89,45
12	Vizija	82,89	100,31	123,87	34	OSK-251/02	105,33	96,86	85,23
13	Matica	64,47	99,23	108,54	35	OSK-236/01	84,53	109,8	97,05
14	KG-100	82,24	94,13	100,25	36	OSK-244/04	101,6	104,1	87,34
15	Edison	75	89,59	102,51	37	Mak. rana	95,71	89,75	82,08
16	Ekvizit	63,82	79,18	108,04	38	Balkanija	97,43	96,15	95,14
17	Ekskluziv	67,11	84,69	107,04	39	Mak. rodna	103,59	95,18	85,45
18	IJZK-13/95	110,38	106,6	101,35	40	Bt 04-069	94,88	93,69	101,0
19	Mihalica	86,75	110,22	114,98	41	Renata	79,76	101,0	91,81
20	Prima	122,52	114	110,49	42	Lucia	90,6	99,74	99,14
21	Zdenka	95,81	95,78	104,49	43	Pipi	105,54	91,05	82,76
22	Tina	109,49	119,11	106,37	44	Felix	87,95	105,2	76,72

Table 2. Average relative yield of wheat cultivars compared to the standard (%)





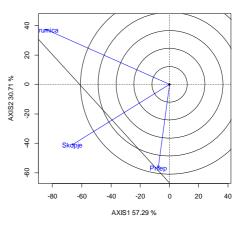
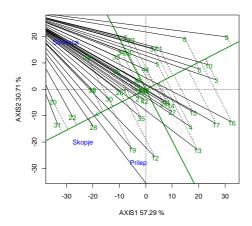


Figure 2. The relationship between the localities where the tests were performed

The relative yields of wheat varieties were used for analyzing the variability of cultivar stability, as well as the representativeness of the locations of wheat testing by using GGE biplot technique. The two main axes of biplot explained 80 % of the total variability that dues to genotype + interaction genotype x external environment, 57.29 % of variability is explained by the first axis and 30.71 % by the second axis (Figure 1). The varieties are indicated by the same ordinal numbers as in Table 2.

The relationship between the localities where the tests were performed is shown in Figure 2. The size of the angle between the individual locations shows the similarity or dissimilarity between the localities. The Figure 2 shows that the localities included in the study are different, with different climatic conditions and therefore, each of them received specific yields.



20 q 23 9 10 AXIS2 30.71 % 3 c 10 15 21 16 17 -20 13 е Р 20 -30 -20 -10 0 10 30 AXIS1 57.29 %

Figure 3. Average yield and stability of the tested wheat varieties

Figure 4. Ranking of the tested varieties according to the relative yield and stability

In the assessment of the stability of tested varieties, based on their relative contribution the varieties positioned on the axis in the direction of the arrow, are characterized by a high average yield, while those that are positioned closer to the axis, independently of the direction, are characterized by higher stability (Yan, 2002). In these studies the greatest stability showed the varieties 31, 22 and 28. The variety 20, although it had high yield on all localities, is less stable than the varieties mentioned above (Figure 3).

At the ranking of the genotypes, the best ranked is the variety that is closest to the center of concentric circles the biplot (Figure 4). In this research, it is the variety 31, followed by 22, 28 and 20.

In assessing the representativeness and discriminatory ability of the locations where the wheat examinations were performed (Figure 5), the locality that is the closest to the arrow biplot is rated as the most representative. In this research, it is the locality of Skopje. The further confirmation of this conclusion is the biplot for ranking localities (Figure 6), where the locality of Skopje is the closest to the center of the concentric circles, representing the ideal location for testing wheat varieties that are included in the survey.

Kamberi *et al.* (2013) examined the representativeness and discriminatory ability of these localities, but with different wheat varieties and found that for those varieties in the appropriate years of testing, the most discriminative location was in Prilep.

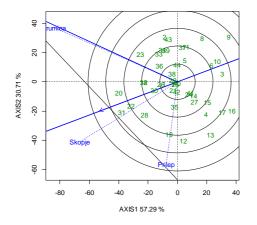


Figure 5. Representativeness and discriminative ability of localities

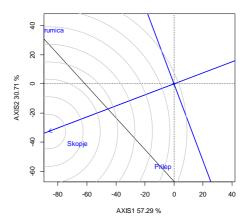


Figure 6. Ranking of the localities where the tests were performed

The Figure 7 presents the best varieties for different location to protruded parts of the polygon in which the appropriate location is. It could be concluded from the graph that each location represents a special outdoor environment in Skopje the best was the variety 31, in Prilep the variety 12 and in Strumica the variety 20.

If these varieties show good results in multiannual examination in the appropriate locations, they could be recommended for mass cultivation in these regions. GGE biplot technique proved successful in assessing the stability of wheat varieties and in the identification of locations and mega external environments for testing wheat in many studies (Kamberi *et al.*, 2013; Mohamed *et al.*, 2013).

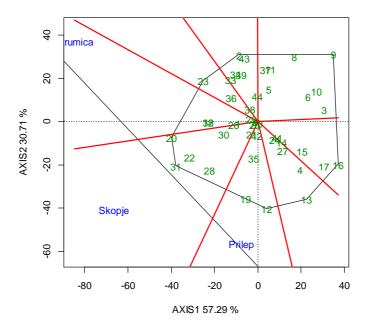


Figure. 7. The best varieties in individual locations

# Conclusions

As a result of the analysis of yield variability of some wheat varieties that are grown in R. Macedonia from 2002 to 2009, the following could be concluded:

The yield of wheat depends on both genetic factors as well as the environmental factors (location and year of cultivation) during the examination.

The average yield of tested wheat varieties ranged from  $3.90 \text{ t} \text{ ha}^{-1}$  in 2003 to 8.10 t ha<sup>-1</sup> in 2004 The lowest average yield in the period from 2002 to 2009 was obtained in Prilep (4.21 t ha<sup>-1</sup>), and the highest in Skopje (7.89 t ha<sup>-1</sup>). At all three sites, in the period from 2002 to 2009, a higher relative yield in terms of the variety Radika (100 %) achieved only varieties Prima, OSK-314/00, Tina, Mambo, IJZK-13/95 and SK-7/98-1/4.

Based on the relative yield the most stable varieties were OSK-314/00, Tina and Mambo. The best results were registered at the variety OSK-314/00 in Skopje, at the variety Vizija in Prilep and in Strumica at Prima variety. The locality of Skopje is an ideal location for testing wheat varieties included in this survey.

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### UDC: 633.11-146.1(497.712) 633.16-146.1(497.712) Original Scientific Paper

# RHIZOSPHERE MICROFLORA AT SOME CEREALS CULTIVATED IN SKOPJE REGION

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### Abstract

These studies were made to determine the number of different groups of microorganisms in soil, rhizosphere and root zone at wheat and barley fields in Skopje agricultural region. Research was conducted over two years, in laboratory and field conditions. The main purpose of these tests was to determine the dynamics (size) of rizosphere microflora in different stages of vegetation of wheat and barley and its effect on the characteristics of wheat and barley. Soil tests were taken during the three stages of vegetative growth of wheat and barley (stage of four leaves, flowering and physiological maturity). Based on the obtained results, it was concluded that the presence of rizosphere microflora, in terms of quantity uring the vegetation increases, with the highest value reaches the stage of intensive development in the flowering stage of culture. Compared with rizosphere microflora presentation of the tested microorganisms in the root zone are greater. In the third phase, compared with the second, rizosphere microflora is more prevalent. Based on this increase in numbers, a major impact had the use of agriculture technical measures. **Key words:** rhizosphere, rhizosphere microflora, wheat, barley, soil.

## Introduction

In the soil are living a big number of microorganisms, which present weight and depends on the type of microorganisms, depth location of the soil samples taken and the type of plant ass (Govedarica et al., 1995).

Accoring to Ranchevikj (2012), the microorganisms produce substances for growth and increase the content of nutrients in soil. They play a significant role in both pedogenetic processes: synthesis of organic matter and transformation of mineral materiel. Great importance for plant nutrition is mineralization process (Hajnal, 2004). There are numerous studies on mutual relation between plants and microflora, also there is a mutual dependence between the root of a plant and microorganisms (Lynch, 1990). The roots plant

affects the concentration of a number of microorganisms in the rhizosphere, which number is greater than in the area which is outside the root system. There are information's that different plants in different vegetation stages have different impact on the rhizosphere microflora. Plants show great effect on rhizosphere microflora in the early period of development.

The greatest effects on the number of microorganisms in the rhizosphere are in the stage before and during flowering of the plants. After the end of vegetation the number of microorganisms is reduced in rhizosphere.

In this paper were made to determine the number of different groups of microorganisms in different variants (control/soil, rhizosphere and root zone) in a variety of cereals (wheat and barley) in the Skopje region.

The main purpose of this study was to determine the dynamics (size) of rhizosphere microflora at different stage of vegetation on wheat and barley in the Skopje region.

## Material and methods

Material research in this paper is wheat variety altana, and barley variety rex, growth in the Skopje region.Studies are conducted on field and laboratory tests.

Soil samples (control/soil, rhizosphere and root zone) for microbiological analysis were taken three times during the vegetation, such as:

I (first) sample in the phase of 4 petals (fall);

II (second) sample at the stage of flowering (May);

III (third) sample under physiological maturity of wheat and barley (end of June).

The microbiological analysis of the control/soil, rhizosphere and root zone includes determining quantitative representation of different group's heterotrophic group of microorganisms.

In this paper were examined following types of heterotrophic microorganisms: amonificators, actinomycetes, yeasts, molds and the total number of bacteria.

Total number of bacteria, were examined on nutritional base mesopopton agar (MPA)(10<sup>-6</sup>);

Amonificators were examined on nutritional base Starch – ammoniacal agar  $(10^{-6})$ 

Actinomycetes were examined on nutritional base Waksman for actinomycetes  $(10^{-5})$ 

Yeasts were examined on nutritional base Sabouraud agar  $(10^{-4})$ 

Molds were examined on food media Chapekagar  $(10^{-4})$ 

The results were calculated in g/absolutely dry soil (a.d.s.).

Analyses on the number of heterotrophic microorganisms were made according to the method of dilution and plating on selective nutrient media. Each group microorganisms is incubated at an appropriate temperature in a thermostat for a period appropriate to the type of the examined microorganisms (Govedarica and Jarak, 1997).

Agrochemical analysis of soil included sampling of soil examined and preparation for laboratory analysis. Laboratory analysis was made of air-dry soil electric mill grind and sieved through a sieve with openings of *1 mm*. Agrochemical analysis of soil were made according to standard methods of chemical analysis of soil (Saric, 1998).

## **Results and discussion**

According to the results of agrochemical analysis of soil it can be concluded that reaction (pH- value) of the soil solution is neutral, soil humus is high and carbonates are low. Examined soil is well supplied with nitrogen and phosphorus and well provided with easily accessible potassium. Established results in the number of microorganisms (heterotrophic) in soil (control/soil, rhizosphere and root zone) under sowing of wheat in Skopje region are shown in Table 1.

Table 1 - Number of different types of microorganisms (heterotrophic) in soil under wheat in the	;
Skopje region	

		Type of microorganisms						
Variants	Stage during the vegetation	Bacteria (10-6)	Amonificators (10 <sup>-6</sup> )	Actinomycetes (10 <sup>-5</sup> )	Yeasts (10-4)	Molds (10-4)		
	Stage of 4 petals	2100	1100	123	35	68		
Control /Soil (0-30 cm)	Stage of flowering	3250	1290	154	40	72		
	Stage of physiological maturity	3220	1210	154	41	71		
	Stage of 4 petals	5220	1310	154	51	52		
Rizosphere	Stage of flowering	6650	3210	215	88	41		
	Stage of physiological maturity	5330	2360	186	69	34		
	Stage of 4 petals	3645	3050	169	66	40		
Root zone	Stage of flowering	12380	3260	262	97	34		
	Stage of physiological maturity	9500	2680	205	79	29		

Total number of bacteria are mostly present, according to the results shown in Table 1 (12.380.000g / a.d.s.) in flowering stage at root zone of soil, while most (2.100.00g / a.d.s.) at stage of four leaves in the control/soil.

The number of amonificators are biggest (3.260.000g / a.d.s.) in the phase of flowering in root zone, while the number is lowest (1.100.000g / a.d.s.) in phase of four petals in control/soil.

From the results, it can be concluded that the most (262.00g / a.d.s.) of actinomycetes are found in root zone, comparing with control/soil in the phase four petals, where the number is lowest (123.000g / a.d.s.).

The greatest representation (97.000g / a.d.s) at the yeasts in root zone are in the flowering stage, while the smallest presents (35.000g / a.d.s.) in the root zone at phase four petals.

Molds are largest (72.000g / a.d.s.) at phase of flowering in control/soil, while the lowest (29.000g / a.d.s.) are in the phase of physiological maturing in root zone.

Established results in the number of microorganisms (heterotrophic) in soil (control /soil, rizosphere and root zone) under sowing of barley in Skopje region are shown in Table 2. According to the results shown in Table 2, the total number of bacteria was highest (8.870.000g / a.d.s.) at phase of flowering in root zone, while the lowest (2.100.000g / a.d.s.) are in phase of four petals in the control/soil. From the results, it can be concluded that the number of amonificators are biggest (4.360.00g / a.d.s.) at stage of flowering of culture in root zone, while the number is lowest (1.100.00g / a.d.s.) in stage of four petals in the control/soil.

The numbers of actinomycetes are highest (317.000g / a.d.s.) in root zone in the flowering stage, while the number is lowest (123.000g / a.d.s.) in control/soil phase of four petals. Most yeasts (104.000g / a.d.s.) are present at root zone in the flowering stage, while the lowest (35.000g / a.d.s.) are present in control/soil phase of four petals.

Most molds (74.000g / a.d.s.) are present in phase of flowering in control/soil, while the lowest (68.000g / a.d.s.) are in the phase of physiological maturing in root zone. Results of the exams are corresponding with available literature data. Accoring to the results of tests of Walter Nelson Osorio Vega (2007), the wheat was observed a higher number of total number of microorganisms in rhizosphere soil, unlike the area of soil that is not under rhizosphere activity.

The largest number of bacteria in examining Govedarica et al., (1999) it was recorded in the end of the vegetation in the rhizosphere. The same sort of results obtained in the examination of Najdenovska et al. (2004).

Table 2 - Number of different types of microorganisms (heterotrophic) in the soil under barley in the
Skopje region

		Type of microorganisms						
Variants	Stage during the vegetation	Bacteria (10 <sup>-6</sup> )	Amonificators (10 <sup>-6</sup> )	Actinomycetes (10 <sup>-5</sup> )	Yeasts (10-4)	Molds (10-4)		
	Stage of 4 petals	2100	1100	123	35	68		
Control /Soil (0-30 cm)	Stage of flowering	2995	1320	258	77	74		
	Stage of physiological maturity	2916	1350	251	73	72		
	Stage of 4 petals	2860	1310	169	64	44		
Rizosphere	Stage of flowering	5590	2650	228	97	32		
	Stage of physiological maturity	4600	2290	198	73	29		
	Stage of 4 petals	3510	1720	177	71	33		
Root zone	Stage of flowering	8870	4360	317	104	30		
	Stage of physiological maturity	6920	3420	256	86	25		

According to the examination results of Đurić et al., (2004), determined to continuously increase the number of multiple groups of microorganisms, including amonificators during vegetation of grown crops.

The number of actinomycetes, according Najdenovska et al., (2004), declined the vegetation.

Yeasts and molds belong to the group of fungi. This group is present in all types of soil, but are dominant in soil with a sour reaction. According to statistic Govedarica (1999), the number of yeast are decreasing during the period of vegetation, the same result were concluded by Najdenovska (2004). The number of fungi reaches its maximum at the beginning of vegetation, and then decreases.

Bases on the results obtained in these studies, it can be conclude the following:

The total number of bacteria in soil is highest in physiological maturing stage in both crops (barley and wheat) and in all three tested areas (control /soil, rizosphere and root zone).

The number of amonificators during the vegetation in the soil with barley in Skopje region is the biggest in the stage of flowering of culture in all three tested areas.

In the control/soil, sowed with barley in the Skopje region, the number of

amonificators is biggest in the stage of physiological maturing of the crop.

When comparing the results of the number of actinomycetes in the soil with wheat and barley, it was found that most of actinomycetes found in flowering sage of the crop in all three studied areas in the Skopje region.

The number of yeasts in soil sown with wheat is the largest in the rhizosphere and root zone in phase of flowering of culture. In the area of control/soil, the greatest number of yeast is found in the phase of physiological maturing of the wheat.

The largest number of molds in the soil sown with wheat was noted in stage of four petals in the rhizosphere and root zone. Also, in the control/soil in the Skopje region, greater amounts of molds are visible in the later period of vegetation in the flowering stage and the stage of physiological maturity.

The number of molds in the soil sown with barley is the largest in the control/soil. In control/soil most molds are found in the flowering stage, while in rhizosphere most molds was noted in phenophase of four petals.

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# CHEMICAL PROPERTIES IN SOME DRY TOBACCO VARIETIES FROM TYPE PRILEP

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### Abstract

The chemical properties have primary significance during the process of tobacco raw material quality determination as well as for the tobacco products manufacture. All researches are being performed in two years period (year 2009 and 2010), on our current tobaccovarieties from type Prilep: P - 23 Ø; P 12- 2/1, NS – 72; P – 66 – 9/7; P -79 – 94 and Prilep Basma-82. The field trials and chemical researches were performed in Scientific Tobacco Institute-Prilep according to the standards and already established methods. The main goal was to determine the content of the most significant chemical properties in dry tobacco, by means of presenting the quality of tobacco raw material of the examined varieties. Once the researches were performed, from the given results we can conclude that the examined varieties have well balanced proportion among the components and they have good chemical properties typical for the tobaccofrom oriental, aromatic type. Also we have established that the genetic constitution of the tobacco varieties has influence on the chemical properties with specific content of certain components (eg. Nicotine in P-23), which is a variety distinction.

Key words: oriental tobacco, varieties, Prilep type, chemical properties.

## Introduction

The tobacco production is significant for the Macedonian economy. Its significance can be noticed in the raw material which is used mainly for export on world's tobacco markets. Our country is known for produce of high quality aromatic tobacco from oriental type, which is included in the blend recipe of some of the world most famous cigarette brands. In the recent years in the Republic of Macedonia the average tobacco production is evaluated around 25 000 tons, out of which 95% are oriental typePrilep and Yaka, smaller percentage are Djebel and Basmak. The demand and consumption of all these tobaccos depends of the quality of the produced raw material. According to Alić-Đemidžić (1999) there are 3 criteria for tobacco quality evaluation: 1. Organoleptic properties – it refers to the visual parameters such as: color, nervature, size, leaf body etc.; 2. Analytical properties – all technological and chemical tobacco characteristics; 3. Smoking properties – given with degustation. Our researches were performed for determination of the chemical properties of the tobacco varieties from type Prilep which are regularly used in the large

scale production, and one newly established variety. Tobacco chemical composition includes over 3 000 chemical components, but we have analyzedonly those which have greater influence on the tobacco quality and the tobacco smoking properties: total nitrogen, nicotine, protein, soluble sugars and mineral matters.

# Material and methods

The following six varieties of tobacco type Prilep were used for this research: Prilep, P - 23 Ø; Prilep, P 12- 2/1, Prilep, NS – 72; P – 66 – 9/7; Prilep, P -79 – 94 and PrilepBasma-82. Field trial was set up at the experimental field of the Scientific Tobacco Institute- Prilep in the period of the years 2009 and 2010 on deluvial-colluvial soil. During tobacco vegetation period, all agrotechnical practices needed for normal growth and plant development, were performed. Previously the plot was fertilized with NPK fertilizer in a combination 8:22:20 with amount of 250 kg/ha. In 2009 the tobacco plots have been watered three times, in 2010 two times (in July and August) with water norm of 250 m<sup>3</sup>/ha water. The harvested and hand strung tobacco is sun dried in a traditional way. During period of two years for the chemical properties (chemical composition) average dry tobacco samples from each variety were tested separately. The tests were performed in the accredited Laboratory for quality control of tobacco and tobacco products - L03 in the Scientific Tobacco Institute-Prilepaccording to the standards and recommended methods. Some of the analyses which we have performed were determination of the total nitrogen using Kjeldahl method; spectrophotometric determination of nicotine; protein determination using Moore method; determination soluble sugars with Bertrand method and mineral matters (ash) determination with dry mineralization (combustion).

### **Results and discussion**

The chemical constituents are the basic component of the dry substance on which depend many smoking properties of the cigarettes as a tobacco's final product. Many scientists have analyzed the chemical constituents and have determined that the tobacco propertiesdepending on the genetic constitution, type, variety, cultivation conditions, the time of harvest, method of drying and fermentation. The research results for each of the properties are presented in the following tables in order to have better preview, varieties comparison and it's easier for making conclusions.

# Total nitrogen content

Total nitrogen content in tobacco is in negative correlation with the quality, especially if it's over 3%. According to Uzunoski (1985) the low nitrogen percentage has positive influence on tobacco smoking. The results from the two years experiments are presented in Table 1.

Tobacco varieties	Year		Average	Index
	2009	2010	Average	muex
P-23 Ø	2.68	2.16	2.42	100.00
P 12-2/1	2.56	1.98	2.27	93.80
NS-72	2.49	2.08	2.28	94.21
P-66-9/7	2.26	2.02	2.14	88.43
P-79-94	2.52	1.94	2.23	92.15
PrilepBasma- 82	2.58	1.97	2.27	93.80

Table1. Total nitrogen content(%)

The average values present that the highest percentage of total nitrogen (2.42%) is being established in the variety P-23, the lowest is in P-66-9/7 (2.14%). Among the rest of the varieties, the total nitrogen content varies from 2.23% in P-79-94 to 2.28% in NS-72. The differences in the total nitrogen during two year analyses are due to rainy 2010 (298 l/m<sup>2</sup>), during tobacco vegetation there is lower percentage starting from 15% up to 20% of total nitrogen among the other tobacco varieties which confirms by Lazaroski (1983) and Hristoski (2006) about the reduction of the total nitrogen content with tobacco watering.

### Nicotine content

The alkaloids are the most characteristic tobacco chemical compound. Tobacco has several alkaloids but the most important of them is the nicotine. It's generated in the root from where it's transported and kept in other plant parts. Highest percentage of nicotine is placed in theleaves, the matured seed doesn't contain any nicotine. According to Avundžijan, cited by Pasoski (1980), the nitrogen fertilizers increase the nicotine percentage in tobacco leaves to a certain level with no further influence to its content. During our analyses we've noticed that the nicotine was varying starting from 0.56% in P 12-2/1, up to 1.52% in P-23 (Table 2). We can see from the table that the investigated oriental varieties from tobacco type Prilep have nicotine starting from 1.00 up to 1.30%. Also it can be noted that the rainy 2010 had influence on lowering of the nicotine percentage among the other tobacco varieties.

Tobacco varieties	Year		Average	Index
	2009	2010	Average	muex
P-23 Ø	1.96	1.08	1.52	100.00
P 12-2/1	0.69	0.44	0.56	36.84
NS-72	1.24	0.96	1.10	72.37
P-66-9/7	1.18	0.83	1.00	65.79
P-79-94	1.46	0.88	1.17	76.97
PrilepBasma- 82	1.66	0.95	1.30	85.53

Table 2. Nicotine content (%)

### Protein content

The proteins are included in the cell structure and have influence on the leaf bodyimprovement as well as on the external look of the tobacco. If the protein content is on lower level the leaves are thinner, smother, with greater elasticity and shinier color. If the protein presence is higher results with thickening of the leaf lamina, lower elasticity and darker leaf color. According to Smuk (1948) high quality tobaccos should contain proteins in percentage of 5-9%. The results of our research are presented in Table 3.

Tobacco varieties	Ŋ	lear	Augraga	Index
	2009	2010	Average	muex
P-23 Ø	6.69	6.82	6.75	100.00
P 12-2/1	6.95	6.11	6.53	96.74
NS-72	6.62	6.62	6.62	98.07
P-66-9/7	6.34	6.83	6.58	97.48
P-79-94	7.22	5.48	6.35	94.07
PrilepBasma- 82	7.04	6.24	6.64	98.37

Table 3. Protein content (%)	Table 3.	Protein	content	(%)
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We can see that the protein content in the tobacco varieties is in the optimal levels. Lowest percentage of protein content is notices in P-79-94 (6.35%), the highest is in the control (6.75%). Also in year 2010 can be noticed that the protein content in the varieties P 12-2/1, P-79-94 and PrilepBasma 82 has lowest percentage, which is a result of the higher quantity of rain.

## Soluble sugars content

The soluble sugars are one of the most active substances in tobacco metabolism. They are used as substance which gives energy during the process of respiration as well as in the processes of construction of other organic compounds. They are significant in the process of tobacco development and yield as well as its quality. According to Veselinov, cited by Uzunoski (1985) high quality oriental tobaccos contain soluble sugars in percentage starting from 14 up to 18%. According to Nuneski (1986) tobacco type Prilep has soluble a sugar on the lower middle leaves with percentage of 18.41%, the middle leaves have 19.11% and the higher middle leaves has 20.69%. The results of our research are presented in Table 4.

Tobacco varieties	Year		Auoroaa	Indou
	2009	2010	Average	Index
P-23 Ø	16.44	24.39	20.41	100.00
P 12-2/1	14.43	24.25	19.34	94.76
NS-72	14.01	17.81	15.91	77.95
P-66-9/7	19.41	22.54	20.97	102.74
P-79-94	12.55	25.08	18.81	92.16
PrilepBasma- 82	12.44	27.30	19.87	94.90

Table 4. Soluble sugars content (%)

The average values show that the variety NS-72 has lower percentage of sugars (15.91%), the variety P-66-9/7 has highest percentage of sugars (20.97%). Control (P-23) has 20.41% sugars. The same conditions for cultivation and agro techniques, the influence of the varieties genetic constitution, the rainy 2010 has also increased the soluble sugars content. If we compare the two years research (year 2009 and 2010) we can see that the varieties P-

79-94 and PrilepBasma 82 have highest percentage of sugars increase of 100%, respectively 119%.

## Mineral matters content

Mineral matters are significant for tobacco's chemicalcomposition and they participate with 9-30% in the total dry weight. They are presented with ash content which is a leftover after tobacco combustion. According to Butorac (2009) the most important ingredients of tobacco mineral complex are potassium, calcium and magnesium, the other elements are represented in minimal amounts. Potassium has positive influence on the combustion, but magnesium negative. Especially negatively affect chlorine salts if they are in a highamount. According to Boceski (2003) the oriental aromatic tobacco from type Prilep depending on the insertion in top leaves has 11.84% mineral matters; higher middle leaves has 15.54%; lower middle leaves has 17.58% mineral matters (raw ash). The author emphasizes the fact that high quality tobacco has less mineral matters. In our research (Table 5) the mineral matters content in percentage starts from 10.73% in P-66-9/7 up to 14.51% in P-23 (Ø). We can see that in other varieties mineral matters are represented with 11.55% in PrilepBasma 82; 12.10% in P-79-94; 13.21% in NS-72 and 13.95% in P 12-2/1 which shows us that these tobacco varieties from tobacco type Prilep have good quality.

Tobacco varieties	Year		A	T. 1.
	2009	2010	Average	Index
P-23 Ø	14.10	14.92	14.51	100.00
P 12-2/1	14.50	13.40	13.95	96.14
NS-72	12.76	13.67	13.21	91.04
P-66-9/7	10.52	10.95	10.73	73.95
P-79-94	13.22	10.99	12.10	83.39
PrilepBasma- 82	12.64	10.47	11.55	79.60

 Table 5. Mineral matters content (%)

# Conclusions

Based on the two year researches of the chemical properties of dry tobacco on examined tobacco varieties of Prilep type we can conclude:

- Total nitrogen content has recommended quantity for oriental tobacco from Prilep type. All varieties the amount of total nitrogen is lower than 3% which has positive influence on smoking raw material.
- Nicotine is represented with minimal percentage of 0.56% in P 12-2/1 and maximum of 1.52% in control variety P-23. We have established the nicotine content depends of the soil climatic conditions, as well as it's a variety distinction.
- The results have shown that the proteins are represented starting from 6.35% in P-79-94 up to 6.75% in control variety, which is a characteristic of tobacco oriental aromatic varieties of tobacco type Prilep and a sign of high quality raw material.
- The soluble sugars have optimal values due to rainy weather conditions during vegetation time in year 2010, which was the reason why their content was higher.

Approximately lowest percentage was seen in NS-72 (15.91%), highest in P-66-9/7 (20.97%). There is a balanced proportion between the soluble sugars and proteins which show us that it's a high quality tobacco.

- Mineral matters vary from 10.73% in P-66-9/7 up to 14.51% in P-23 which is a positive feature because the raw material combustion, organic substances can be easily transferred in carbon dioxide and the ash has white-grey color.
- In the end we can conclude that the analyzed chemical properties of the given varieties are performed on high quality oriental tobacco raw material which also has good degustation features.

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# EFFECTS OF FERTILIZATION AND IRRIGATION OF THE PRODUCTION PROPERTIES ON SEMI ORIENTAL TOBACCO VARIETY OTLJA-ZLATOVRV

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### Abstract

The past few years, Alliance One Tobacco Company made serious efforts for restarting the semi oriental tobacco production in Republic of Macedonia. To improve the sort randman the Scientific Tobacco Institute created new semi oriental tobaccos with high yield and quality. One of them is the variety Otlja-Zlatovrv. The main aim in this investigation is to study the effects of fertilization and irrigation of the production properties on semi oriental tobacco variety Otlja-Zlatovrv. The results of this investigation will be great opportunity for production of this tobacco type. Three-year field experiments were set up on a deluvial-colluvial soil type in 12 variants with three replications. The trial was bifactorial, with three rates of nitrogen fertilizer (25, 35 and 45kg N/ha) and two irrigation regimes (45 and 60% FC). The obtained results confirm that fertilization and irrigation have positive impact on the production properties of this tobacco. The yield has increased by 89.98% (var. 12), the average price by 13.57% (var.7), and the income per unit area by 109.05% compared to the control variety. Fertilization and irrigation have significant impact on reduction of nicotine and proteins and on increase of soluble sugars.

Keywords: semi-oriental tobacco, Otlia, fertilization, irrigation.

### Introduction

Cultivation of tobacco variety Otlja first began in Kumanovo, partly in Skopje and Preshevo area (Горник, 1932). First data from producing of this type are found from 1930. In the period from1950 to 1980 year, average production of semi oriental tobacco type otlja in the Republic of Macedonia is2350tons. At the beginning of this century the production of this type of tobacco is kept to a minimum on limited areas. In the past recent years in Macedonia are cultivated only oriental varieties: Prilep, Yaka, Basmak, Jebel and semi oriental type Otlja. The most abundant type is Prilepwith62.11 %, then comes the variety Yaka with28.06 %, Basmak with9.43 % and negligible production have variety Jebel with 0.34 % and Otlja0.06%. The past few years, Tobacco Company - Alliance One made serious efforts for restarting the semi oriental tobacco production in the Republic of Macedonia, especially in the Kumanovo and Skopje producing area, as most characteristic areas. That is duethe fact that semi oriental types of tobacco are characterized by: fine leafs, delicate fabrics with low content of rib, good factory dressing percentage, while

tasting delivers full, sweet neutral flavor, and smoking it does not scratch the throat (Кочоска, 2008). According to Патче (1979) tobacco raw Otlja issuchafine material with specific underlined tobacco features used to improve the taste properties of mixtures.

In addition to improving the dressing percentage of types of tobacco, Scientific Tobacco Institute-Prilep continuously works on creating new high yielding and quality semi oriental varieties. One of them is semi oriental variety Otlja-Zlatovrv. This type of tobacco as other tobaccos has general biological characteristic of plasticity, i.e. easily react and change in terms of change in eco environment and production technology. Патче (1979) points out that depending on the production conditions, it was noted that in practice, this type of commodity "flavor" changes to "strong" or other ordinary coarse tobacco, thus changing the appearance from bright yellow to red or variegated (brownish-red). Taking all above mentioned things the purpose of this study was the impact of fertilization and irrigation, as indispensable factors in the production technology of tobacco, the changes of product quality and properties of semi-oriental variety Otlja-Zlatovrv.

### Material and methods

Three years filed experiments were done with variety Otlja-Zlatovrv. According to the established methodology it was two factorial experiment with three doses of nitrogen fertilizers (25,30 and 45 kgN/ha), constant amounts of phosphorus (80 kg/ha), potassium (100 kg/ha)and two irrigation regimes (45% and 60% of FWC), in a random block system with three replications as follows:

1. Ø unfertilized, non- irrigated	7. N $_{35}$ P $_{80}$ K $_{100}$ + 45 % of FWC
2. $N_{25}P_{80}K_{100}$	8. N $_{45}$ P $_{80}$ K $_{100}$ + 45 % of FWC
3. $N_{35}P_{80}K_{100}$	9. Øunfertilized + 60 % of FWC
4. $N_{45}P_{80}K_{100}$	10. N $_{25}$ P $_{80}$ K $_{100}$ + 60 % of FWC
5. Øunfertilized + 45 % of FWC	11. N $_{35}$ P $_{80}$ K $_{100}$ + 60 % of FWC
6. N $_{25}$ P $_{80}$ K $_{100}$ + 45 % of FWC	12. N $_{45}$ P $_{80}$ K $_{100}$ + 60 % of FWC

Soil preparation is performed with an autumn (30cm deep) and two spring tillage's (8-20 cm depth). Soil samples were taken for testing agrochemical properties of soil before setting the experiment. Fertilization was done with mineral fertilizer NPK8:22:20 and feeding with27 % KAN. 50% of nitrogen was added with the last plowing before planting, together with phosphorus and potassium, and the remaining 50 % of the nitrogen input was applied at the first digging. Each elementary plot has 4 rows, with 8 plants in row a total of 32plants in the plot. The whole experiment has a total of 1152 plants, with planting distance of 50x25cm. During the vegetation of tobacco all necessary measures to protect the tobacco from diseases, pests and weeds were taken. The quantity of water to maintain the regimes of 45 and 60% of FWC are calculated by the current moisture of the soil. Harvesting is done on dry tobacco, weighing and qualitative assessment by the standards for purchase of dry tobacco (Rules on criteria for qualitative and quantitative assessment of raw tobacco leaves ("Official Gazette" of R. Macedonia, No. 16/2007). Agrochemical soil parameter sand chemical components of tobacco raw materials are determined by standard

methods in accredited laboratories of the Scientific Tobacco Institute-Prilep. The survey results are statistically processed by ANOVA method and LSD test.

# **Results and discussion**

# Soil and climate characteristics

The soil on which the tests were performed is colluvial-delluvial, light loamy with moderately acidic pH (KCl), low humus content and average phosphorus and potassium content (Table 1). Meteorological conditions during the vegetation, especially the amount and distribution of rainfall can have a limiting effect on the yield of tobacco per unit area.

Depth (cm)	рН		Humu	mg/100 g soil		Physical clay
	$H_2O$	KCl	s %	$P_2O_5$	$K_2O$	%
0 - 30	6,64	5,98	0,81	15,69	13,30	24,5
30 - 60	6,46	5,78	0,65	11,81	12,22	26,8
Classification	low acid	moderately acid	low	medium	medium	light loamy

Table 1. Agrohemical propertis of the soil

The temperature as a meteorological element has exceptional importance on the growth and development of tobacco plant. Optimum temperature for the tobacco plant is considered average daily temperature of 22 to 25°C, and border equivalents of shortages and excesses ranging between 18 and 30 °C (Атанасов 1965). The average daily temperature during the three years of research is 19.8°C (Table2) and is typical for the Prilep region as suitable for successful cultivation of semi-oriental tobacco types. Average participation is 203.1 mm. Average participation in 2007 is 229.9 mm. During the vegetation period in 2008 there is 46.6 mm rainfall less than the previous year (183.3mm), while in 2009 this amount is less for 33.9 mm, (196.0mm). The quantities of precipitation are sufficient for normal growth of tobacco, but the schedule of precipitation is unfavorable as it can be seen from the presented data. According to Атанасов (1965), tobacco has greatest need for water 30-40 days after planting. That is the period of intensive growth and is during the July. In this period, during the three years of our investigation, the amount of participation is between 5.3 and 11.0 mm. According to many authors, summer precipitation of 20-25 mm can provide enough soil moisture for 10 days, participation under 10 mm is not enough for normal growth of the tobacco in summer period, while participation of less than 5 mm during a decade brings complete drought (Атанасов, 1965).

Month	0		0	erature (°C)	Precipitation	Days with
Month	Year	Daily	Min	Max	mm	precipitation
	2007	16.9	11.5	22.5	74.3	14
May	2008	16.7	9.7	23.0	41.3	8
	2009	15.8	8.7	25.0	55.0	10
	2007	21.6	15.6	28.0	79.5	11
June	2008	19.9	12.3	29.1	10.0	5
	2009	18.5	11.8	27.6	75.0	10
	2007	25.3	17.1	32.8	5.3	1
July	2008	22.3	13.7	31.3	11.0	4
	2009	21.9	13.4	30.6	8.0	3
	2007	23.7	17.0	30.4	54.2	2
August	2008	23.6	14.1	33.3	11.0	2
	2009	21.4	14.0	29.2	43.0	7
	2007	16.9	10.6	23.5	16.6	5
September	2008	15.8	9.7	23.9	110.0	10
	2009	17.1	11.0	25.3	15.0	5
A	2007	20.9	14.4	27.4	229.9	33
Average/Total (V - IX)	2008	19.7	11.9	28.1	183.3	29
$(\mathbf{v} - \mathbf{I}\mathbf{A})$	2009	18.9	11.8	27.5	196.0	35
Annual	2007-2009	19.8	12.7	27.7	203.1	32
average	1999 - 2008	19.9	13.4	26.5	205.7	34

Table 2 Meteorological data during the vegetation period

The changes of climatic factors in recent years including increased temperatures, reduced rainfall and its erratic distribution during the growth phase led to drought in different growth stages of plant and crop yield levels significantly reduced in dry conditions (Salehzadeh et al, 2009). According to the presented data on the quantity of rainfall and literature data it can be concluded that without irrigation, normal growt hand cost-effective yield of tobacco cannot be ensured. That is why further watering was done, especially during the critical periods when tobacco has the greatest need for water.

To maintain the water level in the soil according to established methodology, during 2007 - 2009 were four irrigations were done, and the quantity of water was different for the two levels of maintained humidity (40% and 60% of FWC), depending on the current moisture content of the soil.

#### Average yieldkg/ha

The average yield of tobacco per unit area is lowest in variant1 (control, unfertilized, nonirrigated) and is 1368 kg/ha. Fertilized variants with 45% and 60% of FWC (var.5 and 9) have an increased yield for 9.65% or 15.35% more than the control, which points out on the positive impact of water as solvent for mineral matter.

These variants are statistically significant on 0.05level. In fertilized variants yield increases accordingly to the dose of the nitrogen 7.27% (var.2) to 26.68% (var.4). Interaction effect of fertilizer and water is most evident invariants 6,7and 8,where the yield

of tobacco increased from45.47 % to 85.16 %. These variants obtained statistical significance of all three levels of probability. Variants 10,11 and 12, where the humidity is maintained at 60% of FWC achieved high yield, but in terms of variants 6, 7 and 8 where the humidity is maintained at45% of the FWC there are not statistically significant differences. Based on the results it appears that by maintaining moisture in the soil of45 % of FWC, in terms of fertilization we ensure high and reliable yield of variety Otlja-Zlatovrv.

According to the presented data and the impact of fertilizing and watering to the average price (Table 4), it can be concluded that only fertilization increases the price of raw material from 4.12 to 7.24 %, and the interaction effect of fertilization and irrigation to 13.57 % (var. 7) compared to the control.

N <sup>O</sup>	Variant		Year		_	%
IN	variant	2007	2008	2009	Х	90
1	Øunfertilized, non-irrigated	1345	1436	1322	1368	100.00
2	$N_{25}P_{80}K_{100}$	1417	1570	1413	1467	107.24
3	$N_{35}P_{80}K_{100}$	1419	1982	1488	1630	119.15
4	$N_{45}P_{80}K_{100}$	1452	2209	1539	1733	126.68
5	Øunfertilized + 45% FWC	1528	1541	1430	1500	109.65
6	$N_{25}P_{80}K_{100}+45\%$ FWC	2019	2141	1809	1990	145.47
7	$N_{35}P_{80}K_{100}$ + 45% FWC	2302	2473	2155	2310	168.86
8	$N_{45}P_{80}K_{100}$ + 45% FWC	2577	2597	2425	2533	185.16
9	Øunfertilized+ 60% FWC	1718	1587	1430	1578	115.35
10	$N_{25}P_{80}K_{100}+~60\%$ FWC	2038	1981	1874	1964	143.57
11	$N_{35}P_{80}K_{100}+~60\%$ FWC	2406	2459	2185	2350	171.78
12	$N_{45}P_{80}K_{100}$ + 60% FWC	2593	2643	2561	2599	189.98
LSD	2007 2008	2009				
0.05	= 125 kg/ha 141 kg/ha	78 kg/ha				
	= 169 kg/ha 191 kg/ha	106 kg/ha				
0.001	= 257 kg/ha 257 kg/ha	143 kg/ha				

Table 3 Yield of tobacco (kg/ha)

Fertilized and irrigated variants with 45 % of FWC have a better effect on the average cost than variants irrigated with 60 % of FWC. In 2007, the results have a statistical significance at the probability levels of 5 % and 1 %, and in 2008 and 2009, irrigated and fertilized variants have statistical significance at level of probability 0.1 %. The obtained results from our investigations are similar to those from Тодороски (1970), Лазарески (1977), Димитриески (2004) etc.

			Year			
N <sup>O</sup>	Variant	2007	2008	2009	x	%
1	Øunfertilized, non-irrigated	31.99	25.32	56.08	37.80	100.00
2	N <sub>25</sub> P <sub>80</sub> K <sub>100</sub>	32.79	27.66	57.32	39.26	103.86
3	N 35 P80 K100	32.53	28.43	57.26	39.41	104.26
4	$N_{45}P_{80}K_{100}$	31.73	27.89	56.08	38.57	102.04
5	Øunfertilized + 45% FWC	32.61	26.03	56.09	38.24	101.16
6	$N_{25}P_{80}K_{100}+45\%$ FWC	35.93	30.47	59.24	41.88	110.79
7	$N_{35}P_{80}K_{100}+45\%$ FWC	35.70	32.17	60.91	42.93	113.57
8	$N_{45}P_{80}K_{100}$ + 45% FWC	34.24	32.67	59.05	41.99	111.08
9	Øunfertilized+ 60% FWC	32.24	26.34	56.91	38.50	101.85
10	$N_{25}P_{80}K_{100}+ 60\%$ FWC	33.66	31.15	60.17	41.66	110.21
11	$N_{35}P_{80}K_{100}+ 60\%$ FWC	34.23	32.20	61.74	42.72	113.01
12	$N_{45}P_{80}K_{100}+~60\%~FWC$	33.72	31.60	58.55	41.29	109.23
LSD	2007 2008	20	09			
0.05 :	= 1.65  den/kg  1.43 den/l	kg 1.64	den/kg			
0.01 :	= 2.24  den/kg  1.94 den/l	kg 2.22	l den/kg			
0.001	= n.s. 2.62 den/k	kg 2.99	den/kg			

Table 4. Average price of tobacco (den/kg)

Gross cash income per hectare (Table5) represents the yield and quality of tobacco for each set of variants. The great yields per hectare and quality of raw tobacco (average price per kg of dry tobacco) the greater economic impact and vice versa. For variety Otlja-Zlatovrv lowest gross income has control variant with 51165 (den/ha), and the highest variant 12 (106960 den/ha), that is 109.05% more than unfertilized and non-irrigated variant.

Variants fertilized and irrigated with 45 % of FWC showed a significant increase in gross income compared to fertilized and not irrigated variants, while compared to variants fertilized and irrigated with 60 % of the FWC there is no significant increase. Analysis of the results for this parameter showed a statistically significant difference between control and fertilized and irrigated varieties.

The chemical composition of the raw material depends on the type of tobacco, agrotechnical measures during vegetation, insertion and technological processing. The presence of the chemical components and their relation are essential for the qualitative value of raw tobacco (Димитриески, 2004). Based on the data it can be concluded that examined agro measures have a dominant influence on the values of important chemical parameters, and thus in their correlation. The increasing doses of nitrogen (var.2-var.4) influenced the increase of the nicotine content to1.41%, the content of protein (8.29 % - 9.82 %), and the mineral matter from12.83 % to 14.20 %. The content of soluble sugars decreases with increasing doses of nitrogen.

N <sup>O</sup>	Variant		Year		_	%
19	v arrain	2007	2008	2009	Х	70
1	Øunfertilized, non-irrigated	42991	36398	74106	51165	100.00
2	N 25 P80 K100	46457	43419	81013	56963	111.33
3	N 35 P80 K100	46142	56397	85174	62571	122.29
4	$N_{45} P_{80} K_{100}$	46078	61588	86174	64613	126.28
5	Øunfertilized + 45% FWC	72498	40140	80226	64288	125.65
6	$N_{25}P_{80}K_{100}+45\%$ FWC	72498	65234	107200	81644	159.57
7	$N_{35}P_{80}K_{100}$ + 45% FWC	82155	79518	131290	97654	190.86
8	$N_{45}P_{80}K_{100}+45\%$ FWC	88185	82231	143183	104533	204.30
9	Øunfertilized+ 60% FWC	55388	41806	81396	59530	116.35
10	$N_{25}P_{80}K_{100}+~60\%$ FWC	68892	61714	112787	81131	158.57
11	$N_{35}P_{80}K_{100}+~60\%$ FWC	83490	79168	134951	99203	193.89
12	$N_{45} P_{80} K_{100}$ + 60% FWC	87420	83502	149959	106960	209.05
LSI	D 2007 2008	3	2009			
0,05	5 = 4538  den/ha  4540 de	en/ha 6	326 den/ha			
0,01	$1 = 6168 \text{ den/ha} \qquad 6171 \text{ det}$	en/ha 8	598 den/ha			
0,00	01 = 8297  den/ha  8301 d	en/ha 11	566 den/ha			

Table 5 Gross income of tobacco (den/ha)

At irrigated and fertilized and irrigated varieties there is great reduction of nicotine content, protein and mineral matter and significant increasing of the content of soluble sugars. The results of our research are within specific variety Otlja (Узуноски 1985).Certain deviation is observed in great percentage of soluble sugars, but these values are close to research of Богданчески (1981) which found that the content of soluble sugars ranged from 15.23 to 25.16%. Лазарески(1982), points out that irrigated tobacco has a lot of carbohydrates, a lower percentage of nicotine and proteins, it is soft, with low flammability and good flavor.

		Nic	cotine	Pro	oteins	Solubl	e sugars	Minera	l matter
N <sup>O</sup>	Variant	$\bar{\mathbf{x}}$	%	$\overline{\mathbf{X}}$	%	$\bar{\mathbf{x}}$	%	$\bar{\mathbf{x}}$	%
1	Øunfertilized, non-irrigated	1.35	100.00	8.82	100.00	11.12	100.00	12.26	100.00
2	$N_{25}P_{80}K_{100}$	1.41	104.07	8.29	93.99	11.91	107.06	12.83	104.61
3	N 35 P80 K100	1.53	113.33	9.70	109.92	11.65	104.77	13.60	110.93
4	$N_{45}P_{80}K_{100}$	1.89	139.63	9.82	111.34	11.17	100.40	14.20	115.82
5	Øunfertilized+45% FWC	0.51	37.78	7.40	83.90	19.68	176.98	10.91	88.99
6	$N_{25}P_{80}K_{100}$ + 45% FWC	0.62	45.93	7.68	87.07	17.46	156.97	12.67	103.34
7	$N_{35}P_{80}K_{100}$ + 45% FWC	0.71	52.59	7.86	89.12	18.22	163.85	13.20	107.67
8	$N_{45}P_{80}K_{100}$ +45% FWC	0.87	64.44	8.08	91.61	19.53	175.63	13.31	108.52
9	Øunfertilized+ 60% FWC	0.49	36.30	7.01	79.48	20.06	180.35	12.12	98.86
10	$N_{25}P_{80}K_{100}$ + 60% FWC	0.60	44.44	6.45	73.07	20.79	186.92	12.52	102.08
11	$N_{35}P_{80}K_{100}$ + 60% FWC	0.61	45.19	7.09	80.33	20.34	182.91	12.55	102.37
12	$N_{45}P_{80}K_{100}+60\%$ FWC	0.82	60.74	7.25	82.14	20.77	186.74	13.67	111.46

 Table 6.Chemical composition of tobacco

# Conclusions

Based on the obtained results the followingconclusions can be made:

- At fertilized variants, due to poor distribution of rainfall it was recorded less effect of fertilizers on the production properties of tobacco, compared with fertilized and irrigated varieties.
- Fertilizing and irrigation have positive effects on the increase of the yield of tobacco, the average price and the gross income per unit area.
- At variants where fertilization is done with 3 doses of nitrogen fertilizer and soil moisture is maintained at a level of 45 % of FWC yield increased from 45.47 % (var. 6) to 85.16 % (var.8).
- Fertilized variants irrigated with 60% of FWC showed no statistically significant differences compared with variants fertilized and irrigated with 45 % of FWC.
- According to obtained data it can be concluded, that fertilized variants with different doses of nitrogen and irrigated with 45 % of the FWC are very successful and economically justified. Higher doses of nitrogen give better effects on lean soil.
- Fertilization and irrigation individually, and their interaction effect have great impact on the chemical composition of the tobacco. This leads to the conclusion that their correct application can significantly influence the content and interrelationship of important chemical indicators that dictate the quality of raw tobacco.
- According to the obtained results from our investigation it can be concluded that fertilization and irrigation should be compulsory measures for growing the semioriental variety Otlja Zlatovrv in the region of Prilep.

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# INVESTIGATION OF VARIATIONS FOR SPIKE CHARACTERISTICS IN F<sub>2</sub>, F<sub>3</sub> AND F<sub>4</sub> GENERATIONS OF WHEAT CROSS POPULATIONS

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#### Abstract

The study was carried out with 20 bread wheat cross populations and 5 varieties with their parents in  $F_2$  (year 2009),  $F_3$  (year 2010) and  $F_4$  (year 2011) segregating generations, in order to evaluate variations, and to determine suitable cross combination with high performance and appropriate selection generation for spike characters in bread wheat. The range of average values across genotypes were 80.67-116.22 cm for plant height, 8.66-10.94 cm for spike length, 41.08-54.96 no for number of grains per spike, 2.07-2.50 no for number of grains per spike length, 41.08-54.96 no for grain weight per spike, 1.89-2.43 for spike density, 69.8-75.85% for spike index and 359.22-547.78 kg/da for grain yield. These ranges were in all cases larger than the same ranges across generations that were 96.28-101.67 cm for plant height; 9.70-10.41 for spike length; 41.38-49.36 no for number of grains per spike, 1.95-2.48 no for number of grains per spikelet, 2.04-2.19 g for grain weight per spike, 1.94-2.26 for spike density, 71.84-74.26% for spike index and 454.15-505.87 kg/da for grain yield. Selection for plant height, spike length, number of grain per spike and spikelet and grain weight per spike would be more efficient in very early generation, and selection for the latter characters should be left to later generations.

Keywords: Bread wheat, reciprocal hybrids, spike characteristics, variation, generation.

## Introduction

Due to the increasing global population together with a growing demand for plant-based food and meat and dairy products, a substantial increase of grain production in the next decades, starting from tomorrow is critical. This is particularly challenging as the basic manageable resources for crop growth and yield (water, nutrients) will not increase (Connor and Mínguez, 2012) and the land available for crop production is likely to decline (Albajes et al., 2013). Among the major crops, wheat is one of the most crucial for warranting human nourishment: it is the most widely crop grown globally and is the primary source of protein for the world population, representing c. 20% of the daily intake for developing countries (Braun et al., 2010). In order to maintain balance between demands and supply alternative ways and means to further raise wheat yield must be found (Chand, 2009). Although it is through re-gaining high rates of genetic gains in yield have

recently been much lower than what it would be required (Reynolds et al., 2012; Fischer et al., 2014). The genetic gain in yield potential since the green revolution has been around 1% per year (Sayre et al., 1997; Shearman et al., 2005; Zhou et al., 2007). The likelihood of accelerating breeding progress would increase with knowledge of genetic variation available for traits putatively determining yield (Slafer, 2003; Reynolds and Borlaug, 2006) in early generations.

The grain yield which is the most important economic trait in wheat improvement is complex quantitative traits controlled by multiple genes and are highly influenced by environmental conditions (Shi et al., 2009). Being the final product of many processes, it is directly and multilaterally determined by yield-component traits, such as productive spikes per unit area, the number of grains per unit area, kernels per spike and kernel weight, where the product of the first two components gives the total kernel number per unit area. Increases in the number of grains per unit area have been the main cause of genetic yield gains in bread wheat during the last century (Austin et al., 1980; Shearman et al., 2005) while no positive correlations have been found between yield improvement and grain weight (Perry and D'Antuono, 1989; Brancourt-Hulmel et al., 2003). Other traits, such as productive tillers per plant, spikelet number per spike, and number of fertile florets per spikelet, could all affect the total kernel number. Genetic and physiological dissection of these components and their relationships would facilitate achieving the maximum yield potential of the crop (Ma et al., 2007). As combination of its length and density, the spike is at the same time a sink and a source for assimilates determining grain yield (Bagnara and ScarasciaMugnozza, 1975). In different wheat cultivars, the total contribution of non leaf green organs, including spikes and peduncles, accounts for about 40-50% of grain mass per spike, which is higher than the total contribution of the flag leaves and penultimate leaf blades (Thorne 1963; Araus et al., 1993 and Wang et al., 2001).

For grain yield is under big influence of spike properties (Martinčić*et al.*, 1996), spike characters such as spike length, number of grains and weight per spike and spikelet, spike index, spike density and plant yield can be considered important selection indices in wheat breeding. So, it is crucial increase of grain yield via increasing those characters.

It was aimed to determine appropriate promising bread wheat cross combinations and selection generation for plant height, grain yield and spike characters at  $F_2$ ,  $F_3$  and  $F_4$  generations in Tekirdağ ecological condition.

## Material and methods

Twenty-five genotypes of bread wheat, including five bread wheat varieties and their 20 crosses were used as genetic material in the study (Table 1). The genetic structure of crosses is population. Any selection process has not been applied.

	Genotypes	
1. Pehlivan/Flamura85	10. Bezostaja1/Krasunia	19. Flamura85/Pehlivan
2. Bezostaja1/Flamura85	11. Flamura85/Krasunia	20. Sana/Pehlivan
3. Krasunia/Flamura85	12. Sana/Krasunia	21. Flamura85
4. Sana/Flamura85	13. Pehlivan/Bezostoja1	22. Sana
5. Pehlivan/Sana	14. Krasunia/Bezostoja1	23. Krasunia
6. Flamura85/Sana	15. Flamura85/Bezostoja1	24. Bezostoja1
7. Bezostaja1/Sana	16. Sana/Bezostoja1	25. Pehlivan
8. Krasunia/Sana	17. Bezostoja1/Pehlivan	
9. Pehlivan/Krasunia	18. Krasunia/Pehlivan	

Table 1. Cross combinations and their parents used as genetic material

The study was carried out during 2009, 2010 and 2011 growing seasons in Namik Kemal University, Agricultural Faculty, Field Crops Department experimental area. Each trial was sown in a randomized complete block design with four replicates. Sowing was done by hand into plots of 4 m<sup>2</sup> (4 rows, 5 m long, spaced 20 cm apart). Genotypes were sown using a seeding rate of 500 seeds per m<sup>2</sup>. Sowing time was in mid-October each year, and a total of 130 kg ha-1 N was applied at sowing, tillering and pre-anthesis stages. Weeds were chemically controlled to avoid a confounding effect. In this study, the characters grain yield, plant height and spike traits such as spike length, number of grain per spike, grain weight per spike, number of grain per spikelet, spike density and spike index were evaluated in three segregating populations (F<sub>2</sub>, F<sub>3</sub> and F<sub>4</sub>).

Combined analyses of variance (ANOVA) across generations for grain yield, plant height and other spike traits were performed according to randomized complete block design. Average values in each generation and in each varieties and Duncan's new multiple range test (DMRT) between the different mean values were calculated using "MSTAT version 3.00/EM" package program.

## **Results and discussion**

The combined analyses of variance (ANOVA) performed on the set of data including three generations for the each character indicated that there were highly significant differences among genotypes averages. To demonstrate these significances, mean comparison for the studied traits shown Table 2-3.

Genotypes Plant Genotype	Plant	Genotypes	Spike	Genotype	s Spike Genotypes Number of Genotypes Number of Genotypes	Genotypes Number of Genotypes Number of Genotypes	pes Nu	mber of	Genoty	vpes Grain	n Genotypes	types	Spike	Genotypes	ypes	Spike	Genot	Genotypes	Grain yield	eld
	height		length		seeds per	II.	se.	seeds per snikelet		weight ner snike			density			index				
	116.22 a	10	10.94 a	18	54.96 a	+	2.5	2.50 a	c	2.46 a			2.43 a	12		75.85 a	22		547.78 a	
	115.56a	18	10.92 a	22	54.12 ab	9	24	2.47 ab	18	2.30 ab	4		2.29 ab	20		75.25 ab	25		541.78 ab	ę
	114.78 ab	14	10.82 a	12	52.89 abc		24	2.44 abc	12	2.29 ab			2.26 abc	Ξ		75.22 ab	12		539.56 ab	ą
	107.56 abc	23	10.74 a	4	52.64 abc	c 18	2.4	2.44 abc	::	2.28 ab			2.25 abc	m		75.01 ab	20		530.56 ab	Ą
	105.78 bcd	e	10.68 a	9	49.98 a-d	1 12	2.4	2.44 abc	21	2.24 ab		14	2.24 a-d	22		74.91 ab	4		515.44 abc	þc
16 10	103.89 cde	24	10.38 ab	3	49.96 a-d	3	2.4	2.42 abc	4	2.21 ab	b 5	14	2.23 a-e	4		74.82 ab	1	•	511.00 a-d	ų
	103.33 cde	6	10.28 abc		49.39 a-f	21	2.4	2.41 abc	23	2.21 ab	b 7	~	2.21 a-e	00		74.57 ab	6		510.00 a-d	ų
	103.33 cd	13	10.21abc	21	48.93 a-g		2.3	2.38 abc	\$	2.18 ab	۹ 8	~	2.21 a-e	17		74.16 abc			507.11 a-e	٩
	103.11 cde	12	10.14 a-d		47.24 a-f	20	2.3(	2.36 abc	10	2.16 ab	b 18	14	2.11 b-f	21		74.04 abc	19		505.89 a-e	٩
	99.78 c-f	11	10.10 a-d	2	47.16 a-f	f 11	2.3	2.32 abc	9	2.16 ab	9 9	~	2.11 b-f	23		73.74 a-d	9		501.44 a-e	٩
	99.67 def	15	10.10 a-d	15	47.10 a-f	19	23	2.31 abc	14	2.15 ab	b 19		2.09 b-g	18		73.71 a-d	8	4	488.79 a-f	4-
	99.22 def	25	9.93 a-d	s	46.84 a-f	23	23	2.31 abc	19	2.14 ab	b 21	.4	2.08 b-g	10		73.53 a-d	ŝ	4	484.33 a-f	ų
	98.33 efg	6	9.91 a-d	11	46.44 a-f	2	2.2	2.28 abc	20	2.13 ab	b 17	14	2.07 b-g	9		73.43 a-d	18	4	476.22 a-g	μ
	96.78 efg	17	9.83 a-d	16	46.03 b-f	14	2.2	2.26 abc	12	2.09 ab			2.06 b-g	19		73.18 a-d	21	4	474.89 a-g	р ц
	95.78 feh	~	9.82 a-d	19	45.47 c-f	~	2.2	2.25 abc	22	2.08 ab		~	2.05 c-g	16		73.09 a-d	S	4	474.22 b-g	) pi
	95.33 feh	21	9.77 a-d	14	44.89 c-f	10	2.2	2.20 abc	25	2.08 ab		14	2.05 c-g	25		72.68 a-d	11		473.00 b-g	) ท่
	93.89 f-i	16	9.77 a-d	20	44.83 c-f	6	2.1	2.14 abc	15	2.06 ab	b 10	~	2.04 c-g	14		72.12 a-d	13		447.00 c-h	ہے،
	93.33 f-i	2	9.59 a-d	S	44.68 c-f	16	2.1	2.11 abc	13	2.05 ab		~	2.03 d-g	-		72.07 a-d	15		444.00 d-h	ą
	93.11 f-i	20	9.58 a-d	24	42.61 def	-	2.1	2.11abc	5	2.04 ab	9 9	~	2.02 d-g	13	(-	71.47 bcd	17	4	438.11 e-h	ą
	92.22 ghi	5	9.57 a-d	25	42.29 def	5 5	2.1	2.11 abc	17	2.02 ab	b 15	(4	2.02 efg	5		71.38 bcd	2	4	423.89 f-1	.7
	91.78 ghi	19	9.54 a-d	7	42.14 def	f 25	2.1	2.11 bc	16	2.01 b	33		2.02 efg	6		71.28 bcd	16	-	414.67 ghi	व
	89.89 hid	4	9.41 a-d	17	41.74 e	13	2.1	2.11 bc	6	1.97 b	=		2.00 fg	7		70.15 cd	14	4	412.33 ghi	व
	87.67 ik	2	9.12 bcd	6	41.41 f	17	2.1	2.10 bc	7	1.95 b			1.95 fg	24		70.13 cd	10	4	400.67 hi	=
	83.67 kl	22	8.80 cd	1	41.12 f	1	2.0	2.07 c	24	1.92 b	6		.93 fg	2		70.02 cd	6		380.44 hi	=
	80.671	1	8.66 d	13	41.08 f	24	5	2.07 c	7	-	14		1.89 g	17	-	69.83 d			359.221	
MSE	17.922	MSE	0.968	MSE	26.663	MSE	_	0.059	MSE	E 0.077	-	MSE	0.018	W	MSE	12.872	MSE	SE	1897.925	25
							ទ	ENERA	GENERATAIONS											
	101.67 a	ц <mark>г</mark>	10.41	£	49.36 a		-1	2.48 a	ц Ц				2.26 a	щ		74.26	F4		505.87 a	ed
щ, I	96.81 ab	μ,	9.71	ម្មី រ	49.17 a	н <sup>с</sup> н	<u>, ,</u>	2.37 ab	ម្មី រ	2.15	н <sup>а</sup> н		2.11 a		ម្មីរ	72.97	њ <sup>с</sup> і		456.31 b	. م
	26.077	r3 MSF	9.70	MCF MCF	175 86	1	+	0.074	MSF MSF	+	ſ	2	0.067	MSF MSF	+ H	11.04	L3	- -	0 CT.4C4	• 5
e,	Significances of	i 9	types acr	oss t	racters		+					3								
	6		-	2 3	4 5	9	7 8	6	10 1	11 12	13 1	14	15 16	17	18 19	20	21 22	23	24	25
Plant height				++	+	⊢	++	⊢		+	+-	$\vdash$	+	+	+	+	⊢	⊢		Γ
Spike length				+				+	+	++	+	+	+		+		$\vdash$	+	+	Γ
Number of seeds per spike	ds per spik	9		++	+	+	+		+	++			+		+		++	+		
Number of seeds per spikelets	ds per spik	elets		++	+	+				++			+		++	+	+	+		
Grain weight per spike	er spike			+	+					+					+		+	+		
Spike density				_	+		++			+		_	+		_	+	+			
Spike index				+	+		+			++				+		+	++			
Grain yield			+		+	+	$\vdash$	+		+					+	+	+	+		+

SECTION 9. FIELD CROP PRODUCTION

The significant difference among genotypes for the traits implies the presence of substantial variation among genotypes which is central to the study of traits and gives an opportunity to plant breeders for improvement of these characters through breeding. The range of average values across genotypes were 80.67-116.22 cm for plant height, 8.66-10.94 cm for spike length, 41.08-54.96 no for number of grains per spike, 2.07-2.50 no for number of grains per spikelet, 1.89-2.46 g for grain weight per spike, 1.89-2.43 for spike density, 69.8-75.85% for spike index and 359.22-547.78 kg/da for grain yield. These ranges were in all cases larger than the same ranges across generations that were 96.28-101.67 cm for plant height; 9.70-10.41 for spike length; 41.38-49.36 no for number of grains per spike, 1.95-2.48 no for number of grains per spikelet, 2.04-2.19 g for grain weight per spike, 1.94-2.26 for spike density, 71.84-74.26% for spike index and 454.15-505.87 kg/da for grain yield (Table 2).

Because biological mass remained unchanged or changed only slightly (Slaferetal.1994), Selection of a short stem contributed most to a higher grain yield (Hay1995). In addition to this, it is preferred that plant height is to be 80-100 cm because of lodging and diseases problems. From the point of view plant height, cross combinations of numbered with 6, 12, 18, 8, 4, 5, 3, 20, 7, 2 and 17 were the best values with the desired ranges. According to Martincic et al. (1996) grain yield is under big influence of spike properties like spike length, number of grain per spike and spikelet, grain weight per spike, spike density and spike index. Islam et al. (1985) stated inclusion of weight per grain in a selection index with grain per spike or per spikelet might have been profitable. When the mean performance of average values across genotypes were evaluated, the promising cross combinations were numbered combinations of 10, 18, 14, 3, 9, 13, 12, 11 and 15 for spike length, 18, 1, 4, 6, 3, 10, 2, 15, 8 and 11 for number of grain per spike, 4, 18, 12, 3, 15, 20, 11, 19 and 2 for number of grain per spikelet, 3, 18, 12, 11 and 4 for grain weight per spike, 4, 12, 16, 5, 7 and 8 for spike density and 12, 11, 3, 4, 8 and 17 for spike index. The results of the evaluation with the aforementioned indicate that numbered of 2, 3, 4, 8, 11, 12, 15 and 18 cross combinations may be promising for spike properties.

The grain yield which is the most important economic trait in wheat improvement is complex quantitative traits controlled by multiple genes and are highly influenced by environmental conditions (Shi et al., 2009). Since non-genetic effects are large (Bernardo, 2003), early generation selection is expected to be ineffective for grain yield. But, screening of segregating populations can give us ideas for the future evaluations. Regarding grain yield, among the cross combinations, numbered of 12, 20, 4, 1, 9, 19 and 6 were remarkable promising cross populations.

Selection for yield and yield components in early generations has produced varying results. Plant characters bearing desirable gene combinations are easily identified and selected for at the early generations preferably at the  $F_1$  before reaching homozygosity in the late generations (Cristina and Hall, 1995). Contrarily, Falcinelli et al., (1983), Alexandra et al. (1984) reported effective selection of grain weight and plant height at  $F_3$  and  $F_4$  generations in wheat. Sing and Singh (1997) reported plant height and kernel weight as showing effective selection in the early generation of  $F_2$  while harvest index, grain yield and dry matter weight were ineffective when selection is made at the early generation in bread wheat. Rasmussen (1987) reported that delaying selection to a later generation of  $F_4$  could lead to loss of such desirable gene combinations. According to Islam et al. (1985),

grain number per spikelet or grain number per spike is more effective than selection for weight per grain and yield *per se* in  $F_2$  generation. Selection for the latter characters should be left to later generations. From the point of view of the characters, when the means of generation from  $F_2$ to $F_4$  were increased for spike index and grain yield, while there were decreases in plant height, spike length, number of grain per spike and spikelet and spike density. Our results indicate selection for plant height, spike length, number of grain per spike and spikelet and grain weight per spike would be more efficient in  $F_2$  or  $F_3$ . In contrary, it would bemore appropriate todelay theselectionforspike density, plant height and grain yield.

# Conclusions

It was concluded from the present research that genotypes showed highly significant variations for all the traits. Numbered crosses of 3, 4, 6, 12 and 20 were the best performing genotypes for plant height, yield and spike properties. Among the parents used in crosses, 22, 23 and 25 varieties showed high performance, hence can be used in future breeding programs aimed to improve yield through spike traits. Based on the generation means, selection for plant height, spike length, number of grain per spike and spikelet and grain weight per spike would be more efficient in very early generation, and selection for the latter characters should be left to later generations.

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# EVALUATION OF YIELD AND YIELD COMPONENTS OF WHEAT GENOTYPES GROWN DURING 1968-2011 YEARS IN THRACE REGION

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#### Abstract

The study was carried out with 36 bread wheat genotypes in three locations (Edirne, Lüleburgaz, Tekirdag) during 2010 and 2011 growing years. The aim of this study was to evaluate grain yield and yield components of selected bread wheat genotypes grown in the Thrace region between 1968-2011 years. The genotypes were examined in 6 different year periods, especially in 1968-75, 1976-83, 1984-91, 1992-1999, 2000-2007 and 2007-after. Compared to those in 2011, higher values were obtained in terms of spike length, grain number per spike, grain weight per spike, harvest index and grain yield, except for plant height. In the locations, the average grain yield changed between 543.06 and 551.36 kg/da, the plant height changed between 92.19 and 97.67 cm, the spike length changed between 8.98 and 9.11 cm, the grain weight per spike changed between 40.21 and 44.40, the grain weight per spike changed between 1.54 and 1.75 g, and harvest index changed between % 38.30 and 40.05. In terms of the characteristics examined, Lüleburgaz location generally gave high values. According to the obtained results, the grain yield and yield components of the bread wheat varieties developed in recent years has significantly improved. When advanced lines and local genotypes compared with bread wheat varieties for grain yield and yield components, it can be said that important improvements have been provided in varieties and advanced lines being developed in recent years.

Key words: Local lines, advanced lines, location, old varieties, new varieties.

## Introduction

Wheat is accepted as the most significant strategic crop because of its importance in human nutrition. While wheat production is made in an amount of 713 million tons in area of nearly 218 million ha in the world, it is made in an amount of 22 million tons in an area of 7.8 million tons in our country (FAO, 2014). In our country, there exist various species of local and foreign wheat in its production. The first wheat development works began in Seed Development Station, known as "Islah- Buzr" (Büzür), in Eskişehir in 1925 in order to improve varieties development in Turkey. While varieties were developed through selection development in wheat in the initial years, varieties were usually developed by means of combination development works, 189 varieties of bread wheat and 62 varieties of durum wheat in country were registered officially (Anonymous, 2015). It has been

observed that wheat cultivation areas have been reduced in recent years. The cultivation area decreased from 9,3 million hectares in 2004 to 7,8 million hectares in 2013. In spite of that decrease, it is seen that not any considerable decrease in production exists. This situation is made possible by means of development of highly yielding species and developments in the practices of growth techniques. Considering such features as spike number in m<sup>2</sup>, plant height, spike length, grain number per spike, grain weight per spike, weight of one thousand grains and harvest index that influence yield and the relationship between these features (Korkut et al., 1993; Saleem et al., 2005; Monouchehr, 2006; Kara and Akman, 2007; Bilgin et al., 2008; Yağdı, 2009; Khan et al., 2010; Laei et al., 2012; Khan and Haqvi, 2012; Zafarnadei et al., 2013), highly yielding species are developed. In this study, it is aimed to determine the effect of year periods on the grain yield and features that are effective on yields and change in these features.

## Material and methods

The study was carried out during two consecutive growing years (2009-2010 and 2010-2011) under rainfed conditions at three different locations in the Thrace Region of Turkey: Edirne (26°35' E, 41°38' N and elev. 32 m), Lüleburgaz (27°16' E, 41°22' N and elev. 41 m) and Tekirdağ (27°34' E, 40°59' N and elev. 10 m). All the locations were on the northern, occidental side of Turkey. Chemical composition of soil and climate conditions differed in the chosen locations. Soil characteristics of experiments were determined by soil analyses and included silty clay (Edirne), fine loamy (Lüleburgaz) and loamy clay (Tekirdağ) textures, with neutral pH and low salt concentrations. Contents of phosphorus, potassium, calcium, magnesium, iron, copper, manganese and zinc were not enough for the crop development. Organic matter was about 1%. In 2009-2010 year, the total rainfall was 610 mm in Edirne, 715 mm in Lüleburgaz and 808 mm in Tekirdağ; the average temperature was 12.1 in Edirne, 11.6 in Lüleburgaz, and 13.2 in Tekirdağ. In 2010-2011 year, the total rainfall was 341 mm in Edirne, 423 mm in Lüleburgaz and 721 mm in Tekirdağ; the average temperature was 11.5 in Edirne, 11.5 in Lüleburgaz, and 13.3 in Tekirdağ. In the study, bread wheat genotypes developed in 1968-2013 and given in Table 1 were used as the material. Genotypes were examined in 6 different year periods, especially in 1968-75, 1976-83, 1984-91, 1992-1999, 2000-2007 and 2007-after. In the period after 2007, advanced lines being developed with cross-breeding took place.

			Year	· Periods		
	1 <sup>st</sup>	$2^{nd}$	3 <sup>rd</sup>	$4^{\text{th}}$	5 <sup>th</sup>	6 <sup>th</sup>
5	(1968-75)	(1976-83)	(1984-91)	(1992-99)	(2000-2007)	(After 2007)
pe,	1. LR-17	7. Bezostoja 1	13. Atilla 12	19. Saroz 95	25. Gelibolu	31. Fl85/Sbosna
Genotypes	2. LR-19	8. Cumhuriyet 75	14. Atay 85	20. Flamura 85	26. Esperia	32. BBBD-7
Jen	3. LR-46	9. Sakarya 75	15. Sadova 1	21. Prostor	27. Sagittario	33. BBBD-13
$\cup$	4. LR-47	10. Orso	16. Marmara 86	22. Pehlivan	28. Krasunia	34. 43 (5)
	5. LR-69	11. Libelulla	17. Flamura 80	23. Golia	29. Aldane	35. Peh /Kate A1
	6. LR-70	12. Kırkpınar 79	18. Kate A1	24. Murat 1	30. Selimiye	36. Bez1/Kate A1

Table 1. Year periods and genotypes used as material in the study.

Experiments were conducted with 3 repetitions in such a way that year periods can make up main plots and genotypes can create sub-plots. Cultivations were made in the first half of November in such a way that 500 seeds exist in plots consisting of 6 rows that were 5 meters long.

In the trials, 20.20.0 compound NPK fertilizer was applied at sowing in such a way that 4 kg of pure nitrogen and phosphorus could exist, Urea fertilizer (% 46 N) was applied at tillering stage in such a way that 7 kg of pure nitrogen could exist, and ammonium nitrate fertilizer (% 26 N) was applied at heading stage in such a way that 3 kg of pure nitrogen could exist. Chemical struggle was carried out against the weeds. The experiments were harvested in the first half of July.

In the study, spike height, spike length, grain number per spike, grain weight per spike, harvest index and grain yield were examined. In the obtained data, the analysis of variance was made, significance of differences between the averages was determined with DUNCAN test by using "MSTAT version 3.00/EM" package programme in accordance with the methods offered by Steel and Torrie (1960).

#### **Results and discussion**

In the study, yield and yield components in genotypes being developed periodically were explored by examining bread wheat genotypes being developed between 1968 and 2013 in 6 different periods.

#### Plant height

In the study, it was found that differences between years, year periods and genotypes were statistically significant while differences between locations were found to be insignificant. It was determined that the average plant height was 103.12 cm in 2011 while it was 86.31 cm in 2011 (Table 2). When the location averages were examined, plant height changed between 92.19 and 97.67. The highest plant height was measured in Tekirdağ location whereas the shortest plant height was measured in Edirne location (Table 2). In the year periods, the plant height changed between 82.48 and 108.79 (Table 2). The values of the highest plant height were determined in the 1<sup>st</sup> and 2<sup>nd</sup> year periods. The shortest plant height was obtained in the 4<sup>th</sup> year period, and the 5<sup>th</sup> year period followed that. In the 3<sup>rd</sup> and 6<sup>th</sup> year periods, the plant heights approximate to each other were measured (Table 2). In 36 bread wheat genotypes researched, the values of the plant height changed between 63.90 and 117.35 cm. Generally, the genotypes whose first development period was 1968-75 and that were developed in the last breeding period as well as the genotype no 36 created through the crossbreeding of two high varieties had the highest plant height (Table 3).

Table 2. The	mean values a	nd significance g			ods and the loc	cations.
	Grain Yield	Plant height	Spike length		Grains	Harvest index
	(kg/da)	(cm)	(cm)	number per	weight per	(%)
				spike	spike (g)	
Years						
2010	562.07 a	86.31 a	9.16	45.44 a	1.82	40.12 a
2011	531.67 b	103.12 b	8.89	40.13 b	1.50	38.29 b
MSE	779.932	85.775	-	3.54	-	3.270
Year periods						
5	619.19 a	89.86 c	8.97 ab	43.47 a	1.85 a	41.38 a
4	601.89 ab	82.48 d	8.24 c	43.02 a	1.63 ab	40.94 a
3	582.82 ab	93.94 bc	9.63 a	45.87 a	1.74 ab	39.96 ab
6	568.32 bc	93.49 bc	8.79 bc	44 58 a	1.77 ab	39.79 ab
2	530.29 с	99.78 ab	9.24 ab	44.08 a	1.58 bc	38.74 b
1	378.73 d	108.79 a	9.27 ab	35.68 b	1.39 c	34.45 c
MSE	4376.009	130.557	7.74	47.886	0.105	7.085
Locations						
Lüleburgaz	551.36	94.28	8.98	43.73 ab	1.75 a	40.05a
Tekirdağ	546.20	97.67	8.99	44.40 a	1.54 b	38.30 c
Edirne	543.06	92.19	9.11	40.21 b	1.68 a	39.28 b
MSE	-	-	-	141.04	0.178	0.512

Table 2. The mean values and significance groups belonging to year periods and the locations.

MSE: Mean Squared Error

Differences coming into existence in the plant height in year periods can result from the fact that the genetic structures of the genotypes used were different. In the studies conducted, it was reported that the plant height in wheat changes according to the genetic structure of the species, sowing density, sowing time, fertilizing, rain condition and soil characteristics (Gençtan and Sağlam, 1987 and Kün, 1996).

## Spike length

The fact that the spike length is long in breeding works and that spikelets on the spike are arrayed loosely are necessary features in terms of resistance to biotic and abiotic stress factors, high yield and quality. It was found that differences between year periods and the genotypes were statistically significant in terms of the spike length while differences between years and the locations were insignificant (Table 2). Compared to those (8.89 cm) in the second year in the study, the longer spikes (9.16 cm) were obtained in the first year. Differences between the locations were insignificant, and the longest spikes were obtained in Edirne location (Table 2). When bread wheat genotypes were examined in 6 years period, the longest spikes were obtained with 9.63 cm in the 3<sup>rd</sup> year, and this was followed by the 1<sup>st</sup> year (9.27 cm) and the 2<sup>nd</sup> (9.24 cm) year periods. The shortest spikes were measured as 8.24 cm in the 4<sup>th</sup> year period, and it was followed by 8.79 cm in the 6<sup>th</sup> year period (Table 2). The reason why the  $5^{th}$  and  $4^{th}$  year periods in which the highest grain yield was obtained did not include the longest spike length was that the spike length has close relationship with the plant height and that the genotypes whose spike length was more were generally those whose plant height was higher. Since it is possible to increase the grain number per spike and the grain weight per spike through the increase of the spike length, choosing plants whose spike length is enough long in grain development carries much importance (Özgen, 1989 and Tunca, 2012). When the mean values of the genotypes were examined, it was observed that the spike length changed between 7.47 and 11.38 cm (Table 3). The longest spikes were determined in the genotypes no 14 (Atay 85) and no 16 (Marmara 86) in the  $3^{rd}$  year period, which were followed in turn by the genotype no 1 (LR-17) with 9.87 cm and the genotype no 8 (Cumhuriyet 75). The shortest spikes were measured in the genotype no 31 (Flamura 85/Saraybosna) in the  $6^{th}$  year genotype, which was followed by the genotype no 19 (Saroz 95) with 7.51 cm and no 24 (Murat 1)(Table 3).

### Grains number and weight per spike

In wheat breeding, the grain number and weight per spike are among the main yield components. The fact that grain number and weight in the spike of newly developed genotypes are high is desired. The higher grain number and weight per spike in the first year were obtained than those in the second year (Table 2).

Tekirdağ location had the highest values in the grain number per spike (44.40), and Lüleburgaz (1.75 g) and Tekirdağ (1.68 g) locations had the highest values in the grain weight per spike. The lowest grain number (40.21) and weight (1.54 g) per spike were observed in Edirne location (Table 2).

While the grain number per spike being obtained changed between 35.68 and 45.87 in year periods, the grain weight per spike changed between 1.39 and 1.85. Out of year periods, the  $3^{rd}$ ,  $6^{th}$ ,  $2^{nd}$ ,  $5^{th}$ , and  $4^{th}$  periods existed statistically in the same group. The lowest values were obtained in the  $1^{st}$  period where only the local populations took place. It was seen that the last periods had higher values in the grain weight per spike (Table 2). The highest weight (1.85 g) was observed in the  $5^{th}$  period, and it was followed with 1.77 g in the last year period. The lowest values were obtained from the  $1^{st}$  year (1.39 g) and the  $2^{nd}$  year (1.58 g) periods in which the genotypes which were mainly developed by selection development existed. Different researchers obtained results that were similar to ours in their studies (Korkut et. al., 1993; Akıncı, 2003; and Tunca 2012).

In 36 bread wheat genotypes examined, the grain number per spike changed between 33.47 and 52.96 while the grain weight per spike changed between 1.15 and 2.12 g. The highest grain number per spike was obtained from the genotype no 14 (Atay 85), and it was followed by the genotype no 12 (Kırkpınar 79) with 51.20. While the highest value in the grain weight per spike was obtained from the genotype no 36 (Bezostoja 1/Kate A1), and it was followed by the genotype no 33 (BBBD-13) with 1.98 g and no 25 (Gelibolu) with 1.90 g. The lowest grain weight per spike was determined in the genotype no 6 (LR-70), and it was followed by the genotype no 3 (LR-46) with 1.16 g which existed in the same statistical group. In our study, it was recognizable that the genotypes in the first year had lower grain number and weight. This situation caused these genotypes to have lower grain yield compared to the genotype in the other year period. Our findings display similarities with those of Bilgin (1997) and Tonkin (2004).

## Harvest index

One of the most important purposes of breeding works in cereals is to enhance economic efficiency within biological yield that is harvest index. Recently, the values of harvest index have been increased to 40% and even 45%. However, the future purpose is to increase harvest index to over 50%. Harvest index being determined as 40.12% in the first

year of our study was found to decrease to 38.29% in the second year. This situation results from the fact that the values of the plant height obtained in the second year was high. While the highest value of harvest index (40.05%) was obtained in Lüleburgaz location where we conducted our study, the lowest value (38.30%) was obtained in Tekirdag location.

The values of harvest index being determined in year periods changed between 34.45% and 41.38%. The highest value of harvest index was observed in the 5<sup>th</sup> year period in which the most yielding genotypes were, and this was followed by the 4<sup>th</sup> year as 40.94%. The lowest values of harvest index were obtained in the 1<sup>st</sup> year period in which the local varieties existed, and this was followed by the  $2^{nd}$  year period (38.74%) in which the local varieties again existed. From 1968s toward today in our study (except for advanced lines), it is recognizable that the values of harvest index have increased. The same statistical rank was shared by both the grain yield of year periods and harvest index (Table 2). This situation supports the fact that harvest index is an important feature in increasing the grain yield. In parallel with our findings, Khush (1996) and Başçifçi et al. (2009) claimed that the feature of harvest index needs to be increased in order to increase the grain yield. When the genotypes averages were explored, it was seen that harvest index changed between 32.06% and 42.87% (Table 3). While the highest harvest index was obtained from the genotype no 23 (Golia) which had the shortest plant, and it was followed by the genotype no 26 (Esperia) with 42.72%. The lowest harvest index was determined in the genotype no 6 (LR-70) in the first year period, and this was followed by the genotypes no 3 (LR-46), no 5 (LR-69), no 2 (LR-19) and no 1 (LR-17) in the same year period.

## Grain Yield

The most important selection criterion in bread wheat genotypes is to develop varieties that have high grain yield. Researchers reported that grain yield arises from the effect of both genotype and the environment together (Özberk et al., 2004; Akçura and Kaya, 2008). Higher grain yield (562.07 kg/da) was obtained in 2010 than that in 2011 (531.67 kg/da). When the locations are examined, it can be seen that the highest grain yield (551.36 kg/da) was determined in Lüleburgaz location, and this was followed by Tekirdağ location while the lowest grain yield (543.06 kg/da) was obtained from Edirne location (Table 2). The values of grain yield determined in year periods changed between 378.73 and 619.19 kg/da. The highest grain yield was obtained from the 5<sup>th</sup> year period, and it was followed by 4<sup>th</sup> year period with 601.89 kg/da. The lowest grain yield was determined in the 1<sup>st</sup> year period where the local varieties existed.

This situation can be evaluated to result from the fact that the varieties being developed in recent years are more superior in terms of their characteristics that are effective on yield. While the highest grain yield in bread wheat genotype was obtained as 655.00 kg/da in the genotype no 32 (BBBD-7) that was the advanced line, and this was followed by the genotypes no 18 (Kate A1) with 654.72 kg, no 27 (Sagittaro with 653.17 kg, no 22 (Pehlivan) with 635.78 kg, and no 28 (Krasunia) with 635.50 kg. The lowest grain yield was obtained from the genotypes no 5 (LR-69) with 357.94 kg/da and no 6 (LR-70) with 358.50 kg/da. It is outstanding that the genotypes from which the lowest grain yield was obtained were the local varieties that were in the first year period.

	* 1		0	ř.			u in 6 years j			CN	OV(1 - 1)
GN	PH (cm)	GN	SL (cm)	GN	GNS	GN	HI (%)	GN	GWS (g)	GN	GY (kg/da)
36	117.35 a	14	11.38 a	14	52.96 a	23	42.87 a	36	2.12 a	32	655.00 a
7	113.38 ab	16	10.08 a	12	51.20 ab	26	42.72 a	33	1.98 ab	18	654.72 a
2	112.21 ab	1	9.87 b	24	49.67 abc	9	42.39 ab	25	1.90 abc	27	653.17 ab
3	111.92 ab	8	9.87 bc	31	48.63 a-d	24	42.28 ab	32	1.89 abc	22	635.78 abc
5	110.27 abc	18	9.84 bcd	36	48.34 а-е	28	41.78 abc	18	1.87 a-d	28	635.50 abc
6	109.90 bc	2	9.83 bcd	16	48.33 а-е	17	42.72 abc	30	1.86 а-е	30	627.72 a-d
1	109.20 bcd	28	9.73 bcd	33	48.04 a-f	25	41.72 abc	26	1.86 а-е	21	624.56 a-e
18	105.18 cde	36	9.70 b-e	25	47.92 a-f	33	41.67 a-d	27	1.85 а-е	25	618.11 a-f
8	104.69 cde	7	9.67 b-e	10	47.13 a-f	16	41.39 a-e	16	1.84 а-е	19	617.22 a-f
14	103.46 def	12	9.58 b-f	13	46.89 a-f	22	41.39 a-e	12	1.81 a-f	16	616.33 b-f
11	102.53 efg	33	9.58 b-f	28	46.48 a-g	32	41.33 а-е	28	1.81 a-f	23	611.33 c-g
35	100.57 e-h	9	9.56 b-g	26	45.03 b-h	10	41.06 b-f	29	1.80 b-f	26	597.66 d-h
12	100.31 e-h	35	9.54 b-g	8	44.30 c-1	30	41.06 b-f	22	1.76 b-g	10	593.83 d-h
33	99.30 ghi	4	9.38 c-g	27	44.26 c-1	15	40.89 c-f	17	1.75 b-g	20	593.50 e-h
4	99.26 ghi	20	9.32 c-h	11	43.98 c-k	29	40.61 c-g	13	1.71 b-h	33	588.11 f-1
	97.61 hij	22	9.27 d-1	15	43.79 c-k	13	40.57 c-g	35	1.70 b-h	13	588.00 f-1
17	97.182 ijk	17	9.14 e-j	18	43.75 d-k	18	40.50 c-g	14	1.70 b-h	34	587.44 f-1
	94.50 jkl	10	9.07 f-k	20	43.70 d-k	27	40.39 d-h	20	1.68 c-1	29	582.94 g-j
	94.11 klm	5	9.07 f-k	21	43.16 d-l	34	40.28 e-h	1	1.67 c-1	17	566.11 hij
16	93.77 klm	30	9.06 f-k	19	43.04 e-m	20	40.11 e-h	7	1.67 c-1	15	565.89 hij
	93.72 klm	29	9.04 f-k	34	42.74 f-m	21	39.89 f-1	23	1.61 d-j	9	561.61 ıj
19	91.51 lmn	25	9.01 g-k	23	42.40 g-n	12	39.39 ghi	21	1.60 d-j	12	552.04 jk
9	89.96 l-o	6	8.82 h-l	32	41.01 h-o	19	39.17 hi	19	1.60 d-j	35	551.07 jk
28	89.79 l-o	32	8.74 1-l	30	40.57 h-o	31	39.17 hı	34	1.60 d-j	24	528.94 kl
25	89.38 mno	3	8.73 1-l	9	39.78 h-p	35	38.791	15	1.57 e-j	31	515.671
21	88.63 nop	15	8.68 jkl	17	39.48 1-r	36	37.44 j	10	1.56 e-j	36	512.001
	87.81 nop	13	8.63 jkl	35	39.32 k-s	11	37.00 j	9	1.56 e-j	11	506.721
	87.69 nop	26	8.56 klm	7	38.61 l-s	4	36.56 j	2	1.54 f-j	14	505.891
	86.16 op	27	8.45 lm	1	37.55 m-s	8	36.56 j	24	1.54 f-j	7	498.28 lm
	84.18 pr	21	8.06 mn	4	37.47 n-s	7	36.22 j	4	1.52 g-j	8	469.22 m
	80.16 rs	23	7.73 no	29	36.56 o-s	14	34.72 jk	8	1.46 hij	4	416.06 n
13	79.84 rs	34	7.72 no	22	36.12 o-s	1	34.64 kl	11	1.44 ijk	2	391.72 no
34	77.59 s	11	7.63 no	2	35.52 o-s	2	34.50 kl	31	1.40 jkl	3	383.56 op
31	71.67 t	24	7.52 o	5	35.02 prs	5	34.33 kl	5	1.22 kl	1	364.61 op
24	70.92 t	19	7.51 o	6	33.86 rs	3	33.44 lm	3	1.161	6	358.50 p
23	63.90 u	31	7.47 o	3	33.47 s	6	32.06 m	6	1.151	5	357.94 p
MSE				MSE			1.341	MSE		MSE	796.198
							ngth CNS: C				

Table 3. The mean values and significance groups concerning characteristics in 36 bread wheat genotypes examined during two years in 3 locations and in 6 years period

GN: Genotype No., MSE: Mean Squared Error, SL: Spike Length, GNS: Grains Number Per Spike, HI: Harvest Index, GWS: Grains Weight Per Spike, GY: Grain Yield

## Conclusions

As a result, in this study which was conducted with 36 bread wheat genotypes being developed for two years in 6 locations in 6 years period, it is seen that significant change occurred in the genotypes, being developed from 1968 until today, in terms of grain yield and yield components. It can be observed that superior genotypes have been developed with advances in the process of plant development, especially in terms of grain number per spike, grain weight per spike, harvest index and grain yield.

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# REACTION OF COMMON WHEAT AND TRITICALE CULTIVARS TO STRIPE RUST P. STRIIFORMIS

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#### Abstract

Stripe rust caused by (*Puccinia striiformis* Westend f. sp. *tritici*) is an important disease on wheat in Bulgaria. Although its occurrence is sporadic and directly dependent on the climatic factors in years which are favorable for its development, it can be severely harmful and cause significant yield losses. The use of resistant cultivars is the best method for control of the disease. This investigation presents the response of 11 triticale and 13 common winter wheat cultivars to natural infection of *Puccinia striiformis*. The study was carried out at Dobrudzha Agricultural Institute, Bulgaria during 2013-2014 cropping season. Among the investigated triticale varieties, cultivar Accord was highly resistant. Another 5 cultivars were identified as resistant: Colorit, Respect, Bumerang, Tvorets and Borislav. Varieties Irnik, Lovchanets and Blagovest were carriers of moderate resistance. In wheat, cultivar Rada was completely immune. High resistance was demonstrated by cultivars Ideal, Bolyarka, Demetra, Neda, Antonovka, Pchelina and Tina. Resistance was found in cultivars Anna, Elena and Lider, while cultivar Kossara reacted as moderately resistant.

Key words: stripe rust; resistance; wheat; triticale; cultivars.

#### Introduction

Rusts are main diseases on cereal crops in Bulgaria. The problem of their control is highly topical under intensive production. A number of reports show that endemic occurrence of stripe rust is predominant in some geographic regions and the damages caused are related to serious financial losses. Stripe rust also affects grain quality and yield. It decreases yield with about 30%, and in some years and regions the losses in susceptible cultivars may reach 100 %. The studies of Gromey (1989) in New Zealand have shown that crop losses are mainly related to the time of infection and the duration of moisture at anthesis. Stripe rust pandemics have been reported from North Africa, the India subcontinent, the Near East, East Africa, China (Saari and Prescott, 2005). Five severe stripe rust endemics were observed in Central Asia in 1998, 2000, 2005, 2009 and 2010 (Ziyaev *et al.*, 2011). Bunta (2005) pointed out that for the last 50 years, stripe rust occurred in Romania with highest intensity in 1961, 1967, 1978, 1985, 1988, 1991 and 2001. It is discussed in publications worldwide that important basis for the occurrence of these epidemics is that the resistance

of genes *Yr2*, *Yr9*, and recently of *Yr27*, too, has been overcome (Mc Intosh, 2009; Sing *et al.*, 2004; Wellings, 2007; Chen *et al.*,2009). Aggressive races of stripe rust have been detected in Sweden and Denmark damaging triticale and affecting wheat, rye and barley (Jorgensen *et al.*, 2010).

The manifestation of stripe rust in Bulgaria is sporadic. It occurs is regions with more humid and cool climate such as the coastal areas and the high fields at the mountain foothills. Stripe rust infections have been recently observed in 2010 and 2014. The favorable conditions of 2014 allowed the wider development of *P. striiformis* in comparison to 2010 and the disease affected the leaves thus causing their fast wilting.

The aim of this study was to assess the adult plant responses of wheat and triticale genotypes of Dobrudzha Agricultural Institute to natural infection of stripe rust.

#### Materials and methods

A research was conducted during 2013-2014 cropping season at Dobrudzha Agricultural Institute to determine the level of resistance in 11 triticale and 13 wheat cultivars to stripe rust (*Puccinia striiformis*) at adult plant stage under natural field infection. Each entry included 5 rows of 1m length, with 25 cm interspacing. Michigan amber was used as a susceptible cultivar. The attacking rate of stripe rust was read as percent of infected leaf area according to the modified scale of Cobb (Peterson, 1948). The attacking rate presented in Fig.1.

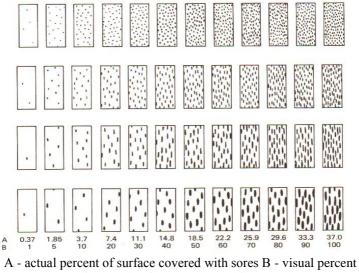
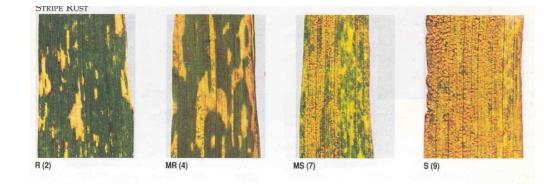


Fig.1 Scale of Peterson-attacking rate

The level of resistance was calculated by comparing relative disease severity (DS) of each of cultivar to that of the susceptible check M. amber. The relative DS was calculated by the formula as follows: Average disease severity (DS) of the cultivar / Average disease severity (DS) of M. amber x 100. The field response of host plant to stripe rust is presented in Table 1.

	Description	Observation	Constant Value
	No visible		
No Disease	infection	0	0
	Visible chlorosis or		
Resistant	necrosis,	R	0,2
	No uredia are present		
Resistant – Moderately Resistant		R-MR	0,3
·	Small uredia		
ModeratelyResistant	surrounded by	MR	0,4
-	chlorotic/necrotic		
	areas		
Mod. Res Mod. Sus.		MR-MS	0,6
	Uredia medium size with no		,
Moderately Susceptible	necrotic	MS	0,8
	margins but possibly some distinct		
	chlorosis		
ModSus Susceptible		MS-S	0,9
*	Large uredia with no necrosis or no		,
Susceptible	chlorosis	S	1

#### Table 1. Field response of host plant to stripe rust



Disease severity and host response data are often combined into a single value called the coefficient of infection (CI) (Table 1). The (CI) is calculated by multiplying the severity times a constant for host response: where immune I= 0.0, R=0.2, MR=0.4, MS=0.8, and S=1(Roelfs *et al.*, 1992). The field responses and the constant values are presented in Table 1.

## The cultivars were classified into:

Very resistant (0-5); Resistant (6-25); Moderately Resistant (26-45); Moderately Susceptible (46-65); Susceptible (66-100). The genotypes with coefficient of infection less than 20 were selected as resistant.

#### **Results and discussion**

The results from the investigation showed that the cause agent of stripe rust on wheat *P. striiformis* is not strictly specialized since it attacks many species belonging to family *Gramineae*. There is evidence of pathogenic races on triticale from Europe, Africa, China, and South America, and of the typical European isolates being highly virulent (Stubbs, 1988; Hovmoller and Justesen, 2007). The climatic conditions of 2014 favored the development and propagation of *P. striiformis* in Bulgaria. The attacks of *P. striiformis* are directly dependent on the climatic conditions; this is a pathogen which prefers low temperatures and high humidity. Johnson (1988) has pointed out that the viability and reproduction of the uredospores are highly dependent on the temperature, light and humidity. Stubbs (1988) has also pointed out that the optimum temperature for germination of the pathogen's spores is 9-12°C, the minimum temperature is 0°C, and the maximum is  $20-26^{\circ}$ C. Figure 2 shows that both the mean minimum (-2.2°C – 16.2°C), and the mean maximum temperatures (4.7°C – 27.8°C) varied within the optimum range for the development of the pathogen during the entire growth season.

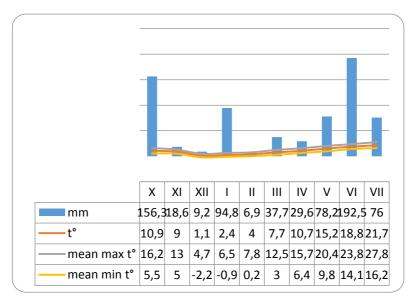


Fig. 2. Climatic characteristics of the period October 2013 - July 2014

The other factor favorable for the development of rust is humidity. During the spring months from March to June, which is the maximum of rust occurrence, the amount of rainfalls was from 37.7 mm to 192 mm. The high humidity in June and the severe occurrence of stripe rust caused fast wilting of leaves, especially in the susceptible cultivars. In our investigation, the attacking rate on the susceptible cultivar Michigan amber was about 60%. The response of the wheat and triticale cultivars involved in the investigation to the natural population of *P. striiformis* varied from immune (I) to susceptible (S) and is presented in Table 2.

Tust I . strujornus		1	<b>.</b> .				
Cultivars Triticale	Reaction	CI	Rating	Cultivars Wheat	Reaction	CI	Rating
Colorit	20 R	6.7	R	Ideal	5R	1.7	VR
Atila	30S	50.0	MS	Bolyarka	5MR	3.3	VR
Accord	10R	3.3	VR	Demetra	5R	1.7	VR
Respect	20R	6.7	R	Anna	10MR	6.7	R
Bumerang	20MR	13.3	R	Neda	5R	1.7	VR
Irnik	40MR	26.7	MR	Elena	10MS	13.3	R
Dobrudzhanets	60S	100	S	Antonovka	5R	1.7	VR
Lovchanets	40MR	26.7	MR	Stoyana	40MS	53.3	MS
Tvorets	20MR	13.3	R	Lider	10MR	6.7	R
Blagovest	40MR	26.7	MR	Kossara	30MS	40.0	MR
Borislav	10MR	6.7	R	Pchelina	5R	3.3	VR
				Tina	5R	3.3	VR
				Rada	0	0	Ι
				M.amber	60S	100	S

Table 2. Response of 11 triticale cultivars and 13 wheat cultivars to a natural population of stripe rust *P. striiformis* 

Six of the triticale cultivars (Bumerang, Irnik, Lovchanets, Tvorets, Blagovest and Borislav) reacted with moderately resistant type of infection, and the percent of attack was within 10-40 %. Cultivar Dobrudzhanets demonstrated highest susceptible reaction equal to the susceptible check Michigan amber - 60%. Resistant type of reaction was exhibited by triticale cultivars Accord, Colorit and Respect. The coefficient of infection for each of the cultivars is given in Table 2. Depending on this coefficient, each cultivar fell within a specific class of resistance. Cultivar Accord fell within the group of the highly resistant cultivars. Cultivars Colorit, Respect, Bumerang, Tvorets and Borislav were referred to the group of resistant cultivars, and cultivars Irnik, Lovchanets and Blagovest - to the group of moderately resistant cultivars. Cultivar Atila was in the group of moderate susceptibility, while cultivar Dobrudzhanets was completely susceptible. In wheat, cultivars Ideal, Demetra, Neda, Antonovka, Pchelina and Tina responded as highly resistant with 5% attack on the leaf surface. Cultivar Rada reacted as immune and no attacking rate was read on it. Cultivars Elena, Stoyana and Kossara responded with a moderately susceptible type of infection and the attacking rate was within 10-40 MS. Moderately resistant type of infection was demonstrated by cultivars Bolyarka, Anna and Lider. According to the coefficient of infection, the wheat cultivars were divided into classes of resistance as follows: cultivar Rada was immune; highly resistant were cultivars Ideal, Bolyarka, Demetra, Neda, Antonovka, Pchelina and Tina; Anna, Elena and Lider fell within the group of the resistant cultivars, and cultivar Kossara was in the group of moderate resistance. Cultivar Kossara reacted with a moderately susceptible type of infection and 30% rate of attack, its coefficient of infection ranking it as a cultivar of moderate resistance. Cultivar Stoyana belonged to the group of moderately susceptible cultivars.

## Conclusions

Among the investigated wheat and triticale cultivars it was found out that a great part of them were carriers of resistance to stripe rust. Triticale cultivars Dobrudzhanets and Atila

are not suitable for growing in the coastal areas of Bulgaria because they are susceptible and carry the risk of epiphytoty of stripe rust. Previous investigations (Ivanova, 2012) found out that a part of the wheat cultivars carry horizontal resistance to leaf rust. All wheat cultivars presented in this investigation which exhibited high to moderate resistance to stripe rust can be used in breeding programs as carriers of resistance, and some of the cultivars – as carriers of the two types of rust, stripe and leaf.

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# MORPHOLOGICAL CHARACTERISTICS OF SIX GRAIN LEGUMES UNDER THE CONDITIONS OF SOUTH-CENTRAL BULGARIA

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#### Abstract

A field experiment was conducted at the experimental base of the Plant Growing Department of Trakia University in Stara Zagora to establish the productive capacity of six annual grain legumes: 2 conventional species: spring pea and wintering pea and 4 new species: common vetch, bitter vetch, grass pea and chick pea, under the environmental conditions of South-Central Bulgaria. The trial was designed by the block method in 4 replications. The following morphological characteristics were measured: pods per plant, grains per plant, grain mass per plant, 1000 grain mass and stem height. Good correlations were found between the morphological characteristics (yield structure characteristics). The values of the morphological characteristics in the studied legumes were determined mainly by precipitation. Studies to determine the effect of tested factors (type of crop and year) showed that the type of crop had stronger influence on the yield structure characteristics (83.42 - 98.49% of the total variation). Stem height was the most important parameter with highest contribution to yield structure and was positive in Factor 1 and Factor 2. 1000 grain mass, grains per plant and total precipitation for the period January - December were the next parameters positive in Factor 1 and negative in Factor 2. The number of pods per plant and number of grains per plant were negative for Factor 1 and positive for Factor 2. Key words: annual grain legume, morphological characteristics, climatic conditions, PC analysis.

#### Introduction

Grain legumes play an important part in natural ecosystems, agriculture and agroforestry, where their atmospheric nitrogen fixation ability makes them excellent predecessors for most crops. Their great economic significance is also determined by the high content of protein in grain, which makes them wholesome concentrated food for people and animals (Duranti, 2006; Graham and Vance, 2003). Despite their valuable properties over the last years, a significant decrease has been observed in the production of protein crops. The main leguminous plants in the European Union, except for soya, have decreased with 30%, while the production of soya decreased with 12% (Häusling, 2011). To reduce the deficit of protein feeds it is necessary to find alternative solutions for efficient production of grain legumes, such as cultivation of drought resistant cultivars with short vegetation period, tolerant to abiotic stress factors (Berger and Turner, 2007; Krouma, 2009). The study of

the structural elements of yield and their correlation with the crops productivity is a certain means to establish the best crops for the specific agroclimatic conditions (Cokkizgin, A., 2012; Vateva and Krumov, 2011; Zhelyazkova et al., 2012). According to many research scientists solving the problem of legumes productivity is directly connected with the issue of structural elements, which precondition it, and the combination of climatic factors (Berger and Turner, 2007; Nenova and Venkova, 2005; Piergiovanni et al., 2011; Tekeli and Ates, 2003).

The aim of this study was to establish the morphological characteristics (yield structure characteristics) of six annual grain legumes: 2 conventional species: spring pea and wintering pea and 4 new species: common vetch, bitter vetch, grass pea and chick pea, under the agroecological conditions of South-Central Bulgaria.

## Material and methods

This study was conducted in the period 2010 – 2013 at the experimental field of Trakia University in Stara Zagora. The following grain legumes were studied: spring pea (*Pisum sativum* L.) cultivar Bogatir, winter pea (*Pisuma rvense* L.) cultivar Mir, common vetch (*Vicia sativa* L.) cultivar Dobrudja, bitter vetch (*Vicia ervilia* L.) cultivar Borina, grass pea (*Lathyrus sativus* L.) cultivar Strandja, and chick pea (*Cicer arietinum* L.) selection line FLIP 06-136.

The trial was set under non-irrigation conditions by the block method, in 4 replications, after a wheat predecessor. To prepare the soil was used soil tillage at 22 cm, after harvesting the predecessor and as pre-sowing cultivation with harrowing. Before the tillage, was applied fertilizer of 60 kg/ha phosphorus, whereas with the pre-sowing cultivation was applied 40 kg/ha nitrogen. For weed control after sowing before emergence was applied the herbicide Afalon 45 SC (Linuron) at a dose rate of 3 l/ha. For pest control against the grain weevils every year were conducted two treatments – at the beginning of flowering and 8 - 10 days later. The insecticide Nurele D (Chlorpiriphosetyl + Cypermethrin) was used at a dose rate of 500 ml/ha.

The soil in the area was Gleic Chromic Luvisols, moderately supplied with humus, with weak acid reaction, poorly supplied with nitrogen and phosphorus and very well supplied with assimilable potassium.

The following morphological parameters were studied: plant height at harvesting, number of pods and grain per plant, grain mass per plant and 1000 grain mass. The obtained data was statistically processed by the variance analysis method. To establish the correlation and regression dependencies was used the software package for statistical data processing StatSoft, STATISTICA for Windows (2000).

During the period of study (2010 - 2013) the mean annual precipitation was 567.8 mm. The annual precipitation sum in 2010 was 725.4 mm, which characterized it as the wettest – the precipitation was 31.3% above the average for the multi-year period (1936 – 2009). The driest year was 2011 when the precipitation sum was 32.3% below the average of the many-year period. The precipitation sum in the vegetation period (March – June) average for the 73-year period (1936 – 2009) was 209.9 mm. The vegetation precipitation sum of the vegetation precipitation was registered in 2010 – 248.2 mm, and the smallest in 2011 –

85.2 mm. During vegetation the precipitation was unevenly distributed, and the highest values were in May and June. This unevenness was particularly well expressed in 2011 and 2013 which were characterized with strong spring drought. As for the air temperature, 2012 and 2013 were warm years – with air temperatures 1.2 - 1.3 <sup>o</sup>C higher than the mean many-year period. The coolest year in the studied period was 2011 when the average annual air temperature was 0.2 <sup>o</sup>C lower than the average many-year value.

## **Results and discussion**

An important parameter, which greatly affected lodging resistance, suitability for mechanized harvesting, quantity and quality of obtained grain, was stem height (Table 1). The comparatively high amount of precipitation in 2010 led to formation of plants with greater stem height for all the studied crops. In these years was observed stem lodging of the pea and common vetch plants. With shortest and strongest stem and most resistant to lodging out of the studied species were distinguished the bitter vetch and chick pea - mean values of 36.2 cm and 41.5 cm, respectively, and with highest stem and most susceptible to lodging was with winter pea. The grass pea and common vetch had almost equally high stem height as spring pea, and the differences in this characteristic between the three crops were not proven. The specific climatic conditions had a significant effect on the stem height of grain legumes, most weakly expressed in grass pea and chick pea. One of the main elements of grain productivity in legumes was the number of pods per plant. Greater number of pods per plant for all studied crops was reported in 2010. The uneven distribution of precipitation in the spring of 2013 created unfavourable conditions for plant development and led to harvesting a smaller number of pods per plant for all studied crops. Spring pea formed an average of 5.4 pods per plant. Winter pea formed more pods per plant but the difference with spring pea was insignificant. The largest number of pods per plant was reported for bitter vetch, followed by chick pea and grass pea, which formed an average of 32.2 and 24.2 pods per plant, respectively. The number of grain per plant of the tested legumes was also highest in 2010 when the climatic conditions during the reproductive stages were more favourable. Spring pea formed an average of 25.3 grains. All the other legumes included in the study formed a greater number of grains per plant. Bitter vetch formed the higher number of grains – an average of 155.1. Almost the same number of grains per plant was formed by chick pea and winter pea -35.7 and 33.7, respectively, and the differences between the two crops by this parameter were not proven. The parameter of grain mass per plant was used as summarizing in order to reveal the productive potential of the plants. It was influenced by the external conditions and varied strongly in both studied years and between the species. The climatic conditions exerted a similar effect on the number of pods and grains per plant. The lowest grain mass per plant was obtained from common vetch and winter pea. The grain mass per plant of bitter vetch was an average of 5.4 g and the difference from spring pea was not significant. The highest grain mass per plant was obtained from chick pea and grass pea – an average of 13.8 g and 10.1 g, respectively. 1000 grain mass is an important parameter for grain quality and is a parameter of genetically based species and varieties. The data showed that the biggest and fullest grain was harvested in 2010, and the smallest was in 2012 when the precipitation in the period of grain filling was weaker. An exception was winter pea where 1000 grain mass in 2013 was higher compared to 2010. As a winter crop, it got in its development ahead of the spring species, which is probably the reason why the dry and hot period of June 2012 did not affect the size and fullness of its grain. Out of the studied crops the bitter vetch had the smallest grain, whereas the chick pea had the largest.

2010-2013, n = 96	1	r	-	r		
Plants / Parameters	2010	2011	2012	2013	Average	
		Stem height,		-		
Pisum sativum L.	85.8	52.4	89.2	44.4	68.0a	
Pisum arvense L.	149.9	123.3	151.1	113.7	134.5***	
Vicia sativa L.	65.9	52.2	98.4	47.0	65.9a	
Vicia ervilia L.	37.9	29.1	49.3	28.7	36.2***	
Lathyrus sativus L.	66.0	54.0	87.3	43.4	62.6a	
Cicer arietinum L.	52.4	36.9	39.0	37.7	41.5***	
Average	76.3	58.0	85.7	52.5	68.1	
		Pods per plant, 1	number			
Pisum sativum L.	6.1	5.4	5.3	4.9	5.4a	
Pisum arvense L.	7.7	7.3	7.1	7.6	7.4ab	
Vicia sativa L.	11.3	11.5	11.3	11.3	11.3*b	
Vicia ervilia L.	90.9	57.9	54.3	48.3	62.8***	
Lathyrus sativus L.	33.3	22.5	21.7	19.4	24.2***	
Cicer arietinum L.	30.9	37.6	32.7	27.6	32.2***	
Average	30.0	23.7	22.0	19.8	23.9	
	(	Grains per plant,	number			
Pisum sativum L.	25.9	26.2	26.0	23.0	25.3	
Pisum arvense L.	35.2	33.9	33.3	32.3	33.7**a	
Vicia sativa L.	49.5	50.7	50.2	47.8	49.5***	
Vicia ervilia L.	228.1	136.8	139.3	116.1	155.1***	
Lathyrus sativus L.	95.3	62.0	54.8	46.4	64.6***	
Cicer arietinum L.	34.8	40.5	36.4	31.3	35.7***a	
Average	78.1	58.3	56.6	49.5	60.6	
		Grain mass per	olant, g	•		
Pisum sativum L.	5.7	5.0	4.8	4.4	5.0a	
Pisum arvense L.	3.5	3.3	3.6	3.1	3.4***b	
Vicia sativa L.	3.2	3.5	3.1	3.0	3.2***b	
Vicia ervilia L.	8.1	5.5	4.8	3.5	5.4a	
Lathirus sativus L.	15.2	9.3	8.4	7.4	10.1***	
Cicer arietinum L.	15.1	15.1	14.9	10.4	13.8***	
Average	8.5	6.9	6.6	5.3	6.8	
		1000 grain ma	iss, g			
Pisum sativum L.	221.4	205.2	186.4	198.3	202.8	
Pisum arvense L.	121.1	116.4	116.0	132.5	121.5***	
Vicia sativa L.	62.2	69.8	54.4	62.5	62.2***	
Vicia ervilia L.	37.0	37.8	34.6	42.5	38.0***	
Lathirus sativus L.	171.7	157.4	140.2	182.5	162.9***	
Cicer arietinum L.	406.0	390.2	370.7	338.0	376.2***	
Average	169.9	162.8	150.4	159.4	160.6	

Table 1. Morphological structure parameters of grain legumes by years, average for the period 2010–2013, n = 96

\* Different letters indicate statistically significant differences among variants at P < 0.05

\*, \*\*, \*\*\* - Statistically significant differences of the variants and control at P< 0.05; 0.01 and 0.001, respectively.

The study aiming to establish the effect of the tested factors (type of crop and year) showed that the strongest, very well proven (P<0.00) effect on the morphological parameters (yield structure characteristics) of grain legumes was exerted by the type of crop - 80.15-98.49% of the total data variance (Table 2). With the parameters of number of pods and grains per plant and grain mass per plant was observed weaker but very well proven interaction of the type of crop with the year conditions -8.13-11.07% of the total variance.

Source of variation	Sum of squares	Degree of freedom	Mean squares	F	P<	%				
Factor analysis for stem height										
Plants	98630.7	5	19726.1	967.96	0.00	80.15				
Year	17367.4	3	5789.1	284.07	0.00	14.11				
Plants*Year	5597.7	15	373.2	18.31	0.00	4.55				
Error	1467.3	72	20.4			1.19				
Factor analysis for number of pods per plant										
Plants	37668.01	5	7533.60	4941.57	0.00	87.94				
Year	1388.00	3	462.67	303.48	0.00	3.24				
Plants*Year	3666.10	15	244.41	160.32	0.00	8.56				
Error	109.77	72	1.52			0.26				
Factor analysis for number of grains per plant										
Plants	186494.01	5	37298.80	3671.13	0.00	83.73				
Year	10861.47	3	3620.49	356.35	0.00	4.88				
Plants*Year	24656.11	15	1643.74	161.78	0.00	11.07				
Error	731.52	72	10.16			0.33				
Factor analysis for grain mass per plant										
Plants	1447.04	5	289.41	863.42	0.00	83.42				
Year	122.47	3	40.82	121.80	0.00	7.06				
Plants*Year	141.02	15	9.40	28.05	0.00	8.13				
Error	24.13	72	0.34			1.39				
Factor analysis for 1000 grain mass										
Plants	1192352.1	5	238470.4	222084.8	0.00	98.49				
Year	4745.6	3	1581.9	1473.2	0.00	0.39				
Plants*Year	13470.2	15	898.0	836.3	0.00	1.11				
Error	77.3	72	1.1			0.01				

Table 2.Influence of factors on the yield structure characteristics, average for the period 2010 - 2013, n=96

F- ratio among the variables; P- statistical signifficance

The power of influence of the climatic conditions over the years on the morphological parameters of the studied legumes was also very well proven (P<0.00), but lower. The reasons for this may be the great differences in the weather conditions of the year. The specific climatic conditions over the years as a factor affected to a greater extent the stem height (14.11% of the total variance). Good correlations existed between the yield structure characteristics of grain legumes (Table 3). The number of pods per plant had a good correlation dependency with the number of grains (r = 0.583 - 0.990) and the grain mass per plant (r = 0.793 - 0.987) for spring pea and common vetch, bitter vetch, grass pea and chick pea. 1000 grain mass was in a good correlation with the number of grains per plant for spring and winter pea, common vetch and chick pea (r = 0.585 - 0.795). The values of the yield structure characteristics for the studied legumes were determined to a

great extent by precipitation. Strongest influence on the 1000 grain mass of spring pea was observed by the precipitation in April and May (r = 0.601 and -0.652, respectively), of winter pea in March (r = 0.800), and of common vetch, bitter vetch and grass pea in May r = -0.795; -0.798 and -0.922, respectively. The number of pods per plant of winter pea was most affected by the precipitation in March (r = 0.798), of bitter vetch and grass pea – in April (r = 0.574 and 0.563, respectively), and of chick pea in the period March – June (r = -0.717). The grain mass per plant of winter pea was most affected by the precipitation in May (r = 0.645), of common vetch – the precipitation in the period March – June (r = 0.645)0.590), of grass pea – in April (r = 0.559), and of chick pea – in March (r = 0.509). The established correlation coefficients between the listed morphologic parameters and annual precipitation were lower and statistically insignificant, which can be explained with the fact that these are spring crops with comparatively small vegetation period. The Principal Component Analysis (PCA) conducted to determine the power of influence and contribution of the studied morphological parameters (yield structure characteristics) on the grain yield, showed that the principal components with greatest effect on grain yield can be determined (Table 4). These components described from 37.72% to 31.21% of the changes. Stem height and 1000 grain mass were the parameters with greatest extent of influence on the grain yield (0.560 - 0.547, respectively). Stem height was the most significant morphological parameter for the studied legumes, because it was positive in the first and second factor (Figure 1). The other parameters had no significant effect on grain yield. The integrated PC analysis on the influence and contribution of the studied morphological parameters of yield showed that they can be divided into three groups, as follows: first - stem height - positive in F1 and F2; second - 1000 grain mass, grain mass per plant and annual precipitation sum - positive in F1, negative in F2; third - number of pods and grains per plant – negative in F1 and F2.

## Conclusions

The strongest influence on the morphological parameters of grain legumes was exerted by the type of crop - 83.42 - 98.49% of the total data variance. Good correlations existed between the yield structure characteristics. The number of pods per plant had a good correlation dependency with the number of grains and the grain mass per plant for spring pea and common vetch, bitter vetch, grass pea and chick pea. 1000 grain mass was in a good correlation with number of grains per plant for spring and winter pea, common vetch and chick pea. The values of the yield structure characteristics for the studied legumes were determined to a great extent by precipitation. Strongest influence on the 1000 grain mass of spring pea was observed by the precipitation in April and May, of winter pea in March, and of common vetch, bitter vetch and grass pea in May. The number of pods per plant of winter pea was most affected by the precipitation in March, of bitter vetch and grass pea – in April, and of chick pea in the period March – June. The stem height was the most important parameter with highest contribution to yield structure and positive in F1 and F2. 1000 grain mass, number of grains per plant and total precipitation for the period January – December were the next parameters positive in F1 and negative in F2. Number of pods per plant and number of grains per plant were negative for F1 and positive for F2.

Pods per plant, number         Pods per plant, number           tig         0.583*           umber         0.583*           0.710*         0.733*           tig         0.710*           uber         1.000           tig         0.710*           uber         1.000           tig         0.710*           uber         1.000           tig         0.517*           uber         0.617*           uber         0.617*	Grains per plant, number 1.000 0.699* 0.123 0.123 0.123	Grain mass per plant, g	1000 grain mass, g	Precipitatio n I VII mm	Precipitatio n	Precipitati	Precipitati on	Precipitati	Precipitati	Precipitatio
plant, number 1.000 0.583* 0.793* 0.710* 0.710* 0.374 0.374 0.517* 0.633*	plant, iumber 0.123 0.123 0.123 0.123	mass per plant, g	mass, g	n T VII mm	n	5	on	u o		
number           1.000           1.000           0.583*           0.710*           1.000           0.710*           1.000           0.7110*           1.000           0.7110*           1.000           0.7110*           0.517*           0.517*           0.633*	umb <u>er</u> 0.123 0.123 0.123 0.123 0.123 0.123 0.123 0.100	plant, g		T VII mm		110			IIO	п
t 0.583* 0.583* 0.793* 0.710* 0.710* 0.710* 0.374 0.311 0.517* 0.464 0.633*	1.000 1.000 0.123 0.123 0.123			1-A11, IIIII	III-VI, mm	III, mm	IV, mm	V, mm	VI, mm	X-VI, mm
t 0.583* 0.583* 0.793* 0.793* 0.710* 0.710* 1.000 0.374 0.311 0.517* 0.633*	1.000 1.000 0.123 0.123 0.123		Pisum sativum	um L.						
t 0.583* 0.710* 0.710* 0.710* 0.374 0.374 0.311 0.517* 0.633*	1.000 0.123 0.123 0.123 0.123 0.123 0.100			0,270	0.257	0.137	0.314	-0.039	0.209	
0.793* 0.710* 0.710* 0.374 0.374 0.311 0.517* 0.517* 0.464 0.464	0.123 0.123 1.000 0.334 0.334			0.117	-0.221	-0.479	-0.339	0.395	-0.422	
0.710* 0.374 0.311 0.517* 0.517* 0.464 0.633*	0.123 1.000 0.334 -0.406	1.000		0.358	0.255	0.154	0.315	-0.057	0.220	
1.000 0.374 0.311 0.517* 0.517* 0.464 0.464	1.000 0.334 -0.406	0.674*	1.000	0.027	0.231	0.579*	0.601*	-0.652*	0.554*	
1.000 0.374 0.311 0.517* 0.517* 0.464 0.464 0.633*	1.000 0.334 -0.406		Pisu	Pisum arvense L.						
0.374 -0.311 0.517* 1.000 0.464 0.633*	1.000 0.334 -0.406			0.330	0.501*	0.798*	0.773*	-0.649*	0.779*	-0.025
-0.311 0.517* 1.000 0.464 0.633*	0.334			0.246	0.125	0.064	0.205	-0.053	0.114	-0.066
0.517* 1.000 0.464 0.633*	-0.406	1.000		0.439	0.178	-0.381	-0.157	0.645*	-0.248	-0.326
1.000 0.464 0.633*		-0.575*	1.000	0.088	0.418	0.800*	0.638*	-0.654*	0.732*	-0.010
1.000 0.464 0.633*			Vicia sativa	a L.						
0.464 0.633*				-0.166	-0.244	-0.109	-0.141	-0.090	-0.144	
0.633*	1.000			-0.128	-0.292	-0.423	-0.361	0.279	-0.405	
	0.573*	1.000		-0.507*	-0.590*	-0.312	-0.372	-0.153	-0.387	
1000 grain mass, g 0.219 0	0.079	0.582*	1.000	-0.662*	-0.616*	0.101	-0.108	-0.725*	-0.067	
			Vicia erviliaL	liaL.						
Pods per plant, number 1.000				0.382	0.472	0.397	0.574*	-0.197	0.474	
Grains per plant, number 0.984* 1	1.000			0.398	0.524*	0.375	0.573*	-0.117	0.469	
0.962*	0.944*	1.000		0.251	0.287	0.180	0.364	-0.077	0.254	
-0.328	-0.377	-0.482	1.000	-0.248	0.037	0.649*	0.401	-0.798*	0.513*	
			Lathyrus sativus L.	ivus L.						
Pods per plant, number 1.000				0.359	0.475	0.381	0.563*	-0.172	0.462	
	1.000			0.288	0.383	0.326	0.496	-0.184	0.395	
ant, g 0.987*	0.992*	1.000	0.179	0.324	0.445	0.389	0.559*	-0.212	0.461	
1000 grain mass, g 0.154 0	0.126	0.179	1.000	-0.010	0.350	0.921*	0.762*	-0.922*	0.828*	
			Cicer arietinum L	um L.						
Pods per plant, number 1.000				-0.516*	-0.716*	-0.679*	-0.681*	0.233	-0.717*	
Grains per plant, number 0.971* 1	1.000			-0.474	-0.655*	-0.653*	-0.640*	0.253	-0.680*	
	0.718*	1.000		0.054	-0.210	-0.509*	-0.336	0.432	-0.438	
0.510*	0.514*	0.795*	1.000	0.035	-0.062	-0.161	0.002	0.066	-0.109	

#### SECTION 9. FIELD CROP PRODUCTION

Characters	P C 1	P C 2	P C 3	P C 4
Yield, kg.ha <sup>-1</sup>	0.690	-0.384	-0.338	0.406
Height, cm	0.560	0.561	-0.385	0.329
Legume, number	-0.836	-0.448	-0.213	0.158
Grain per plant, number	-0.913	-0.103	-0.303	0.244
Grain per plant, g	0.094	-0.953	-0.051	0.052
1000 grain mass, g	0.547	-0.776	0.164	-0.126
Precipitation I-XII, mm	0.102	-0.043	-0.888	-0.445

Table 4. Principle component analyses, average for the period 2010 - 2013, n=96

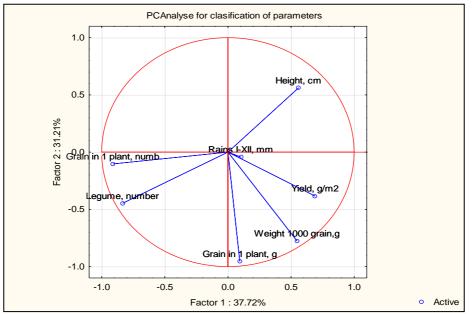


Figure 1. Principal component analysis for the distribution of parameters of yield structure characteristics of annual legume grain.

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#### UDC: 633.71-152.61(497.775) Professional paper

# TECHNOLOGICAL PROPERTIES OF TOBACCO IN SOME ORIENTAL VARIETES OF TOBACCO TYPE PRILEP

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#### Abstract

Based on the technological properties of tobacco leaves is determined the so-called technological-commercial tobacco quality. Researches were performed in 2014 on patented and current tobacco varieties from the type prilep such as: prilepP-23 Ø; prilepP-66-9/7; prilepNS-72 and prilepP-79-94. In the accredited Laboratory for control of authenticity and quality of raw tobacco - L04, at the Scientific Institute of Tobacco–Prilep were analyzed the most important technological properties of dry tobacco leaves from the middle and upper harvest belt: the presence of the midrib, materiality and thickness of the leaves. The results from the research, shows that the subject varieties from the type Prilep are characterized by very good technological properties and they have high technological-commercial quality.

Key words: tobacco varieties, technological properties, type Prilep.

## Introduction

The tobacco as an agriculture crop is being grown for its leaves. Oriental tobaccos are well known in world markets, given the fact that the raw material is intended for export. Our tobacco types with its quality are involved in making the highest quality cigarette brands. Total tobacco production in Macedonia is around 22 - 26000 tones with a tendency to increase in future. Into the structure of the total export of oriental tobacco, the dominate tobacco type is Prilep with 62.5%, followed by the type Jaka with 25.5% and the type Basma with 10.4% (Mitreski, 2012). it's well known that this tobacco types are characterized with nice and delicate fabrics, good elasticity and pronounced veins. Technological properties of tobacco leaves are very important because through them the tobacco assess can be made and they are also very important for the processing and processing for the final product. Through them, organoleptic and taste properties of tobacco are presented. These studies aim to identify the important tobacco properties (representation of the first rib, representation of shale, thickness of tobacco leaves and materiality) through which is determined the so-called technological-commercial quality.

## Materials and method

The trial was set up in 2013 on delluvial-colluvial soil in the field of Tobacco Institute-Prielp with four varieties of Prilep tobacco (P-23 Ø, P-66-9/7, NS-72 and P-79-94). The second spring ploughing was followed by fertilization with NPK 10:30:20 in a rate of 250 kg/ha. The experiment was designed in randomized blocks with four replicates. All necessary agro-technical measures for normal growth and development were applied in the field during the growing season. In July and August watering of the trial with wing sprinklers system was applied with 25  $1/m^2$  water. After harvesting, tobacco was sun-cured in barns specially designed for drying of oriental tobacco.

The researches were concentrated midrib presence; thickness and the leaves material are performed in the accredited laboratory – L04 in the Scientific Tobacco Institute-Prilep.

#### **Results and discussion**

Quality of tobacco raw material is determined by several external indicators, which also includes the technological properties.

Boceski, quoted by Mitreski (2012), separates the technological properties in 3 groups: morphological, physical and organoleptic. From morphological properties he mentions the insertion, stem of the leaf, the size and scale and also the shape of the leaf. From the physical properties he marks materiality, the thickness of the leaf, volume weight of tobacco leaf and its elasticity. As important organoleptic properties, he includes the color of the leaf, the tenderness and the smell of tobacco.

In Table 1 we present the results of our research for each property separately, for better visibility comparison between varieties and of course for making appropriate conclusions.

Tuble1: Teennologieu	i properties of tootae	es ieures (ureiuge i	araes)	
Varieties for the	Presence of the	Presence of the	Thickness	Materiality
tobacco type Prilep	midrib	leaf	(µm)	$(g/m^2)$
	(%)	(%)		
P - 23 Ø	18,13	81,87	81,15	52,35
P-66-9/7	17,94	82,06	80,46	50,79
Р-НС-72	18,76	81,24	81,74	48,83
P-79-94	18,95	81,05	83,92	49,17

Table1. Technological properties of tobacco leaves (average values)

## Presence of the midrib in the leaves

Stem leaf constitute the nerve and secondary nerves. The content of the rib relative to the total weight of the sheet, on one side points on the fineness of leafy fabrics, but on the other hand points the thickness of the sheet which negatively reflects in fabrication.

Lazaroski (1976), concluded that the midrib from the type Prilep occupies 18,89% of the total weight of the leaf in the lower middle leaves, 18,75% in the actual middle leaves and 17,83% in upper secondary leaves.

Pecijareski (1959), says that the weight of the midrib for Macedonian tobacco types Prilep, Jaka and Otlja is relatively low and that in the total weight of leaf participates with 16,38% in the type Jaka, 17,38% in the type Prilep and 18,70% in the type Otlja.

In our tests (Table 1), we can see that on average, the highest percentage content of midrib we have variety in P-79-94 (18.95%), while the lowest percentage is distinguished

variety P-66-9/7 (17.94%). In the control variety P-23 the average proportion of the midrib is 18.13%.

#### Thickness of tobacco leaves

The thickness of tobacco leaves is an important property for the raw tobacco. With the rising of the thickness the quality of tobacco is getting low, but also very thin leaves have bad quality as in the case with the immature and overripe tobacco leaves.

The thickness of the leaves is approval of type characteristic, but it changes a lot depending on climatic conditions, insertion, applied agrotechnics, level of ripeness etc.

Uzunoski (1985), states that thickness of the leaf is inversely proportional to the quality of tobacco. With increasing thickness, the quality of the tobacco decreases. An exception is making tobaccos which thin leaf fabrics is due to immature or overripe. According to thickness, tobacco leaves are divided to three groups: thin, malnourished, low material leaves, comprehensive and wooden leaves.

Mitreski (2012), based on two years of research found that the thickness of the leaves on the type Prilep is moving average from 75,91  $\mu$ m in average harvests and 93,50  $\mu$ m in the above harvests.

Uzunoski (1985), provides data where the average thickness of the leaves type Prilep is  $110,40\mu$ m, the type Jaka  $86,30\mu$ m and the type Otlja 105 micrometers.

In our comparative researches in question varieties (Table 1), the average thickness of the leaves varies from  $83,92 \mu m$  in the variety P-79-94, to 80.46 micrometers in the variety P-66-9/7 which has the most thin leaves.

## Tobacco leaves materiality

The materiality of the sheet indicates the amount of dry material concentrated per unit area of the sheet. As a physical indicator of quality, this is the most outstanding feature summary outward sign of inner content of the leaf, and primarily serves for determination by way of indirect smoking. Materiality increases from the lower to upper insertions.

Veselinov, quoted by Patche and Georgiveski (1987), emphasizes that the materiality in practice is identified with the term content, but this is not always correct because this two terms are depending on use values of tobacco. In oriental aromatic types of tobacco it is very important to dominate chemical components that dictate taste and aroma (carbohydrate, resins and essential compounds).

Uzunoski (1985), concluded that the Macedonian tobacco brands are characterized by moderate materiality. According to him that materiality that reflects the highest quality for high harvests in some types of tobacco in Macedonia ranges: from 61-67 g/m<sup>2</sup> type Prilep; type Jaka 48-57 g/m<sup>2</sup> and type Otlja 56-57 g/m<sup>2</sup>.

Average materiality (medium and upper harvests) in the tested varieties can be seen in Table 1. The table clearly shows that the control variety (P-23) features the highest average materiality of 52,35 g/m<sup>2</sup>, while it is lowest the variety NS-72 (48,83 g/m<sup>2</sup>). In the other two varieties (P-66-9/7 and P-79-94) materiality average is 50.79 or 49,17 g/m<sup>2</sup>.

## Conclusions

From our research on technological properties and the results obtained, we can bring the following conclusions and findings:

1. The presence of the midrib against the shale is relatively low. The average content of the midrib ranges from 17.94% (P-66-9/7), to 18.95% (P-79-94). In the control variety it is 18.13% and is somewhat higher among the variety NS-72 (18.76%).

2. The thickness of the tobacco leaves is in the optimal range of aromatic oriental tobacco. In the tested varieties with average heaviest leaves is P-79-94 ( $83,92\mu m$ ), and a variety slimmest P-66-9/7 ( $80,46\mu m$ ).

3. The results of the research showed that the tobacco varieties with are with quality raw material. Namely, the highest average materiality features control (P-23) with 52,35 g/m<sup>2</sup>, followed by the variety P-66-9/7 with 50,79 g/m<sup>2</sup>. Slightly lower average materiality has P-79-94 (49,17 g/m<sup>2</sup>), while the lowest value for this technological capacity has seen the variety NS-72 (48,83 g/m<sup>2</sup>).

4. From the results and conclusions presented, we conclude that the tested varieties such as Prilep are characterized by good technological properties, that have high-tech commercial quality.

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# COMPARATIVE TESTING OF THE BULGARIAN VARIETIES OF DURUM WHEAT (TRITICUM DURUM DESF.)

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#### Abstract

The experiment is performed at the Agricultural University of Plovdiv, according to the block system method, with four repetitions, with plot dimensions of  $15 \text{ m}^2$ , during the period 2010 - 2013. The following new Bulgarian varieties of durum wheat are tested: IPK Elbrus, IPK Deni, Zvezditsa and Impuls, in order to analyze their productive capacity. The variety Progress is used as a standard. As a result of the performed experiment, it was concluded that: the productivity of the new Bulgarian varieties of durum wheat is higher than that of the standard variety Progress. The grain yield by variety IPK Elbrus is 420 kg/ha (10.7%), variety Zvezditsa with 350 kg/ha (8.9%), variety IPK Deni with 230 kg/ha (5.8%), and variety Impuls with 130 kg/ha (3.3%) more than variety Progress. **Key words:** Bulgarian varieties durum wheat, productivity.

## Introduction

The increase of the productivity and quality of the durum wheat grains is a result of the interaction of a complex factors including variety, ecological conditions and technology of growing (Delibaltova & Kirchev 2010); (Kirchev 2014); (Kolev *et al.*, 2000). The optimization of these factors leads to the achievement of high results in the production of durum wheat (Kolev *et al.*, 2008); (Yanev & Kolev 2008); (Semkova *et al.*, 2007).

Every variety with its genetic capacity is an effective mean for increasing the yield and grain quality (Kolev *et. al.*, 2004); (Yanev 2006). By growing appropriate varieties for a certain region we can reduce the negative impact of ecological conditions (Clarke, 2000). The objective of the study is evaluating the productive capacity of some Bulgarian varieties of durum wheat in the conditions of Southern Bulgaria.

## Material and methods

The experiment is conducted at the Agricultural University of Plovdiv, according to the block system method, with four replications, and plot dimensions was 15 m<sup>2</sup>, on alluvial-and-meadow soil (Molic Fluvisols according to FAO), which is characterized with average sandy and clayish mechanical composition, humus content 1-2 %, pH 7.7, carbonate content up to 7.4 %. The content of the main nutritional elements in the soil layer 0-20 cm is the following: N – 15.6 mg/1000 g, P<sub>2</sub>O<sub>5</sub> - 32 mg/100 g, K<sub>2</sub>O - 47 mg/100 g (Popova &

Sevov 2010). The following Bulgarian varieties of durum wheat were tested, IPK Elbrus, IPK Deni, Zvezditsa and Impuls, which have been developed in the Institute of field agricultural plants in the town Chirpan. Variety Progress is used as a standard. The production of the durum wheat is done according to the generally accepted technology (Yanev *et al.*, 2008). The plants are sown within the optimal period, i.e. from 20.X. to 10.XI., and with applied fertilization 80 kg/ha  $P_2O_5$  and 120 kg/ha N, which is done in the following way: before sowing – the entire quantity of phosphorous fertilizer and 1/3 of the nitrogenous fertilizer, while early in the spring – the remaining part of the nitrogenous fertilizer as a nutrition.

The following indicators are analyzed: plant height (cm); wheat-ear length (cm); number of small ears in the wheat-ear (pieces); number of grains in the wheat-ear (pieces); grain weight in the wheat-ear (g); weight of 1000 grains (g); hectoliter weight (kg); grain yield (t/ha). The physical characteristics of the grains are determined according to the methodology specified in the Bulgarian National Standard – BDS.

The statistical analyses of the obtained data were performed according to the method of the dispersion analysis.

#### **Results and discussion**

During the experimental period more significant deviations are observed in the quantity of rainfalls for the different years, as well as during the critical phases of the vegetation of the durum wheat, compared to the thirty-year period. The quantity of rainfalls from sowing to harvesting (X-VI) of the durum wheat is as follows: 2010/2011 - 388.5 mm; 2011/2012 - 517.4 mm, and 2012/2013 r. -462.6 mm; while this value for the long-term period is 419.0 mm. The distribution of the precipitation during the vegetation period is most beneficial for the development of the durum wheat during the third experimental year. The registered quantity of rainfalls during the harvest in 2012 for the months March and April, was less respectively with 33.1 mm and 18.8 mm, compared to the long-term period. Due to the less rainfalls accompanied by higher temperatures compared to the normal for the month of May, the normal blossoming was delayed, as well as the normal pollination and formation of the grains in the wheat-ear. These climatic characteristics during the harvest 2012 played a negative effect on the grain yield of the durum wheat for all tested varieties.

The analysis of the meteorological data during the critical periods of the development of this agricultural crop allows us to characterize the experimental years as follows: first one – dry; second – averagely dry; and third one – normal.

Table 1 presents the average data for the obtained values based on the biometrical observations of some of the structural elements of the yield and the physical characteristics of the grain.

The tested new varieties of durum wheat have shorter stems of the plants from 2.5 cm for variety IPK Elbrus to 5.8 cm for variety IPK Deni, compared to the wheat variety Progress. Variety IPK Elbrus is characterized with the longest wheat-ear, which exceeds the standard with 0.11 cm. For the other tested varieties the length of the wheat-ear is less compared to Progress; variety Inpuls is characterized with the shortest wheat-ears. In each wheat-ear of the tested new varieties there are formed averagely from 24.4 small ears for Impuls to 25.6 small ears for IPK Elbrus, whiles for the standard they are 23.5 pieces.

The wheat-ears of the newly selected varieties of hard wheat form larger number of grains: with 0.1 pieces for Impuls; with 1.7 pieces for IPK Deni; with 2.0 pieces for Zvezditsa; and with 5.7 pieces for IPK Elbrus more that those formed by the variety Progress. Regarding grain weight in the wheat-ear the tested varieties IPK Elbrus – 2.52 g, Zvezditsa – 2.50 g and IPK Deni 2.48 g surpass the standard - 2.46 g. Among the explored physical characteristics of the grain we observe more significant variation in the weight of 1000 grains. With regards to this indicator the highest values are registered for variety Zvezditsa 51.4 g, followed by the standard Progress with 50.7 g.

			Variety		
Indices	IPK Elbrus	Impuls	IPK Deni	Zvezditsa	Progress
Height of the plants, cm	92.8	90.2	89.5	90.2	95.3
Lenght of the spike, cm	8.81	7.0	7.4	8.4	8.7
Number of the spikelets per spike	25.6	24.4	24.8	25.0	23.5
Number of the grains per spike	53.8	48.2	49.8	50.1	48.1
Mass of the grains per spike, g	2.52	2.40	2.48	2.50	2.46
Mass of 1000 grains, g	48.4	47.0	47.6	51.4	50.7
Mass hl., kg	81.0	79.4	79.8	80.0	80.4

Table 1. Biometrics data (mean of the period 2010 - 2013)

The other varieties have lower weight of 1000 grains compared to the standard. Regarding the indicator hectoliter weight there are no significant changes between the standard and the tested varieties. The new varieties of durum wheat surpass in productivity the standard variety Progress (Table 2). For the different years the highest grain yield is achieved during the normal in climatic aspect year 2013, followed by the yield of 2011. Due to the drought during the spring months of 2012, the productivity of all tested varieties is lower.

Variety	2010-2011	2011-2012	2012-2013	Aver	age
variety	t/ha	t/ha	t/ha	t/ha	%
IPK Elbrus	4.29	4.08	4.69	4.35	110.7
Impuls	4.03	3.85	4.30	4.06	103.3
IPK Deni	4.11	3.96	4.41	4.16	105.8
Zvezditsa	4.23	4.03	4.58	4.28	108.9
Progress	3.89	3.77	4.13	3.93	100.0
GD 5%	0.40	0.31	0.56		

Table 2. Grain yield, t/ha

The data in Table 2 shows that highest grain yield is achieved by variety IPK Elbrus for the three-year experimental period. During the harvest year 2011, yield of this variety is 4.29 t/ha (10.3 %), during 2012 - 4.08 t/ha (8.2 %), and during 2013 - 4.69 t/ha (13.6 %) or averagely for the experimental period 4.35 t/ha (10.7 %), while yield of standard Progress is 3.93 t/ha. The higher yields of grain for the different years varies from 400 kg/ha during the first year to 560 kg/ha during the third year, or averagely with 420 kg/ha more than the standard. Averagely for the experimental period the varieties Zvezditsa, IPK Deni and Impuls produce respectively 4.28 t/ha (8.9 %); 4.16 t/ha (5.8 %) and 4.06 t/ha (3.3 %), which means with 350 kg/ha, 230 kg/ha and 130 kg/ha more than variety Progress.

# Conclusions

The productivity of the new Bulgarian durum wheat varieties is higher than the standard variety Progress. The grain yield is higher for variety IPK Elbrus 420 kg/ha (10.7 %); variety Zvezditsa 350 kg/ha, variety IPK Deni 230 kg/ha, and variety Impuls 130 kg/ha than variety Progress. The plants of the new durum wheat varieties form larger number of grains, with bigger weight of the grains in the wheat-ear. Regarding the indicator weight of 1000 grains, the highest values are registered for variety Zvezditsa 51.4 g, followed by the standard Progress with 50.7 g. The other varieties have lower weight of 1000 grains compared to the standard. Regarding the indicator hectoliter weight there are no significant differences between the standard and the tested varieties.

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# QUINOA - PROSPECTS FOR ITS PRODUCTION AS A NEW CROP IN SERBIA

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#### Abstract

Quinoa (*Chenopodium quinoa* Willd.) is a seed crop of the *Chenopodiaceae* family, traditionally cultivated in the Andean region for several thousand years. Quinoa is considered as a multipurpose agricultural crop that is also tolerant to different stress factors. The aim of the present paper was to test the possibility for introducing quinoa as a new crop in Serbia. Several field trials in Serbia (localities Stara Pazova and Surduk) were conducted in rainfed conditions during period of 2010-2011 year. For testing quinoa varieties KVL37 and KVL52 were used. They were selected for European climatic conditions and provided by the University of Copenhagen (Denmark). Yield of both varieties depended on the year, but the average value was at around 1,500 kg/ha. Assessment of the seeds of field grown plants confirmed its significant nutrient values (high content of proteins, essential amino acids, oil, starch and minerals - K, Mg, Fe, Cu, Zn and Mn).

Key words: quinoa, yield, seed quality.

## Introduction

Quinoa (*Chenopodium quinoa* Willd.) is a pseudocereal which originates from the Andean highlands of South America. Due to its healthy nutritional characteristics and its high yield potential under adverse soil and climate conditions, quinoa has gained an increasing amount of international attention over the past few decades. In contrast to most plant protein sources, quinoa seed protein contains a balanced level of essential amino acids. Quinoa also has higher levels of minerals compared to other grains and contains significant levels of vitamins and essential fatty acids (Vega-Gálvez *et al.*, 2010). Additionally, it contains high levels of polyphenols and flavonoids which are beneficial for human health (Repo-CarrascoValencia *et al.*, 2010). Attention has been also given to quinoa for people with celiac disease (allergy to gluten), as an alternative to the cereals wheat, rye and barley, which all contain gluten (Jacobsen, 2003).

In the last decade, the quinoa supply has diversified in terms of both varieties and products available. Today, in addition to the basic seed quinoa (with the saponin removed, ready for consumption), there is a wide variety of quinoa-based products, such as breakfast cereals biscuits, healthy snacks, noodles, instant soups, beverages, beers and ice creams.Our recent study of bread supplemented with quinoa seeds could enable the development of a range of new baking products with enhanced nutritional value (Stikic *et al.*, 2012). Quinoa also exhibits remarkable agronomic characteristics because plants show tolerance to frost, salinity and drought, and have the ability to grow on marginal soils (Jacobsen *et al.*, 2003). Thus, significant efforts are increasing to adapt and grow quinoa in various parts of the world (Jacobsen, 2003). Recently, quinoa's potential as a nutritious and resilient crop was recognized by the FAO which declared 2013 as the International Year of Quinoa (FAO, 2013). Although quinoa has been tested in Macedonia with good yield (Prof. D. Bosev, pers. com.) and in one locality in Serbia (Stikic *et al.*, 2012), the potential of growing this crop in South East Europe has not been fully exploited. Thus, the aim of presented study was to further test the possibility for growing and utilizing quinoa in Serbia by assessing potential yield quantity and quality under rainfed field conditions.

#### Material and methods

The experiments were carried out during the 2010 and 2011 growing season in two localities (Stara Pazova and Surduk) in Serbia. The experiments were organised as field trials in rainfed conditions as randomized complete block design with four replications. Plots size were 4 rows of 4 m, with an inter-row spacing of 0.5 m. The soil in both fields had optimal mineral content and was classified as chernozem according to IUSS working group soil classification (IUSS, 2006). Quinoa varieties used for investigation were KVL37 and KVL52, provided by the University of Copenhagen. The varieties have been selected for adaptation to European conditions. Sowing rate of quinoa for both trials and seasons was 10 kg/ha at a final density of 600.000 plants /ha. Sowing was done in April, while harvest at physiological maturity (in August). Physiological maturity was defined as the date when seeds from the main panicle become resistant when pressed (Bertero et al., 2004). During the investigated seasons, plants were treated against weeds. From both localities and both seasons ten plants from the central rows in each replication were randomly chosen for seed yield quantity. The seeds of 40 plants were bulked and weighed and the seed yield/ha was calculated. The seed quality was checked from one locality (Stara Pazova) and during 2010 season. Assessment of seed quality was done on the base of the analyses the content of proteins, oils, starch and minerals - K, Mg, Fe, Cu, Zn and Mn according to the procedure explained by Stikic et al., (2012).

#### **Results and discussion**

The results for quinoa yield obtained during both investigated seasons and on both localities are presented in Table 1.

Table 1. Yield of quinoa varieties KVL37 and KVL52 (kg/ha) grown in localities Stara Pazova and Surduk during the 2010 and 2011 seasons.

Varieties	Seasor	n 2010	Seasor	n 2011
	Stara Pazova	Surduk	Stara Pazova	Surduk
KVL37	1,495	1,671	1,508	1,629
KVL52	1,224	1,533	1,425	1,383

Yield results demonstrated that yield of variety KVL37 in both trials and seasons (average 1,575 kg/ha) was for 22% higher comparing to the average yield of KVL52 (1,392 kg/ha). Comparison of the effects of season and locality showed that the yield of variety KVL37 was slightly (but not significantly) higher in 2011 comparing to the 2010 season, and in locality Stara Pazova than in locality of Surduk. For variety of KVL52 the trend was opposite. The influence of climatic conditions on yield of quinoa varies depending on the stage of plant development. The climatic characteristic for the years of 2010 and 2011 when the quinoa experiments were conducted was a warm springs (March and May) and an hot summers (June and August). Both seasons were similar in average temperature values for the period April and September (18.4°C for 2010 and 19.2°C for 2011), while the sum of seasonal precipitation for this period was higher in 2010 season (509 mm) compared to 2011 (270 mm).

High germination of quinoa seeds required adequate contact of seeds with soil at the optimal moisture level (Darwinkel and Stolen, 1997). Sowing depths of 2 to 5 cm were recommended because quinoa germination rates were found to drop steeply at depths greater than 5 cm (Darwinkel and Stolen, 1997). According to these recommendations sowing depth in our experimental fields was 5 cm. Although soil temperature has a strong effect on successful germination, reports on optimum temperature vary. In Denmark, seed is usually planted when the soil temperature is 7-8°C, while in UK soil temperatures of 5-8°C are suitable for planting quinoa in the spring (Jacobsen and Bach, 1998). To avoid the effect of low temperature on seeds germination, our seed planting was done in April. In this period of both seasons the soil water content in our rainfed trials was sufficient for germination. Our results did not confirm that smaller total participation during season 2010 compared to 2011 had a decreasing effect on the yield of our quinoa plants.

Quinoa is considered to have remarkable drought tolerance. It has been reported to grow with as little as 200 mm in annual precipitation in pure sand (Aguilar and Jacobsen, 2003). Our very recent results indicate that reaction of quinoa plants to drought are based on drought avoidance mechanism: reduced transpiration and sustained water uptake (Stikic *et al.*, 2015). However, the effect of drought on yield of quinoa depend the stage of plant development. When drought occurred during the pre-flowering stage up until the dough stage, significant decreases in yield were noticed. Jensen *et al.* (2000) found decreases in yield when drought was applied during flowering and seed fill. Less rainfall during the flowering season of 2010 compared to the same period in 2011 may explain differences in yield among these two seasons.

Yield in our experimental trials was lower than yields in some of the reporting data. Quinoa yields in Denmark typically range from 2,000-3,000 kg/ha (Jacobsen *et al.*, 2010), while the variety Baer yielded 5,140 kg/ ha (Risi and Galwey, 1991). Our experimental fields were under rainfed conditions and one of the recommendations for growers to increase yield is 25-38 cm of combined irrigation and rainfall (Johnson and Croissant, 1985). The addition of organic matter has also been shown to be useful in increasing quinoa yield under arid conditions (Martínez *et al.*, 2009).

In our experiment with the variety KVL52 grown in the locality of Stara Pazova and during the season of 2010, we also assess the quality of quinoa seeds. In Table 2. the results for following parameters are presented: proteins, essential amino acids (lysine and tyrosine), oil, starch and minerals (K, Mg, Fe, Cu, Zn and Mn).

Table 2. Quality parameters of KVL 52 variety seeds.	
Parameter	Quantity
Moisture (%)	10.90
Protein (%)	17.43
Essential amino acids-lysine and tyrosine	3.8 and 3.6
(g/100g proteins)	
Oil (%)	4.92
Starch (%)	63
K (g/kg)	9
Mg (g/kg)	1.3
Fe (mg/kg)	48
Cu (mg/kg)	1.8
Zn (mg/kg)	15
Mn (mg/kg)	19

Table 2. Quality parameters of KVL 52 variety seeds.

In general, seed quality similarly to our previous results (Stikic *et al.*, 2012) and results of Koziol (1992) confirmed that the protein content in quinoa seeds was higher than in cereals (maize 10%, rice 8%, wheat 14%). Similar was for oil content, while the content of essential amino acid and minerals were in the range of mineral content reported for other quinoa genotypes (Vega-Galvez *et al.*, 2010). Our results also confirmed that quinoa seeds were particularly high in essential amino acids, especially lysine (3.8) which is the limiting amino acid in most cereals.

# Conclusions

The results of field trials showed that prospect for introducing quinoa production in Serbian agro-climatic conditions are good. Yield of both varieties depended on the year, but the average value was at around 1,500 kg/ha. Assessment of the seeds of field grown plants confirmed its significant nutrient values (high content of proteins, essential amino acids, oil, starch and minerals - K, Mg, Fe, Cu, Zn and Mn). Further testing other quinoa varieties and cultivars will be worthwhile.

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# THE IMPACT OF GUAPSIN AND TRICHOPHYTE ON THE QUANTITATIVE TRAITS OF AN EAR OF WHEAT

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#### Abstract

The paper examines the impact of Guapsin and Trichophyte biological preparations on the quantitative traits of an ear of wheat. The experiment was conducted during 2014 in the municipality of Aleksinac, in the village of Dobrujevac. The soil at the experimental plot produced a slightly acidic reaction, with 3% of humus, poorly secured with available phosphorus and well secured with potassium. The applied biological preparations were Guapsin, in the amount of 5 1/t of seed, and Trichophyte, in the amount of 2 1/t of seed immediately before planting, while for the purposes of the experiment the Balaton variety of wheat was used. By means of a random selection, 40 ears of wheat from each repetition had been taken, which provided the following indicators: the length of an ear, the number of spikelets, the number of grains per spike and the weight of grains per spike. A treatment with the said biological preparations has had a positive impact on the examined quantitative traits of an ear of wheat. The results of the research have shown that these biological preparations have a significant impact on an ear's length and the weight of grains per spike, while the number of spikelets and the number of grains per spike have not exhibited specific significant differences.

Key words: biological preparations, wheat, quantitative traits of an ear of wheat.

## Introduction

The use of microbiological fertilizers or biofertilisation in modern agricultural production represents the introduction of living microorganisms to the soil with the aim of improving the plant supply with essential nutrients. Biofertilisers are preparations which contain specially selected cultures of microorganisms which are either used for the purposes of inoculation of seeds and seedlings, or introduced in the soil in order to intensify certain microbiological processes which have the ability to increase the amount of available nutrients to the plant (Jarak & Čolo, 2007).

The impact of biofertilisers is manifested through an improved supply of plants with nitrogen, phosphorus, potassium, iron, sulphur, as well as through stimulated root growth. By introducing bacteria to a plant rhizosphere one accelerates the processes of transformation of organic substances and the plant is supplied with essential nutrients (Jarak & Durić, 2006). Fungi are heterotrophic microorganisms which, due to their

differentiated enzymatic system, play a significant part in the soil, since they participate in the degradation of varios organic substrates and are widely distributed in the plant rhizosphere (Jarak & Đurić, 2008).

Associative nitrogen-fixing bacteria live in a close association with a plant and are most frequently applied to the seed before sowing, or introduced in the soil during the growing season. This group comprises the bacteria of the genera *Acetobacter*, *Azospirillum, Bacillus, Pseudomonas, Rhizobium*, etc. The ability of these bacteria is to fix nitrogen from the atmosphere and produce growth stimulators (Jarak & Čolo, 2007). It is well-known that the bacteria of the genus *Pseudomonas* have the ability to transform organic forms of sulphur to inorganic, thus making it more available to plants. Furthermore, these bacteria have the ability to synthesise plant hormones (such as gibberellin and auxin), which additionally stimulate a plant growth and influence a plant resistance. By introducing microbiological fertilizers to soil one influences the flow and direction of microbiological processes in the soil, which consequently influences the growth and development of plants, as well as the quality of soil. The aforementioned indicates that the use of microbiological fertilizers has its place in modern, conventional, as well as organic agricultural production.

#### Material and methods

The paper examines the impact of Guapsin and Trichophyte biological preparations on the quantitative traits of an ear of wheat. The experiment was conducted during 2014 in the municipality of Aleksinac, in the village of Dobrujevac. The soil at the experimental plot produced a slightly acidic reaction, with 3% of humus, poorly secured with available phosphorus and well secured with potassium. The preceding crop was wheat, the ploughing was done at a depth of 20 to 25 cm, the fertilisation preceding ploughing was performed with 130 kg of the MAP fertilizer and 100 kg of urea. The seedbed preparation was carried out by means of a disc harrow and harrowing, while sowing was carried out by means of a seed drill with the spacing of 15 cm between the feeders. The sowing was performed on October 15<sup>th</sup> 2013, with the sowing density of 200kg/ha. The applied biological products were Guapsin, in the amount of 5 1/t of seed, and Trichophyte, in the amount of 2 l/t of seed, immediately before the planting, while for the purposes of the experiment the Balaton variety of wheat was used. During the growing phase the crop was treated against weeds, diseases, and vermin by using the following preparations: the Decis (0,25 l/ha), the Impact (0,3 l/ha), the Sekator (0,15 l/ha)1/ha) and the Cikosel (0,5 1/ha). In the part of the plot where seed treatment was conducted this mixture was enriched with Guapsin (6 l/ha) and Trichophyte (3 l/ha). In the flowering stage, in the third decade of May, the second treatment was carried out by using the Fastak preparation (0,2 l/ha). The experiment was conducted within four repetitions. By means of a random selection, 40 ears of wheat from each repetition had been taken, which provided the following indicators: the length of an ear, the number of spikelets, the number of grains per ear and the weight of grains per ear.

On the basis of the effect of the *Pseudomonas aureofaciens* B-306 and B-111 bacterial strains, it can be asserted that biopreparation Guapsin protects the plants from vermin and disease, and encourages their growth and development. The results of scientific

research show that these bacteria are modifiers of poorly soluble soil compounds, whereby the physiological activity of plants is increased.

The biopreparation Trichophyte, an aqueous suspension of soil spores and mycelia and the *Trichoderma lignorum* fungus, shows successful results in the battle against fungal diseases. Biologically active substances secreted by *Trichoderma lignorum* stimulate the growth and development of plants and increase their resistance to various diseases. Due to high biological activity of the fungus the organic compounds included in the processes of ammonification and nitrification decompose, phosphorus and potassium mobilisation increases, while the soil becomes enriched with nutrients.

#### **Results and discussion**

Microbiological fertilizers help the plants to collect substances more effectively by using natural resources, thus avoiding the soil pollution. They help the nutrition of plants by converting organic and poorly soluble components into milder forms, which thus become more available in the root system. The use of microbiological fertilizers in the form of biofertilizers in the production of different plant species enables a better use of their potentials for the purposes of developing both plants and microorganisms (Avies et al., 2008).

Repetitions	Ear len	gth (mm)	Number	of spikelets
	untreated	treated	untreated	treated
Ι	80,65	83,13	16,66	18,36
II	80,86	82,11	16,57	17,43
III	81,96	81,45	16,63	17,13
IV	79,21	82,80	15,76	17,07
Average	80,67	82,37	16,40	17,50
LSD 005		1,75		1,16
001		2,68		1,52
Cv		4,23		

Table 1. The values of quantitative indicators of the ear length and number of spikelets

The ear length of untreated variants was between 79,21 mm and 81,96 mm, whereby the average value was 80,67 mm. The ear length in treated variants was between 81,45 mm and 83,13 mm, the average value being 82,37 mm. The average number of spikelets was 16,40, i.e. its span across repetitions was from 15,76 to 16,66. In the treated seeds the average number of spikelets was 17,50, while its span across repetitions was from 17,07 to 18,36. Between the examined treatments there were no ascertained significant differences, bearing in mind the genetic prospects of these traits.

Tuble 2. The fulle	os or quantitative ma	ieutors or the grunn	number und weight pe	i cui
Repetitions	Grain num	ber per ear	Grain weigh	t per ear (g)
	untreated	treated	untreated	treated
Ι	35,51	41,55	1,525	1,871
II	34,30	40,03	1,432	1,792
III	34,73	40,06	1,488	1,691
IV	34,55	40,60	1,417	1,742
Average	34,77	40,56	1,466	1,774
LSD 005		5,68		0,223
001		6,37		0,305
Cv		4,65		

Table 2. The values of quantitative indicators of the grain number and weight per ear

Regarding the number of grains per ear, the values in the untreated variant went from 34,30 to 35,51, i.e. the average value was 34,77. The number of grains per ear in the treated variant across the repetitions went from 40,03 to 41,55, the average value being 40,56. The grain weight in the treated variant across the repetitions went from 1,417 to 1,525 g, the average value being 1,466 g. Regarding the grain weight per ear in the treated variant, the values went from 1,691 to 1,871 g, the average value being 1,774 g. Between the examined treatments the authors established significant and extremely significant differences. The results of Kovačević and associates (2007) show that the combination of organic and microbiological fertilizers in the organic system of growing resulted in significantly higher yields of different types of winter wheat. Kovačević et al (2009, 2011) suggest that they had obtained significantly higher yields of different alternative sorts of winter wheat by means of foliar nutrition treatments with microbiological fertilizer. During the experiments with winter rye, row naked oats, buckwheat and spelled they also established that different microbiological fertilizers in the combination with soil improvers provide one with optimal results (Oljača et al., 2012; Dolijanović et al., 2013a).

Differences between treated and untreated variants show that in all examined indicators positive results were obtained after a treatment with microbiological fertilizers. The largest percentage increase occured in indicators dealing with grain weight per ear (21%), and the smallest in indicators dealing with the number of spikelets (7%), whereby the number of grains per ear is by 17% larger in the treated variant. The results of Hilali et al. (2001) show that the yield of wheat grain inoculated with *Rhizobium legumonisarum bv*. *Trifoli* was by 25% larger than with non-inoculated wheat.

#### Conclusions

On the basis of the results obtained in this paper one can safely assert that the applied biological preparations Guapsin and Trichophyte significantly influenced the quantitative traits of wheat ear. Particularly significant is the impact of these preparations on the increase of grain weight per ear by 21 %, and on the increase of grain number per ear by 17%. Regarding the ear length and number of spikelets, the impact of the examined biological preparations is less significant, bearing in mind the genetic prospects of these traits.

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## THE INFLUENCE OF SOME FACTORS ON THE LEVEL OF ACCEPTANCE OF TOBACCO SEEDLINGS

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#### Abstract

Tobacco has widespread areal of distribution and great adaptation towards outdoor conditions. During the production, the acceptance of seedlings is different as a result of biological potential of the varieties, technical measures and agro-environmental factors at the time of transplanting as well as quality of seedlings, as a the main factor. The aim of the study was to find out how certain factors influence on the acceptance of tobacco seedlings. The experiment was conducted in random block system, in four replications on two varieties of oriental tobacco (*prilep NS 72*, and *yaka JV 125/3*) in three variants: variant 1- control; variant 2 - Float Tray N (TERRA STAR/22:11:22+2Mg) and variant 3 - Float Tray P (CHELAN/ 11:49:12+2Mg). The results show different level of acceptance of the tobacco seedlings as a result of different meteorological conditions in the studied years. The results also show a statistically significant difference between numbers of accepted plants due to the different technology of production of tobacco seedlings, Therefore, for successful tobacco production it is necessary putting into practice appropriate, modern and profitable technology for production of seedlings.

Key words: oriental tobacco, acceptance, tobacco seedlings.

#### Introduction

The production of oriental tobacco in Macedonia has a very long tradition. Tobacco is one of the most economically important industrial crops in the country. To produce highquality tobacco, growers must begin with healthy seedlings. The ideal seedling is disease free and resistant to survive transplanting shock. The seedlings should be available on time, in order to start with transplanting earlier in season. In general, earlier transplanted seedlings give better yield than late-transplanted tobacco, (Smith et al., 2003). During the vegetation, a large number of factors have an impact on tobacco plant and interfere to express its biological and production potentials. Except the biological potential of the varieties, the largest influences have taken the scientific farming methods and agro ecological conditions during the growing period. Each type of tobacco variety, requires separate intervention depending on the intensity of biotic agents, soil and climatic

conditions, as well as cultural, traditional practices. As extremely adaptable, tobacco could be grown on different soil type; therefore the yield and quality received will highly dependent on the type of soil and its physical and chemical properties. The influence of soil conditions is correlated with a complex of other environmental factors. Thus, the same soil type but with different climatic conditions, we get the raw tobacco material with different yield and quality. The production of oriental tobacco in Macedonia is located almost in all regions, mostly soils with weaker productivity. Usually, for growing oriental tobacco loamy-sandy soil with weaker productivity are preferred (Georgievski, 1990), where rarely other agricultural plant cultivation would be cost-effective. On the other hand, for successful tobacco production good quality seedlings are needed, to obtain uniformity according to morphological and biological characteristics of tobacco at field. Traditionally, seedlings in Macedonia are produced at the open field, in *cold beds*, covered with polyethylene. The most critical period in development of tobacco occurs immediately after transplanting. The plants are torn away from the previous, favorably living environment and immediately underwent to a radically different and many times unfavorable external conditions, resulting in stress in young plants. This change is significant in terms of seedlings produced by classical technology of production, where there is an extremely large imbalance between the root system and the new conditions that often result in extended periods of stress, poor reception and slow plant growth (post transplanted shock). This shock is caused by the loss of most of the root system in the process of uprooting the seedlings from the beds (Hoyert, 1979). Climatic conditions also cause the severity while transplanting (from cold and wet spring, into a rapid shift to hot and dry summer). Applied practice and scientific research in the production of tobacco seedlings indicate that on the Balkans; the production technology is quite conservative and it is very demanding to implement a new one. According to Chaluhov, (1987), in spite of great importance of changing the traditional production technology, there is a lack of interest by the majority of tobacco growers to introduce new technologies for the production of oriental tobacco seedlings. The seed quality is also an important factor in production of tobacco seedlings. Bigger seed has a higher energy of germination, which is guarantee of getting healthy and quality plants with well-developed root system. This is precondition for higher yield and quality of tobacco. The production of healthy seedlings, with certain length and thickness of the stalk, is the first step in the process of excellence tobacco production. The last decade has resulted in increased awareness of tobacco producers about the importance of quality tobacco seeds. Although the seeds with a declaration of minimum 90% germination, seedling producers fail to get 90% germination during nursery stage according to Clarke et al., 2001. It is believed that there is no method that guarantees the safe production of seedlings, although today there are many computer programs for the establishment and management of parameters and analysis of the results of the process. For this reason, education and human responsibility at work are decisive factors. Improving tobacco production requires good agricultural practice which among other things involves implementation of new production technologies. The introduction of new varieties of tobacco in the structure of production of tobacco in the country, with higher yield and quality is of great importance as well, considered Kabranova & Arsov (2009).

#### Material and methods

The experiment was placed in the region of Veles. It was conducted in random block system, in four replications on two varieties of oriental tobacco prilep NS 72 and yaka JV 125/3: Variant 1-control (traditional production); Variant 2 - Float Tray (TERRA STAR 22:11:22+2Mg with microelements: Fe-0,0335 %, Cu-0,017 %, Mg-0,1 %, B-0,01 %, Mn-0,017 %, Mo in traces, Zn-0,01 %, Co in traces, +EDTA and Auxin) and Variant 3-Float Tray (CHELAN 11:49:12+2Mg with microelements: Fe-0,0335 %, Cu-0,017 %, Mg-0,1 %, B-0,01 %, Mn-0,017 %, Mo in traces, Zn-0,01 %, Co in traces). The total quantity of fertilizer was added in water beds (0.001 % solution). The required quantity of certificated tobacco seed (granulated tobacco seed for FT variant) sown in polystyrene trays (589 alveolus per tray) that were filled with peat 50% and perlite 50%); polyethylene for covering water beds; agril (as a protector towards condensation) and adequate protection against diseases, insects' damage. Conductivity of water into the pool was followed regularly for keeping the concentration of fertilizer. Thermoregulation above the pool was conducted with uncovering of the tunnel. For traditional technology of seedlings production (control) additional fertilizers were needed (1% solution is applied to seedlings), followed with application of fungicides and insecticides in order to get vigorous seedlings. All agro-technical measures for proper development of plants were made for both technologies of seedlings production, in order to obtain maximum healthy, usable seedlings per unit area. Common agro-technical measures for successful production experiment were conducted. Transplantation of tobacco was done manually. Transplanting time was in close connection with the meteorological conditions during the test period. After transplantation of tobacco seedlings, the number of accepted plants (%), and the crisis period (from transplanting to acceptance) were analyzed. The results were processed by SPSS for Windows, procedure Sum of squares, Model III.

#### **Results and discussion**

The period after transplanting is considered crucial for the overall production at the end of vegetation. Therefore, after transplanting, the plants should have enough available moisture well to embrace and develop the first strip of leaves. Thus, precipitation in this period should be in amount of 60 mm. In case of insufficient moisture and high temperature, however, the growth and development of plant are reduced and the plant suffers from drought. The temperature regime for oriental tobacco, together with precipitation in the initial stages of growth and development of tobacco should give a stimulating effect. Preferred temperature throughout the vegetation period is between 22 - 25 °C. Thus, the optimal temperature has positive effect on tobacco seedlings acceptance. But it is very rare to get such a temparature in our conditions. Distribution of rainfall was inadequate during the growing season, while the air temperature values were relatively high. There is statistically significant difference (\*\*) between percentage of accepted plants between 2007-<sup>th</sup> compared to 2008-<sup>th</sup> and 2007-<sup>th</sup> compared to 2009<sup>th</sup> (\*), shown below, in Table 2. The tobacco requirements in the field significantly differ from those in the nursery. Therefore, plant experiencing a biological stress as result of the drastically negative changes in the environment and manifested by the reduced growth and

development of plants with altered biological functions. Depending on the intensity changes of permanent damage caused by low or high temperature, drought, etc. may occur. If this stress is lower, the plants will begin with normal growth and development faster (Vukadinovic, 1999). Production of tobacco continues with the transplantation of tobacco seedlings in the field, depending on the seedlings and weather conditions at the time of the year. The level of acceptance of tobacco seedlings depends on agro-environmental features at the time of transplanting, agro-technical measures, but mostly from the quality of seedlings. Plants are often competitive in terms of external factors: light, CO<sub>2</sub>, H<sub>2</sub>O, oxygen, minerals, substrate (in the area where the roots develop). Competition does not always bring negative consequences, because according Vukadinovic (1999), it affects the formation of the size of the habitus of plants. During the uprooting of seedlings, part of the root system is damaged, much reduced, and most parts of the roots are excluded from the soil particles. In the early days after transplanting, a plant falling into crisis and is unable to satisfy their need for water. Therefore, wilted leaves lead to reduction of surface assimilation of plants. In this stage, the processes of degradation exceed synthesis processes. At this point, the special role of stem should be pointed out as the main reservoir of food that supports the viability of the plant. Plants that have a thicker stem, are easier to accept than those who have a thin stem that often, if not die giving underdeveloped plants with weak productive opportunities. In this phase, the elevated parts of the plant almost do not grow. After successful acceptance of plants in the field, a strong root system is developing and the plant is ready for further rapid increase (Uzunovski, 1989). The seedlings derived from the classical mode of production (variant 1 -control), have a critical period after transplantation in the average range of 7 to 10 days in both examined varieties (prilep NS 72 and yaka JV 125/3). In that period, many of the plants suffer due to poorly developed root that after transplantation, it adapts to the new, often less favorable conditions for growth and development. Therefore it is necessary to add, replace with new plants between. Table 1 shows the variations in % of acceptance after transplanting in the field.

				Variant			
Year	variant 1	- control	variant	2-FT, N	varian	t 3-FT, P	Year,
I Cai	prilep NS	yaka JV	prilep	yaka JV	prilep	yaka JV	average
	72	125/3	NS 72	125/3	NS 72	125/3	average
2007	68.5	72.5	94.8	90.8	90.0	88.0	84.1
2008	79.3	80.5	95.0	93.8	89.8	89.3	87.9
2009	77.9	75.8	94.2	91.9	91.5	89.7	86.8
Variety,	75.2	76.3	94.7	92.1	90.4	89.0	
average	15.2	70.5	94.7	92.1	90.4	89.0	
Technology,	7	5.7	0	3.4	89.7		
average	/.	5.1		J. <del>4</del>	0	17.1	

Table 1. Acceptance of tobacco seedlings after transplanting, %

Acceptance of tobacco seedlings (Table 1) is different during the three-years trials and presented as an average of all tested varieties is: 84.1% for the 2007-<sup>th</sup>, 87.9% for 2008-<sup>th</sup> and 86.8% for 2009-<sup>th</sup>. The three-year results show the highest average of accepted plants in variant 2-FT –N (93,4%), while 89.7% in the variant 3-FT –P. The lowest

acceptance shows variant 1-control (75.7%). In the same technology of production (FT), variant 2-FT -N shows greater acceptance of tobacco seedlings which is certainly the result of the tested fertilizers. Choice of the appropriate combination of macro-and micronutrients is an additional factor that contributes to the normal development of seedlings. The seedling, because of good care and support with the necessary ingredients (nutrition, water, air, etc..) and not existence of mutual competition, were equal, with balanced growth and development of plants (strong roots, well-developed stem and number of leaves) giving the higher acceptance of plants in the field. Increased percentage of acceptance of the FT seedling was as a result of the short period of time needed for their acceptance after transplantation. Accepted plants in the field, did not respond to altering conditions, means that tobacco seedlings are completely healthy and strong enough to resist the shock of transplantation. Acceptance in the field depends on tobacco seedlings quality, expressed above all with a well developed root system and morphological uniformity in terms of its dimensions (Pearce ve Palmer, 2005). As for the differences due to the variety, the results showed that percentage of accepted plants variant 2-FT, N was 94.7% in the variety prilep NS 72, or 92.1% of the variety vaka JV 125/3. The average of accepted plants in variant 3-FT, P was 90.4% in the variety prilep NS 72, or 89.0% of the variety yaka JV 125/3. The results show greater acceptance and minimal crisis period indicate that seedling of this quality have no need of additional transplantation in rows, so the plants continue their rapid development equally. These plants are morphologically identical during growth and development in terms of height, number of leaves, the size of the leaves that are almost identical (Turshic, 2000). Statistical analysis of the values of factors: interaction between year and technology tested with F-test shows statistically significant difference at the level P = 0.01 (Table 2).

Dependent variant	Ye	ar	Difference of mean values between years	Standard error	Significance
o/ 1	2007	2008	-3,8333**	0,99121	0,004
% accepted plants	2007	2009	-2,7400*	0,99121	0,022
plants	2009	2008	-1,0933	0,99121	0,299
Dependent variant	Techn	ology	Difference of mean values between variants	Standard error	Significance
0/ 1	1	2	-22,6600***	0,99121	0,000
% accepted plants	1	3	-18,9633***	0,99121	0,000
plants	3	2	-3,6967**	0,99121	0,005

Table 2. Statistical analysis of the values of factors

From the data presented in Table 2, there is a statistically significant difference between percentage of accepted pants in  $2007^{th}$  compared to  $2008^{th}$  and  $2007^{th}$  compared to  $2009^{th}$ . Also, in terms of production technology can be seen that there is a statistically significant difference between the percentage of accepted plants in variant 1-control and variant 2-FT -N, the variant 1-control and variant 3-FT -P as well as variant 3-FT, P and variant 2-FT –N.

## Conclusions

Accepting tobacco in the field depends on its quality expressed above all with a well developed root system and morphological uniformity in terms of its dimensions.

As primary limiting factor for successful transplanting, and thus better accepting, the quality of seedlings should be emphasized, or the way how tobacco seedlings are produced.

Selecting the right technology for tobacco seedling production along with the selection of varieties, are key factors for getting high yield and quality of tobacco raw material, with respect to the impact of agro-environmental conditions and agro-technical measures implemented during the vegetation of tobacco.

Agro ecological conditions have strong influence on its biological, morphological and technological characteristics as well.

These results indicate that tobacco production deserves more attention when it comes to adaptation measures in order to achieve better quality of seedlings.

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# INVESTIGATION ON PRODUCTIVITY PARAMETERS OF NEW DURUM WHEAT VARIETIES BY GENOTYPE AND NITROGEN FERTILIZATION

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#### Abstract

The aim of the study was to investigate the productivity parameters of 5 varieties of durum wheat by genotype and 4 levels of nitrogen fertilization. The study was performed at the Institute of Field Crops in Chirpan during the period 2004-2007. Varieties Progress, Neptun2, Beloslava Saturn1 and Prosperity were tested. Structural elements of the crop - number of tillers, number of spike stems, productive tillering and plant height were taken as components of yield and as components of spike - spike length, number of grains and grain weight per spike. By increasing of fertilizer rate up to N18 productive tillering increases as well as structural elements of the spike of all tested varieties of durum wheat during the first two years of study, while under the conditions of 2007 fertilizer rate N12 adversely affects these indicators. Nitrogen fertilization is a factor with strong influence on the formation of leaf area of durum wheat. Compared to the impact of the variety, the effect of nitrogen fertilization on leaf area formation of durum wheat is more pronounced in the three phenological phases (phase of stem, ear formation and lactic ripeness).

Key words: durum wheat, nitrogen fertilization, productive tillering, plant height.

#### Introduction

Modern production of durum wheat provides highly productive varieties, technology based on mechanization as well as strong and effective application mineral fertilizers and plant protection products.

Durum wheat is highly responsive to fertilization (Lalev al., 2000). Higher grain yield is obtained as a result of increasing of productive tillering, the number of seeds per spike and the grain mass in spike at optimal interaction between the factors N kg / ha and sowing rate (Dzhugalov, 2010). Fertilization affects the weight of seeds in a spike and the weight of 1000 grains as well as on the weight of the seeds in the spike and the number of productive plants and seeds of spike m<sup>2</sup> (Duplak et al., 2002). Nitrogen fertilization of durum wheat influences the formation of solids which continues throughout the growing season (Panaiotova, 2004). Different dates of sowing and temperature conditions in the autumn affect phenological development of durum wheat (Kirchev, 2011). According to Aparicio et al., (2002) the size of the seed is most closely associated with the development

and biomass of seedlings, as well as the overall development of the first two to four leaves. This stage of growth and development is one of the most important stages, playing a decisive role in the structure of the crop.

#### Material and methods

The experiment was conducted during the period 2004/2007 at the Institute of Field Crops in the town of Chirpan. The perpendicular method in 4 repetitions was used at predecessor cotton. Two diskings were performed - the first of 10-12 cm in the direction of the lines of its predecessor, and the second - at 6-8 cm perpendicular to the previous one.

Sowing was carried out with 550 germinating seeds / m2. The study included standard Progress, Vozhod, Beloslava, Saturn 1 and Neptune 2. Four levels of nitrogen fertilization - N0, N6, N12, N18 were applied, once in early spring in the form of ammonium nitrate. The following structural elements of the crop were examined- number of tillers, number of spike stems, productive tillering as well as plant height, as well as the components of the spike - the spike length, number of grains and grain weight per spike. To establish a statistically significant influence of the studied factors and differences between the tested variants it was used the dispersive analysis of variance /Shanin 1977/. To calculate the qualitative and quantitative relationships the correlative analysis was used. Statistical treatment of the results was carried out with the product "BIOSTAT ©".

#### **Results and discussion**

The magnitude of yield is closely linked with productive tillering of durum wheat and the other cereal crops. In this study, density of stems in the crop of durum wheat varies quite widely over the years of study, depending on the region, genotype and nitrogen fertilization. Because the third (2007) harvest year is special in terms of climate, we consider it separately.

	N norms kg/da	Progres	Neptun2	Beloslava	Saturn1	Vozhod	GD A B
Number of tillers /m <sup>2</sup>	0 6 12 18	485 542*** 628*** 642***	461 ns 634*** 672*** 698***	497 ns 542*** 610*** 672***	483 ns 582*** 665*** 728***	512### 634*** 686*** 743***	5% 14.1 12.6 1% 18.7 16.7 0.1% 24.3 21.8
Number of spike stems /m <sup>2</sup>	0 6 12 18	502 573*** 636*** 638***	502 ns 666*** 652*** 652***	521## 566*** 580*** 715***	504 ns 614*** 691*** 731***	531### 615*** 652*** 721***	5% 14.1 12.6 1% 18.7 16.7 0.1% 24.3 21.8
Productive tillering, %	0 6 12 18	64.5 63.5 70.2 70.0	67.1 74.7 75.2 76.2	68.6 73.2 75.3 75.2	66.4 69.7 73.2 81.2	71.8 74.3 78.0 78.1	

Table 1. General and productive crop density - average for two years (2005-2006)

Factors (according to Material and Methods): A - Variety; C - Nitric norm; \* \*\* \*\*\* - Proven increase in the number of tillers and the number of spike stems / m<sup>2</sup> to N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences; # ## ### - Proven increase in the number of tillers and the number of spike stems / m<sup>2</sup> to control variety Progress in fertilization N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences

The data from two-factor analysis of variance showed that genotype differences in the number of tillers and the number of spike stems are well defined and statistically significant in 2005-2006 .With the highest average number of tillers is variety Vozhod and the highest average number of spike stems - variety Saturn 1. The nitrogen fertilization positively influences the fundamental indicators of productive crop, which is apparent when applying the norm N<sub>6</sub>. In all tested varieties of durum wheat in the first and second harvest year the maximum tillers and spike stems are formed at fertilization with the highest tested fertilizer rate (N<sub>18</sub>). Productive tillering of surveyed durum wheat varieties during the two harvest years increased with the increasing of nitrogen fertilizer rate.

Table 2. Analysis of variance of general and productive crop density – 2007								
	N norms kg/da	Progres	Neptun2	Beloslava	Saturn1	Vozhod	GD A B	
Number of tillers /m <sup>2</sup>	0 6 12 18	441 431 ns 512*** 559***	520### 578*** 622*** 494 ns	381 ns 486*** 617*** 591***	473### 609*** 606*** 648***	473### 452 ns 617*** 593***	5% 6.6 5.9 1% 8.8 7.9 0.1% 11.4 10.2	
Number of spike stems /m <sup>2</sup>	0 6 12 18	252 297*** 181 ns 194 ns	410### 368 ns 305 ns 236 ns	318### 307 ns 129 ns 129 ns	399### 417*** 179 ns 139 ns	381### 257 ns 189 ns 84 ns	5% 6.6 5.9 1% 8.8 7.9 0.1% 11.4 10.2	
Productive tillering, %	0 6 12 18	57.1 68.9 35.4 34.8	68.9 63.6 49.0 47.9	83.5 63.2 20.9 21.8	84.4 68.5 29.4 21.5	80.6 57.0 30.6 14.2		

Table 2. Analysis of variance of general and productive crop density – 2007

Factors (according to Material and Methods): A - Variety; C - Nitric norm; \*\*\*\*\*\* - Proved increase in the number of tillers, the number of spike stems / m<sup>2</sup> to N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences; # ## ### - Proven increase in the number of tillers and the number of spike stems / m<sup>2</sup> to control variety Progress at fertilization N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences.

The third (2007) harvest year is characterized by abnormalities in terms of climate, due to insufficient soil moisture necessary for the formation of a high yield in studied durum wheat varieties. The overall and productive tillering is higher in non - fertilized varieties and varieties of fertilization  $N_6$  unlike variants fertilized with high nitrogen fertilization rates -  $N_{12}$  and  $N_{18}$ . Due to the peculiarity of the year, high fertilizer rate proved unfavorable to the spike formation as well to the quality and quantity of yield. As a result of the strong drought, especially at the end of the vegetation, influenced the spike formation at high fertilization rates or their poor seed formation.

Using the results of the dispersion analysis of 2007 it was concluded that the genotype differences in the number of tillers in varieties Beloslava and Saturn 1 are well defined and statistically significant. In varieties Progress and Prosperity there are no proven differences in types of fertilization  $N_6$  regarding the number tillers / m<sup>2</sup>, and in variety Neptune 2 in maximum tested fertilizer rate  $N_{18}$ . As for the other important indicator number spike stems / m<sup>2</sup> were statistically significant only in the standard variety Progress at level  $N_6$ . In the other varieties at all levels of fertilization there are no proven differences.

Weather conditions of the year affect plant height indicator. Without application of nitrogen fertilization the height of studied durum wheat varieties vary during the years as follows: between 66.5 and 84.8 cm in the first (2005); between 72.3 and 87.1 cm in the second (2006) and between 38.1 and 47.8 cm in the third (2007) (Table. 3).

As expected, the lower height is the variety in options without application of nitrogen fertilization, during the first and third year variety Saturn 1 studied and during the second year - Vozhod. The highest height is the standard variety Progress for the first, second and third year of study. Average for all durum wheat varieties and levels of fertilization, the lowest seed harvest was reported in 2007 (47.6 cm), in 2005 - 82.6, and the highest in the second year with the highest grain yield (86.8 cm).

	Ν	-			~ .	
	norms	Progres	Neptun2	Beloslava	Saturn1	Vozhod
	kg/da					
	0	84.8	71.0	76.0	66.5	70.0
2005	6	90.2	84.3	78.3	78.2	73.8
	12	98.9	86.2	82.1	83.1	77.4
	18	101.4	89.1	84.2	87.5	89.3
	0	87.4	73.7	74.0	72.9	72.3
2006	6	90.5	85.1	87.2	87.6	86.9
2006	12	101.9	87.2	87.5	90.4	86.3
	18	102.5	92.0	88.1	94.0	88.0
	0	47.6	47.8	43.9	38.1	38.3
2007	6	64.5	54.5	47.5	53.7	43.2
2007	12	55.4	48.2	43.1	44.1	41.5
	18	60.5	51.9	46.7	40.1	42.2
	0	73.7	64.2 ns	64.6 ns	59.2 ns	60.2 ns
Average	6	81.7***	74.6***	71.0***	73.2***	68.0***
for	12	84.6***	73.9***	70.9***	72.5***	69.2***
period	18	88.1***	77.7***	73.0***	73.9***	73.2***
		A B				
	5%	0,4 0,3				
GD	1%	4,8 4,3				
	0.1%	6,3 5,7				

Table 3. Height of crop of durum wheat varieties

Factors (according to Material and Methods): A - Variety; C - Nitric norm; \*\*\*\*\*\* - Proven increase in height to  $N_0$  for levels 5, 1 and 0.1%; NS - unproven differences from the control variety Progress in fertilization  $N_0$  - NS - unproven differences.

In the present study the performance of the structural elements of the spike - the spike length, number of grains in a spike and weight of the grains in a spike were examined in order to track their changes by year and nitrogen rates.

Nitrogen fertilization affects significantly average data of spike structural elements o in the durum wheat for the period, as changing them positively (Table 4).

The longest average spike for the two years (2005-2006) of study is formed in Saturn 1 and the shortest in variety Vozhod at fertilizer tested at the maximum rate. With regard to the average mass of grain in a spike it is higher in variety Neptune 2 due to the higher mass of the grain. By number of grains in spike variety Neptune 2 is outweighs the other varieties having on average for the period 58 grains in a spike at fertilization  $N_{18}$ .

	Ν			Variety			GD
	norms kg/da	Progres	Neptun 2	Beloslava	Saturn1	Vozhod	A B
Length of class, cm	0 6 12 18	6.4 6.5 ns 7.2*** 7.5***	6.7 ns 7.1** 7.4*** 7.7***	6.1 ns 7.0*** 7.4*** 7 6***	5.9 ns 6.4** 7.0*** 7 8***	6.0 ns 6.6*** 6.8*** 7.0***	5% 0.4 0.3 1% 0.5 0.4 0.1% 0.6 0.6
Number of Grains in class	0 6 12 18	32 35*** 36*** 40***	36### 46*** 55*** 58***	34### 39*** 43*** 47***	33 ns 40*** 49*** 56***	39### 39 ns 39 ns 45***	5% 1.2 1.0 1% 1.6 1.4 0.1% 2.0 1.8
Weight of the grains in 1 class, g	0 6 12 18	1.8 2.2*** 2.3*** 2.5***	1.8 ns 2.2*** 2.6*** 2.9***	1.5 ns 1.9*** 2.0*** 2.3***	1.6 ns 1.8* 2.1*** 2.4***	1.7 ns 2.0** 2.1*** 2.4***	5% 0.3 0.2 1% 0.4 0.3 0.1% 0.5 0.4

Table 4. Struktural elements of spike average for 2 years (2005-2006)

Factors (according to Material and Methods): A - Variety; C - Nitric norm; \*, \*\*, \*\*\* - Proven increase in the length of the spike, cm, the number of seeds in the spike and the mass of the grains in spike 1, g N<sub>0</sub> relative levels of 5, 1 and 0.1%; NS - unproven differences # ## ### - Proven increase in the length of spike, cm, number of seeds in the spike and mass of seeds in one spike, g to control variety Progress at fertilization N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences.

	Ν		GD				
	norms kg/da	Progres	Neptun 2	Beloslava	Saturn 1	Vozhod	A B
	0	5.3	5.8 ##	4.5###	5.4 ns	4.9#	5% 0.4 0.3
Length of	6	7.0***	6.5***	6.8***	6.4***	5.7***	1% 0.5 0.5
class, cm	12	7.4***	6.6***	6.9***	7.3***	6.8***	
	18	7.5***	7.4***	7.4***	7.5***	6.9***	0.1% 0.7 0.6
Number	0	17	20###	21###	16 ns	16 ns	5% 1.0 0.9
of	6	30***	32***	34***	26***	21***	
Grains in	12	29***	19 ns	16 ns	13 ns	12 ns	1% 1.3 1.2 0.1% 1.7 1.5
class	18	28***	23***	18 ns	10 ns	20***	0.1% 1.7 1.3
Weight of	0	0.9	0.6 ns	0.8 ns	0.5 ns	0.7 ns	5% 0.3 0.3
the grains	6	1.5***	1.1***	1.1*	0.9**	0.8 ns	1% 0.4 0.4
in 1 class,	12	1.2***	0.8 ns	0.4**	0.4**	0.4 ns	
g	18	1.2***	0.8 ns	0.5***	0.4**	0.7 ns	0.1% 0.6 0.5

Table 5. Variance analysis of the structural elements of spike during 2007

Factors (according to Material and Methods): A - Variety; C - Nitric norm; \*, \*\*, \*\*\* - Proven increase in the length of the spike, cm, the number of grains in the spike and the mass of the grains in the spike 1, g to N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences; ###### - Proven increase in the length of spike, cm, number of seeds in the spike and the mass of grains in one spike, g to control variety Progress at fertilization with N<sub>0</sub> for levels 5, 1 and 0.1%; NS - unproven differences

With regard to the indicator number of grains in the third year spike again shows variations in the results, unlike the previous two years, which is due to changes in climatic terms, so we consider it separately.

The number of seeds in the spike during the third harvest year increases in variants of fertilization  $N_6$  while, increasing the fertilizer rate, it decreases. Thus, this increase compared to not-fertilized variations in percentage terms by variety is as follows: Progress variety - with 76.5%; Neptune variety 2-60%, Beloslava - 61.9%; Saturn 1 - 62.5% and in variety Vozhod - 31.3%.

Fertilizing with  $N_6$  also proved to be the most positive tasted fertilizer rate regarding the mass index of a spike in 2007 as the standard variety Progress shows the highest values of 1.5 g, while the lowest were reported in variety Vozhod - 0.8 g.

## Conclusions

1. Weather conditions of the year affect plant height indicator. During the three years of study, highest plant height has the standard variety Progress. Average for all durum wheat varieties and levels of fertilization, the lowest seed harvest was reported in 2007 due to droughts, while the highest is in the second year, with highest grain yield.

2. Increasing nitrogen fertilization positively influences productive tillering almost at all investigated durum wheat varieties increasing the percentage of productive tillers in both years. The third (2007) harvest year is characterized by abnormalities in terms of climate, due to insufficient soil moisture necessary for the formation of a high yield in durum wheat varieties studied. The overall productive tillering is higher at not fertilized variants and variants of fertilization with N<sub>6</sub> unlike variants fertilized with high nitrogen fertilization rates - N<sub>12</sub> and N<sub>18</sub>. Due to the peculiar conditions of the year, high fertilization rates turned out to be unfavorable for yield and yield components.

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## RESULTS OF INVESTIGATIONS OF INTRODUCED AND DOMESTIC VARIETIES AND LINES OF VIRGINIA TOBACCO IN THE REGION OF PRILEP IN 2012 AND 2013

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#### Abstract

Investigations were carried out in 2012 and 2013 on the Experimental field of the Scientific Tobacco Institute - Prilep. They included four introduced fertile varieties and three domestic promising hybrid lines in CMS form created in the Institute, with the standard American variety K-394 used as a check. The trial was set up in randomized blocks with 4 replications. The highest average yield per stalk (143.8 g) and per hectare (3197 kg) was achieved in line V-79/09 CMS F<sub>1</sub>. The lowest yield per stalk (84.2 g) and hectare (1871 kg) was recorded in the variety Delcrest. The highest quality of tobacco raw, expressed through the average price, was obtained in the line V-79/09 CMS  $F_1$  (70.70 denars/kg) and lowest in the variety Delcrest (56.20 denars/kg). Also, the gross income was the highest in line V-79/09 CMS  $F_1$  (226.158 denars/ha) and the lowest in the variety Delcrest (104.990 denars/kg). All varieties and lines except for the variety Delcrest showed statistically significant differences at 1% level compared to the check in both years of investigation for the traits yield per stalk and hectare and gross income. Statistical difference for the average price was observed only in line V-79/09 CMS F<sub>1</sub>, which appeared to be dominant over the other varieties and lines with regard to these traits. Key words: tobacco, Virginia, yield, income.

#### Introduction

The raw material of Virginia tobacco is inevitable component in the manufacturing of blended cigarettes. Good variety, strictly controlled production and adequate drying conditions are the requirements that should be met to get a good yield and quality of this type of tobacco. According to Beljo (1996) and Uzunoski (1985), Virginia belongs to the group of large-leaf, high tobaccos (200 cm). Its growth and development requires precisely determined agro-ecological conditions and cultural practices, and specific way of curing (flue-cured). This tobacco was grown in certain regions of the Republic of Macedonia until 2002, after which the production has stopped and now Macedonian cigarette factories is fully dependent on imports of this type. To overcome this situation it is necessary to restart the production according to world standards and to create raw material similar or equal to the imported one. In production of Virginia tobacco variety is a very important chain which has a large impact on yield and quality of the raw material. In recent years, many

hybrid varieties (lines) in CMS form have been created in Tobacco Institute - Prilep. The results of comparative trials show that the introduced varieties might be also interesting for tobacco growers.

## Material and methods

Seven varieties, four of them in fertile form, were used as material for work: K 394 (USA) - used as a check, Delcrest (Canada), TL 33 (Zimbabwe), V-519 (Bulgaria) and the malesterile hybrid lines V-11/11 CMS F<sub>1</sub>, V-12/11 CMS F<sub>1</sub> and V-79/09 CMS F<sub>1</sub> created in Tobacco Institute-Prilep. The trial was set up on colluvial soil in randomized block system with four replications at 90×50cm planting density, on previously prepared site (one autumn and two spring ploughings, fertilization with 300 kg/ha NPK 8:22:20 and application of herbicide). Before the second hoeing, manual feeding of stalks was made with 3 g/stalk 26% KAN. The plants were also treated with chemicals for their protection from pests and diseases. Harvested tobacco was stringed, yellowed and then dried. Qualitative assessment of cured tobacco was made according to the Rules for assessment of quality of dry Virginia tobacco (Rules on criteria for qualitative and quantitative assessment of raw tobacco leaves "Official Gazette" of R. Macedonia, No. 16/2007, amended and supplemented No. 144/2010 and No. 20/2011). Corrected yield per stalk and hectare was calculated by the method of Rimker and the gross income (denars/ha) by multiplying yield per hectare and the average price of 1 kg raw tobacco. The results were statistically processed using the analysis of variance and LSD test.

## **Results and discussion**

Tobacco yield mainly depends on leaves, their number and size. This trait is genetically controlled in each variety (genotype), but it is also highly affected by the environmental conditions during the growing season. Dražic (1986) explains that the yield is directly influenced by the genotype and environment. Carriers of the yield and quality of raw material in Virginia tobacco are the middle belt leaves, their size and color obtained after curing. According to Beljo (1996), tobacco yield and quality also depend on cultural practices applied during the growing season (fertilization, irrigation, harvesting time, yellowing, curing etc.). Results of our investigation of the above traits are presented in Tables 1,2,3 and 4.

## Yield per stalk

Data on the obtained yield per stalk from the investigated varieties and lines are presented in Table 1.

Variety	Year		Average	Dif	ference	D 1
5		g/stalk	2012/2013	Absolute	Relative	Rank
K-394 Ø	2012	96.9	99.7	/	100.00	6
K-394 Ø	2013	102.5	99.1	/	100.00	0
Delcrest	2012	80.5	84.2	-15.5	84.37	7
Delciest	2013	87.9	04.2	-13.3	04.37	/
T.L.33	2012	$119.5^{++}$	127.5	+27.8	127.88	4
1.L.33	2013	135.5++	127.5	+27.0	127.00	4
V-519	2012	$111.1^{++}$	123.2	+23.5	123.57	5
V-319	2013	135.3++	125.2	+23.3	125.57	5
V-11/11	2012	$118.0^{++}$	127.9	+28.2	128.28	3
CMS F <sub>1</sub>	2013	137.8++	127.9	+20.2	120.20	3
V-12/11	2012	119.3++	128.5	+28.8	128.88	2
CMS F <sub>1</sub>	2013	137.6++	126.5	+20.0	120.00	Z
V-79/09	2012	131.3++	143.8	+44.1	144.23	1
CMS F <sub>1</sub>	2013	$156.4^{++}$	143.0	<b>+++++++++++++</b>	144.23	1
20	012 5%	$b^+ = 5.63 \text{ g/s}$	talk	2013	$5\%^+ = 6.22$ g/stalk	
	1%	$^{++} = 7.72 \text{ g/s}$	talk		1% <sup>++</sup> = 8.53 g/stalk	-

Table 1. Corrected yield per stalk (g/stalk)

According to above data, the highest average yield per stalk was recorded in line V-79/09 CMS  $F_1$  (143.8 g) and the lowest in the variety Delcrest (84.2 g). The check variety N 394 achieved an average yield of 99.7 g. In other varieties in the trial, the average yield per stalk ranged from 123.2 g in variety V- 519 to 128.5 g in line V-12/11 CMS  $F_1$ . It can be also noted that higher yields were achieved in the more humid conditions of 2013. In both years of investigation, all varieties and lines, except for the variety Delcrest, achieved statistical significance at 1% level compared to the check. Drazic et al. (2012) made investigations with 13 domestic and introduced varieties and lines in 2011 at various locations in Serbia and found that the yield per stalk ranged 105 - 257 g/stalk in Nova Pazova and 101 - 298 g/stalk in Storchevo. Risteski (1999) reported that the stalks of MV-1, grown at nutritional area of 0.25 m<sup>2</sup> achieved an average yield of 79.7 g/stalk and those grown at 1 m<sup>2</sup> achieved 198.2 g.

## Yield per hectare

Data on the obtained yield per hectare are presented in Table 2.

Variety	Year	kg/ha	Average	Diff	ference	Rank
		Kg/IId	2012/2013	Absolute	Relative	Kalik
K-394 Ø	2012	2153	2215	/	100.00	6
K-394 Ø	2013	2278	2213	/	100.00	0
Delcrest	2012	1790	1871	-344	84.46	7
Delciest	2013	1953	10/1	-344	04.40	/
T.L.33	2012	2655++	2833	+618	127.90	4
1.L.33	2013	3011++	2033	+018	127.90	4
V-519	2012	$2470^{++}$	2738	+523	123.61	5
V-319	2013	3007++	2738	+325	125.01	5
V-11/11	2012	2627++	2843	629	128.35	3
CMS F <sub>1</sub>	2013	3063++	2845	+628	128.55	3
V-12/11	2012	2653++	2955	. (10	100.00	2
CMS F <sub>1</sub>	2013	3058++	2855	+640	128.89	2
V-79/09	2012	2919++	3197	+982	144.33	1
CMS F <sub>1</sub>	2013	3476++	5197	+902	144.33	1
2		$^{+} = 125.18$ kg		2013	5% <sup>+</sup> = 136.74 kg	
	1%	$^{++} = 171.68$ k	kg/ha		1% <sup>++</sup> = 187.52 kg	g/ha

Table 2. Corrected yield per hectare (kg/ha)

Data in this table reveal a very close relationship between yield per stalk and per hectare. The highest average yield was obtained in line V-79/09 CMS F1 (3197 kg/ha) and the lowest in variety Delcrest (1871 kg/ha). The yield of the check variety K-394 was 2215 kg. In other varieties investigated, the yield ranged from 2738 kg/ha in variety V- 519 to 2855 kg/ha in line V-12/11 CMS  $F_1$ . In both years of investigation, all varieties and lines, except for the variety Delcrest, achieved statistical significance at 1% level compared to the check.

Devcic et al. (1982) reported that by application of good cultural practices, Croatian hybrid varieties H-30, H-31 and H-32 can reach over 2000 kg/ha. Hawks (1978) presented the yields of Virginia tobacco in the United States in different periods of time. According to the data, the average yield in the period 1934-1938 was only 959 kg/ha and in 1964-1967 it increased to 2224 kg/ha. Risteski et al. (2012) reported that higher yields were achieved in Virginia tobacco lines created in the Scientific Tobacco Institute – Prilep, reaching up to 3549 kg/ha in line V-53 CMS F1.

#### - Average price

The quality of tobacco raw expressed in monetary value gives the average price per kg. It is closely related to the variety, proper and timely applied cultural practices, yellowing, curing, etc. Data on average price in the varieties investigated are presented in Table 3.

Variety	Year	denars/kg	Average	Dif	ference	Rank
		uenars/kg	2012/2013	Absolute	Relative	Nalik
К-394 Ø	2012	60.29	61.27	/	100.00	6
K-394 Ø	2013	62.25	01.27	/	100.00	0
Delcrest	2012	58.79	56.20	-5.07	91.72	7
Deletest	2013	53.61	50.20	-5.07	91.72	1
T.L.33	2012	64.86	64.31	12.04	104.06	4
1.L.33	2013	63.67	04.31	+3.04	104.96	4
V-519	2012	63.73	63.73	+2.46	104.01	5
V-319	2013	63.72	03.75	+2.40	104.01	5
V-11/11	2012	66.57	67.79	+6.52	110.64	2
CMS F <sub>1</sub>	2013	69.02++	07.79	+0.32	110.04	Z
V-12/11	2012	63.83	65.95	+4.68	107.64	3
CMS F <sub>1</sub>	2013	$68.08^{++}$	03.95	+4.08	107.04	5
V-79/09	2012	70.71++	70 70	.0.12	115 20	1
CMS F <sub>1</sub>	2013	70.69++	70.70	+9.43	115.39	1
2	012 5%	$^{+} = 6.81 \text{ den}$	ars/ kg		$5\%^+ = 3.67$ denars/k	
	1%	$^{++}=9.34$ den	ars/kg		$1\%^{++} = 5.03$ denars/k	(g

Table 3. Average price (denars/ha)

According to the above data, the average price ranged from 56.20 denars/kg in variety Delcrest to 70.70 denars/kg in line V-79/09 CMS F1, compared to the check K-394 with 61.27 denars/kg. In the other varieties and lines, the average price ranged from 63.73 denars/kg in variety V-519 to 67.79 denars/kg in line V-11/11 CMS F1. Statistically significant differences at 1% level in both years of investigation were observed only in line V-79/09 CMS F1. Statistical significance at 1% was also achieved in lines V-11/11 CMS F1 and V-12/11 CMS F1 but only in 2013. Kochovska et al. (2004) in investigations with six varieties and lines in the region of Prilep during 2002 and 2003 reported that the highest average price was achieved in line V-53 (65.83 denars/kg) and the lowest in line V-69 (57.08 denars/kg).

## Gross income

The data presented in Table 4 determine the average price per 1 kg of raw tobacco and yield per hectare achieved by the investigated varieties and lines.

Variety	Years	denars/ha	Average	Dif	ference	Rank
		denars/na	2012/2013	Absolute	Relative	Kank
К-394 Ø	2012	119 538	130 646	/	100.00	6
K-394 Ø	2013	141 755	150 040	/	100.00	0
Delcrest	2012	105 225	104 994	-25 652	80.36	7
Delciest	2013	104 763	104 994	-23 032	80.30	/
T.L.33	2012	172 390++	182 085	+51 439	139.37	4
1.L.33	2013	$191\ 780^{++}$	162 065	+31 439	139.37	4
V-519	2012	157 555++	174 750	+44 104	133.76	5
V-319	2013	191 946++	174 750	+44 104	155.70	5
V-11/11	2012	174 618++	192 993	+62 347	147.72	2
CMS F <sub>1</sub>	2013	211 368++	192 993	+02 547	147.72	2
V-12/11	2012	169 530++	188 759	+58 113	144.48	3
CMS F <sub>1</sub>	2013	207 988++	100 759	+56 115	144.40	5
V-79/09	2012	206 446++	226 158	+95 512	173.10	1
CMS F <sub>1</sub>	2013	245 870++	220 138	+75 512		
20	012 5%	$^{+} = 22\ 244\ de$	nars./ ha	2013	5% <sup>+</sup> = 13 923 d	
	1% ++	$= 30\ 506\ den$	ars./ ha		1% <sup>++</sup> = 19 095 d	enars./ ha

Table 4. Gross income (denars/ha)

Compared to the check variety K-394 (130 646 denars/ha), the highest gross income was recorded in line V-79/09 CMS F1 (226 158 denars/ha) and the lowest in variety Delcrest (104.994 denars/ha). In other varieties and lines, gross income ranged from 174 750 denars/ha in V-519 to 192 993 denars/ha in line V-11/11 CMS F1. Except for Delcrest, all other varieties and lines in both years of investigation achieved statistical significance at 1% level compared to the check.

## Conclusions

- The highest average yield per hectare and stalk was recorded in line V-79/09 CMS F1 (143.8 g/stalk and 3196 kg/ha) and the lowest in variety Delcrest (84.2 g/stalk and 1871 kg/ha).

- The highest average price of raw tobacco was achieved in line V-79/09 CMS F1 (70.70 denars/kg) and the lowest in variety Delcrest (56.20 denars/kg).

- The highest gross income was obtained in line V-79/09 CMS F1 (226 158 denars/ha), and the lowest in variety Delcrest (104 994 denars/ha).
- The above data lead to a conclusion that genotype has high influence on the investigated traits and therefore more attention should be paid to the selection of varieties for mass production.

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# EFFECT OF FOLIAR FERTILIZATION ON CORN GRAIN QUALITY

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#### Abstract

The presented data were collected at the experimental fields of the Institute of Agriculture, in Skopje, R. of Macedonia, during 2008 and 2009. The experiment with corn plant was conducted to study the effect of fertilizer Megagreen on some elements in corn grain (hybrid ZP 677, FAO 600). Megagreen is ecological foliar fertilizer, made of calcite, micronized by a new tribomechanical technology. Main components are: CaCO<sub>3</sub> 82.3 %; SiO<sub>2</sub> 5.56 %; MgO 3.02 %; CaO 41.7%; Fe 8783 mg/kg; Mn 156 mg/kg; Se 0.24 mg/kg. Foliar treatments consisted of three levels of Megagreen with in a concentration of 0.3, 0.6 and 0.9 percent and control variant (without fertilizing). The experiment was carried out on two different experimental sites on alluvial soils. Soil samples (0-20 and 20-40cm) were collected to determinate the chemical properties of the experimental field. During the harvest, the samples of corn grain were taken for chemical analysis from each variant. The soil and plant analysis were carried out according officially adopted international methods. The statistical analyses showed significant positive effect of foliar applications of Megagreen in concentration of 0.9 % on the content of K, Mn and Zn in corn grain. The concentration of these elements in grain samples for the 2008 and 2009 growing seasons was as follows: K- 0.27 and 0.29%; Mn - 5 and 6 mg/kg and Zn - 22 and 26.33 mg/kg. The findings showed that effects of Megagreen on the P (0.18-0.20 %), Ca (0.13-0.15 %), Mg (0.07-0.08 %) and Fe (23.5-25.66 mg/kg) concentration in the grain were insignificant. Results showed the high relationship between some macro and microelements in corn grain. A significant positive correlation was found between concentration of Ca (like main fertilizer component) and Fe and Cu. Megagreen applications in the both years of our research showed significant negative correlations between Ca and N concentration. Key words: corn, grain foliar, fertilizer, macro, micro, element

## Introduction

Mineral fertilization is one of the most important yield and quality factors. Arable crops yields and their quality can be improved by adequate soil and crop management practices or through the process of plant breeding creation of new varieties resistant to unfavorable production conditions (Bukvić *et al.* 2003). Approximately 60% of the world arable land is considered to be difficult for plant production due to mineral stress caused by the

deficiency, unavailability, or toxicity of some essential nutritive elements (Foy, 1983). Besides the root nutrition from the soil, plants can absorb nutrients through the organs which are above ground (leaves and stem) (Anon, 1995). Foliar fertilization researches are lasting for 4-5 decades.

Mayer and Bohm are the first who treat the problem with practical application of foliar nutrition (cit. Khoury and Boutros, 1978). Increasing yield and quality are significant positive effects of foliar application (Anonymous, 2003). Ling and Silberbush (2002) during their research, noted that foliar fertilization gives better results than soil fertilization, in terms of the length of root, leafy area, fresh and dry weight of plants and content of chlorophyll, nitrogen, phosphorus and potassium in the leaves. Foliar application with zinc is stage of 3-4 leaves, have an positive effect on the increasing uptake on nitrogen and yield on corn grain (Grzebisz *et al.* 2008). Tejada and Gonzalez (2006) noted that corn fields foliar threated with "beet vinasse" (by-product; spin off from sugar industry), compared with un-foliar-threated corn fields, have increased content of microelements Fe, Mn, Cu, Zn and macroelements N and K in leaves, chlorophyll amount *a* and *b*, proteins in grain by 30%, numbers of grains per cob by 20% and increased corn yield by 13%. Main evidence for the positive impact of Megagreen are the numerous research programs carried out in different regions around the world: Europe, Asia, South America and Australia (www.ecoideas.ca).

The objective of this work is to determine the impact of applying foliar fertilizer Megagreen on grain quality.

### Material and methods

This research was conducted at the experimental fields of the Institute of Agriculture, in Skopje region, R. of Macedonia, during 2008 and 2009. The Skopje valley is in the continental-submediteran climatic region, where the influence of submediteran climate is weaker than the continental climate (Filipovski *at al.* 1996). The growing seasons 2008 and 2009 were favorable for corn growing (Tab.2).

The experimental design was a randomized complete block, with four variants and three replications. Maize, hybrid ZP 677 belongs to the group of late hybrid, FAO 600 (http://mrizp.rs/zp-677). Megagreen is ecological foliar fertilizer, made of calcite, micronized by a new tribomechanical technology. Main components of fertilizer are: CaCO<sub>3</sub> 82.3 %; SiO<sub>2</sub> 5.56 %; MgO 3.02 %; CaO 41.7%; Fe 8783 mg/kg; Mn 156 mg/kg; Se 0.24 mg/kg.

Foliar applications were made by back sprayer, with 4.8l solution for each variant. The foliar fertilizer was applied four times during the growing period, starting from the stage of 7-8 leaf (V7), in a intervals of 10-15 days.

Experimental variants of foliar treatments were as follows:

Variant 1 – control (without fertilizer);

Variant 2 – fertilizing with concentration of 0.3 %;

Variant 3 – fertilizing with concentration of 0.6 %;

Variant 4 – fertilizing with concentration of 0.9 %.

During the harvest, the samples of corn grain were taken for chemical analysis from each variant. The experiment was carried out on two different experimental sites on alluvial

soils. Soil samples (0-20 and 20-40cm depths) were collected to determinate the chemical properties of the experimental field (Tab.1).

The soil and plant analysis were carried out according officially adopted international methods: available N in soil samples was measured by method of Bremner (Dzamić *et al.*, 1996); available forms of  $P_2O_5 \mu$  K<sub>2</sub>O in soil samples was measured by Al method (Manojlović *et al.*, 1969); the total amount of N in corn grain was measured by method of Kjeldahl (Sarić *at al.*, 1986); total amount of K in grain samples was determined by phlamefotometar (Sarić *at al.*, 1986); with AAS (type Varian Spectra AA800) was determined mobile fraction of Ca and Mg in soil after extraction by ammonium acetate solution, and Mn, Fe, Cu and Zn with DTPA method (Jones, 2001); the total amount of P, Ca, Mg, Mn, Fe, Cu and Zn in corn grain was measured by ISP-AES technique after their microwave digestion using concentrated HNO<sub>3</sub> + H<sub>2</sub>O<sub>2</sub>. Plant and soil analysis was made in the laboratorys of Institute of Agriculture and Institute of Chemistry, University "Sc.Cyril and Methodius University, Skopje.

Analysis of variance was performed using SPSS 14.0 software (SPSS Inc., 2005), and correlation was calculated at 0.05 level.

	Dept	CaC	p	Н	Hum		Availab g/100g				Availa			
Year	h	$O_3 \%$	H <sub>2</sub>	Κ	us %	N	P <sub>2</sub> O	K <sub>2</sub> O	Ca	Mg	ppn Zn	Fe	Mn	Cu
2008	0-20	13,97	8.0	7.0	1.58	11.	26.3	16.5	2218.	239.	2.6	9.58	34.4	3.5
2008	20-	15,85	8.0	7.0	1.16	2.8	13.3	14.9	2304.	218.	2.1	10.3	24.3	3.0
avera	0-40	14.91	8.0	7.0	1.37	7.0	19.8	15.7	2261.	228.	2.3	9.94	29.3	3.2
2009	0-20	12.74	8.0	7.2	1.85	10.	40.5	54.0	2428.	386.	10.	12.0	17.9	7.1
2009	20-	15.28	8.1	7.1	1.94	2.5	42.4	30.8	2516.	430.	6.6	12.1	18.9	7.0
avera	0-40	14.01	8.0	7.1	1.89	6.6	41.5	42.4	2472.	408.	8.4	12.0	18.4	7.0

Table 1. Some soil chemical characteristics

Table 2. Average monthly temperature (°C) and monthly rainfalls (mm)

			Mo	nth			A variance via setation	
Year	IV	V	VI	VII	VIII	IX	Average vegetation temperature °C	Average annual temperature °C
		Averag	e monthly	/ temperat	ure °C		temperature C	temperature C
2008	13.7	18.0	22.4	24.2	26.0	17.9	20.36	13.90
2009	13.4	18.0	21.0	24.1	23.8	19.3	19.93	13.00
<b>'</b> 91/05	12.2	17.6	22.3	24.5	24.2	18.9	19.95	13.48
		Ν	Ionthly ra	infalls mr	n		Vegetation amount of rainfalls mm	Annual amount of rainfalls mm
2008	18.7	40.7	46.9	57.8	24.8	78.5	438.5	267.4
2009	65.3	70.2	104.3	10.2	50.2	11.1	660.5	311.3
<sup>•</sup> 91/05	47.9	48.7	41.1	37.1	31.8	39.1	486.3	245.7

### **Results and discussion**

The basic precondition for obtaining high quality corn yields is provision of sufficient available quantities of necessary nutrients during the whole vegetation. All elements indispensable for normal life cycle are named as, necessary'' elements. This group includes: C, O, H, N, P, K, Ca, S, Mg, Fe, B, Mn, Zn, Cu, Mo and Co (Sarić *at al.*, 1991).

Significant amount of N during R2 stage is transferred from the leaves to the grain (Alley *et al.*, 2009). Nitrogen is accumulated in the grain and its absolute content increases till the end of the wax maturity. In our experiment the average of N content (Tab.3) in grain is the highest at the control variant, where it was noticed the highest average content from this element in the leaf at the end of the vegetation. The average content is 3.09 %, which confirms the statement of Barker *et al.* (2007) for the content of this element in seed which ranges from 2 to 7 %, with optimal amounts of >2 % dry weight.

From the data presented (Tab.3) it can be concluded that foliar fertilizing had positive influence on the P content in corn grain. From the research variants, the highest average content of P in grain compared to the control variant (0.191 % P) has the variant 4 with measured 0.204% and variant 3 (0.198 %), but without significant difference. Xingkai and Wang (2007) detected statistically significant 2.35 mg/g P of dry matter in fertilized corn variant and 2 mg/g P in control variant.

At the beginning of stage R3, the amount of K in plant tissues is reduced, at the end of the vegetation content of this element continues to grow until the physiological maturity of grain (Sprague, 1955). Variant 4 in both years of field experiment has the highest K content. Its average content is 0.28 % and its significantly different from the control variant where 0.24 % K was obtained. The obtained values are in the ranges given by Jevtić (1977). According to him, the content of potassium in corn grain is in the ranges of 0.28 to 0.34 % dry matters.

When fertilizing with 60 and 12 kg of N ha<sup>-1</sup> at corn, an increased Ca content in grain (0.36-0.38 g kg<sup>-1</sup>) compared with the control variant (0.35 g kg<sup>-1</sup>) was noticed (Hussaini *at al.*, 2008). From the results shown in Tab. 3, it can be concluded that at the end of the vegetation in R6 stage of experimental variant, the average content of Ca is highest at variant 4, with 0.0151 %. Although, foliar fertilization at variant 4 has an influence on the increasing content of this macro element compared to the control variant (0.0138 % Ca), this difference is not statistically significant at 0.05 level.

In the corn grain, Mg is always higher than Ca content (Jevtić, 1977), which can be confirmed by our result. With analyzing of the data from our research (Tab. 3) we determined that variant 4 has the highest average content of Mg, 0.0853 %. At variant 2, the lowest average Mg content in dry matter was determined, with 0.0764 %, lower than control variant, with average content of 0.0772 %. Variant 3 and 4 showed that foliar application of Mg has a influence on the increasing content of this element in corn grain, but this increasing is not significant. The investigations of the percentage of mineral elements in different corn organs, showed that corn grain contains 0.08% Mg in dry matter (Sprague, 1955).

Revealing the importance of microelements for plant life cycle, pave the way for explanation of whole set of processes responsible for the appearance of certain anatomical

and morphological deformations, which was believed to be due to a plant disease. Data collected for the content of microelements in corn grain are presented in Tab.4.

Out of the performed analyses, it can be concluded that the highest average content of Fe is 25.6667 mg/kg, at variants 2 and 4. Although, there is a difference between fertilizing variants and control variant in terms of the average Fe content in corn grain, there is no significant statistical difference. According to Hongxing and Yu-kui (2011) Fe content in corn grain is 28.89 mg/kg. The results from the Tab. 4, indicate that the control variant and variants 2 and 4 have the highest average content of Cu, of 1.5 mg/kg.

The average content of Mn in plants is in ranges of 50 to 250 ppm. The average quantity of Mn in wheat grain and barley are 34 and 17 ppm, respectively, while in corn grain the average quantity of Mn is 6 ppm (Vukadinović and Loncarić, 1997). According the presented results in Tab. 4, at all variants with different concentration of foliar fertilizer a higher average content of Mn is determined, compared to the control variant. The highest average content of 6 mg/kg Mn is determined at variant 4, and the lowest average content of 5 mg/kg Mn at control variant. Results in Tab. 4, shows that variant 4 has the highest content of Mn in grain in both experimental years. Although, all three fertilized variants have a higher average content of Mn, which shows that foliar fertilization has positive influence on Mn content, but only variant 4 is significantly different from the control variant. Hongxing and Yu-kui (2011) in their research concluded that Mn content in corn grain in that region is 4.40 mg/kg. From all microelements, Zn has the greatest impact on the corn growth. In a line with the average Zn content in the corn grain, treated variants 3 and 4 have the higher content, in compare to the control variant (21.1667 mg/kg Zn). The variant 4, which contains 24.1667 mg/kg Zn has the highest average content on this element in the grain, compared with other variants of the experiment (Tab. 4). The average Zn content in corn grain at 15% moisture, according to Benson (cit. Jevtić, 1977) is 17 mg/kg. The treated variants have the highest Zn content in grain, which shows that foliar fertilizing has a positive influence on Zn content in this part of plant. Nevertheless, only the differences between variant 4 and control variant are statistically significant at the 0.05 level of probability. Table 5 showed the high relationship between some macro and microelements in corn grain. A significant positive correlation was found between concentration of Ca (like main fertilizer component) and Fe and Cu. Megagreen applications in the both years of our research showed significant negative correlations between Ca and N concentration.

## Conclusion

The results of our investigations for the effects of foliar fertilizer Megagreen over macro and microelements, which was applicated in three different concentrations, showed the influence of: P (0.204 %), K (0.28 %), Ca (0.0151 %), Mg (0.0853 %), Fe (25.6667 mg/kg), Mn (6 mg/kg) and Zn (6 mg/kg) at variant 4, fertilized with concentration of 0.9%. Results showed the high relationship between some macro and microelements in corn grain. A significant positive correlation was found between concentration of Ca (like main fertilizer component) and Fe and Cu. Megagreen applications in the both years of our research showed significant negative correlations between Ca and N concentration.

1						
		average	0,0772 a	0,0764 a	0,0813 a	0,0853 a
	Mg	2009	0,0854	0,0709	0,0748	0,0826
		2008	0,0690	0,0818	0,0878	0,0880
		average	0,0138 ab 0,0690 0,0854	0,0149 a 0,0818 0,0709	0,0130 b 0,0878 0,0748	0,0151 a 0,0880 0,0826
	Ca	2009	0,0080	0,0079	0,0087	0,0083
		2008	0,0196	0,0219	0,0172	0,0218
		average	0,24 ð	0,23 <sup>b</sup>	0,27 ab	0,28 a
	K	2009	0,20	0,20	0,27	0,27
		2008	0,28	0,26	0,27	0,29
P, K, Ca and Mg in com grain (%)		average	0.191 a	0.182 a	0.198 a	0.204 a
Ag in cor	Р	2009	0.216	0.175	0.188	0.216
Ca and <b>N</b>		2008	0.165	0.190	0.208	0.191
		average	3,09 a	2,60 <sup>b</sup>	2,59 ð	2,71 ð
content c	N	2008 2009	3,44	3,03	3,09	3,07
verage (		2008	2,75	2,16	2,10	2,35
Table 3. Average content of N		v anant	1	2	3	4

\*  $2.35 + 3.07 + 2.71^{\circ} + 0.191 + 0.216 + 0.204^{\circ} + 0.29 + 0.27 + 3.300$  \* yallyes followed by the same letter are not statistically significant at the probability level p <0.05

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Table 4.

Wariant		Fe			Cu			Mh			Zn		
	2008	2009	average	2008	2009	average	2008	2009	average	2008	2009	average	_
1	26,0000	24,0000	25,0000 4	2	1	1,5 a	4	6	5,0 8	21,6667	20.6667	21,1667 ð	
2	31,3333	20,0000	25,6667 a	2	1	1,5 a	9	5	5,5 ab	24,6667	16.6667	20,6667 <sup>à</sup>	
3	25,3333	21,6667	23,5000 4	1	1	1,0 ð	6	5	5,5 ab	24,0000	20.0000	22,0000 ab	
4	28,6667	22,6667	25,6667 a	2	1	1,5 a	6	6	6,0 ª	26,3333	22.0000	24,1667 a	
* values follow	ved by the same	e letter are not	t statistically sign	ufficant at	the probab	the probability level p <0.05	0.05						

TAULE J. COLL	Laule 2. Collelauon maulx 10	JI HIACTO ALLO TIL	or macro and microelements in com grain	COIN grain					
Element	N	Ъ	K	Ca	Mg	Fe	Сц	Mh	Zn
z	1	.223	394(*)	830(**)	153	683(**)	522(**)	084	533(**)
Ъ		1	.289	398(*)	.870(**)	083	-321	.882(**)	.393(*)
K			1	.410(*)	.457(*)	.354	.505(**)	.337	.734(**)
Ca				1	.030	.849(**)	.832(**)	104	.601(**)
Mg					1	.279	.021	.944(**)	.696(**)
Fe						1	.759(**)	.195	.683(**)
Cu							1	055	.587(**)
Mn								1	.578(**)
Zn									1
** Correlation is	** Correlation is significant at the (	0.01 level (2-tailed)	(1)			*	* Comelation is significant at the 0.05 level (2-tailed	ficant at the 0.051	evel (2-tailed

# SECTION 9. FIELD CROP PRODUCTION

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## LOW TEMPERATURE EFFECT ON SEED GERMINATION OF SOME ORIENTAL TOBACCO VARIETIES INVESTIGATED IN LABORATORY CONDITIONS

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#### Abstract

Investigations on temperature effect on seed germination were conducted in 2012 in thermostats, in the accredited laboratory (LO1) for quality control of seed in Scientific Tobacco Institute – Prilep. Seed of four oriental tobacco varieties produced in 2010 was used as material for investigation (Prilep P -66-9/7, Yaka YV - 125/3, Djebel Dj-38 and Basmak MK – 1). The temperature effect on seed germination was studied in five variants (5°C, 10°C, 15°C, 20°C and 25°C). Our research included monitoring of the energy of germination and total germinability of tobacco seed. Total germinability at 10°C ranged from 3.50 % in variety Basmak MK - 1, 11.00 % in DjebelDj - 38, 16.00 % in Prilep P - 66-9/7, up to 37.00% in Yaka YV - 125/3. The highest tolerance to low temperatures was observed in the seed of Yaka YV 125/3 and the most susceptible was the seed of Basmak MK-1. The optimum temperature for seed germination of the varieties investigated in laboratory conditions was 20 – 25°C and the tolerance temperature ranged between 15° and 25°C. Temperatures below15°C and higher than 30°C donot give a real picture on tobacco seed quality and its germination ability.

**Keywords:** tobacco seed, germination energy, total germinability, Prilep P - 66-9/7, Yaka YV - 125/3, Djebel DJ - 38 and Basmak MK – 1.

#### Introduction

In order for the seed material to be marketed and distributed, it must be properly packaged, labeled and certified. Among the other factors, in the seed certificate is presented the seed germination energy and the total germinability. Germination ability of tobacco seed is important parameter, obtained by laboratory testing, using standard and accepted methods. Ambient temperature plays a very important role in estimation of germinability. Investigation on the temperature effect on tobacco seed germinability is expected to give practical knowledge that can be used to reduce the test time and to obtain higher quality tobacco seedlings, with possibility to shorten the period of production. Beside its scientific value, the results of investigation will be of interest especially to the accredited laboratories of this kind. It is also expected to obtain information which temperature has stimulating effect on seed germinability and which has an inhibitory effect. The aim of the investigation was to study the temperature effect on seed germinability in some oriental

tobacco varieties in order to determine the most favorable temperature regimes for germination in laboratory conditions.

### Materials and methods

The investigation was performed with seed of four oriental tobacco varieties created in Scientific Tobacco Institute Prilep in 2010: Prilep P-66-9/7; Yaka YV-125/3; Djebel Dj-38 and Basmak MK-1. Testing was conducted in the Accredited Laboratory for quality control of agricultural seed L01 during 2012, in separate thermostats for each variant.

The seeds were placed in Petri dishes, 4 x 100 seeds on filter paper with two replicates. The temperature impact on seed germinability was investigated in five variants, at 5° C, 10° C, 15° C, 20° C and 25° C.

The temperature of 25° C was taken as a check and it is considered an optimal temperature for tobacco seed germinability in lab conditions (Карајанков, 2007).

The prepared Petri dishes were placed in separate thermostats, depending on the treatment, and germination of seed was monitored on daily basis.

In our investigation, we monitored the germination energy and the total germination ability of tobacco seed, as the most important feature of the seed quality (Čirkovski, 1954).

Germination energy is speed with which the seeds germinate, expressed as a percentage of the seeds germinated within the first week of analysis with respect to total germination. Overall germinability is expressed as percentage of normally sprouted seeds with respect to total number of seeds placed to germinate, determined after the time allowed for the final assessment, which for tobacco seed is 16 days.

#### **Results and discussion**

Average results on the temperature impact during our investigation of seeds of four oriental tobacco varieties depending on the variant will be presented in Table 1.

Variety		Trea	tments (varia	nts)		Augrago
variety	5°C	$10^{0}$ C	15 <sup>°</sup> C	$20^{0}$ C	25 <sup>0</sup> C	Average
Prilep P-66-9/7	0	16.00	87.50	85.50	85.50	54.90
Yaka YV 125/3	1	37.00	90.75	94.75	88.75	62.45
DjebelDj-38	0	11.00	96.25	96.50	94.25	59.60
Basmak MK-1	0	3.50	96.50	97.50	92.00	57.90

Table 1. Seed germination energy, in %

The energy of germination of tobacco seeds tested at a temperature of 30° C is lower compared to the values obtained at 25° C. At this temperature, the germination energy ranged from 84.00% in variety Prilep 66-9/7 to 93.75% in variety Djebel-38. By increasing the temperature to 35° C, the germination energy is significantly reduced (Богданчески, 1972), especially in the variety BasmakMK-1 where it is only3.25%. This temperature also has a strong impact on germination energy of the seed of PrilepP-66-9/7, where it reaches 50.00%. The germination energy in the other two varieties is some what lower than in the variant at 30°C and ranges from 82.25% in variety Yaka YV-125/3 to88.00% in DjebelDj-

38. With temperature increase at 40° C, the germination energy continues to decrease, especially in the variety PrilepP-66-9/7, where it achieves only 7.50%, while in the other varieties it ranges between 2.25% in Basmak MK-1, 57.75% in Yaka YV-125/3 and 78.50% in DjebelDj-38.

In parallel with germination energy is the total germination of tobacco seed in the varieties investigated tobacco. Data on temperature impact on the total germination of tobacco seeds are presented in Table 2.

Verieter		Trea	tments (varia	nts)		A
Variety	5 <sup>0</sup> C	$10^{0}C$	15 <sup>0</sup> C	$20^{0}$ C	25 <sup>0</sup> C	Average
Prilep P-66-9/7	1	16.00	87.50	85.50	85.50	55.10
Yaka YV 125/3	2	37.00	90.75	94.75	90.25	62.95
DjebelDj-38	1	11.00	96.25	96.50	96.75	60.30
Basmak MK-1	0	3.50	96.50	97.50	99.50	59.40

Table 2. Total germination in %

Total germination of tobacco seed drops gradually when the temperature is lowered from 25° to 20° and 15°C. At 10° C the total germination is dramatically reduced, especially in Basmak MK-1 (3.50%) and DjebelDj-38 (11%), while in Prilep P-66-9/7 and Yaka YV 125/3 it reached 16% and 37%, respectively. The above analysis confirms the importance of temperature on tobacco seed germination in a laboratory environment (Преспаноски, 1997). The decrease of temperature at 25, 20 and 15°C greatly reduced the seed germination. Especially sensitive to the low temperatures was the variety Basmak MK-1.

Some resistance to low temperatures was recorded in the seed of Yaka YV-125/3. Generally, the low temperatures cause the seeds to weakly germinate and are dormant until the conditions are satisfactory (Ророvić, 1962). Temperatures below 10°C are not recommended (Младеновски, 1996). Temperatures over 5°C have an inhibitory effect on germination. According to Узуноски (1985), seed germination consists of three substages, of which the first stage, at low temperatures is prolonged, and the second, in which in-depth biochemical processes are occurring is stopped. There from comes the natural (genetic) resistance of varieties to low temperatures during the seed germination. Tobacco varieties, depending on their genetic constitution show different reactions to temperature in terms of the energy of germination. Thus, Basmak MK-1 and Djebel Dj-38 are more susceptible to low temperatures than the varieties Prilep P-66-9/7 and Yaka YV-125/3.

#### Conclusions

Based on the results obtained during investigation, the following statements can be made:

- Temperature is one of the major factors in investigation of tobacco seed germinability.
- At a temperature of 25°C, the seed of investigated tobacco varieties showed high values of germination energy and overall germinability, which points out to a supreme quality seed material.
- Temperatures below15°C reduce the germinability of tobacco seed.
- Thetemperature of 10°C significantly reduces the germination energy and overall germinability of seed. Especially susceptible to this temperature is the seed of the

BasmakMK-1 variety. Similarly, theseed of DjebelDj-38 also showed much lower values of germination energy and overall germinability, compared to varieties Prilep P-66-9/7 and Yaka YV125/3, which are quite resistant to this temperature.

- The temperature of 5°C showed inhibitory effect on the seed of BasmakMK-1 and DjebelDj-38. Much higher resistance to this temperature was recorded in the seed of Prilep P-66-9/7, and the highest resistance showed the seed of Yaka YV-125/3.
- The temperature impact on the energy of germination and total germination of tobacco seed also depends on variety of tobacco. The most resistant to low temperatures was the seed of variety Yaka YV-125/3, followed by that of the variety Prilep P-66-9/7. The most susceptible to low temperatures was the seed of the variety BasmakMK-1, followed by that of the varietyDjebelDj-38.

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## ACCUMULATION AND REUTILIZATION OF DRY MASS IN BULGARIAN BARLEY GENOTYPES

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### Abstract

The accumulation and reutilization of dry biomass in Bulgarian malting barley genotypes were studied. Four varieties such as Obzor (state standard), Emon, Kaskadior and Kramiand ten perspective breeding lines, were investigated in conditions of long-term fertilizer trial at the Department of Genetics and Breeding at the Agricultural University-Plovdiv.It was found that the biomass of barley varieties Obzor, Kaskadior, Emon, Kramiandthe studied lines at heading/anthesis was higher compared to the mass of vegetative plant parts at maturity, respectively in 2013. The results showed that the genotypes synthesized from 1,26 to 54,7 times more biomass before anthesis compared after this phase and remobilized and reutilized reserves accumulated before anthesis. The highest amount of translocated pre-anthesis biomass (DMT) was reported in line 29 (429,3 kg/da) and line 44 was characterized with the lowest amount (29,4 kg/da). The efficiency of dry mass translocation (DMTE) varied significantly and was from 2,8 % in line 44 to 36,8 % in line 29. The relative contribution of synthesized pre- and post-anthesis assimilates in grain yield varied widely, and some of the studied genotypes were characterized by the fact that during grain filling except assimilates formed by current photosynthesis, they reutilized assimilates accumulated before anthesis. CAVG values varied from 5,3% in line 44 to 114,4% in line 29. In variety Kaskadior, line 13,18 and 24 were calculated negative values for the three parameters-DMT, DMTE and CAVG. Key words: barley genotypes, reutilization, dry mass.

## Introduction

Current scientific and applied priority in the physiology of cereals is to obtain new knowledge about the response of grain yield to changes in the availability of assimilates during different periods of vegetation (Dordas, 2009). In anthesis the accumulated reserves are an important source of carbohydrates for grain filling when the current photosynthesis is inhibited by drought, high temperatures or diseases. Remobilization and reutilization of assimilates is derived from plant senescence, active in barley involves accumulation of carbohydrates from two sources: current assimilates, moved and sequential process that involves the translocation of stored reserves from stems, leaves and roots to the grain (Gan and Amasino, 1997; Zhang et al., 1998). Many factors during vegetation are influencing source-sink relationhip, the most important of which are genotype, fertilization,

temperature, rainfalls (Borraset al., 2004; Miralles and Slafer, 2007). According to Asseng and van Herwaarden (2003), the relative participation of remobilization in grain yield is mainly determined by source-sink relations during the grain filling, regardless of the environmental conditions of cultivation.

Barley is usually grown in non-irrigated fields where stressful conditions during grain filling can limit productivity and increase the dependence of spare vegetative assimilates in grain yield. During their life cycle plants survive periods when yield is mainly limited by the power and capabilities of the source for assimilates, the capacity of the sink to accept and use assimilates, or a combination of both (Borras et al., 2004). The better knowledge of source-sink relations in barley is important to establish the physiological and agrochemical characteristics suitable for genetic selection and modification of grain yield. The purpose of the present study was to establish the accumulation and the dynamics of dry mass in malting barley genotypes.

### **Material and Methods**

The study includes ten straight lines two-row winter barley, a selection of the Department of Genetics and Breeding at the Agricultural University -Plovdiv and varieties such as Obzor (state standard), Emon, Kaskadior and Krami(widely spread and included in production by the Executive Agency for Variety Testing, Field Inspection and Seed Control system of variety testing as a control). Genotypes were grown in competitive variety trials under irrigated conditions on yield plots of 10m<sup>2</sup> each in three replications. The study was conducted in the educational and experimental base of Agricultural University-Plovdiv during 2012-2013 year on Molic fluvisoil. More important agrochemical indicators of soil were: pH water=7.2; mineral nitrogen 39.2 mg Nkg; available phosphorus 22.8 mg  $P_2O_5/100$  g and available potassium 50mg  $K_2O/100$ g soil. A standard agrotechnics was applied in the region of South Bulgaria. Sowing fertilization with 60 kg N/ha was provided. Climatic conditions during barley vegetatiton for the investigated period were considered to be favourable for the air temperature and the sum of rainfalls. In heading/anthesis of barley the aboveground plant parts were analyzed. The samples were divided into leaves, stems and growing spikes and at maturity the samples were separated into grains and leaves, stems and chaffs. They were weighed and dried to constant weight at 60 °C. The parameters referring to dry mass accumulation and reutilization within barley genotypes were calculated as follows: dry mass accumulation in anthesis and maturity (kg/da); dry mass translocation (DTM) = dry mass at anthesis-dry mass of straw at maturity without grain(kg/da); dry mass translocation efficiency (%) = (dry mass translocation/dry mass at anthesis) x 100; contribution of pre-anthesis assimilates to the grain (%)  $CAVG = (dry mass translocation/grain yield) \times 100; harvest$ index (HI) = grain yield/total aboveground biomass at maturity. Other researchers were used the same approach (Przulj and Momcilovic, 2001; Abeledo et al., 2008). Analysis of variance (ANOVA) is attached for statistical processing of the results and statistical significance was determined by Duncan's multiple range test. Only differences in the level of significance  $\alpha = 0.95$  are accepted to be proven.

#### **Results and discussion**

The studied barley varieties and lines differed in accumulation of pre-anthesis biomass (Table 1). Lines 17 and 33 were distinguished with the highest values for this parameter, respectively 1266 and 1288 kg/da. Variety Kaskadior and line 18 accumulated the lowest amount of dry mass in anthesis. The mass of the vegetative plant parts (straw+chaff) at maturity differed significantly in the studied genotypes. Line 17 accumulated the highest content of dry mass in anthesis(1212 kg/da), with 249 kg/da more than variety Obzor (state standard). Cultivar Kaskadior, Emon and line 29 were characterized with the lowest values for this parameter. After anthesis the studied genotypes synthesized average net dry mass from 396,7 kg/da butthere were no proven mathematical differences between the genotypes. Line 29 was distinguished with no accumulation of net dry mass after anthesis.

The relative amount of accumulated dry biomass before and after anthesis was represented by the ratio of dry mass in anthesis to dry mass synthesized after anthesis (Table 1) and as a percentage of the two parts of the dry mass at maturity (Fig. 1). The results showed that the genotypes synthesized from 1,26 to 54,7 times more biomass before anthesis compared after this phase. The accumulation of biomass after anthesis was highest in line 24 and lowest in variety Krami-79,5% and 1,8% of the total biomass at maturity, respectively (Fig. 1). The selected malting barley genotypes in the Department of Genetics and Breeding in AU-Plovdiv accumulated more than 70% of the total biomass at maturity phase to anthesis. The obtained values for the ratio pre-anthesis dry mass / post-anthesis dry mass were higher than the cited in the literature for 20 genotypes of spring barley in which high values were established (more than 600 kg/da) of biomass accumulation after anthesis and variety differences (Przulj and Momcilovic, 2001).

Genotype	DM anthesis	DM straw+chaff	NetDM after	DM anthesis/DM
	(kg/da)	(kg/da)	anthesis	after anthesis
			(kg/da)	
Obzor	1188 ab	963cde	302	3,93
Kaskadior	733 e	747 f	484	1,51
Emon	1090 b	795ef	134	8,13
Krami	1148 ab	812ef	21	54,67
5	1152 ab	863cdef	227	5,07
13	1156 ab	1173 ab	599	1,93
16	1079 b	1004bcd	422	2,56
17	1266 a	1212 a	534	2,37
18	722 e	855cdef	527	1,37
24	861 de	1019bcd	685	1,26
29	1168 ab	738 f	-	-
31	912 cd	837def	431	2,12
33	1288 a	976cde	268	4,81
44	1054bc	1024bc	524	2,01

Table 1. Dry mass at heading/flowering and maturity (without grain), net mass after flowering, and ratio of pre- to post anthesis accumulated mass

Genotype	Grain yield (kg/da)	DMT (kg/da)	DMTE (%)	CAVG (%)
Obzor	527 abc	225,1	18,9	42,7
Kaskadior	470 bcd	-14,0	-1,9	-3,0
Emon	428 cde	294,8	27,0	68,8
Krami	357 e	335,8	29,3	94,2
5	517 abc	289,3	25,1	56,0
13	582 a	-17,2	-1,5	-3,0
16	497 abc	74,7	6,9	15,0
17	588 a	53,5	4,2	9,1
18	395 de	-132,7	-18,4	-33,6
24	527 abc	-158,0	-18,4	-30,0
29	375 de	429,3	36,8	114,4
31	507 abc	75,6	8,3	14,9
33	580 a	312,0	24,2	53,8
44	553 ab	29,4	2,8	5,3

Table 2. Grain yield, dry mass translocation (DMT), dry mass translocation efficiency (DMTE), and contribution of pre-anthesis assimilates in the grain (CAVG)

The grain yield in barley genotypes differed significantly- from 588 kg/da in line 17 to 357 kg/da in variety Krami (Table 2).Biomass in anthesis in all studied varieties and lines was higher compared to the mass of vegetative plant parts at maturity (straw+chaff) or genotypes reduced the weight of leaves+stems after anthesis, which means that they remobilized and used reserves accumulated before anthesis. On this basis the values of DMTE and CAVG were calculated.

The distribution of biomass was determined by the number and activity of the source and the number of grains was closely related with the presence of assimilates in anthesis (Wardlaw, 1990). Proper selection of varieties in addition to the cultivation of crops could increase the efficiency of reutilization of dry mass (Cox et al., 1985). The amount of translocated pre-anthesis biomass DMT was in the range from 29,4 kg/da to 429,3 kg/da in the studied genotypes, except from variety Kaskadior, line 13,18 and 24, which were characterized with negative values of DMT (Table 2). The efficiency of dry mass translocation (DMTE) is a parameter indicating the percentage of translocated biomass. Its values were in the range from 2,8% in line 44 to 36,8 % in line 29, indicating that the varieties and lines differed in pre- and post-anthesisphotosynthesis in grain yield. Variety Kaskadior, line 13,18 and 24 were distinguished with negative values of DMTE. Similar values to those found in this study were cited by other authors for winter wheat (Papakosta&Gagianas, 1991), durum wheat (Dordas, 2009), barley (Przulj&Momcilovic, 2001a). The participation of pre-anthesis assimilates in grain might be crusial for maintaining grain yield under unfavourable weather conditions, reduced photosynthesis, water availability and utilization of mineral elements (Arduini et al., 2006). It is known that winter wheat transfers a high percentage (6-73%) of pre-anthesis assimilates reserves into the grain (Papakosta&Gagianas, 1991) and for durum wheat the percentage may be in the range from 71.5-90% (Dordas, 2009). The relative contribution of synthesized pre- and post-anthesis assimilates in grain yield differed significantly, and some of the studied genotypes were characterized by the fact that during the grain filling except from ssimilates formed by current photosynthesis, they reutilized assimilates accumulated before anthesis. CAVG values varied from 5,3% (line 44) to 114,4% (line 29). In variety Kaskadior, line 13,18 and 24 were calculated negative CAVG values.

IIIdex				
Genotype	Mass of spike anthesis % of mass in anthesis		HI	
	(kg/da)			
Obzor	256def	100	0,354 f	
Kaskadior	147 h	61,7	0,386 c	
Emon	183gh	91,8	0,350 b	
Krami	275cde	96,6	0,305 k	
5	290cde	97,0	0,375 de	
13	396 b	97,3	0,331 i	
16	328 c	90,8	0,331 i	
17	515 a	106,6	0,327 i	
18	199fgh	60,8	0,316 j	
24	296 cd	72,5	0,341 h	
29	305 cd	98,3	0,338 a	
31	227efg	76,8	0,377 d	
33	309 cd	108,4	0,373 e	
44	248 def	88,7	0,351 g	

Table 3. Dry mass of spike in anthesis, partof aboveground biomass in anthesis and yield harvest index

The biomass of the spike in anthesis and the amount of nitrogen in it could be seen as a consequence of two processes: accumulation of biomass and nitrogen in aboveground plant parts in pre-anthesis period and their distribution between the vegetative plant parts and the spike (Demotes-Mainard&Jeuffroy, 2001). The mass of the spike varied widely in studied barley genotypes (Table 3). Line 17 was characterized with highest value of this parameter (515 kg/da) and the lowest value was calculated in cultivar Kaskadior (147 kg/da). The highest harvest index was established in variety Kaskadior (0,386) and the lowest-in variety Krami (0,305).

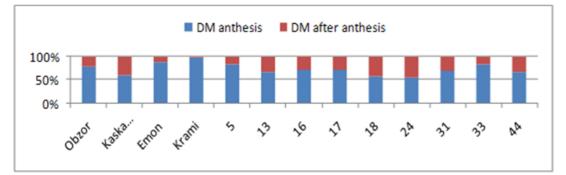


Fig. 1. Percent of accumulated pre- and post-anthesis mass from total aboveground mass at maturity.

## Conclusions

The biomass in heading/anthesis in varieties Obzor, Kaskadior, Krami, Emon and the ten new studied breeding lines was higher compared to the mass of vegetative plant parts at maturity. The studied genotypes synthesized from 1,3 to 54,7 more biomass before anthesis than after that phase and reutilized reserves accumulated before anthesis. The efficiency of biomass translocation and the participation in pre-anthesis assimilates in grain were the highest in variety line 29 (36,8% and 114,4%, respectively) and the lowest in line 44 (2,8% and 5,3%, respectively). The relative contribution of synthesized pre- and post-anthesis assimilates in grain yield differed significantly, and some of the studied genotypes were characterized by the fact that during the grain filling except from assimilates formed by current photosynthesis, they reutilized assimilates accumulated before anthesis. CAVG values varied from 5,3% (line 44) to 114,4% (line 29). In variety Kaskadior, line 13,18 and 24 were calculated negative values for the three parameters-DMT, DMTE and CAVG.

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## EFFECT OF SOURCE-SINK RATIO ON THE AMINO ACID COMPOSITION OF WHEAT GRAIN

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#### Abstract

The aim of this study was to examine the effect of source-sink ratio after anthesis on the amino acid composition of wheat grain. The durum wheat (*Triticum Durum* Desf.) variety Vazhod was grown under greenhouse conditions in pots containing 4 kg sandy soil. Dissolved nutrients were added to the pots before sowing. The source-sink relationship was changed by removing the upper half of the ear at anthesis and at 10 and 20 days after anthesis. It was established that with reducing the spike size the remaining grains accumulated more nitrogen and the magnitude of this effect depended on the development stage of the grain and on the date of halving. The nitrogen content in the grain increased more with early (anthesis and 10 days after anthesis) than late (20 days after anthesis) halving. The ear halving let to increasing of the relative amount of Glutamic acid+glutamine in wheat grain. The relative amounts of threonine and lysine in the grain of halved spikes decreased significantly in the later stages of grain development – twenty days after anthesis and at maturity. Slight effect of reducing of ears size was observed for relative content of proline, valine, isoleucine, leucine, tyrosine and histidine at maturity. **Key words:** wheat, source-sink, amino acids.

## Introduction

Wheat protein quality is mainly dependent on the protein content and the balance of amino acid composition in the wheat grain (Li and Zhang, 2000). The changes in the composition of free and total amino acids in wheat grain during development were consistent with an increase in the proportion of storage proteins (Martín Del Molino *et al.*, 1988). For increasing the yield potential and quality of wheat it is important to determine the physiological factors limiting them. The main step towards this is to asses whether the growth of harvesting organs is limiting by the availability of substrates (source limited) or by the capacity of organ to assimilate and utilize the available substances for growth (sink limited) (Patrick, 1988; Cruz-Aguado *et al.*, 1999). Grain protein concentration and composition are major determinants of grain nutritional value (Feil, 1997; Liu *et al.* 2002). The concentration of lysine in grain, the most limiting amino acid in cereals for human and monogastric animals, increases with increasing grain protein concentration (Feil, 1997) despite the decrease of its concentration in total protein (Mossé *et al.*, 1985). Grain protein

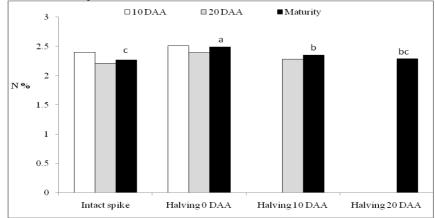
concentration and composition are also the major determinants of flour functional properties (Weegels *et al.*, 1996; Shewry and Halford, 2002). An increase in grain protein content may come from either improved capacity of the grain to accumulate nitrogen (N) or through greater N supply to the grains (Triboï and Triboï-Blondel, 2002). The aim of this study was to examine the effect of source-sink ratio after anthesis on the amino acid composition of wheat grain.

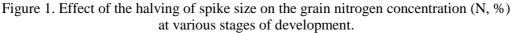
## **Material and Methods**

The durum wheat (*Triticum Durum* Desf.) variety Vazhod was grown under greenhouse conditions in pots containing 4 kg sandy soil. Dissolved nutrients were added to the pots before sowing -2,137 g Ca(NO<sub>3</sub>)<sub>2</sub>, 1,204 g KNO<sub>3</sub>, 388,5 mg KH<sub>2</sub>PO<sub>4</sub>, 999 MgSO<sub>4</sub>.7H<sub>2</sub>O, 1014 µg MnSO<sub>4</sub>.H<sub>2</sub>O, 733,5 µg ZnSO<sub>4</sub>.7H<sub>2</sub>O, 112,5, µg CuSO<sub>4</sub>.5H<sub>2</sub>O 370,9 µg H<sub>3</sub>BO<sub>3</sub>, 181,5 µg Na<sub>2</sub>MoO<sub>4</sub>.H<sub>2</sub>O, 45 µg Fe-EDTA. The sourse-sink ratio was changed by removing the upper half of the ear at anthesis and at 10 and 20 days after anthesis. At each date the size of the ear was reduced in 3 pots (10 plants.pot<sup>-1</sup>), leaving another 3 pots with intact ears. At 10 and 20 days after anthesis, and at maturity, samples of three shoots per pot were taken. Grains in halved ears and in the lower half of the intact spikes were used for analysis. In dry samples total nitrogen was determined following Kjeldahl digestion and amino acids content by hydrolysing with 6 M HCl. The analysis of variance and Duncun multiple range tests were carried out.

### **Results and discussion**

With reducing the ear size the remaining grains accumulated more N (Figure 1). The magnitude of this effect depended on the stage of development of the grain and on the date of halving. At 10 days after anthesis the grains in ears halved at anthesis accumulated more N than those in intact spikes.





At the interval between 10 and 20 days after anthesis the most amount of N were accumulated in ears halved at anthesis (Figure 1). The same was observed during the

period between 20 days after anthesis and maturity - the higher N content was in the halved ears at anthesis. In maturity the effect of late reducing of ears (20 days after anthesis) was not significant. When the size of the ear sink was decreased, the N content in grain increased more with early (anthesis and 10 days after anthesis) than late (20 days after anthesis) halving. This is partly due to the fact that reducing the sink decreased the absorption of exogenous N (Marschner, 2012).

protein) at 10 and at 20 days after antnesis (DAA).					
Amino acid	10 days after anthesis		20 days after anthesis		
	Halving (DAA)			Halving (DAA)	
	Intact ears	0	Intact ears	0	10
Asx**	8.40	7.75	6.51 a*	5.78 b	5.88 b
Thr	4.31	4.10	3.94 a	3.73 b	3.74 b
Ser	6.62	6.59	6.56	6.54	6.60
Glx	18.01	21.20	22.78 b	25.01 a	24.21 a
Pro	6.98	7.34	9.50	10.14	9.66
Gly	8.30	8.04	8.20	7.80	8.13
Ala	9.85	9.30	7.81	7.41	7.87
Val	5.90	5.73	5.63	5.60	5.61
Met	1.77	1.81	1.49 b	1.53 b	1.82 a
Ile	3.90	3.80	3.79	3.79	3.86
Leu	7.60	7.39	7.40	7.44	7.37
Tyr	2.61	2.46	2.42 a	2.31 a	2.25 b
Phe	3.30	3.02	3.47	3.53	3.47
Lys	5.01	4.74	4.00 a	3.62 c	3.77 b
His	2.06	1.96	1.91	1.92	1.93
Arg	4.78	4.63	3.79	3.87	3.90

Table 2. Effect of the halving of spike size on the amino acids compositions of grain (g.100  $g^{-1}$  protein) at 10 and at 20 days after anthesis (DAA).

\*The same letters in the rows means no significance.

\*\*Asx – Aspartic acid+asparsgine; Thr – Treonine; Ser – Serine; Glx – Glutamic acid+glutamine; Pro – Proline; Gly – Glycine; Ala – Alanine; Val – Valine; Met – Methionine; Ile – Isoleucine; Leu – Leucine; Tyr – Tyrosine; Phe – Phenylalanine; Lys – Lysine; His – Histidine; Arg – Arginine.

The amino acid composition of the grain also varied when the spike size was reduced (Tables 1 and 2). Ten days after halving it was observed a tendency that the relative content of glutamic acid+glutamine was increased and the amounts of aspartic acid+asparsgine, tyrosine and lysine were decreased, compared to its content in the intact leaves. The relative amount of glutamic acid+glutamine significantly increased in halved spikes and this was demonstrated twenty days after anthesis and at maturity. The increasing of phenylalanine was established only at maturity (Table 2). Our results showed that the relative amounts of threonine and lysine in the grain of halved spikes decreased significantly in the later stages of grain development – twenty days after anthesis and at maturity. Slight effect of reducing of ears size was observed for relative content of proline, valine, isoleucine, leucine, tyrosine and histidine at maturity. The higher differences of amino acids content in comparison to the intact leaves were demonstrated when the ears were halved at anthesis or 10 days after anthesis.

Amino acid	Intact ears	Halving (days after anthesis)		
		0	10	20
Asx	5,83	5,4	5,70	5,95
Thr	3,72 a	3,42 b	3,46 b	3,50 b
Ser	6,11	6,04	6.00	6,01
Glx	26,60 c	29,71 a	28,36 b	27,70 b
Pro	10,31	10,54	10,25	10,04
Gly	8,30 a	7,39 c	7,87 b	8,03 ab
Ala	6,54	5,91	6,19	6,16
Val	5,60	5,42	5,50	5,53
Met	0,95	0,76	0,82	0,89
Ile	3,67	3,71	3,74	3,70
Leu	7,13	7,24	7,39	7,29
Tyr	2,01	1,97	2.00	2.00
Phe	3,40 c	3,77 a	3,72 a	3,66 b
Lys	3,39 a	3,01 c	3,10 c	3,23 b
His	2,12	2.00	2,05	2,08
Arg	4,30 a	3,77 c	3,94 b	4,13 b

Table 3. Effect of the halving of spike size on the amino acids compositions of grain (g.100  $g^{-1}$  protein) at maturity.

\*The same letters in the rows means no significance.

\*\*Asx – Aspartic acid+asparsgine; Thr – Treonine; Ser – Serine; Glx – Glutamic acid+glutamine; Pro – Proline; Gly – Glycine; Ala – Alanine; Val – Valine; Met – Methionine; Ile – Isoleucine; Leu – Leucine; Tyr – Tyrosine; Phe – Phenylalanine; Lys – Lysine; His – Histidine; Arg – Arginine.

## Conclusions

With reducing the spike size of wheat the remaining grains accumulated more nitrogen and the magnitude of this effect depended on the development stage of the grain and on the date of halving. The nitrogen content in the grain increased more with early (anthesis and 10 days after anthesis) than late (20 days after anthesis) halving. The ear halving let to increasing of the relative amount of glutamic acid+glutamine in wheat grain. The relative amounts of threonine and lysine in the grain of halved spikes decreased significantly in the later stages of grain development – twenty days after anthesis and at maturity. Slight effect of reducing of ears size was observed for relative content of proline, valine, isoleucine, leucine, tyrosine and histidine at maturity.

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## CHEMICAL PROPERTIES OF THE RAW OF SOME BASMA TOBACCO VARIETIES BYBELTS

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#### Abstract

During the three-year investigations (2009, 2010, 2011), comparative trials were carried out in the experimental field of Tobacco Institute – Prilep with tobacco varieties YK 7-4/2  $\emptyset$ , MK-1, MB-2 and MB-3 and the check variety YK 7-4/2, using randomized block system with 5 replications. Chemical analyses were made in the accredited laboratory of the Institute, according to standard and internationally recognized methods in order to study the following chemical properties: nicotine, protein and soluble sugars contents in % and Shmuk's quality index. According to the chemical composition of dry tobacco in the investigated varieties, the content of nicotine increases from the lower to the upper belts, ranging from 0.59% in the check variety to 2.34% in MK-1. The soluble sugars content ranged from 8.66% in the lower belt of MB-2 variety to the maximum 22.65% in the upper belts of MK-1variety. The lower protein content in the investigated Basma varieties indicates a better quality of this tobacco compared to the check. Shmuk's index of quality is positive in all the varieties and its highest value was estimated in the upper belts. Keywords: tobacco, Basma, Chemical properties, nicotine.

#### Introduction

Chemical composition is one of the main indicators of tobacco quality. It depends on the content of certain components and their relationships. The chemical composition of tobacco is a type and varietals characteristic which depends on environmental conditions, applied cultural practices during the growing season (primarily fertilization and irrigation), after-harvest processing etc.

According to Uzunoski (1985), chemical composition of tobacco is manifested through the external appearance of the leaves, especially through their color and substantiality.

Baylov (1965) reports that tobacco quality does not depend so much on chemical components but on their relationship and on the changes caused by the conditions of growing and method of curing.

## Material and methods

The three-year investigation (2009, 2010 and 2011) included four varieties: YK 7 - 4/2 as a check ( $\emptyset$ ) and Basma varieties MK–1, MB - 2 and MB-3. The seedling was produced in traditional way, under polyethylene, in the field of Scientific Tobacco Institute - Prilep. Soil preparation before transplanting consisted of one autumn and three spring ploughings. Soil was fertilized with 300 kg/ha NPK (8:22:20). Investigations were made on previously prepared soil in randomized block design with 5 replications at 45 × 12 cm planting density. Harvesting and stringing were performed manually, by insertions, in a stage of technical maturity and tobacco was sun-cured in horizontal racks. After the qualitative assessment of cured tobacco in accordance with applicable regulations. Rules on criteria for qualitative and quantitative assessment of raw tobacco leaves ("Official Gazette" of R. Macedonia, No. 16/2007, amended and supplemented No. 144/2010 and No. 20/2011), tobacco leaf samples were separated by belts, fermented and then ground and prepared for chemical analysis.

Chemical analysis was performed in the accredited laboratory of Tobacco Institute by standard and internationally recognized methods. The following chemical characteristics were tested: nicotine, total nitrogen, soluble sugars and Shmuk's quality index.

Nicotine was determined spectrophotometrically, (ISO 2881:1992.-Tobacco and tobacco products - Determination of alkaloid content - Spectrometric method). Total nitrogen content was determined by the method of Foster, proteins by the method of Moor, soluble sugars by the method of Bertrand. Shmuk's index of quality presents the ratio between soluble sugars in % and proteins in %.

### **Results and discussion**

### *Nicotine content* (%)

Nicotine is the most important alkaloid in tobacco plant and the main reason for which tobacco leaf is used for smoking. It is distributed in all parts of the plant, except for the mature seed. It is synthesized in roots and transported in highest percentage in tobacco leaves. According to some authors, Uzunoski (1985) reported that the nicotine content increases from the lower to the upper insertions, but it is not always the rule, because this traitis largely affected by climate conditions. Thus, heavy rainfalls reduce the content of nicotine and the drought increases it. According to the obtained average values, Basma tobacco contains higher content of nicotine compared to the check variety of the type Yaka, which can be interpreted as a type-specific trait and reaction of the variety to soil and climate conditions and agricultural practices applied during the growing season. The nicotine content increases from the lower to the upper belts, ranging from 0.59% in the lower belt of the check YK 7-4 to 2.34 in the upper belt leaves of the variety MB-2 (Table 1, Figure 1).In varieties MK-1 and MB-3 this pattern does not exist. In MK-1 the lower belt has 1.77% average content of nicotine, the middle belt 2.23% and the upper belt 2.19%. In MB-3 nicotine content ranges from 1.64% in the lower to 1.97% in the middle and 1.81% in the upper belt. It can be concluded that the investigated varieties of Basma tobacco contain a higher percentage of nicotine in all belts, compared to the check. Baylov

and Popov (1965) reported that the same variety of tobacco in dry conditions can yield double nicotine content than in wet conditions. Topping of the flower bud also affects the increase of nicotine content. According to Grabuloski (1969), the content of nicotine in Prilep tobacco ranges from 12.59% to 0.84%. Lazaroski (1984) found that irrigation has significant effect on the decrease of nicotine in tobacco. Depending on the variant of irrigation, nicotine content ranged from 0.80% to 1.14%.

Variati	Vaar		BELTS	
Variety	rear	Year         Lower         Mid           2009         0.45         0.3           2010         0.55         0.7           2011         0.78         1.4           Average         0.59         0.8           Index         100.00         100           2009         1.29         1.5           2010         2.12         2.5           2011         1.89         2.6           Average         1.77         2.2           2010         2.12         2.5           2011         1.89         2.6           Average         1.77         2.2           Index         300.00         259           2009         1.18         1.4           2010         1.99         2.2           2011         1.56         2.2           Average         1.57         2.0           Index         266.10         244           2009         1.34         1.2	Middle	Upper
	2009	0.45	0.34	0.35
	2010	0.55	0.78	0.88
YK 7-4/2 Ø	2011	0.78	1.46	2.05
	Average	0.59	0.86	1.09
	Index	100.00	Middle 0.34 0.78 1.46	100.00
	2009	1.29	1.53	1.52
	2010	2.12	2.54	2.34
МК-1	2011	1.89	2.63	2.73
	Average	1.77	2.23	2.19
	Index	300.00	259.30	200.92
	2009	1.18	1.49	1.415
	2010	1.99	2.28	2.825
MB-2	2011	1.56	2.26	2.785
	Average	1.57	2.01	2.34
	Index	266.10	244.19	214.68
	2009	1.34	1.25	1.16
	2010	1.87	2.13	1.82
MB-3	2011	1.69	2.53	2.44
	Average	1.64	1.97	1.81
	Index	277.97	229.07	166.06

Table 1. Nicotine content by belts (%)

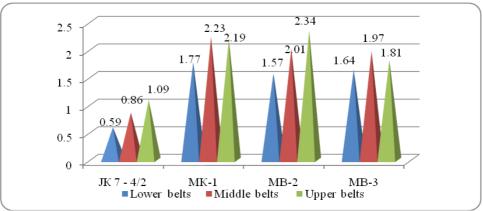


Fig.1. Nicotine content by belts (%)

#### Soluble sugars content

Soluble sugars are the basis for many other organic compounds in photosynthesis. They have an essential role in formation of tobacco quality.

The soluble sugar content by belts is presented in Table 2 and Fig. 2. The lowest percentage was found in the lower belt of check variety YK 7-4/2 in 2011 (4.85%) and the highest in the upper belt of the variety MB-3 in 2009 (29.42%). The average values increase from the lower to the upper belts, ranging from 8.66% in the lower belt of the variety MB-2 to 22.65% in the upper belt of the variety MK-1, which is 12.46% higher than the check.

According to Gyuzelev et al., (1965), oriental tobaccos containing less than 5% soluble sugars are of poor quality, those with 10-11% are of medium quality and those with 11-16% are of high quality. Tobaccos which soluble sugars content exceeds 16% have a neutral and monotonous taste.

Lazareski (1967) reported that soluble sugars content of Prilep tobacco in the region of Bitola ranges from 18.42% to 22.26%, increasing from the lower to the upper insertions. Nuneski (2008) reported that the average soluble sugars content of Izmir Basma tobacco produced in Turkey ranged from 12.92% in lower belts to 18.37% in the upper and 19.33% in the top leaves.

	Ŭ	III by bens (%)	BELTS	
VARIETY	Year	Lower	Middle	Upper
	2009	12.95	16.36	24.23
	2010	11.84	15.43	21.55
ҮК 7-4/2 Ø	2011	4.85	8.82	14.65
	Average	9.88	13.54	20.14
	Index	100	100	100
	2009	11.24	18.02	25.33
	2010	9.89	15.49	26.91
MK-1	2011	5.89	14.16	15.72
	Average	9.01	15.89	22.65
	Index	91.19	117.36	112.46
	2009	9.40	14.35	26.49
	2010	10.55	14.99	22.15
MB-2	2011	6.04	8.91	16.19
	Average	8.66	12.75	21.61
	Index	87.65	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	107.30
	2009	15.04	23.73	29.42
	2010	8.61	14.64	19.06
MB-3	2011	5.94	11.64	16.65
	Average	9.86	16.67	21.71
	Index	99.80	123.12	107.79

Table 2. Soluble sugars content by belts (%)

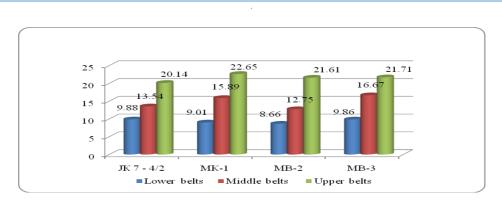


Fig. 2. Average content of soluble sugars by belts (%)

## Proteins content

Proteins have a negative impact on tobacco and give an unpleasant taste in smoking. The higher percentage of soluble carbohydrates in tobacco raw reduces or neutralizes the negative effect of proteins. According to Shmuk (1948), the optimum content of proteins is 5-10%. When it is below 5%, the flavor components are neutral and over 10% has a negative effect on tobacco taste. Lazaroski (1976) reported a protein content of 8.01 to 8.85% in Prilep tobacco in the region of Bitola in 1973 and 1974. The same author (1983), in his three-year investigations, reported a protein content of 6.18%- 7.39%. He found that irrigation reduces the protein content to 16.73%.

Nuneski (2008) reported that the average protein content in all insertions of Izmir Basma tobacco was 7.18% and 7.65% in 2004 in 2005 respectively, which is an average of 7.41% for the two years.

According to Arsov et al. (2011), the presence of proteins below 5.5% of tobacco dry matter is an indication of monotonous taste and content over 7% indicates poor taste and quality of tobacco.

Mitreski (2012) reported that protein content in some varieties of Prilep tobacco in 2009 ranged from 6.34 in P 66-9/7 to 7.22 % in P-79-94, while in 2010 it ranged from 6.35% in P-79-94 to 6.75% in P-23.

The protein content by belts and years ranged from 5.17% in the middle belt of MB-3 in 2009 to 8.50% in the upper belt of MB-2 in 2009 (Table 3, Fig. 3.)

The average content ranged from 13.6% in the middle belt of MB-3 to 7.40% in the upper belt of MB-2. Here from it can be concluded that they increase from the lower to the upper belt, with small deviation in the middle belt of varieties MB-2 and MB-3, which has a lower protein content. The above data reveal that the protein content of the varieties MK-1 and MB-3 is lower compared to the check variety, which is a characteristic of a good quality tobacco.

VADIETV	Voor	BELTS						
VARIETY	Year	Lower	Middle	Upper				
	2009	6.43	5.91	8.02				
	2010	6.74	7.09	5.92				
YK 7-4∕2 ∅	2011	6.89	7.56	7.64				
	Average	6.68	6.86	7.19				
	Index	100.00	100.00	100.00				
	2009	6.28	5.87	6.64				
	2010	6.73	7.01	6.12				
МК-1	2011	6.08	6.35	7.81				
	Average	6.36	6.41	6.86				
	Index	95.21	93.44	95.41				
	2009	6.45	6.09	8.50				
	2010	7.11	6.60	6.84				
MB-2	2011	6.52	7.21	6.86				
	Average	6.69	6.64	7.40				
	Index	100.15	96.79	102.92				
	2009	7.30	5.17	7.31				
	2010	6.54	6.95	6.09				
MB-3	2011	5.44	6.28	6.59				
	Average	6.42	6.13	6.67				
	Index	96.11	89.36	92.77				

Table 3. Protein content by belts (%)

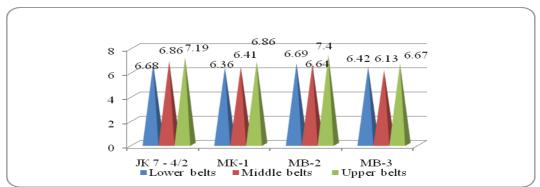


Fig. 3.Average protein content by belts (%)

### Shmuk's quality index

For objective estimation of quality and use value of tobacco, it is very important to know the components of its chemical composition and their mutual relationship. Various authors suggest a number of factors to determine the quality, including the Shmuk's index. Table 4 and Figure 4 present the Shmuk's index of the varieties investigated in this trial by belts and years. The lowest index was recorded in the lower belt of MB-2 (0.93 in 2011) and the highest in the middle belt of MB-3 (4.64 in 2009). The average Shmuk's index ranged from 1.30 in the lower belt of MB-2 to 3.42 in the upper belt of MK-1, which is 18.75% higher than the check. According to the presented data, the Shmuk's index increased from the

lower to the upper belt and the highest quality with regard to this parameter was achieved in tobacco from the middle and the upper belts. The same is valid for the check variety YK 7-4/2, where the Shmuk's index ranged from 2.05 in the middle belt to 2.88 in the upper. In variety MK-1 the index ranged from 2.51 in the middle belt to 3.42 in the upper, in MS-2 from 2.01 in the middle belt to 3.00 in the upper and in MB-3 from 2.90 in the middle belt to 3:26 in the upper. With regard to the Shmuk's index, especially in the middle and upper belts, the investigated varieties are characterized by a high quality tobacco raw.

VADIETV			BELTS	
VARIETY	Year	Lower	Middle	Upper
	2009	2.09	2.80	3.03
	2010	1.76	2.19	3.68
YK 7-4∕2 ∅	2011	0.71	1.16	1.92
	Average	1.52	2.05	2.88
	Index	100.00	100.00	100.00
	2009	1.84	3.07	3.84
	2010	1.47	2.19	4.40
МК-1	2011	0.99	2.27	2.03
	Average	1.42	2.51	3.42
	Index	94.08	122.44	118.75
	2009	1.48	2.36	3.17
	2010	1.48	2.39	3.45
MB-2	2011	0.93	1.27	2.37
	Average	1.30	2.01	3.00
	Index	85.53	98.05	104.17
	2009	2.06	4.64	4.13
	2010	1.32	2.14	3.13
MB-3	2011	1.10	1.91	2.53
	Average	1.49	2.90	3.26
	Index	98.03	141.46	113.19

Table 4. Shmuk's quality index

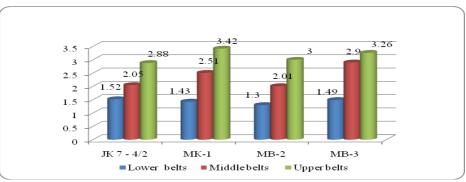


Fig. 4. Average Shmuk's index by belts

# Conclusions

- The content of nicotine increases from the lower to upper insertions from 0.59% in the lower belt of the check variety YK 7-4/2 to 2.34% in the upper belt of the variety MB-2. In Basma tobaccos, the nicotine content is somewhat higher.
- The content of soluble sugars increases from the lower to the middle and upper belts. Basma tobaccos have higher content of soluble sugars (up to 21.71% in MB-3), which is an indication of positive properties of the raw.
- Proteins increase from the lower to the upper belts. In variety MB-3 the percentage of proteins is lower and ranges from 6.67% to 6.13% in the upper belts.
- Due to their quality, Basma tobaccos have a higher Shmuk's index compared to the check, which increases from the lower to upper belts (from 1.30 to 3.42).
- Chemical properties of the investigated Basma varieties are typical for the oriental tobacco and confirm the good quality of the raw material.

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# INFLUENCE OF THE GROWTH REGULATOR "IMMUNOCYTOPHIT" ON THE DEVELOPMENT AND PRODUCTIVITY OF OIL RAPESEED, VARIETY "ELITE"

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### Abstract

The experiment is carried out in the period 2009 - 2012 in the study, research and implementation base of Department of Plant Growing at Agricultural University of Plovdiv. The experiment is performed according to the block method, repeated four times, with size of the experimental field of  $20 \text{ m}^2$ , without irrigation system, after precursor wheat, with hybrid "Elite". The influence of the growth stimulator "Immunocytophit" is studied in the following variants: Control – untreated variant; Treatment in the phases of rosette (6-8 leaves), buttoning and blossoming, in the following dosages:  $\frac{1}{2}$  tablet, one tablet, and 1 and  $\frac{1}{2}$  tablet, dissolved in 20 1 water /50m2. The highest mathematically proven yield compared to the control variant is reported in 2011, for the variant treated in the phase 6-8 leaves (with 6.0 to 9.6%) and dosage 1.5 tablets (5000 kg/ha). **Keywrds:** rapeseed, bio-stimulator "Immunocytophit", phases, doses.

## Introduction

The bio-stimulator "Immunocytophit" is a new generation product, for 100% ecological agriculture, which stimulates the growth and development of the plants. The growth biostimulator is universal means enhancing the immune system of plants as a result of which plants more easily overcome the unfavorable climatic conditions and become more resilient to diseases and pests. The positive effect of the growth regulator "Immunocytophit" and of other biological stimulators on durum wheat, rye, triticale, asters, roses, basil and other cultures is proven by the research work of Voineac., (2003); Kolev et al., (2005); Belcheva et al., (2007) Atanasova et al., (2008); Kolev, (2008); Stankov et al., (2008); Todorov et al., (2010); Ivanov I Yanchev., (2011, 2013). The research abroad and in our country related to studying the effect of the growth stimulator "Immunocytophit" on rapeseed are very scarce. Therefore we set ourselves the goal to establish the effect of this stimulator on the development and productivity of the winter oilseed rape.

#### Materials and methods

The experiment is carried out in the period 2009 - 2012 in the study, research and implementation base of Department of Plant Growing at Agricultural University of Plovdiv. The experiment is performed according to the block method, repeated four times, with size of the experimental field of 20  $m^2$ , without irrigation system, after precursor wheat, with hybrid "Elite". The influence of the growth stimulator "Immunocytophit" is studied in the following variants: Control - untreated variant; Treatment in the phases of rosette (6-8 leaves), buttoning and massive blossoming, in the following dosages: 1/2 tablet, one tablet, and 1 and  $\frac{1}{2}$  tablet, dissolved in 20 l water /50m<sup>2</sup>. Immunocytophit is a mixture of polyunsaturated fatty acids with active substance 5% ethyl ester of the arachidonic acid. One tablet of Immunocytophit is 3g and is 0.16 g/kg active substance ethyl arachidonate. The following parameters are reported: height of the plants (cm); number of branches of one plant (pce); Number of fruit of one plant (pce); length of the fruit (cm); number of seeds of one plant (pce); weight of the fruit of one plant (g); weight of the seeds of one plant (g) and yield of seeds kg/ha. The data from the obtained yields are mathematically processed according to the method of analysis of variance, while the differences between the variants are determined by the multi-rank test of DUNKAN (Dunkan., 1999). In a vascular experience there are planted seeds treated for a period of 1 to 3 hours with different dosage of the preparation (1/2, 1, 1 <sup>1</sup>/<sub>2</sub> tablets). After 14 days we register the number of sprouting plants, height of the stems and length of the root system.

### **Results and Discussion**

The variance in the meteorological factors, temperature and rainfall during the experimental period influences both the structural elements of the rapeseed, and the seed yield. During the three years of the experiment there are no significant deviations from the values of the average day and night temperature compared to the requirements of the culture and the long-term period (Fig.1).

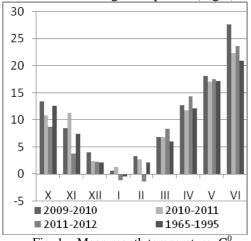


Fig. 1. Mean month temperature,  $C^0$ 

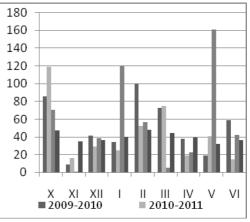


Fig.2. Quantity of rainfalls, mm

Differences are observed with regards to the quantity and distribution of rainfall during the experimental period – one of the main ecological factors that limit the yield amounts of rapeseed in our country is water. The largest quantity of rainfalls during the vegetation of rapeseed (from sprouting to harvesting) is registered during the period 2011-2012 (520,8mm). They exceed with 67.8 mm the rainfall of the long-term period (453.0 mm).

Less than the usual long-term rainfall are the rainfalls during the period 2010 -2011 (401.2 mm). Despite this, characteristic for this year is the even distribution of rainfalls during the period of cultivation and the presence of enough moisture in the critical phases of its development (Fig.2). Phenological observations during the years of the experiment are presented in Table 1 and they show that the treatment of the plants with Immunocytophit does not influence the course of the development phases of the rapeseed.

Differences in the course of the phases are observed only due to the climatic factors during the experimental years. The seeds sown in the autumn of 2010 sprout fastest (or 22 days), which is due to the larger quantity of rainfalls during the period – sowing – sprouting (58.8 mm). The lesser rainfalls consisting of 25,9 mm in this period of 2011 prolong the period of sprouting to 28 days. One month after sprouting the plants reach the phase 6-8 leave, and in this stage they spend the winter most successfully.

With lowering of the temperature during December a gradual slowing of the growth processes is observed and suspension of the vegetation. At the constant increase of the temperatures above  $5^{\circ}$  C there are conditions for resuming the vegetation growth. This usually happens in the end of February to the middle of March, depending on the climatic conditions. The higher temperatures in the months of February and March of 2009 ( $8-9^{\circ}$  C) and rainfalls of 100.3 mm create conditions for resuming the vegetation even in the beginning of March 2010 (05.03), while this period in 2011 and 2012 starts with 8-10 days later (15.03; 18.03). In the three years the rapeseed reaches the phase of stem-formation in the end of March (24, 27, 29 March). In 2011 and 2012 the phase of buttoning occurs with 7 and 8 days later compared to 2010. The phase of massive blossoming occurs in 2010 (19.04), while with 4 days later, in 2011 and 2012 (23.04).

Phenological observation	Years						
Γ	2009-2010	2010/2011	2011-2012				
Sowing	17.09	21.09	15.09				
Prouing	10.10	13.10	13.10				
6st-8st leaf	13.11	17.11	18.11				
Finish vegetation	24.12	17.12	23.12				
Beginning vegetation	05.03	15.03	18.03				
Stem development	24.03	27.03	29.03				
Bud stage	8.04	15.04	16.04				
Full flowering	19.04	23.04	23.04				
Wax ripeness	07.06	12.06	15.06				
Full maturity	26.06	26.06	30.06				
Vegetation period, days	255	256	261				

Table 1. Phenological observation

The larger quantity of rainfalls in the end of May and the beginning of July in 2012 prolongs the phases of wax and full ripeness and they occur latest (15.06; 30.06) compared

to the other two years. As a result of this the vegetation period is longest during this year (261 days). The differences in the temperature and rainfalls during the period of the experiment influence on the values of the structural elements of the yield of rapeseed (Table 2).

The data about the structural elements of the yield prove the positive effect of the growth stimulator Immunocytophit depending on the phase and dosage of treatment.

Thanks to the influence of the preparation on the growth and development all treated variants form plants higher than these of the control.

The most stimulating effect with regards to all parameters is observed when treating the rapeseed in the phase 6-8 leave, when the organs determinants of the yield are formed and differentiated (number of flower and axil buds on the top of the growing cone).

	Height and structural elements/plants											
	Height	Number of	Number	Length	Number	Weight	Weight					
Variants	/plants	branches/pl	of	of pod,	of	of	of					
	(cm)	an	pods/pla	cm	seeds/po	pods/pl	seeds/pla					
			nt		d	ant, g	nt, g					
			6st-8st le	eaf								
Control	152.0	9.0	203	7.8	24.0	25.8	15.4					
0,5 pill	153.2	9.1	218	7.9	26.0	25.9	15.5					
1 pill	155.4	9.2	223	7.9	27.0	26.0	15.7					
1,5 pill	157.0	10.1	234	8.0	30.0	27.7	16.6					
			Bud sta	ge								
Control	150.4	8.8	199	7.5	22.0	24.7	14.8					
0,5 pill	151.4	8.9	210	7.5	23.3	24.9	14.9					
1 pill	153.2	9.1	216	7.7	23.5	25.0	15.0					
1,5 pill	155.6	9.7	223	7.7	26.0	26.1	15.9					
			Full flowe	ering								
Control	148.9	8.4	195	7.2	19.0	24.4	13.4					
0,5 pill	149.2	8.6	196	7.3	19.5	24.5	13.5					
1 pill	150.3	8.7	200	7.5	20.0	24.7	13.6					
1,5 pill	152.6	8.9	210	7.5	22.0	25.0	14.0					

Table. 2 Structural analysis of yield element, mean for the period 2009-2012

The longer this period is the more formed generative organs and higher yields obtained. The climatic conditions in the period of rosette formation to termination of the autumn vegetation determine 70% of the estimated yields. The effect of the growth stimulator on the measurements of the structural elements when treating the plants in the phase of buttoning is less, while the phase of massive blossoming – weakest. In testing the different dosage treatment variants of the plants with Immunocytophit, the strongest stimulating effect with regards to the height of the plants and the structural elements is reported for the variant – treatment with 1.5 tablet. The values of the parameters such as number of fruit on 1 plant (15,3%); number of seeds in one plant (25,0%); fruit weight (7,4%) and seed weight (7,7%) of one plant, in the variant treated with 1,5 tablet in the phase 6-8 leaves exceed mostly the control (with the percentages shown after the names of the parameters) compared to the rest variants and phases of treatment. The studied growth regulator has

positive effect also regarding achieve yields (Table 3). The good quantity of rainfall during the critical periods of the development of the rapeseed during 2010/2011 is a prerequisite for achieving the highest yields. During this year the yields vary from 4300 kg/ha to 5000 kg/ha, depending on the treated variants.

Despite the larger quantity of rainfall during 2009/2010 and 2011/2012, the uneven distribution of the rainfalls during the vegetation is a reason for the lower yields.

In 2009/2010 the yields vary from 4120 kg/ha to 4560 kg/ha, while in 2011/2012 from 3900 kg/ha to 4380 kg/ha.

Depending on the phases of development, highest yields are achieved when treating the plants in 6-8 leaves, followed by the phases of buttoning and massive blossoming.

During the three years of the experiment, as well as averagely for the period of the experiment, the seed yield for all treated variants has higher values.

	Years										
	201	0 г.	201	Ιг.	2012 г.		Ср	едно			
Variants	kg/ha	%	kg/ha	%	kg/ha	%	kg/ha	%			
6st-8st leaf											
Control	4300a	100.0	4560a	100.0	4110a	100.0	432	100.0			
0,5 pill	4440b	103.2	4680b	103.3	4250b	103.4	446	103.2			
1 pill	4480b	104.2	4790c	105.0	4330c	104.6	453	104.9			
1,5 pill	4560c	106.0	5000d	109.6	4380d	106.6	465	107.6			
LSD 5%	76.8		37.2		27.9						
			Bu	d stage							
Control	4200a	100.0	4480a	100.0	4100a	100	426	100.0			
0,5 pill	4250b	101.1	4540b	101,3	4150b	101,2	431	101.2			
1 pill	4300c	102.3	4600c	102.6	4180c	101.9	436	102.3			
1,5 pill	4350d	103.5	4650d	103.7	4210d	102.6	440	103.2			
LSD 5%	14.8		13.0		11.5						
			Full t	flowering							
Control	4120a	100.0	4300a	100.0	3900a	100.0	411	100.0			
0,5 pill	4160b	100.9	4320ab	100.5	3930b	100.7	413	100.5			
1 pill	4200c	101.9	4350b	101.2	3970c	101.7	417	101.4			
1,5 pill	4230d	102.6	4410c	102.5	4000d	102.5	421	102.4			
LSD 5%	31.5		52.6		32.6						

Table 3. Seed yield, kg/ha

This is due to the ability of the preparation to hydrolyze the ethyl ethers in the lipid membranes of the cells to the arachidonic acid and its metabolites. As a result of this the preparation enhances the natural immunity of the plants to the unfavorable climatic conditions such as frost, spring droughts and excessive moisture, which reflects on the productivity of the crop. The stimulating effect of the preparation is expressed in higher yield during the different years from 2,5% to 9,6%, depending on the phase and dosage of treatment. The highest mathematically proven yield compared to the control is reported in 2011, with the variant treated in the phase 6-8 leaves (with 6,0 to 9.6%) and dosage of 1,5 tablets.

Source of Variation	Sum of Square	DF	Mean Square	Sig of	Partial ETA
				F	Sqd
					$\eta^2$
Does - A	709022.22	3	236340.74	,000	95.0
Years- B	4511488.89	2	2255744.4	,000	99.0
Phases – C	2656822.22	2	1328411.1	,000	98.7
A x B	33844.44	6	5640.74	,000	49.9
A x C	167977.78	6	27996.30	,000	83.2
B x C	128177.78	4	32044.44	,000	79.1
A x B x C	37155.56	12	3096.30	,000	52.3
Residual	33936.00	108	314.22		

Table 4. Dispersion analysis (Anova) of seed yield

The lowest is the effect of the preparation at this dosage on the yield achieved in the phase of buttoning with 2,6% to 3,7%. The weakest and unproven is the stimulating effect when treating the plants in the phase of massive blossoming with 1,5 tablets (with 2.5 - 2.6%). The dispersion analysis made of the yield of seeds shows strong statistically proven effect during the years of the experiment (99%) and strongly, phases of treatment (98,7%), followed by doses (95%) on the variableness of the values of this parameter. The interaction between the two factors (phases and doses) however is most powerful (83,2%) (Table 4).

Table 5. Vascular experience

Variants	Spring up plants /				Root length / cm/	
	1 h	3 h	1 h	3 h	1 h	3h
Control	33	33	10,5	10,5	1,4	1,4
¹∕₂ pill	90	39	6,9	5,5	1,7	1,5
1 pill	95	42	9,1	6,3	2,5	2,1
1 <sup>1</sup> / <sub>2</sub> pill	70	60	7,5	6,8	2,0	1,9

The data from the vessel experiment show that the values of the reported parameters in the event of three-hour treatment are with lower values compare d to the variant with 1-hour treatment (Table 5). Both in the one-hour and in the three-hour treatment the parameters like number of sprouted seeds and length of the root system exceed the values of the control variant. The highest stimulating effect with regards to the parameter number of seeds and length of the roots of seeds that have been 1 hour in the preparation is reported for the variant with concentration of 1 tablet. From 100 seeds in this variant, 95 have sprouted and the longest roots have been measured (2.5 cm). For the parameter height of the above-ground mass for all tested variants, the values are under these of the control variant (10.5 cm). All this gives us the grounds to consider that the treatment of the rapeseed seeds with smaller or higher dosage of Immunocytophit influences positively the sprouting of a larger number of plants and stronger development of the root system of the corp which is a prerequisite for the better resilience of the crops to the drought and lower temperatures.

# Conclusions

The treatment of the plants with Immunocytophit does not influence the course of the different phases of the rapeseed development.

For all treated variants the values of the structural elements of the yield are higher compared to the control.

The strongest stimulating effect with regards to the structural elements and the yield is observed when treating the rapeseed in the phase 6-8 leaves with 1.5 tablets.

The highest mathematically proven yield compared to the control variant is reported in 2011, for the variant treated in the phase 6-8 leaves (with 6.0 to 9.6%) and dosage 1.5 tablets (5000 kg/ha). In the vessel experiment, the treatment with Immunocytophit influences positively the sprouting of larger number of plants and stronger development of their root system. The highest stimulating effect for the parameter number of seeds and length of the roots for the seeds that have spent 1 hour in the preparation is reported for the variant with concentration of 1 tablet.

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# STUDY ON HYDRO PHOBIA OF MAIZE SEED, AS A METHOD OF EARLIER TERMS OF SOWING

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#### Abstract

The possibility of earlier sowing of maize before the optimal time has come  $(12-15^{\circ}C)$ relieves the manufacturers, which leads to more rational use of machineries, ensures early germination and fuller use of the moisture from the winter-spring period. This opportunity is achieved through a peculiar preserving of the seeds, considered as an additional part of their pre sowing preparation or complex preparation including the pesticides. Hydro phobia represents a construction of a protective water-impermeable layer of polymers with a different thickness and different time of degradation at definite conditions. The treatment of seeds with pesticides is done separately or in conjunction with the creating of the polymer covering. The exposure of the seeds in cold and moist soil to their germination is directly dependent on the thickness of the coating and the time for its breaking under the influence of the soil microbial activity. A correlation between the surface of corn from different fractions and their mass is established. A relationship between the thickness of the polymer coating and the period's duration of its degradation is established. A function between the mass of grains, the thickness of the coating, the amount of polymer and the amount of the solvent is established. The period of stay of the seeds in the soil depends on the thickness of the coating and the time of sowing should end with the coming of minimum conditions  $(7-9 \ ^{0}C)$  for germination.

Keywords: maize, hydro phobia, polymer, coating, terms of sowing.

#### Introduction

The main purpose of hydro phobia, as the name itself points, is to reduce the inflow of moisture into the seeds by creating a polystyrene insulation coating around their surface. Besides this, this coating has to insulate also from penetration of pathogenic microflora into the seeds under the terms of their prolonged stay in the soil. Hydro phobia during the Seventies of the  $20^{\text{th}}$  century was based on the use of: Chloroform – 17 liters, Polystyrene - 0,5 kilograms and Fenthioram- 2 kg per ton of maize kernel. Both ingredients are comparatively well known today and they can be supplied in industrial quantities without serious restrictions. Cloroform-trochloromethane CHCL<sub>3</sub>. Fenthioram represents a combination of chemical substances, including: 40% Thiram–presently it is used mainly for decontamination of vegetable seeds and 20% gamma-isomer of hexachlorocyclohexane (HCH), a chemical substance that was awarded a Nobel Prize because it saved people from

the widely spread fleas among people, but consequently its use was banned due to found out negative effects on people and the long duration of its adverse aftereffect; and finally the last ingredient of Fenthioram is the chemical substance with 10% content of copper trichlorophenolate – a red-and-brown powder with sharp carbolic scent, which is also forbidden for independent use. The necessity of hydro phobia arises from land consolidation, i.e. the increase of the size of the cultivated fields, which requires more agricultural equipment with higher efficiency. Also a problem arises related to the longer time for their transportation due to the significant distances between the separate cultivated fields. The structure of the cultivated crops is changed both due to economic reasons and to the permanent change of climate caused by the cyclicity of the annual periods against the background of global warming. The increased fields sown with rapeseed due to the unlimited market and the high price led to rapeseed occupying about 2 million dca, and due to the same reasons the fields with sunflower increased to 7 million dca. Problems occurred with the correct rotation of the cultures due to the fact that both cultures do now allow subsequent sowing after it as well as one after another for a period of at least two years. Among the cereal crops, main predecessors for them are wheat and maize each having its own advantages and disadvantages. The first one is an excellent predecessor, covers large land areas, it is harvested early, but it does not allow further sowing after two subsequent years, after which the resources necessary for preserving the yields get higher and then occurs the problem with the crop rotation of rapeseed and sunflower. Maize which occupies large pieces of lands reaching 3-4 million dca tends to increase, particularly in North-West Bulgaria and it is commonly used as predecessor of the main cultures and of rapeseed as well. During the years of normal distribution of rainfalls maize forms high yields, allows mono-crop cultivation, which makes it the Queen of the field. Main aspect of the technology is the opportunity for earlier sowing which generally is related to the better use of moisture from the winter reserve and the early spring rainfalls, formation of good root system and earlier phases of growth, slower in the beginning, but much intensive with the rising temperature. Hydro phobia of the seeds is an opportunity for earlier sowing of maize seeds in the cold and moist soil. Also, significant is the fact that maize sprouts endure even light short-term frost. Hydro phobia of maize seeds in industrial quantities is known and used in the 80-ies of the  $20^{th}$  century, but the stated prerequisites in the current economic conditions are a significant motive for its revival and further improvement. The purpose of this study was to find out the relation between the quantity of seeds and their surface with the help of which to determine the quantity of polymer necessary for their hydrophobization which might differ depending on the time of sowing and their stay in the soil.

### Material and methods

Decontaminated maize seeds are used, of the hybrid F-38 by the company Pioneer, with fraction of the kernels 3.5 and 4.5 mm / Table1 and Function /. By wrapping the kernels with aluminum foil and then by superimposing their imprints on the foil over graph paper we determine their surface. In order to make a correction of the measurements the entire quantity of seeds is immersed in water, the water displacement is equated to a dense sphere that shows the surface. Five samples have been studied, repeated two times, each

consisting of 10 kernels, which weight and surface were measured, from two fractions. The calculated quantity of polystyrene is dissolved in chloroform and while constantly stirring the seeds in a chamber, it is added to them. The exposure takes from five to fifty minutes depending on the necessary quantity and it is related to the time of stirring and homogenization. The time for preparation depends also on the size of the solvent which subsequently we lose due to the fact that the chamber is opened while the stirring continues until the evaporation of the solvent. Its optimal amount will be determined by further studies related also to the drying of the prepared material. Sowing is made in the end of February and beginning of March with soil temperature of -3-4 C<sup>0</sup> reaching for short periods from 3 to 5 C<sup>0</sup>, a period during which from ten years we usually have eight or nine with suitable sowing conditions.

	Weight / g /	Surface /mm <sup>2</sup> /	Weight / g /	Surface /mm <sup>2</sup> /	Weight / g /	Surface /mm <sup>2</sup> /	Weight / g /	Surface /mm <sup>2</sup> /	Weight / g /	Surface /mm <sup>2</sup> /
Sample №		I	I	I	III		Ι	V		V
1	0,38	469	0,43	427	0,38	411	0,45	460	0,42	539
2	0,40	683	0,43	326	0,41	471	0,38	433	0,35	521
3	0,34	549	0,40	399	0,31	394	0,33	547	0,44	545
4	0,44	558	0,33	404	0,37	244	0,36	506	0,39	534
5	0,37	437	0,42	408	0,39	453	0,36	580	0,40	561
6	0,37	378	0,43	479	0,39	459	0,37	513	0,45	531
7	0,42	258	0,34	340	0,43	289	0,37	459	0,43	486
8	0,33	416	0,40	314	0,38	422	0,42	584	0,45	559
9	0,40	326	0,42	432	0,42	494	0,27	260	0,34	479
10	0,40	449	0,34	398	0,36	286	0,39	421	0,34	522
Total	3,85	4522	3,94	3927	3,84	3923	3,70	4763	3,91	5277
Average	0,385	452,2	0,394	397,2	0,384	392,3	0,370	476,3	0,391	527,7
Sample №		Ι	1	Ι	III		IV		V	
1	0,41	454	0,38	580	0,41	425	0,41	589	0,43	483
2	0,43	451	0,42	591	0,38	336	0,36	409	0,37	376
3	0,51	527	0,36	577	0,36	375	0,45	503	0,36	402
4	0,41	465	0,41	461	0,43	322	0,43	275	0,41	511
5	0,41	531	0,32	382	0,41	464	0,34	368	0,46	563
6	0,37	400	0,38	579	0,40	338	0,39	411	0,37	448
7	0,39	527	0,35	374	0,35	275	0,35	330	0,45	412
8	0,36	567	0,39	489	0,39	275	0,46	344	0,35	385
9	0,35	295	0,34	307	0,43	319	0,47	477	0,44	403
10	0,43	387	0,39	535	0,41	247	0,44	401	0,43	396
Total	4,07	4704	3,74	4875	3,97	3376	4,10	4089	4,07	4379
Average $\mathbf{X} = \mathbf{X} + \mathbf{Y}$	0,407	470,4	0,374	487,5	0,397	337,6	0,410	408,9	0,407	437,9

 $\begin{array}{l} X_{ave} = X_1 + X_2 + .... X_n = 0,3919 \ /g / \\ y_{ave} = y_1 + y_2 + .... y_n = 438,35 \ /mm^2 / \end{array}$ 

 $\delta_{x} = \frac{(X_{ave} - X_1)^2 + (X_{ave} - X_2)^2 + (X_{ave} - X_n)^2}{(X_{ave} - X_1)^2 + (X_{ave} - X_2)^2 + (X_{ave} - X_n)^2} = 0.0004614 / g/$ 

n.(n-1)

 $\begin{array}{l} \delta_{y} = & (\underline{V_{ave}} - \underline{V_{1}})^{2} + (\underline{V_{cave}} - \underline{V_{2}})^{2} + (\underline{V_{ave}} - \underline{V_{n}})^{2} = 1,6586392 / mm^{2} / \\ n.(n-1) \\ y = ax + b \end{array}$ 

## **Results and discussion**

Moistened samples soaked for 24 hours are set to germinate – treated and untreated seeds. After a period of 20 days at temperature regime of about -3, -4 <sup>o</sup>C they are inserted in thermostat at temperature of 12-15 degrees. The seeds treated with polystyrene in the beginning after the third day and in the end of the eighth day are not germinating, except in the cases when some of them might not be very well coated and admit water. Regarding the untreated seeds the germination rate is 95-97%. After 20 days' stay in a refrigerator in a container at  $+5^{\circ}$  C with high humidity of 80-90%, the germination rate of the untreated seeds is reduced and we notice signs of rotting, while in the same time we do not observe any germination of the treated seeds. When putting the seeds in planters with moist soil and at low temperature, the untreated seeds for 20 days' period at temperatures of-3,-4 °C rot - 30-50 % of them, while the treated ones preserve low germination rate of about 20-30 %, without rotting seeds. In the conditions of the soil the germination rate of the treated seeds at the end of the period, on the 30<sup>th</sup> day starts to increase on the basis of the starting destruction of the polystyrene coating. On the 30<sup>th</sup> day after the sowing the germination rate reaches 75% while this percentage is only 25% for the untreated sown seeds. We found out the average weight of the kernel and its surface which are presented by the limits of the average values at P - 5%, 1% and 0.1%, which are respectively: 0.318-0.392; 0.390-0.393; 0.391- 0.394 g and the respective surface within the limits : 43.51- 48.62; 43.41-44.26; 43.29- 44.38mm<sup>2</sup>. We found out the thickness of the polystyrene coating when using different quantities of polystyrene per 1 ton of kernels and a function of the thickness of the coating depending on the surface of the kernel. The measured germination rate of moistened untreated and treated seeds set to germinate, after the end of the period of stay at low temperature and its increase to 15 °C, as well as those set to germinate as a control batch show the following results:

Untreated with a period of stay at low temperature -57 %

Treated with a period of stay at low temperature- 75 %

Untreated, set to germinate as a control batch - 98 %

The limits /LSD/ of the average values are calculated at P- 5% and 1%.

A correlation analysis of the results was done according to Duncan.

А	В	C	D	E	F	G
			C*1000 /(100000/B)		E/1000000	D/A/F*1000
mm <sup>2</sup> /1kernel	gram /1 kernel	Polystyrene [kg] /1 ton kernel	Polystyrene [gr] /1 kernel	Polystyrene [gram/dm3]	Polystyrene [gram/mm3]	H[mkm] coating thickness in micrometers
226	0,34	2	0,00068	1061	0,001061	2,8
260	0,33	2	0,00066	1061	0,001061	2,4
261	0,32	2	0,00064	1061	0,001061	2,3
265	0,31	2	0,00062	1061	0,001061	2,2
267	0,3	2	0,0006	1061	0,001061	2,1
282	0,32	2	0,00064	1061	0,001061	2,1
292	0,31	2	0,00062	1061	0,001061	2,0
263	0,28	2	0,00056	1061	0,001061	2,0
249	0,29	2	0,00058	1061	0,001061	2,2
274	0,31	2	0,00062	1061	0,001061	2,1
262	0,32	2	0,00064	1061	0,001061	2,3
283	0,33	2	0,00066	1061	0,001061	2,2
267	0,33	2	0,00066	1061	0,001061	2,3
294	0,36	2	0,00072	1061	0,001061	2,3
281	0,34	2	0,00068	1061	0,001061	2,3
281	0,31	2	0,00062	1061	0,001061	2,1
290	0,36	2	0,00072	1061	0,001061	2,3
274	0,33	2	0,00066	1061	0,001061	2,3
269	0,32	2	0,00064	1061	0,001061	2,2
269	0,33	2	0,00066	1061	0,001061	2,3

Table 2. Thickness of the coating as a function of the weight and surface of the kernels

\*Only the values of columns 1, 2 and 3 are variable.

legend

 $A = mm^2/1$  grain

B=gr/1grain

C=polystyrene for 1 t

D=C\*B/1000=polystyrene for 1 kg

E=factor 1061polystyrene/gr/dm<sup>3</sup>

F=factor 1.061 polystyrene  $\text{gr/cm}^3$ 

 $G=C*B/mm^2$  on 1 grain\*polystyrene gr oncm<sup>3</sup>

G= polystyrene for 1 t\*gr.grain/mm<sup>2</sup>grain\*polystyrene gr /cm<sup>3</sup>

H (mkm) -coating thickness in micrometers

## Conclusions

There is an opportunity to make a hydrophobization coating of the seeds with different thickness depending on their weight, surface and the duration of their stay in cold and moist soil. There is an opportunity for very early sowing, in which case the plants are rooted earlier and their growth happens faster, while in the same time they utilize better the winter reserve of moist and the early spring rainfalls. The seeds subjected to hydrophobization endure much better the low temperatures compared to the untreated ones, in all germination conditions at reduced temperature.

A function was found out that helps to determine the different thickness of the polystyrene coating in the cases of different quantities of the treated seeds and their surface.

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# RESPONSE OF BULGARIAN WINTER BARLEY VARIETIES TO DIFFERENT TYPES OF STRESS

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### Abstract

The long phenological development of cereals is the reason for the exceptionally big diversity of factors that have influence on their vegetation and productivity. Depending on the growth stage of the plants, their mechanism of reaction is specific. General criteria for evaluation of the tolerance of a particular genotype towards adverse conditions of the environment is the yield. The aim of this investigation was to study the response of Bulgarian winter barley to different types of stress. The trial was carried out during 2010 – 2014 at Dobrudzha Agricultural Institute. Meteorologically, the years of investigations differed considerably. This allowed evaluating the tested varieties for their resistance to different types of biotic and abiotic stress. The investigated 13 malting and 8 feed cultivars of winter barley were very well differentiated by their productivity, date to heading, yield components, resistance to net blotch and lodging. From the group of malting barley, cultivars Zagoretc, Yaspis, Asparuh, Lardeia and Ahat possessed the highest productivity, and from the group of feed barley these were cultivars Tangra, Hemus and Bori. The structure of yield was specific and was determined not only by the systematic affiliation but also by the peculiarities of the individual genotypes.

Key words: Winter barley, yield, abiotic stress, biotic stress.

## Introduction

Among the cereals, barley is the crop second in distribution in Bulgaria. The territory of the country is of variable and contrasting micro climates. This makes the higher adaptability a main focus of the breeding programs. The problems of barley production are related mostly to its successful over wintering, its resistance to lodging and in the southern regions of Bulgaria – to its drought tolerance. In the recent years, the high infection rate of net blotch is becoming a limiting factor. There are several centers in Bulgaria, where barley is subjected to breeding and improvement work. The main center is the Institute of Agriculture – Karnobat. As a result from the accumulated traditions, rich genetic resources, the methods developed for assessment and selection, cultivars have been realized in different directions of breeding. They occupy a large part in the varietal list of the Republic of Bulgaria. The cultivars developed at the Agrarian University – Plovdiv and at the

Institute of Plant and Genetic Resources – Sadovo are of the malting barley type. They possess short stem, resistance to lodging and high quality. The breeding program of Dobrudzha Agricultural Institute (DAI) was the last to start, and during 2012 - 2015 four new cultivars were released. The conditions in this region allow selection of forms with very good level of winter resistance, which is the main direction of work. The aim of this investigation was to evaluate the response of Bulgarian winter malting and feed barley varieties to different types of stress.

## Material and methods

The investigation was carried out in the trial field of Dobrudzha Agricultural Institute – General Toshevo during 2010 - 2014. The working collection included

- Winter malting barley: Ahat, Asparuh, Emon, Imeon, Kaskadyor 3, Kuber, Lardeia, Obzor, Orfej, Perun, Phlavij, Yaspis and Zagorets;
- Winter feed barley: Aheloj 2, Bori, Hemus, Izgrev, Panagon, Radul, Tangra and Veslets.

The accessions were planted according to the block method in four replications. The size of the plot was 10 m<sup>2</sup>. The sowing norm was 450 germinating seeds/m<sup>2</sup>. At the beginning of February fertilization was done with 0.04 t.ha<sup>-1</sup> active matter of nitrogen. The biometrical measurements were in accordance with the methodology of IPGRI (1994) and UPOV (2003). The following indices were analyzed: days to heading (DH); number of days (from  $1^{st}$  January); plant height (PH), cm; productive tillers of  $1 \text{ m}^2$  (NPT), number; grains per spike (NGS), number; thousand kernel weight (W<sub>1000</sub>), g; test weight (TW), kg; field winter resistance (FWR), score (1 - very low, 5 - moderate, 9 - very high); lodging resistance (L), score (1 - susceptible, 3 - intermediate, 9 - resistant); grain yield (YG) t.ha<sup>-</sup> <sup>1</sup>. The attacking rate of net blotch (Nb) (*Drecheslera teres* Shoem.) was evaluated in field according to a 9-degree scale (1 - very susceptible; 9 - no symptoms). The experimental data were processes with the help of XLSTAT, version 7.5.2. North-east Bulgaria, where Dobrudzha Agricultural Institute is situated, is characterized with soil and climatic conditions favorable for the development of cereals. The low temperatures without snow cover during the winter months are critical. The absolute minimum temperature for this region is - 29.4° C, and the absolute maximum +41.1° C. Due to the frequent flows of cooling ground-level air currents coming from the sea, the spring here is late with 10-15 days. The summer is cool, and the autumn is long, with gradual cooling of the weather. There are two distinct periods of drought, in March – April and July – August. The mean annual sum of rainfalls is 510 mm. The leached chernozem soils are predominant in the region. Due to the heavy composition of soil, the values of the hydrological indices are comparatively high.

## **Results and discussion**

The effects from the main risk factors for the development of winter barley in the region of Dobrudzha have been summarized in previous reports of the research team (Mihova, 2012; Mihova, 2013). They were evaluated on the basis of: differences in the phenological development, level of variation of productivity and its structural components. The obtained

mean yield from feed barley for 9 harvest years (2004 - 2012) was 7.41 t.ha<sup>-1</sup>, and in the group of malting barley it was 6.71 t.ha<sup>-1</sup>.

Growing season	Extreme min t, °C	Extreme max t, °C	Sum of rainfalls (X- VI), mm	Differentiation factors
2010- 2011	-15.6	+30.2	371.1	Late emergence, unfavorable conditions for hardening, occurrence of net blotch
2011- 2012	-19.8	+35.0	469.6	Unsuitable stage for hardening and winter- survival, severe winter conditions, favorable conditions during grain filling
2012- 2013	-14.6	+34.5	320.8	Drought during emergence and beginning of tillering, conditions for stem lodging
2013- 2014	-17.5	28.5	623.8	Favorable winter conditions, occurrence of net blotch, heavy rainfalls reason for mass lodging

Table 1. Differentiation factors by growing season.

Table 2. Mean yield (t.ha<sup>-1</sup>) and variation (CV, %) of winter malting and feed barley\*.

2011	2012	2013	2014	Mean, t.ha <sup>-1</sup>	CV, %				
Malting barley									
5.49 b	6.71 b	9.32 a	5.97 a	6.87 a	24.87				
5.26 b	7.01 a	9.65 a	6.19 a	7.03 a	26.90				
4.52 c	6.12 c	8.87 b	5.82 a	6.33 c	28.89				
5.29 b	6.62 b	9.00 b	5.34 b	6.56 b	26.48				
5.21 b	6.75 b	9.10 b	5.50 b	6.64 b	26.71				
5.25 b	6.25 c	9.24 b	6.26 a	6.75 b	25.54				
5.33 b	7.05 a	9.36 a	5.93 a	6.91 a	25.71				
4.79 c	6.36 c	9.17 b	4.94 c	6.31 c	32.16				
5.51 b	6.57 b	9.63 a	5.43 b	6.78 b	28.98				
4.94 c	6.55 b	9.02 b	4.41 c	6.23 c	33.30				
5.49 b	6.96 a	8.50 c	5.58 b	6.63 b	21.33				
5.89 a	6.85 a	9.48 a	5.91 a	7.03 a	24.05				
5.71 a	6.37 c	9.55 a	6.69 a	7.08 a	23.96				
5.28	6.63	9.22	5.69	6.70					
		Feed barley							
5.81 a	7.20 a	8.28 b	5.51 b	6.70 b	19.23				
5.32 b	6.90 b	9.41 a	6.38 a	7.00 a	24.80				
5.74 a	6.98 b	9.26 a	6.10 a	7.02 a	22.53				
5.27 b	6.92 b	8.39 b	5.81 b	6.59 b	20.92				
5.61 a	6.71 b	8.20 b	6.65 a	6.79 b	15.72				
5.65 a	7.18 a	8.46 b	5.85 b	6.78 b	19.27				
6.01 a	7.21 a	8.80 b	6.15 a	7.04 a	18.31				
5.25 b	6.64 b	8.59 b	5.13 b	6.40 b	25.22				
5.58	6.97	8.67	5.95	6.79					
	$\begin{array}{c} 5.49 \text{ b} \\ 5.26 \text{ b} \\ 4.52 \text{ c} \\ 5.29 \text{ b} \\ 5.21 \text{ b} \\ 5.25 \text{ b} \\ 5.33 \text{ b} \\ 4.79 \text{ c} \\ 5.51 \text{ b} \\ 4.94 \text{ c} \\ 5.49 \text{ b} \\ 5.89 \text{ a} \\ 5.71 \text{ a} \\ 5.28 \\ \hline \\ 5.81 \text{ a} \\ 5.28 \\ \hline \\ 5.81 \text{ a} \\ 5.28 \\ \hline \\ 5.61 \text{ a} \\ 5.65 \text{ a} \\ 6.01 \text{ a} \\ 5.25 \text{ b} \\ \hline \end{array}$	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Malting barley $5.49 \text{ b}$ $6.71 \text{ b}$ $9.32 \text{ a}$ $5.26 \text{ b}$ $7.01 \text{ a}$ $9.65 \text{ a}$ $4.52 \text{ c}$ $6.12 \text{ c}$ $8.87 \text{ b}$ $5.29 \text{ b}$ $6.62 \text{ b}$ $9.00 \text{ b}$ $5.21 \text{ b}$ $6.75 \text{ b}$ $9.10 \text{ b}$ $5.25 \text{ b}$ $6.25 \text{ c}$ $9.24 \text{ b}$ $5.33 \text{ b}$ $7.05 \text{ a}$ $9.36 \text{ a}$ $4.79 \text{ c}$ $6.36 \text{ c}$ $9.17 \text{ b}$ $5.51 \text{ b}$ $6.57 \text{ b}$ $9.63 \text{ a}$ $4.94 \text{ c}$ $6.55 \text{ b}$ $9.02 \text{ b}$ $5.49 \text{ b}$ $6.96 \text{ a}$ $8.50 \text{ c}$ $5.89 \text{ a}$ $6.85 \text{ a}$ $9.48 \text{ a}$ $5.71 \text{ a}$ $6.37 \text{ c}$ $9.55 \text{ a}$ $5.28$ $6.63$ $9.22$ Feed barley $5.81 \text{ a}$ $7.20 \text{ a}$ $8.28 \text{ b}$ $5.32 \text{ b}$ $6.90 \text{ b}$ $9.41 \text{ a}$ $5.74 \text{ a}$ $6.98 \text{ b}$ $9.26 \text{ a}$ $5.27 \text{ b}$ $6.92 \text{ b}$ $8.39 \text{ b}$ $5.61 \text{ a}$ $6.71 \text{ b}$ $8.20 \text{ b}$ $5.65 \text{ a}$ $7.18 \text{ a}$ $8.46 \text{ b}$ $6.64 \text{ b}$ $8.59 \text{ b}$	Malting barley $5.49 \text{ b}$ $6.71 \text{ b}$ $9.32 \text{ a}$ $5.97 \text{ a}$ $5.26 \text{ b}$ $7.01 \text{ a}$ $9.65 \text{ a}$ $6.19 \text{ a}$ $4.52 \text{ c}$ $6.12 \text{ c}$ $8.87 \text{ b}$ $5.82 \text{ a}$ $5.29 \text{ b}$ $6.62 \text{ b}$ $9.00 \text{ b}$ $5.34 \text{ b}$ $5.21 \text{ b}$ $6.75 \text{ b}$ $9.10 \text{ b}$ $5.50 \text{ b}$ $5.25 \text{ b}$ $6.25 \text{ c}$ $9.24 \text{ b}$ $6.26 \text{ a}$ $5.33 \text{ b}$ $7.05 \text{ a}$ $9.36 \text{ a}$ $5.93 \text{ a}$ $4.79 \text{ c}$ $6.36 \text{ c}$ $9.17 \text{ b}$ $4.94 \text{ c}$ $5.51 \text{ b}$ $6.57 \text{ b}$ $9.63 \text{ a}$ $5.43 \text{ b}$ $4.94 \text{ c}$ $6.55 \text{ b}$ $9.02 \text{ b}$ $4.41 \text{ c}$ $5.49 \text{ b}$ $6.96 \text{ a}$ $8.50 \text{ c}$ $5.58 \text{ b}$ $5.89 \text{ a}$ $6.85 \text{ a}$ $9.48 \text{ a}$ $5.91 \text{ a}$ $5.71 \text{ a}$ $6.37 \text{ c}$ $9.55 \text{ a}$ $6.69 \text{ a}$ $5.28$ $6.63$ $9.22$ $5.69$ Feed barley5.81 a $7.20 \text{ a}$ $8.28 \text{ b}$ $5.32 \text{ b}$ $6.90 \text{ b}$ $9.41 \text{ a}$ $6.38 \text{ a}$ $5.71 \text{ b}$ $6.92 \text{ b}$ $8.39 \text{ b}$ $5.81 \text{ b}$ $5.61 \text{ a}$ $5.61 \text{ a}$ $6.71 \text{ b}$ $8.20 \text{ b}$ $6.01 \text{ a}$ $7.21 \text{ a}$ $8.80 \text{ b}$ $6.01 \text{ a}$ $7.21 \text{ a}$ $8.80 \text{ b}$ $6.13 \text{ b}$ $6.64 \text{ b}$ $8.59 \text{ b}$	2011201220132014Mean, t.ha <sup>-1</sup> Malting barley $5.49 \text{ b}$ $6.71 \text{ b}$ $9.32 \text{ a}$ $5.97 \text{ a}$ $6.87 \text{ a}$ $5.26 \text{ b}$ $7.01 \text{ a}$ $9.65 \text{ a}$ $6.19 \text{ a}$ $7.03 \text{ a}$ $4.52 \text{ c}$ $6.12 \text{ c}$ $8.87 \text{ b}$ $5.82 \text{ a}$ $6.33 \text{ c}$ $5.29 \text{ b}$ $6.62 \text{ b}$ $9.00 \text{ b}$ $5.34 \text{ b}$ $6.56 \text{ b}$ $5.21 \text{ b}$ $6.75 \text{ b}$ $9.10 \text{ b}$ $5.50 \text{ b}$ $6.64 \text{ b}$ $5.25 \text{ b}$ $6.25 \text{ c}$ $9.24 \text{ b}$ $6.26 \text{ a}$ $6.75 \text{ b}$ $5.33 \text{ b}$ $7.05 \text{ a}$ $9.36 \text{ a}$ $5.93 \text{ a}$ $6.91 \text{ a}$ $4.79 \text{ c}$ $6.36 \text{ c}$ $9.17 \text{ b}$ $4.94 \text{ c}$ $6.31 \text{ c}$ $5.51 \text{ b}$ $6.57 \text{ b}$ $9.63 \text{ a}$ $5.43 \text{ b}$ $6.78 \text{ b}$ $4.94 \text{ c}$ $6.55 \text{ b}$ $9.02 \text{ b}$ $4.41 \text{ c}$ $6.23 \text{ c}$ $5.49 \text{ b}$ $6.96 \text{ a}$ $8.50 \text{ c}$ $5.58 \text{ b}$ $6.63 \text{ b}$ $5.89 \text{ a}$ $6.85 \text{ a}$ $9.48 \text{ a}$ $5.91 \text{ a}$ $7.03 \text{ a}$ $5.71 \text{ a}$ $6.37 \text{ c}$ $9.55 \text{ a}$ $6.69 \text{ a}$ $7.00 \text{ a}$ $5.28 \text{ b}$ $6.63 \text{ b}$ $9.22 \text{ c}$ $5.69 \text{ c}$ $6.70 \text{ b}$ $5.32 \text{ b}$ $6.90 \text{ b}$ $9.41 \text{ a}$ $6.38 \text{ a}$ $7.00 \text{ a}$ $5.27 \text{ b}$ $6.92 \text{ b}$ $8.39 \text{ b}$ $5.81 \text{ b}$ $6.79 \text{ b}$ $5.27 \text{ b}$ $6.92 \text{ b}$ $8.39 \text{ b}$ $5.81 \text{ b}$ </td				

\*Values with the same letter do not differ significantly

Source of Variation	SS	df	MS	F	P-value	F crit				
	Malting barley									
Total	130,401	51								
Genotype	3,920	12	0,326	2,730	0,009	2,032				
Year	122,174	3	40,724	340,389	1,82E-26	2,866				
Error	4,307	36	0,119							
		Fee	ed barley							
Total	50,027	31								
Genotype	1,445	7	0,206	2,623	0,055	2,487				
Year	46,081	3	15,360	128,974	1,1E-13	3,072				
Error	2,501	21	0,119							

Table 3. Analysis of variance for grain yield.

The harvest years involved in the investigation were characterized with specific combination of meteorological conditions allowing good differentiation of the working collection (Table 1). In three of them the forage forms demonstrated higher productivity, the variation by cultivars being within a narrow range (Table 2).

The year had a significant part in the total variation (Table 3). Lowest mean yield was realized in 2011. The main reasons were the late germination and the slow development of the crops in the autumn of 2010. The mean temperatures dropped down below the biological minimum for this period and at the beginning of winter most cultivars had formed only one tiller. The expectations that there would be second tillering in spring were not fulfilled. In practice, the coefficient of tillering remained low, being close to 1 in feed barley (Tables 4 and 5). At a later stage the amount of rainfalls was sufficient and evenly distributed. Nevertheless, the plants could not compensate the low productive tillering. Productivity was considerably above the average in 2013. Spring was cool and in spite of the longer photoperiod barley did not enter the next phenophase. The conditions favored active spring tillering. As a result sufficient biomass was formed to reduce the negative effect of the retarded development in autumn.

In malting barley the variations of grain yield were highly significant. Highest productivity was demonstrated by cultivars Zagotets, Yaspis, Asparuh, Lardeia and Ahat. The applied k-means clustering placed cultivar Zagorets in the most favorable group in three of the harvest years. Only in 2012 the mean value was low, which was a result from the lower number of grains, while the levels of the other yield components were moderate. During this year there was good differentiation by rate of over wintering, cultivar Zagorets demonstrating lower resistance. Cultivar Yaspis was with stable yield considerably exceeding the mean level. A shortcoming of this cultivar was its susceptibility to net blotch, but even in years with high infection rate such as 2014 it managed to realize its potential. Highest absolute yield during the investigated period was registered in cultivars Asparuh and Orfej, 9.65 and 9.63 t.ha<sup>-1</sup>, respectively.

Variety	,	DH	PH	NPT	NGS	W <sub>1000</sub>	TW	FWR	Nb	L
Ahat	mean	127.5	89.8	694	24.6	46.4	69.3	7.0	8.5	8.0
Allat			09.0		24.0 23.7-	40.4	65.5-			
	min-max		73-110	568-844	25.6	40.3- 53.0	72.0	7	8-9	7-9
Asparuh	mean	129.3	86.0	794	23.1	44.7	67.7	7.5	8.0	7.0
	min-max	123-141	72-104	544- 1012	20.8- 25.7	34.2- 53.9	63.1- 69.8	7-8	7-9	5-9
Emon	mean	127.3	89.8	662	27.0	43.0	70.0	5.0	6.0	6.5
Linon					25.7-	38.9-	69.4-			
	min-max	122-137	68-112	472-836	27.8	46.7	70.5	5	5-7	4-9
Imeon	mean	129.0	86.8	745	28.8	37.9	67.9	5.5	6.0	8.0
	min-max	125 140	73-105	512-996	25.9-	29.1-	62.3-	5-6	5-7	7-9
	mm-max		/5-105	512-990	30.0	48.5	72.0	3-0	5-7	7-9
Kaskadyor 3	mean	129.0	83.8	765	25.8	42.0	66.1	7.5	5.5	5.0
	min-max	125 130	73-101	464-	25.4-	35.4-	61.2-	7-8	4-7	3-7
	шш-шах	125-139		1048	26.0	52.3	70.5			3-7
Kuber	mean	127.5	90.3	736	23.1	45.4	68.8	5.5	8.5	6.5
	min-max	122 138	75-112	552-	20.8-	39.7-	65.3-	5-6	8-9	6-7
	шш-шах			1100	24.4	53.1	72.3	5-0		0-7
Lardeia	mean	127.5	92.3	850	22.4	43.3	68.5	6.0	8.5	6.0
	min-max	122 138	78-110	548-	22.2-	36.1-	65.5-	6	8-9	5-7
	ппп-тах	122-138		1152	22.6	48.2	70.4			
Obzor	mean	129.0	93.0	739	25.0	42.3	66.1	7.5	5.5	3.5
	min-max	125 140	72-106	468-	23.2-	33.7-	60.4-	7-8	4-7	1-6
	ппп-тпах			1000	26.5	49.5	68.4			
Orfej	mean	127.8	88.3	829	26.6	36.8	66.9	7.0	7.5	6.5
	min-max	123 130	75-108	516-	24.1-	31.0-	64.3-	7	7-8	4-9
	ппп-тпал			1164	27.6	42.6	70.9			
Perun	mean	126.8	95.8	758	22.3	47.1	68.9	5.5	7.0	5.5
	min-max	122-138	78-113	508-	20.3-	39.6-	64.6-	5-6	7	4-7
	IIIII-IIIax			1112	24.3	54.1	71.1			
Phlavij	mean	129.5	76.5	673	26.1	43.3	66.8	3.5	4.0	8.5
	min-max	125-140	66-91	448-940	23.6-	35.5-	64.3-	3-4	1-7	8-9
					29.0	50.4	69.1			
Yaspis	mean	128.5	92.8	695	22.8	48.1	68.7	7.5	5.5	5.5
	min-max	124-140	75-112	528-892	20.4-	39.5-	62.5-	7-8	4-7	4-7
	iiiii iiiuX				27.4	55.7	69.0			
Zagorets	mean	128.3	82.8	767	21.9	43.8	69.8	6.0	7.0	8.5
	min-max	124-139	72-97	536-	20.8-	36.3-	6572.8	6	8-6	8-9
2010-2011	mean	139.1	78.3	1020 512.6	23.2 25.7	53.0 43.8	69.3	6.2	7.8	_
2010-2011		124.5	73.3	586.5	23.7	50.8	70.9	6.4		-
	mean			983.1	23.4				-	
2012-2013	mean	125.7	95.2			42.5	67.9	-	-	7.9
2013-2014	mean	123.5	106.2	904.6	24.2	36.4	64.3	-	5.7	5.2

Table 4. Mean, minimum and maximum values for characters in winter malting barley.

Variety		DH	PH	NPT	NGS	W <sub>1000</sub>	TW	FWR	Nb	L
Aheloj 2	mean	127.0	97.0	664	45.8	33.5	64.0	8.0	5.0	2.0
	min-max	122-139	78-114	464-868	43.1-	30.0-	60.1-	7-9	3-7	1-3
D :		100.0	01.0	501	47.6	39.1	67.8	0.5	0.0	7.0
Bori	mean	128.3	91.8	591	50.0	35.4	64.8	8.5	8.0	7.0
	min-max	125-138	67-112	412-776	48.7- 51.2	30.2- 39.9	60.0- 68.2	8-9	7-9	7
Hemus	mean	127.8	96.0	648	41.5	35.3	63.8	8.0	5.0	2.0
	min mor	122-139	77-111	472-840	38.4-	30.5-	59.5-	7-9	3-7	1-3
	min-max	122-139	//-111	472-840	47.5	40.9	67.4		5-7	1-5
Izgrev	mean	128.0	93.0	613	43.6	36.8	63.0	8.0	6.0	4.5
	min-max	123-139	72-117	436-868	41.1-	32.7-	61.0-	7-9	5-7	3-6
	ппп-тах	123-139	/2-11/	430-808	45.7	41.3	66.8		5-7	3-0
Panagon	mean	125.8	93.3	647	48.2	31.4	65.8	6.5	5.5	3.0
	min-max	121-138	74-114	440-876	47.2-	28.5-	62.1-	6-7	4-7	3
	Шп-шах	121-150	/11-	+0-070	49.5	36.8	69.6		<b>-</b> -7	5
Radul	mean	128.0	89.5	662	46.5	33.6	60.4	8.5	6.0	2.0
	min-max	123-139	73-113	460-840	40.5-	28.9-	58.0-	8-9	4-8	1-3
	ппп ппах				49.8	40.3	65.6		-	-
Tangra	mean	128.5	93.5	666	44.3	38.1	65.0	8.0	7.0	7.0
	min-max	124.140	75-110	476-820	42.7-	31.5-	60.6-	7-9	5-9	7
	ппп ших	12 10			46.8	43.4	69.4			
Veslets	mean	126.5	93.0	654	40.7	35.5	64.8	6.5	4	2.0
	min-max	122-138	76-112	420-860	36.0-	30.6-	60.9-	6-7	3-5	1-3
					45.4	38.3	68.8			15
2010-2011	mean	138.8	82.0	447.5	47.6	34.3	67.1	7.3	7.4	-
2011-2012	mean	123.1	74.0	541.5	43.7	40.0	67.8	8.3	-	-
2012-2013	mean	125.3	104.6	811.0	44.7	35.1	60.2	-	-	4.4
2013-2014	mean	122.8	112.9	772.5	44.4	30.5	60.7	-	4.3	3.0

Table 5. Mean, minimum and maximum values for characters in winter feed barley.

In the group of feed barley, cultivars Tangra, Hemus and Bori had productivity above the average. According to its systematic affiliation, Bori belongs to var. parallelum. Peculiarities of these forms are the shorter and more compact spike, and the higher number of grains with lower absolute weight. The biometrical analysis revealed successful combination of traits in this cultivar - 1000 kernel weight was closer to the weight of the other genotypes from var. *pallidum*. The advantage of Tangra is its high resistance to net blotch and lodging, which is a serious problem of production in this region. Cultivar Hemus did not have these advantages but under the conditions of North-east Bulgaria it formed high stable yield determined by the balanced combination of its components. Averaged for the period, lowest yield was read in cultivar Veslets. This cultivar is of winter-spring type of development, which allows planting in autumn or in early spring. It has shorter period to heading, and its major shortcomings are susceptibility to the economically important diseases and to lodging. Nevertheless, cultivar Veslets is among the most successful feed cultivars included in the list of Republic of Bulgaria. It is especially suitable for growing in regions and in years with frequent droughts during heading and formation and filling of grain (Valcheva et al., 2010). The comparison between the two groups of barley showed that the malting accessions had lower stem, higher number of productive tillers, formed lower number of grains but with higher kernel

and test weight, and possessed resistance to lodging. The feed cultivars were of poly-rowed spike type and formed highest number of grains. The differences were determined by the biological peculiarities of the respective systematic affiliations, although varietal specificity was also observed.

Averaged for the period, cultivars Panagon, Veslets and Perun had the earliest dates to heading. Under the conditions of the region, the beginning of the heading stage often occurs at the beginning of May. In such accessions, damages caused by late frosts are often observed in some years resulting in sterility (Valcheva et al., 2010). The reason for the longer period to heading can be unfavorable winter conditions as in 2011. The height of the plant is conditioned by the combination of the meteorological factors during the spring months when vegetation growth is resumed and booting occurs. In 2014 this was the reason for the great positive deviation of the trait. Disturbed correlation was observed with the lodging rate in the new cultivars Ahat, Asparuh, Bori, Imeon and Tangra. The brief rainfalls, which were however accompanied with strong winds at the end of the vegetation period of 2013, allowed better differentiation of the investigated accessions according to the rate of lodging. Year 2014 was especially problematic. The mass lodging severely deteriorated the flow of assimilates to the grain and it remained with low absolute weight and poor physical properties. The following forage barley cultivars were with low resistance to lodging: Aheloj 2, Hemus, Radul and Veslets, and among the malting cultivars - Obzor and Kaskadyor 3. The structure of yield is a dynamic value which is formed by a large number of factors but is also genetically determined. The lower number of productive tillers in some malting cultivars was related to higher values of number of grains per spike (Emon, Imeon) or higher 1000 kernel weight (Ahat, Kuber, Perun and Yaspis). Depending on the conditions, a very good compensatory mechanism between the traits was observed in Orfej and Lardea which guarantied stable productivity over years. Similar correlations were found in the group of forage barley. The greater number of grains in Panagon was in negative correlation with their absolute weight. Bori was characterized with lower productive tillering but the formed tillers were uniform and with similar contribution to yield. In harvest years 2011 and 2014 the occurrence of net blotch was a key factor for the level of yield. The defoliation of the plants reduced the level pf photosynthesis and deteriorated the grain quality. Unfortunately the greater part of the cultivars was with different susceptibility. Lowest scores were referred to Phlavji, Veslets, Aheloj 2, Hemus, Kaskadvor 3, Obzor, Yaspis and Panagon.

## Conclusions

The investigation was carried out in the region of North-east Bulgaria where the soil and climate conditions are very favorable for the development of the winter cereals. The study encompassed four years different by the level of abiotic and biotic stress. The tested winter barley cultivars were very well differentiated by productivity, date to heading, yield components, and resistance to net blotch and to lodging. In the group of malting barley, cultivars Zagorets, Yaspis, Asparuh, Lardeia and Ahat possessed high productivity, and in the group of forage barley these were Tangra, Hemus and Bori. The structure of yield was specific and was determined not only by the systematic affiliation but also by the specificity of the individual genotypes.

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# EFFECTS OF DIFFERENT CONCENTRATIONS OF SOME FOLIAR FERTILIZERS ON THE YIELD OF SEEDS OF OILSEED RAPE (Brassica napus L.)

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## Abstract

Experiment was performed to determine the influence of different concentrations of secondary microelement magnesium sulphate and microelement manganese chelate, on three oilseed rape hybrids: Hybrirock, Petrol, and Speed. The trial was set up in the region of Skopje, beginning of autumn 2013, in 5 variants: Variant 1. Control $\emptyset$ , Variant 2. MgSO<sub>4</sub>-25 %, Variant 3. MgSO<sub>4</sub>-35 %, Variant 4. Mn chelate-20 % and Variant 5. Mn chelate-30 %.The aim of the study was to determine the effects of different concentrations of fertilizers on the yield of seeds of oilseed rape. The following parameters were monitored: plant height (cm), number of branches per plant, number of pods per plant, length of the pods (cm), number of seeds per pods, and seed yield. The selection of these fertilizers and the equivalent concentrations are relevant to the needs of oilseed rape in the vegetation. The results showed that the use of foliar fertilizers with a higher concentration provides a higher yield on the investigated hybrids.

Keywords: concentrations, foliar fertilizers, oilseed rape, seed yield.

## Introduction

Oilseed rape is the second edible oil resource in the world, with high seed oil content of about 40-45% (Sovero, 1993). Winter rape as a plant with high nutritional requirements responds with a significant increase in seed yield both to soil and foliar fertilizer application. This applies not only to nitrogen, potassium and magnesium, but also to other mineral nutrients (Kwiatkowski, 2012). The highest nutrient uptake occurs during the intensive growth of above-ground biomass, 3-4 weeks before flowering, and subsequently during the period of silique and seed formation (Czuba et al., 1995). During this time, nutritional requirements can be met by applying foliar fertilization (Pais, 1983; Harasim and Filipek, 2009). Properly balanced fertilization affects the quality of yield and acts asan element enhancing winter hardiness of winter rape, in particular by providing good magnesium supply to plants (Czuba et al., 1995). The application of different concentrations of foliar fertilizers has different impacts on the seed yield of oilseed rape. The need for and the presence of NPK as primary macro elements are crucial for high yield in *Brassica napus* L. The most important secondary macroelementis magnesium (Mg),

which is present in central atom in the molecule of chlorophyll, and is essential for the absorption of light energy. It also assists in the use of the nutrient substances, and neutralizes acids and toxic compounds created in plants. Magnesium deficiency appears on lower leaves, which turn yellow and white, while the veins remain dark green. Manganese (Mn) as micronutrient works withenzymes to reduce nitrates beforeprotein development in the plant. The deficit of manganese appears on the top sheet as necrotic yellow spots (Omidi H., 2011). Lack of manganese in oilseed rape in terms of soil occurs in soils high in limestone content, and sandy soils (Orlovius K., 2003). According to Kwiatkowski (2012), the application of foliar fertilizers would have a beneficial effect on winter rape productivity, at the same time maintaining high quality of the raw material, even if the rates of soil-applied mineral NPK fertilizers are reduced by ¼. The application of foliar fertilizers is to what extent it relates. Therefore, the objective of this research was to study the effect of different levels of Mg and Mn in applied foliar fertilizers on yield and some yield components of rapeseed (*Brassica napus* L.).

### Material and methods

The experiment with 3 oilseed rape genotypes: Hybrirock, Petrol and Speed was carried out in the season 2013. It was placed on a surface of 600  $m^2$  in surroundings of village Stajkovci (Skopje region -  $42 \circ 02'08.7"$ N;  $21 \circ 30'42.0"$  E), with 27 plots ( $10m^2$ ) deployed in 3 replications, with Random Block System design(RDB) in relation with investigated hybrids and split plot design in relation to the investigated elements. Sowing was carried out on September 10th. Seed norm was 5 kg/ha (5g/plot). Basic fertilization NPK (8:16:24)420 kg/ha wasperformed, of which 2/3 was applied in autumn, before sowing, and 1/3 in spring, at the beginning of growth. Different concentration of fertilizers contains different percentage of sulfur (magnesium 5% and manganese 7%). Total application of MgSO<sub>4</sub> -25% was 5.6 l/ha (variant 2), MgSO<sub>4</sub> -35% was 7.8 l/ha (variant 3), Mn chelate -20% was 4.4 l/ha (variant 4), Mn chelate -30% was used 6.6 l/ha (variant 5). Plots without foliar fertilization(only 100% of NPK) were the control Ø, variant 1. The foliar application is performed on two occasions: I) stage of flower bud development (4<sup>th</sup> April) and II) stage of early flowering; after a period of 15 days. One-phase harvest of oilseed rape was June 27<sup>th</sup>untilJuly10<sup>th</sup>, 2014. At the technical maturity stage, 10 performed from representative plants were collected from each plot and the following yield component were determined: height of the plant (cm), number of branches per plant, number of pods per plant, length of the pod, and number of seed per pod. The results were statistically processed using the analysis of variance and LSD test with SPSS (PSAW 18).

### **Results and discussion**

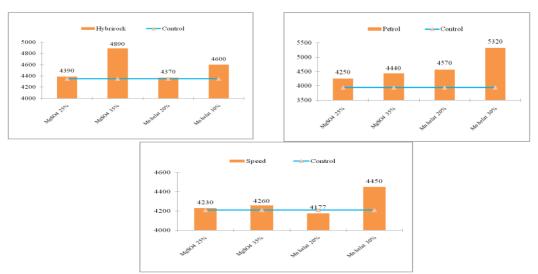
The data presented in Table 1 show mean values of yield components: plant height, number of branches per plant, number of pod per plant, length of the pod, and number of seed per pod. Upon comparison of genotypes, plant height data revealed that Speed was the tallest, followed by Petrol and Hybrirock with mean values of 168cm, 166cm, and 164cm, respectively. These differences between canola hybrid plant height, although minor, might

be due to the differences in genetic background and genetic x environment interaction effects. El-Nakhlaway and Bakhashwain (2009), (according by Maestro 1995; Reddy and Reddy, 1998), reported that brassica varieties differed significantly regarding their plant heights. Certain cultivars might be sensitive to environmental factors while others may be tolerant (Sana et al., 2003). The mean value of plant height under the foliar fertilizer rates showed some differences in plant height among investigated genotypes compared with control variants. However, only in the case of Speed the used treatments showed that maximum plant height - 180 cm, with Mn chelate (20%) (Variant 4), and Mn chelate (30%) (Variant 5), (Table 1). These results might be due to the positive effect of manganese on the growth development of the stem and leaf of certain hybrids (Speed) which was reflected on taller plants. For the number of branches, the mean values in Table 1 revealed that the number of branches was 4.6 on the hybrid level (Hybrirock and Petrol), while Speed showed lower number of branches (3.9). Analysis of the applied fertilizers revealed that the best results were obtained from higher concentrations of magnesium sulphate and manganese chelate. Mean values for the number of pods per plant showed that the higher concentration of foliar application (variants 3 and 5) resulted in a higher number of pods/plant. Increasing foliar application rate increased number of pods per plant, but the increase was not statistically significant. Similar results were obtained on each of the examined hybrids (Table 1). Foliar application fertilizer had no significant effect on the length of pod. The length of pod ranged from 6.6cm to 7.4cm indifferent variants and hybrids. Petrol has the longest pod (average 7cm) among hybrids. The obtained results might be due to the positive effect of foliar application of different concentrations and environment on the development of pods which was exhibited as longer pods, but the differences were not statistically proved. The length of the pod, besides the effect of environment and fertilizer application, largely depends on genetic constitution of examined hybrids. Number of seeds per pod (Table 1) has not been affected by application of different concentrations of fertilizers on any of investigated hybrids.

Hybrid	Variant	Concentrations/	Plant	Number	Number	Length	Number
		fertilizer	height (cm)	branches/plant	of pods/plant	of pod (cm)	of seed / pod
	1	Control Ø	170	4.4	127.5	6.9	36.9
×	2	MgSO <sub>4</sub> 25%	160	4.8	116.9	6.8	34.4
Hybrirock	3	MgSO <sub>4</sub> 35%	150	4.5	87.2	7.0	29.9
ybri	4	Mn chelate 20%	170	4.3	84.5	6.5	31.5
Ηì	5	Mn chelate 30%	170	5.2	131.0	6.6	33.9
	average		164	4.6	109.4	6.8	33.3
	1	Control $\emptyset$	160	4.1	99.0	7.0	31.1
	2	MgSO <sub>4</sub> 25%	170	4,5	95.5	7.4	32.8
Petrol	3	MgSO <sub>4</sub> 35%	170	5.2	130.4	6.9	37.7
Pet	4	Mn chelate 20%	160	4.0	102.4	7.0	32.3
	5	Mn chelate 30%	170	5.0	114.3	6.9	31.7
	average		166	4.6	108.3	7.0	33.1
	1	Control $\emptyset$	150	4.0	86.5	7.0	30.1
	2	MgSO <sub>4</sub> 25%	170	3.3	70.3	6.9	30.4
Speed	3	MgSO <sub>4</sub> 35%	160	4.2	88.3	7.0	28.9
	4	Mn chelate 20%	180	3.6	81.5	6.6	31.1
	5	Mn chelate 30%	180	4.5	94.3	7.1	36.5
	average		168	3.9	84.2	6.9	31.4

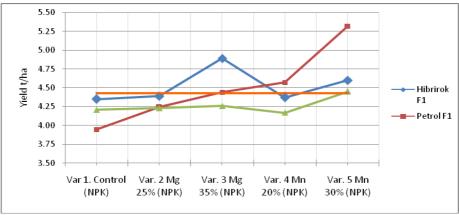
Table 1. Morphological characteristics

Seed yield of oilseed rape(t/ha). The average value of seed yield from foliar applications for all examined hybrids and various concentrations in relation to Control  $\emptyset$  is represented in Graph 1. Hybrirock was point out with applied concentration of MgSO4 35% and gave the highest yield of 4.89 t/ha. All other applied foliar applications gave lower yield /ha, but still above the control variant. Previously carried survey in the region showed lower results (2.6 t/ha) at Hybrirock, but without foliar application of fertilizers (Spirkoska M., 2013). Petrol is on the border, with 4.44 t/ha, but nevertheless comes before Speed, which gave yield of 4.26 t/ha.Referring to the variant 4, all hybrids have higher yield compared with ControlØ, where Petrol with 4.57 t/ha and Hybrirock with 4.37 t ha are prominent. The higher concentration (30%) of Mn chelate - variant 5, also positively influenced the return of the oilseed rape yield compared to Control $\varnothing$ . Thus, it can be observed from the data that Petrolhad the best yield (5.2 t/ha), followed by Hybrirock with 4.6 t/ha, and the lowest yield at this concentration of foliar fertilizers was obtained from Speed with 4.45t/ha. From the values presented in Graph 1, it should be emphasized that Speed showed better results of seed yield compared with ControlØ, except in variant 4, where lower results were obtained.



Graph. 1.Progress of the seed yield for each hybrid per variants

The effect of the application fertilizers by variants of investigated hybrids in relation to the mean values obtained from all varieties (Graph. 2), show a positive impact in relation to the set concentrations and an increase in terms of seed. Thus values of all varieties are higher than the Control $\emptyset$ . Foliar fertilizers with MgSO<sub>4</sub> in trials in Germany showedyield of up to 1.40 t/ha, depending onsoil conditions and sulfur deficiency (Orlovius K., 2003). In the survey, results showed that the highest yield was obtained with higher concentrations of fertilizers, or Mg SO<sub>4</sub> 35% and Mg chelate 30%. Nabipoura A. (2009), has cited Bybordi et al. (2001) and Malakouti (2002) where it showed the highest seed weight obtained by applying 15 kg/ha Mn and 200 kg/ha urea. The lower concentrations of the applied fertilizers (MgSO<sub>4</sub> 25% and Mn chelate 20%), gave the same positive effect in relation to the Control $\emptyset$ , but their impact is to some extent lower than the average of all varieties valued.



Graph. 2. Average yield per hybrids vs.variants of all hybrids

The higher concentrations  $MgSO_4$  35% and Mn chelate 30% provide the highest seed yield per unit area (Graph. 2).

14010 11 202 1001 001				
(I) Variant	(J) Variants	Mean Difference (I-J)	Std. Error	Sig.
Control	Mg SO <sub>4</sub> 25%	-121 0000	233.71759	0.616
	Mg SO <sub>4</sub> 35%	-361.0000	233.71759	0.153
	Mn chelate 20%	-203.3333	233.71759	0.405
	Mn chelate 30%	-621.0000*	233.71759	0.024
LSD	0.52			

Table 2. LSD test between variants

\*The mean difference is significant at the .05 level.

Statistical data processing showed significance only at variant 5 (Mn chelate 30%), compared to the Control $\emptyset$ , thus producing a higher yield of seed (0.62 t/ha) compared to the Control  $\emptyset$ . Significance has been confirmed by the result obtained from LSD test (0.52).

## Conclusions

From the research in relation to the application of foliar fertilizers, both elements magnesium and manganese show solid results in both applied doses. The results showed that effects of  $MgSO_4$  35% and Mn chelate 30% concentration on yield were significant (variant 3 & variant 5). Hence one can conclude and recommend to the producers of oilseed rape to optimize utilization of fertilizers, using efficiently balanced nutrition byfoliar application, i.e. not use only traditional microelements NPK, but place a special emphasis on the microelements. It should be emphasized that these are preliminary results, and that the research will continue in the future in order to determine the adequate concentration of fertilizers for foliar application.

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#### UDC: 633.11(497.2)"2011/2014" Original scientific paper

# COMPARATIVE STUDY ON COMMON WHEAT (*Triticum aestivum* L.) CULTIVARS GROWN UNDER THE AGROECOLOGICAL CONDITIONS OF SOUTH CENTRAL BULGARIA

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#### Abstract

During the period of 2011-2014 in South Central Bulgaria a field experiment was researched. The cultivars common wheat 'Enola', 'Miryana', 'Andino', 'Azimut' and 'Trakiyka', were studied. The experiment was applied in block design with 4 replications and 15 m<sup>2</sup> plot size, after predecessor sunflower. The growing of plants was performed in compliance with the standard technology. The aim of the present investigation was to carry out a comparative study of the yield and quality of some common wheat cultivars grown in South Central Region of Bulgaria. The analysis of the results showed that the highest grain yield was obtained from Andino variety (7300 kg ha<sup>-1</sup>), followed by Azimud (7000 kg ha<sup>-1</sup>) and the lowest one – Trakiyka (4000 kg ha<sup>-1</sup>). Among the studied common wheat cultivars, the highest values of 1000 kernels weight and the test weight were reported for 'Miryana' (50,3 g and 81,0 kg, respectively) and the highest wet gluten content was established in 'Azimut' cultivar (27,0%). The lowest value of the test weightwas reported for 'Trakiyka' cultivar (73,3 kg); of the 1000 kernels weight – for 'Azimut' cultivar (34,3 g) and of the wet gluten content for 'Andino' cultivar (22,0%).

Key words: wheat, cultivars, grain yield, thousand kernel (grain) weight, test weight, gluten.

#### Introduction

One of the major ways of increasing the yield and improving the wheat grain quality under the current market conditions is the establishment of new highly productive cultivars and their introduction into practice. Establishing the proper cultivar structure depending on the concrete agroecological conditions of the region can significantly increase grain yield and quality. That requires a very good understanding of the characteristics of the different cultivars, in order to be able to make the right choice (Barzegar, 2002); (Capouchová & Petr, 2004); (Delibaltova&Kirchev, 2010); (Ilieva, 2011); (Ivanova *et al.*, 2009);(Williams *et al.*,2008). Studies of a number of authors show that the amount of grain yield is closely related to the cultivar, the use of farming machinery and the soil and climatic conditions of the region (Ivanova *et al.*, 2010); (Kirchev&Stoeva, 2003); (Nankova&Penchev, 2006); (Stoeva *et al.*,2006). Therefore, in order to use the full productive potential of the cultivar, the proper choice of suitable cultivars for each agroecological region is a decisive factor for obtaining high yields. That necessitates systemic studies of the cultivars in the different regions of the country (Garg *et al.*, 2005); (Tonev *et al.*, 2005); (Yanchev& Ivanov, 2012). The aim of the present investigation was to carry out a comparative study of the yield and quality of some common wheat cultivars grown in South Central Region of Bulgaria.

# Material and methods

A field experiment with common wheat was carried out on the experimental field of the village of Carimir, (South Central Bulgaria) in the period 2011 - 2014. The test was performed by means of a block method with four replications; experimental field area -15 $m^2$ , after the predecessor sunflower. The following cultivars were tested; 'Enola', 'Miryana', 'Andino', 'Azimut' and 'Trakiyka'. All the stages of the established technology for wheat growing were followed. The period of the research (2011-2014) is characterized with variety of temperature and rainfall conditions which enables to evaluate the reaction of the studied varieties in accordance with their yields and quality characteristics under different climatic conditions. Rainfalls in autumn and during the critical spring period are decisive for the development of the wheat plants. The mean annual precipitation sums during October - March, which formed the autumn-and-winter moisture reserves in soil during the experimental years 2011-2012, 2012-2013 and 2013-2014 were higher with 61, 101 and 7 mm, respectively, than the mean sums of the long - term period. During April-May when plants were at stages booting and heading, the mean annual precipitation sum in 2012 and 2013 was lower than the mean long - term value, while in 2014 this sum was higher with 119 mm. In June and July (during grain filling-maturation) rainfalls in harvest year 2014 was higher with 62 mm, in comparison to the long - term period, while in 2012 and 2013 they were with 44.3 and 14.0 mm, respectively lower. The most favourable for plant growth and development was the third experimental year (2013-2014), followed by the second (2012-2013), and unfavourable was the first year (2011-2012), of the experiment, having an effect on yield and grain quality of common wheat. Soil tillage included single disking (10-12 cm) after harvesting of the previous crop, and double disking after the main fertilization. The area was treated by  $N_{120}P_{80}$  and the whole quantity of the phosphorous fertilizer and 1/3 of the nitrogenous fertilizer were applied before main soil tillage. The remaining amount from the nitrogen norm was applied before the beginning of permanent spring vegetation. Triple super phosphate and ammonia nitrate were used. Sowing was completed within the agrotechnical term optimal for this region at sowing norm 550 germinating seeds/m<sup>2</sup>. Control of weeds, diseases and pests was done with suitable pesticides when necessary. Harvesting was done at full maturity. The grain yield is determined with standard grain moisture of 13%. The indices grain yield (kg ha<sup>-1</sup>); kernels weight (g), test weight (kg); wet gluten content (%) were determined. For the purpose of determining the quantity dependence between the studied indicators, the experimental data was processed according to the Anova Method of dispersion analysis, and the differences between the variants were determined by means of the Dunkan's Multiple Range Test (Dunkan, 1995).

### **Results and discussion**

Data about the grain yield (Table 1) show that 'Andino' cultivar surpassed the other cultivars included in the experiment, when reporting the harvest by years and in average for the period of study. In result of the better moisture provision of plants during the vegetation period and in the period of grain formation and ripening, higher yields were obtained in 2013-2014 compared to 2011-2012 and 2012-2013. Under the conditions of that season they varied from 5000 kg ha<sup>-1</sup> for 'Trakiyka' cultivar to 8400 kg ha<sup>-1</sup>for 'Andino'. The cultivars 'Miryana', 'Enola' and 'Azimut' yielded by 2900, 2300 and 200 kg ha<sup>-1</sup>less than 'Andino' and by 500, 1100 and 3200 kg ha<sup>-1</sup>more than 'Trakiyka', respectively. All the differences were statistically significant. The lowest grain yields during the studied period were reported in the first year of the experiment (2011-2012) and it was due to the insufficiency of moisture during the critical stages of growth and development of the wheat plants. In that season the lowest yield was obtained from 'Trakiyka' cultivar – 3000 kg ha<sup>-1</sup>, which was by 16,7% less compared to 'Miryana', by 83,3% less compared to 'Enola', by 190% less compared to 'Azimut' and by 200% less compared to "Andino' cultivar.

Variety		Years of study		Average for the
	2011-2012	2012-2013	2013-2014	period
	(kg/ha)	(kg/ha)	(kg/ha)	(kg/ha)
Enola	5500°	6400 <sup>c</sup>	6100 <sup>c</sup>	6000
Miryana	3500 <sup>b</sup>	4500 <sup>b</sup>	5500 <sup>b</sup>	4500
Andino	6000 <sup>e</sup>	7500 <sup>e</sup>	8400 <sup>e</sup>	7300
Azimut	5700 <sup>d</sup>	7100 <sup>d</sup>	8200 <sup>d</sup>	7000
Trakiyka	3000 <sup>a</sup>	4000 <sup>a</sup>	5000 <sup>a</sup>	4000
LSD <sub>5%</sub>	98.3	263.5	180.4	

Table1. Grain yield (kg ha<sup>-1</sup>)

In 2012-2013 the yields obtained ranged from 4000 kg/ha to 7500 kg/ha. The differences between the studied cultivars were statistically significant. The grain yield during the second experimental year was by 24,5% higher in average than in 2011-2012 and by 12,5% lower than in 2013-2014. The highest grain yield, in average for the three years of the study, was reported for 'Andino' cultivar – 7300 kg/ha, surpassing the cultivars 'Azimut', 'Enola', 'Miryana' and 'Trakiyka' by 4,2; 16,7; 55,6 and 75,0%, respectively. The results of the dispersion analysis of the grain yield data of five common wheat cultivars (Table 2) show the significant effect of the studied factors:  $\eta$  99 of the cultivar and  $\eta$  96 of the year. Interactions between the studied factors were also significant  $\eta$  87 for the grain yield.

Source of Variation	SumofSquare	Df	Mean Square	Sig of F	%η <sup>2</sup>
Variety	118762026.70	4	29690507.0	.000	99
Years	36100480.00	2	18050240.0	.000	96
2-way Interactions	8551253.33	8	1068906.7	.000	87
Residual	1338800.00	45	29751.1		

The results of the quality characteristics of the studied cultivars are presented in Table 3. The thousand kernel weightis a cultivar specific trait influenced by the agroecological conditions and the growing technology. The results show that thanks to the favourable climatic conditions during the wheat vegetation period in the third year, the values of that characteristic were significantly higher compared to the other experimental years. In 2013-2014 'Miryana' cultivar produced the largest grains (52.0 g of 1000 grains), followed by 'Enola' (46,0g), while 'Azimut' had the smallest grains (38,0 g). The differences between the cultivars were statistically significant. The thousand kernel (grain) weightof 'Andino' and 'Trakiyka' cultivars had similar values (42,0and 43,0 g, respectively) and the difference was statistically insignificant.

Drought weather combined with high air temperatures at the stage of grain formation and ripening in 2011-2012 had an effect on grain weight. The lowest weight of thousand kernel (grain)was reported for 'Azimut' cultivar (31,0 g). The cultivars 'Miryana' and 'Enola' surpassed in weight of thousand kernel (grain) the cultivars 'Andino' and 'Trakiyka' by 30,5% and 17,75%, respectively, the differences being significant.The largest grains in average for the period 2011-2014 were reported for 'Miryana' cultivar (50,3 g), followed by the cultivars 'Enola' (46,3 g), 'Trakiyka' (41,7 g) and 'Andino' (39,3 g). The lowest values of that characteristic were established for 'Azimut' cultivar.

Test weight is a commercial indicator showing grain quality and it plays an important role in determining the market price. That characteristic of the studied cultivars in the years of the experiment varied from 69,0 to 82,0 kg. The test weight of the cultivars 'Enola', 'Andino' and 'Miryana' had similar values in all the three years of the study, which is an evidence that for those cultivars, the characteristic is slightly influenced by the climatic conditions in the separate years and it depends more on the cultivar. In contrast to 'Enola', 'Andino' and 'Miryana', the test weight of the cultivars 'Azimut' and 'Trakiyka' is significantly influenced by the climatic conditions and less by the cultivar.

Index	Variety		Years of study					
		2011-2012	2012-2013	2013-2014	(2011-2014)			
	Enola	45.0 <sup>d</sup>	48.0 °	46.0 °	46.3			
Th	Miryana	49.0 <sup>e</sup>	50.0 °	52.0 <sup>d</sup>	50.3			
Thousand kernel	Andino	36.0 <sup>b</sup>	40.0 <sup>b</sup>	42.0 <sup>b</sup>	39.3			
(grain) weight, g	Azimut	31.0 <sup>a</sup>	34.0 <sup>a</sup>	38.0 <sup>a</sup>	34.3			
	Trakiyka	40.0 °	42.0 <sup>b</sup>	43.0 <sup>b</sup>	41.7			
	LSD <sub>5%</sub>	4.62	3.70	2.80				
	Enola	77.0 <sup>b</sup>	79.0 <sup>b</sup>	80.0 <sup>b</sup>	78.7			
	Miryana	80.0 °	81.0 °	82.0 °	81.0			
Test weight, kg	Andino	78.0 <sup>b</sup>	79.0 <sup>b</sup>	80.0 <sup>b</sup>	79.0			
	Azimut	72.0 <sup> a</sup>	74.0 <sup> a</sup>	79.0 <sup>ª</sup>	75.0			
	Trakiyka	69.0 <sup>ª</sup>	73.0 <sup>ª</sup>	78.0 <sup>ª</sup>	73.3			
	LSD <sub>5%</sub>	1.80	1.98	1.11				
	Enola	25.0 <sup>b</sup>	29.0 <sup>b</sup>	26.0 <sup>b</sup>	26.6			
Wet gluten, %	Miryana	23.0 <sup> a</sup>	25.0 <sup>a</sup>	28.0 °	25.3			
-	Andino	25.0 <sup>b</sup>	27.0 <sup> a</sup>	14.0 <sup>a</sup>	22.0			
	Azimut	27.0 °	29.0 <sup>b</sup>	25.0 <sup>b</sup>	27.0			
	Trakiyka	22.0 <sup> a</sup>	26.0 <sup> a</sup>	30.0 °	26.0			
	LSD <sub>5%</sub>	1.93	3.07	1.82				

Table 3. Quality of the grain

The highest test weight of wheat grain, in average for the period of the study, was reported for 'Miryana' cultivar (81,0 kg.), followed by 'Andino' and 'Enola' (79,0 and 78,7 kg, respectively), and the lowest values were reported for the cultivars 'Azimut' and 'Trakiyka' (75,0 and 73,3 kg, respectively). The results obtained about the wet gluten content showed that in the studied cultivars, this characteristic was greatly influenced by the production year. In the first year of the experiment the lowest values of that characteristic were reported for the cultivars 'Trakiyka' (22,0%) and 'Miryana' (23,0%), while the highest values of that characteristic for the same two cultivars were established in the third experimental year (30,0 and 28,0%, respectively). In 2014 a comparatively low wet gluten content was reported for 'Andino' cultivar (14,0%), i.e. the significant amount of rainfall at the ripening stage led to washing out of the wet gluten, while in 'Trakiyka' cultivar, rains caused an increase of the values of that characteristic (30,0%). In the second experimental year (2012-2013), the wet gluten content in the cultivars 'Miryana', 'Trakiyka' and 'Andino' varied from 25,0 to 27,0%, the differences being slight and statistically insignificant, while the cultivars 'Enola' and 'Azimut' produced grains with 11,5% higher wet gluten content. The highest wet gluten content, in average for the whole study period 2011-2014, was established in 'Azimut' cultivar (27,0%) and the lowest - in 'Andino' cultivar (22,0%). The two-factor dispersion analysis of grain quality characteristics (Table 4) reveal that the commercial year with its specific climatic conditions had the greatest influence on the wet gluten content  $-\eta$  97. The climatic conditions during the production year had a significant, although not so strong effect on the hectoliter weight ( $\eta$  80) and the weight of 1000 grains ( $\eta$  57), while the genetic characteristics of the cultivar were crucial for those two characteristics  $-\eta$  90 and  $\eta$  94, respectively. The effect of the cultivar is significant for the wet gluten content  $-\eta$  61.

Index	Source of Variation	SumofSquare	DF	Mean Square	Sig of F	$\eta^2$
Thousand	Variety	1857.07	4	464.27	.000	94
kernel	Years	151.23	2	75.62	.000	57
(grain) weight, g	2- Way Interactions	68.93	8	8.62	.004	38
	Residual	112.50	45	2.50		
	Variety	458.42	4	104.61	.000	90
Test	Years	218.00	2	109.00	.000	80
weight, kg	2- Way Interactions	84.9	8	10.61	.000	61
	Residual	55.38	45	1.23		
	Variety	235.07	4	58.77	.000	61
Wet	Years	88.23	2	44.12	.000	37
gluten, %	2- Way Interactions	494.43	8	61.80	.000	77
	Residual	148.75	45	3.31		

Table 4. Analysis of variance for quality of the grain

The relationship between the production year and the cultivar is statistically proven both for the wet gluten content ( $\eta$  77) and the hectoliter weight ( $\eta$  61), as well as for the weight of 1000 grains ( $\eta$  38).

# Conclusions

The highest grain yield, in average for the experimental period 2011-2014, under the climatic conditions of South Central Bulgaria, was obtained from 'Andino' cultivar (7300 kg/ha), followed by 'Azimut' (7000 kg/ha), and, the lowest one – from 'Trakiyka' cultivar (4000 kg/ha). 'Andino' cultivar produced by 300; 1300; 2800 and 3300 kg ha<sup>-1</sup>higher grain yield than the cultivars 'Azimut', 'Enola', 'Miryana' and 'Trakiyka', respectively. Among the studied common wheat cultivars, the highest values of the thousand kernel (grain) weight and the test weight were reported for 'Miryana' (50,3 g and 81,0 kg, respectively) and the highest wet gluten content was established in 'Azimut' cultivar (27,0%). The lowest value of the test weight was reported for 'Trakiyka' cultivar (73,3 kg); of the thousand kernel (grain) weight– for 'Azimut' cultivar (34,3 g) and of the wet gluten content – for 'Andino' cultivar (22,0%).

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# THE APPLICATION OF MINERAL FERTILIZERS IN DRIP FERTIGATION IN SUGAR BEET PRODUCTION

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## Abstract

Physiological role of boron is especially important in nutrition of sugar beet. While the boron is necessary in protein synthesis, it plays an important role in the metabolism of carbohydrates. Boron also has an important role in cell division as well as in the development of the root system. In order to reduce the difference between the yield of beet root and sugar depending on irrigation and fertilization, field trial was set on farm Topolik in Baranja in 2014. The drip irrigation and fertigation with NPK + boron was used in this investigation. A significantly higher yield and sugar content are obtained by applying of fertigation.

Key words: sugar beet, fertigation, boron, digestion.

# Introduction

Irrigation with drainage, soil management and application of the appropriate tillage adjusted to pedoclimatic conditions are required agrotechnical measures in agricultural production (Madar, S., 1986., Tomić, F., 1988, Turšić, I., et al. 1999.). However, modern intensive agricultural production in Croatia is very often oriented mainly on the use of the complex and concentrated mineral fertilizers (macro elements) without the application of organic manure. Due to their physiological importance, microelements are also essential for the plant development and may become a limiting factor in the production and quality of agricultural products, and are neglected (Anić, J., 1968). Previous research has shown that the application of macroelements, especially on soils which do not contain enough available microelements, has given good results in the production of sugar beet (Bergmann, W., 1992). Boron helps in the transport of carbohydrates and reduces transpiration coefficient which is very important during arid years, especially in the soils of the smaller water regime. Boron reduces the intensity of breathing, which is also especially when sugar beet root is kept in storage. The first symptoms of insufficient boron nutrition in sugar beet are appearing on the younger leaves which first become brown, then turn black, and then they wilt. In case of higher boron deficiency, the top part of the sugar beet root starts to rot, after which rot affects the root itself (Picture 1). This "disease" is also known under the name of rot or "black heart" and significantly reduces the yield or the percentage of sugar in the healthy parts of the "diseased" sugar beet (Pospišil, M. 2013).



Picture 1. Boron deficiency symptoms on sugar beet leaf and root (Bergmann, W. 1992)

Light textured (sandy) soils are more prone to nutrient leaching, especially nitrate nitrogen and boron. With the soil, climate also affects the boron nutrition in sugar beet (Kristek, A. 1982). A very small number of agricultural areas in the Republic of Croatia have been irrigated and Croatia is on one of the last places in Europe in irrigated surfaces.

The lack of systematic irrigation is a significant risk factor of production, so in the past few years it has happened that drought (a lack of water and dissolved biogenic elements) significantly reduced the yield of sugar beet root, and especially of sugar yield per hectare (Šoštarić, J. et al. 1997, Šimunić, I. et al. 2007).

# Material and methods

The effects of drip irrigation were investigated on Belje d.d. and Topolik farms in Baranja in 2014. Beside control variant (without fertigation), effect of fertigation on yield and quality of sugar beet was investigated. Area of investigated plot was  $8.000 \text{ m}^2$ . Half of the plot was fertigated. The investigation was carried out in four randomized repetitions  $(8 \times 1000 \text{ m}^2)$ .



Picture 2. Soil monolith, trial location and used water for irrigation (Topolik farm, 2014)

Depending on content of available water in the soil,  $11-27 \text{ m}^3\text{ha}^{-1}$  of water was added to soil. 480 g of NPK 20+20+20 (Polyfeed) and 330 ml ha<sup>-1</sup> of Folibor L were added to the water. The sugar beet root yield and sugar content were measured in four repetition and the results were processed by analysis of variance.

# Results

Samples for analysis of basic physical and chemical characteristics, as well as the evaluation of the soil suitability for irrigation were taken from the soil profile before starting the experiments on experimental plots. Physical and chemical characteristics of the soil are shown in tables 1, 2, 3, 4. According to the mechanical composition (Škorić, A., 1965), the soil is of silty clay texture, with the average water and air capacity (Gračanin, M., 1950). Soil type is humogley (Husnjak, S. 2014).

r										
Depth cm	Soil mechanical properties in Na- pyrophosphate, % particle content, diameter mm									
Deptirent	Coarse sand	Fine sand	Coarse silt	Fine silt	Clay					
	2.0-0.2	0.2-0.063	0.063-0.02	0.02-0.002	< 0.002					
0-35	3.2	18.0	27.2	33.5	18.1					
35-58	3.1	118.024.735.718.5								

Table 1. Soil mechanical properties of field trial

Table 2. Soi	l physical	properties	of	field trial

Donth	Water con	Water canacity		Vater capacity Air capacity		Total porosity		Bulk density	
Depth	Water capacity		All capacity		Total polosity		Vol.	Real	
cm	vol %	Mark	vol %	Mark	vol %	Mark	g cm <sup>-3</sup>	g cm <sup>-3</sup>	
0-35	39.1	Medium	10.3	Medium	49.4	Porous	1.30	2.58	
35-58	40.8	Medium	9.1	Medium	49.9	Porous	1.31	2.62	

	Soil water rete	Physiologically		
Depth cm	15 bar*	6.25 bar*	0.33 bar*	active water**
	% vol.	% vol.	% vol.	% vol.
0-35	16.9	24.1	39.1	22.2
35-58	16.6	25.7	40.8	24.2

Table 3. Soil water retention of field trial

<sup>\*</sup>15 bar= wilting point; 6.25 bar = lento capillary water; 0.33 bar = soil water capacity

\*\* difference between soil water capacity and wilting point

With regard to the chemical properties of the soil (Table 4), it is of neutral reaction, of high organic content, moderately supplied in phosphorus and potassium, and poorly to moderately supplied in boron (Berger, K.C. and Truog, E., 1944, Jackson, M.L., 1958).

Table 4. Soil chemical properties

							Available 1	nutrients	В
Depth	pН			Humus			conte	ent	ppm
cm			Description	%	Descriptio	on	mg/100	g soil	
	H <sub>2</sub> O	KCl					$P_2O_5$	K <sub>2</sub> O	
					rather h	high	14.1	20.6	0.30
0-35	7.85	7.08	neutral	3.65	organic				
					content				
					rather h	high	8.2	15.3	0.18
35-58	7.65	6.91	neutral	3.01	organic				
					content				

The applied drip irrigation, with the application of mineral Polyfeed fertilizers NPK 20:20:20 and boron dissolved in water for irrigation, and adapt to the amount of precipitation (Table 5 and 6).

Table 5 Monthly precipitation and mean monthy air temperatures in 2014

						Month						
Ι	II	III	IV	V	VI	VII	VIII	IX	Х	XI	XII	Total
				P	recipita	tion, mm						
22.3	30.3	40.5	68.9	251.4	58.7	117.6	84.0	119.7	86.3	6.7	52.7	939.1
			]	Mean mo	nthly ai	r tempera	ature, °C	2				Year
4.1	5.9	10.0	13.3	16.1	20.8	22.3	21.2	17.3	13.8	9.0	4,2	13.2

Polyfeed fertilizer 20-20-20 contains 10,3 % amid nitrogen, 5,8 % nitrate and 3,9 % amonium. It also contains 1000 ppm Fe, 500 ppm Mn, 200 ppm B, 150 ppm Zn, 110 ppm Cu and 70 ppm Mo.

Date of irrigation	Water (mm)	Fertilizer quantity i	n irrigation water
		NPK 20:20:20 (g)	Boron (ml)
17/07	11 (+ rain)	480	330
18/07	26	480	330
26/07	27	480	330
02/08	22	480	330
09/08	10 (+ rain)	480	330
26/08	25	480	330
Total	121	2880	1980

Table 6. The applied quantity of water in irrigation (mm), the amounts of dissolved mineral fertilizers and boron micronutrient.

The number of plants per hectare, the yield of sugar beet per hectare and sugar percentage (digestion) in sugar beet root was measured. The percentage of sugar in sugar beet root was measured at the Faculty of Agriculture in Zagreb and the College of Agriculture in Križevci. Analyses have been conducted by refractometric method in four repetitions. The results obtained have been processed by analysis of variance (table 7).

Variation			Repetitions					
		Ι	II	III	IV			
No fertigation	Yield t/ha	85.20	87.60	82.55	96.30	87.91		
(control)	% sugar	14.50	16.80	14.00	15.00	15.08		
Fertigation	Yield t/ha	112.95	125.10	110.01	111.30	114.84*		
	% sugar	17.20	16.00	16.40	15.80	16.35*		

Table 7 The influence of	of fortigation on an	or boot root wield (t/l	na) and direction (0/ sugar)
rapie 7. The influence (	n terugation on sus	2a Deel tool vieta $(1/1)$	ha) and digestion (% sugar)

<sup>\*</sup>LSD, 5 %

A significantly higher yield and sugar content are obtained by fertigation with macro and micro nutrients (especially Boron).

# Conclusions

Application of the fertigation in 2014 has increased the root yield by 14.6% and digestion for 8.42%. Sugar beet which was not treated with boron had lower digestion for 1.27% in relation to the treated one.

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# THE EFFECT OF DIFFERENT FERTILIZER FORMS IN PEA (*Pisum sativum* L.) ON YIELD AND YIELD COMPONENTS

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## Abstract

This study carried out to investigate the effect of different fertilizer forms and dosses on yield and yield components in pea. Research carried out at Diyarbakir of Turkey 2014 growing season. Two pea cultivars (Utrillo and Cambados) used from seed companies. Organo-mineral fertilizers, nitrogen + organo-mineral fertilizers, chicken manure, chicken manure + nitrogen, bacteria, nitrogen + bacteria, triple super phosphate ( $P_2O_5$ ), nitrogen + triple super phosphate ( $P_2O_5$ ), ammonium sulfate, ammonium nitrate and diammonium phosphate (DAP) fertilizers used as fertilizer forms. Fertilization was applicated on soil and only one time at sowing. Experiment setup two factor randomized complete block design with three replications. Sowing was making in March. Plot will be design 4 m length and 5 rows. Seed amount will be 50 seeds per square meter. Every parcel was space of 1 m to avoid interference with fertilizer. The effect of fertilizer forms and dosses on number of pods plant<sup>-1</sup>, number of seeds plant<sup>-1</sup>, biological yield and grain yield was significant.

Keywords: Pea, Pisum sativum L., fertilizer, fertilization.

# Introduction

Pea (*Pisum sativum* L.) is an important nutritive vegetable and is mainly cultivated as cool season crop, but is also cultivated in summer in hilly areas. It is considered as an important cultivated legume next to soybean, groundnut and beans. Pea is a leguminous crop belonging to the family leguminoseae, which contains higher amount of protein and is an excellent human food. In the World, peas are grown in an area of 10 379 535 hectares, with total production of 10 979 946 tons (FAO, 2014). In 2013 growing season in Turkey, it occupies an area of 1200 ha with production of 3200 metric tons. In Turkey the cultivation and production of peas are very low as compared with most of the countries of the world (TUIK, 2014). The application of fertilizers is one of the primary methods for improving the availability of soil nutrients to plants. Fertilizing can positively change the plant traits, but heavy uses of chemical fertilizers have created a variety of economic, environmental,

and ecological problems. The use of fertilizer is considered to be one of the most important factors to increase crop yield. The use of inorganic fertilizers in combination with organic materials is able to give the desired and sustainable crop yields than the sole use of inorganic fertilizer or animal manure (Masarirambi et al., 2011). Application of chicken manure, which is the most widely used for growing crops, increases the growth of crops. Bio-fertilizers can help meet the demands of sustainable, productive agriculture at low cost. Rhizobial inoculants have been applied to legume crops for over 120 years as bio-fertilizers, and inoculants carrying plant-growth-promoting rhizobacteria (PGPR) have been used in agriculture for over half a century. Phosphorous is to be an essential element and its application is to be important for growth, development and yield (Servani et al., 2014). The nitrogen (N) is a vital nutrient for the activity of plant organs. It is a fraction of many components, so plant growth can be affected by the amount of nitrogen. The present study was under taken to verify the effect of different fertilizer forms on the performance of pea varieties.

# Material and methods

The experiment was conducted at the experimental farm of the Faculty of Agriculture, Dicle University, Diyarbakir, Southeastern Anatolia of Turkey, in the spring of 2014. The soil was a clay-loam, moderate in organic matter content (1.2%), pH:7.6 and low in phosphorus contents (16.15 kg P ha-<sup>1</sup>). Normal P content South East Anatolia of Turkey soils is insufficient for phosphorus by 65% (30-60 kg ha<sup>-1</sup> to 60-90 kg ha<sup>-1</sup>). P fertilizer application is necessary in experiment area soil (16.15 kg P ha-<sup>1</sup>) due to deficit P. Soil properties were given in Table 1.

	The son properties of study site								
Deep (cm)	CaCO <sub>3</sub>	Н	$\begin{array}{c} P_2O_5\\ (kg ha^{-1}) \end{array}$	K <sub>2</sub> O (%)	Organic matter (mg kg <sup>-1</sup> )	Fe	Cu	Zn	Mn
0-20			0.132	12.1	0.79	3.769	1.316	0.415	3.84
20-40	10.26	7.24	0.166	12.6	0.71	3.879	1.312	0.620	4.35

Table 1. The soil properties of study site

Two pea cultivars (Utrillo and Cambados) used from private seed companies. Organo-mineral fertilizers, nitrogen + organo-mineral fertilizers, chicken manure, chicken manure + nitrogen, bacteria, nitrogen + bacteria, triple super phosphate ( $P_2O_5$ ), nitrogen + triple super phosphate ( $P_2O_5$ ), ammonium sulfate, ammonium nitrate and DAP fertilizers used as fertilizer forms. Fertilizer forms and its dosses were given in Table 2.

Table 2. Pertilizer and dosses	
Fertilizier forms	Dosses
Control	0.0
Rhiziobiumleguminoserum	1/100
Diamonyumfosfat (NH <sub>4</sub> ) <sub>2</sub> HPO <sub>4</sub>	$30 \text{ kg N ha}^{-1}$
Ammonium Sulfate (NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	$30 \text{ kg N ha}^{-1}$
Ammonium Nitrate NH <sub>4</sub> NO <sub>3</sub>	$30 \text{ kg N ha}^{-1}$
Triple Superfosfat ( $P_2O_5$ )	$50 \text{ kg ha}^{-1}$
Ammonium Nitrate + Triple Super Phosphate	$30 \text{ kg N ha}^{-1} + 20 \text{ kg ha}^{-1}$
Chicken Manure	2000 kg ha <sup>-1</sup>
Chicken Manure + Triple Super Phosphate	$2000 \text{ kg ha}^{-1} + 50 \text{ kg ha}^{-1}$
Chicken Manure + Ammonium Nitrate	$15 \text{ kg N ha}^{-1} + 2000 \text{ kg ha}^{-1}$
Organo-Mineral (Vermi-Compost)	4000 ml ha <sup>-1</sup>
Organo-Mineral + Ammonium Nitrate	$15 \text{ kg N ha}^{-1} + 4000 \text{ ml ha}^{-1}$
Ammonium Nitrate + Triple Super Phosphate	$15 \text{ kg N ha}^{-1} + 50 \text{ kg ha}^{-1}$

Table 2. Fertilizer and dosses

Fertilization was applicate on soil and only one time at sowing. Experiment set up two factors randomized complete block design with three replications. Cultivars were assigned to main plots and fertilization forms to sub-plots. Sown was in the second week of March, 2014. Plot will be design 4 m length and 5 rows. Seed amount will be 50 seeds per square meter. Every parcel was space of 1 m to avoid interference with fertilizer. Plants were harvested in the first week of July 2014. Supply irrigation water applied two times at pre-flowering and podded stage. Observations were recorded on plant height, number of branches, pods and seeds plant-<sup>1</sup>, 100 seed weight and grain yield kg ha<sup>-1</sup>. The data were statistically analyzed by using MSTATC computer package program.

#### **Results and discussion**

Analysis of variance and mean values of the effect of different fertilizer forms and dosses on agronomic traits in pea were given at Table 3, 4 and 5.

Sources of Variation	Seed weight plant <sup>-1</sup>	Plant height	No. of pods plant <sup>-1</sup>	No. of seeds plant <sup>-1</sup>	No. of seeds pods <sup>-1</sup>	100 seed weight	Pod length	Pod width	Biological yield	Seed yield
Variety Fertilizer Variety x		89.924** 11.221 10.705	0.333 1.714** 1.388*	8.986 28.667** 38.838**	0.289 0.487 0.590	5.388 3.890 5.407	36.957 287.388* 67.669	7.683** 2.173** 6.813**	16003.7** 3729.402* 3696.749*	62.69** 712.67** 399.5**
fertilizer Errror	1.350	10.378	0.607	3.905	0.590	2.951	64.530	0.941	1668.159	70.500

Table 3. The analysis of variance (mean of square).

\*\* P<0.01, \*P< 0.05 significant

Analysis of variance revealed that the effect of fertilization forms on seed weight plant<sup>-1</sup>, number of pods and seeds plant<sup>-1</sup>, pod length and width, biological yield and seed yield was significant. Variety x fertilizer forms interaction was significant for seed weight plant<sup>-1</sup>, number of pods and seeds plant<sup>-1</sup>, pod width, biological yield and seed yield (Table 3).

Mean values of the effect of different fertilizer forms and dosses on agronomic traits in pea were given at Table 4 and Table 5.

The effect of fertilizer forms on seed yield  $\text{plant}^{-1}$  was significant, and seed yield  $\text{plant}^{-1}$  ranged from 6.79 g to 8.86 g, the highest value was obtained from 30 kg ha<sup>-1</sup> ammonium nitrate (AN) + 20 kg Triple Super Phosphate (TSP) ha<sup>-1</sup>, followed by 2000 kg ha<sup>-1</sup> chicken manure + 50 kg ha<sup>-1</sup> TSP, and these values higher than control group. The lowest seed yield plant<sup>-1</sup> was 2000 kg ha<sup>-1</sup> only chicken manure.

The effect of fertilizer forms on number of pods plant<sup>-1</sup> was significant, and the highest value with 5.83 pods/plant was in 15 kg AN ha<sup>-1</sup> + 50 kg TSP ha<sup>-1</sup> application, the lowest value with 4.13 pods/plant was in 30 Ammonium Sulfate (AS) application. Some researchers was reported that different fertilizer forms and dosses on number of pods plant<sup>-1</sup> (Akhtar et al., 2003, Achakzai and Bangulzai 2006 and Kumar 2011) were significant. For example, Akhtar et al., (2003) 46 kg P ha<sup>-1</sup> Achakzai and Bangulzai (2006) 100+60+40 kg NPK ha<sup>-1</sup>, Alam et al., (2010) N<sub>50</sub>P<sub>26</sub>K<sub>42</sub>S<sub>12 +</sub> 1 kg ha<sup>-1</sup> Mo, B and Zn, Kumar (2011) 12 kg ha<sup>-1</sup> phosphate and bacteria applications had significantly effect for high number of pods plant<sup>-1</sup>. However, Hamed Mir et al., (2014) reported that biomanure for this traits was no gignificant.

significant, but Fayetörbay et al., (2014) reported that separately chicken manure and bacteria applications were decreased the number of pods plant<sup>-1</sup>, conversely, was chicken manure + bacteria application was increased up this trait.

Fertilizer forms	Seed	No. of	No. of	Pod	Pod
and dosses	weight	pods	seeds	Length	Width
	plant <sup>-1</sup> (g)	plant <sup>-1</sup>	plant <sup>-1</sup>	(mm)	(mm)
Control	7.43 bcd	4.41 cd	14.94 f	78.16 ab	16.24 b
Bacteria	7.33 bcd	5.15 a-d	20.20 a-d	77.26 ab	18.19 a
AS (30 kg ha <sup>-1</sup> )	7.97 a-d	4.13 d	18.17 cde	72.91 bc	16.69 ab
$DAP (30 \text{ kg ha}^{-1})$	7.94 a-d	5.06 a-d	22.03 ab	87.85 a	16.14 b
AN (30 kg ha <sup>-1</sup> )	8.43 abc	4.73 a-d	19.50 b-e	64.62 c	16.08 b
TSP $(50 \text{ kg ha}^{-1})$	7.43 bcd	4.53 bcd	17.66 def	83.81 ab	15.92 b
AN+TSP (15 kg ha <sup>-1</sup> +50 kg ha <sup>-1</sup> )	8.67 ab	5.83 a	22.83 a	83.93 ab	15.97 b
AN+TSP $(30 \text{ kg ha}^{-1}+20 \text{ kg ha}^{-1})$	8.86 a	5.36 abc	20.78 abc	85.56 a	16.22 b
Chicken manure (2000 kg ha <sup>-1</sup> )	6.79 d	4.48 bcd	16.95 ef	88.30 a	15.89 b
Chicken+TSP (2000 kg ha <sup>-1</sup> + 50 kg ha <sup>-1</sup> )	8.83 a	4.86 a-d	19.74 b-e	88.66 a	16.39 b
Chicken+AN (2000 kg ha <sup>-1</sup> + 15 kg ha <sup>-1</sup> )	7.31 cd	4.70 a-d	21.39 ab	82.08 ab	16.08 b
OrganoMineral (4000 ml ha <sup>-1</sup> )	8.40 abc	5.65 ab	19.77 a-e	85.29 ab	16.05 b
$OM+AN (4000 \text{ ml ha}^{-1}+ 15 \text{ kg ha}^{-1})$	7.66 a-d	4.18 cd	18.21 cde	84.21 ab	16.31 b
Mean	7.93	4.85	4.321	81.74	16.32
LSD					
Fertilizer	1.347*	1.205**	3.055**	12.42**	1.5**
Variety x fertilizer	1.975*	1.278*			2.121**

Table 4. The effect of fertilizers and dosses on agronomic traits in pea

\*\* P<0.01, \*P< 0.05 significant

Number of seeds  $plant^{-1}$  was affected by fertilization forms, and the highest no. of seeds  $plant^{-1}$  was obtained by 15 kg AN ha-<sup>1</sup> + 50 kg TSP ha-<sup>1</sup> applications according to control group and other applications. It was determined that no. of seeds  $plant^{-1}$  was increase up with the forms and applications when compared with control group.

Pod length and width were affected by fertilization forms, and the longest pod with 87.85 mm was obtained from 30 kg DAP ha<sup>-1</sup> application, followed by 2000 kg chicken manure +

15 kg AN ha<sup>-1</sup> application with 88.30 mm. the shortest pod with 64.62 mm was obtain from only 30 kg AN ha<sup>-1</sup> application. Maximum pod (18.19 mm) width was obtained from bacteria application, and minimum pod width (15.89 mm) was in chicken manure application, but ammonium sulphate and bacteria application except, other fertilizer forms were in the same statistic group. Several other researchers also revealed that the effect of different fertilizer dosses and forms on pod traits was significant (Akhtar et al., 2003, Achakzai and Bangulzai 2006, Kumar 2011). Kakar et al., (2002) reported that maximum pod length (8.49 cm) were recorded in plots with received 75+120+120 or 75+120+0 kg NPK ha-1, respectively, while Carr et al., (2000) noted that no advantage in pea plant stand on seed yield, or seed quality resulted from application of N and P fertilizers The effect of different fertilizer dosses and forms on biological yield was significant, ranged from 3909 kg ha<sup>-1</sup> to 4688 kg ha<sup>-1</sup>. Maximum biological yield was in 15 kg AN ha<sup>-1</sup>+ 50 kg TSP ha<sup>-1</sup> application, minimum value was in 4000 ml ha<sup>-1</sup> organo-mineral application. Similarly, Khorgamy and Farnia (2009) reported that phosphate and zinc fertilizer applications were increased biological yield.

Fertilizer forms	Biological	Seed	No. of	Plant	100
and dosses	yield	yield	seeds	height	seed
	$(\text{kg ha}^{-1})$	$(\text{kg ha}^{-1})$	pods <sup>-1</sup>	(cm)	weight
					(g)
Control	4579 abc	893.5 abc	4.08	28.67	22.09
Bacteria	4107 cde	711.6 ef	3.91	31.88	23.75
$AS (30 \text{ kg ha}^{-1})$	4346 a-de	888.6 abc	4.41	31.38	21.93
$DAP (30 \text{ kg ha}^{-1})$	4394 a-d	879.9 abc	4.45	30.17	21.06
$AN (30 \text{ kg ha}^{-1})$	4048 de	820.7 b-e	4.37	30.33	23.05
$TSP (50 \text{ kg ha}^{-1})$	4688 a	990.9 a	3.91	29.25	22.59
AN+TSP (15 kg ha <sup>-1</sup> +50 kg ha <sup>-1</sup> )	4238 а-е	1003 a	3.87	28.75	22.46
$AN+TSP (30 \text{ kg ha}^{-1}+20 \text{ kg ha}^{-1})$	4623 ab	932.7 ab	3.87	31.92	21.67
Chicken manure (2000 kg ha <sup>-1</sup> )	4373 а-е	749.4 def	3.70	30.42	22.31
Chicken+TSP (2000 kg ha <sup>-1</sup> + 50 kg ha <sup>-1</sup> )	3976 de	817.0 b-e	4.54	28.29	22.94
Chicken+AN (2000 kg ha <sup>-1</sup> + 15 kg ha <sup>-1</sup> )	4413 a-d	848.4 bcd	3.95	28.50	21.76
OrganoMineral (4000 ml ha <sup>-1</sup> )	3909 e	621.2 f	3.91	30.63	23.28
$OM+AN (4000 \text{ ml ha}^{-1}+15 \text{ kg ha}^{-1})$	4151 b-e	774.6 cde	4.41	28.04	23.63
Mean	4296	838.4	4.11	29.86	22.50
LSD					
Variety x fertilizer	66.98*	**			
Fertilizer	47.36*	**			

Table 5 the effect of fertilizers and dosses on agronomic traits in pea

\*\* P<0.01, \*P< 0.05 significant

Maximum seed yield (1003 kg ha<sup>-1</sup>) was obtained from 15 kg AN ha<sup>-1</sup>+ 50 kg TSP ha<sup>-1</sup> application followed by 50 kg TSP ha<sup>-1</sup> with 990.9 kg ha<sup>-1</sup>. Minimum seed yield (621.2 kg ha<sup>-1</sup>) was in 4000 ml ha<sup>-1</sup> organo-mineral application. Some researchers reported that grain yield increased by the phosphate and nitrogen applications (Servani et al., 2014, Fayetörbay et al., 2014). Stevovicet al., (2006) revealed that high grain yield was in 40 kg N ha<sup>-1</sup> application, Chen et al., (2006) noted that 135 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> application was produced greater yield than 10% as compared with control. Bacteria application was no signifiant on

seed yield, this finding are in agreement with the findings of Ngenove ark. (2012). However, Drobereiner and Campelo(1976) and Kaya et al., (2002) noted that grain yield of pea was increased by bacteria application. Our findings, chicken manure had low grain yield, are in disagreement with the findings of Balkcomet al., (2003),Göksu(2012), Fayetörbayet al., (2014).

The effect of fertilizer forms on plant height, number of seeds  $\text{pod}^{-1}$  and 100 seed weight was no significant. However, some earlier researchers reported that fertilization in pea for the traits had positive and significant response (Akhtar et al., 2003, Amjad et al., 2004, Kumar 2011,). For example, Akhtaret al., (2003) reported that phosphate application in pea on number of seeds  $\text{pod}^{-1}$  had a significant effect, and 46 or 69 kg  $P_2O_5$  ha<sup>-1</sup> doses of phosphate and 12 kg  $P_2O_5$  ha<sup>-1</sup> + bacteria (Kumar (2011)) were advisable dose.

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# IMPACT ASSESSMENT OF THE SOME HERBICIDE FORMULATIONS ON THE PRODUCTIVITY OF 6 COMMON WHEAT VARIETIES BY CLUSTER ANALYSIS

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#### Abstract

The purpose of this development is to examine the effect of the treatment with several herbicides and herbicide mixtures upon main biometrical indicators and the productiveness of six common wheat varieties. The seed treatment is displayed with the following preparations for weed control: Axial one (pinoxaden + florasulam) - 100 ml/da; Axial 050 EC (pinoxaden) - 90 ml/da; Traksos 045 EC (pinoxaden + clodinafop) - 120 ml/da; Logran 20 WG (triasulfuron) - 3.75 g/da; Lintur 70 WG (triasulfuron + dicamba) - 15 g/da. The field experiment was conducted between 2012 - 2014, in the conditions of meadowcinnamon soil. It was made a comparison with the control option and the treatment options through a cluster analysis. It was determines that the control option, in relation to all indicators, was most distant from: 1) option 7, with a coefficient of 28.180 for Illico variety, followed by Ingenio variety with a coefficient of 19.984; 2) option 4 for Enola variety with a coefficient of 17.769, followed by Apolon and Diamond varieties with coefficients 10,345 and 9,640 respectively; 3) option 2 was most strongly expressed for Bologna variety with a coefficient of 18.503, followed by diamond and Apolon varieties with an equal coefficient of 17.153. The classification and grouping of the options through a hierarchical cluster analysis allow a higher objectivity in the evaluation of the complex impact of the treatment options upon the structural elements for the examined wheat varieties.

Key words: wheat, cluster analysis, yield.

# Introduction

The yield and the grain quality are the result of complex interactions between the specific genetic systems that combine the best in specific growing conditions. The productive capacity of wheat varieties are genetically determined and are the result of many years of breeding work. Wheat is the crop that is highly dependent on the specific microclimate conditions. This arises from its special conditions of growth and its stepwise development. The introduced wheat varieties are continuously explored in Bulgaria. Emphasis is placed on the quality of the grain, the greater ecological plasticity, and the resistance to drought and frost of the plants. The accumulated extensive research experience shows that varieties

exhibit their productive capacities to varying degrees, depending on the specific soil and climatic characteristics of the region (Tzenov et al. 2009; Ivanova, 2009; Ilieva, 2011). The ecological plasticity and stability of are optimizing the share of the variety as a factor. (Cheleev et al. 1993; Delibaltova et al. 2014). The determining role of agrotechnics for wheat is arises from the need of weed control. The right choice of plant protection measures ensures the production of high quality products (Dixit et al. 2011; Delchev, 2012; Delchev et al. 2014;). The herbicides are the primary factor in modern integrated technologies for weed control.

Cluster analysis is a method for evaluating and shaping natural groups based on several indicators simultaneously (Ilchovska et al. 2014). This method of approach was applied to evaluate the lines and varieties, where pooling is a method of assessment according to defined, in this case, biometric indicators. The merger is carried out per similarities and differences, and promotes comprehensive assessment of the studied herbicides. Cluster analysis is a comparative analysis which assesses and forms natural groups based on several signs simultaneously. The purpose of the study is to assess the impact of some herbicides and herbicidal mixtures on major biometric indicators for six varieties of common wheat and their grouping by applying cluster analysis.

## Materials and methods

The field experiment was conducted between 2012 and 2014 in the area of the training - experimental terrain of Agricultural Faculty at Thracian University, Stara Zagora. The soil type was characterized as a typical meadow-cinnamon soil. The profile power was 103 - 105 cm with well formed horizons.

Ten treatment options of crops were examined:

- Control without herbicide treatment
- Axial 1 1000 ml/ha
- Lintur + Tracsos 150 g/ha + 1200 ml/ha tank mixture
- Logran + Tracsos 37.5 g/ha + 1200 ml/ha tank mixture
- Lintur + Axial 150 g/ha + 900 ml/ha tank mixture
- Logran + Axial 37.5 g/ha + 900 ml/ha tank mixture
- Lintur + Axial 150 g/ha + 600 ml/ha separate treatment
- Lintur + Tracsos 150 g/ha + 1200 ml/ha separate treatment
- Logran + Axial 37.5 g/ha + 600 ml/ha separate treatment
- Logran + Tracsos 37.5 g/ha + 1200 ml/ha separate treatment

Grouping the ten examined treatment options was performed by a hierarchical cluster analysis (Ward, 1963, Dyuran, B., P. Odelly, 1977). The results from the clustering were presented graphically by dendrograms, showing sequence of unification between the options and the formed clusters. The data processing was performed through the statistical program SPSS.

# **Results and discussion**

In the conducted examination it was made an assessment of the similarity and the remoteness of the control's impact and the impact of other treatment options of common wheat (varieties: Diamond, Enola, Apolon, Bologna, Illico and Ingenio) and their grouping on the base of main

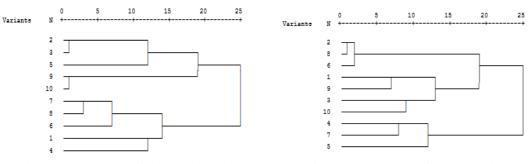
biometrical indicators through the application of a cluster analysis. Experimental data from the 3-rd field test were analyzed, which included 10 treatment options with herbicides and herbicide mixtures. The evaluation of the tested products was made on the base of the following indicators: plants height, wheat-ears length, ears number, grains number in an ear, grains mass in an ear, mass of 1000 grains and hectoliter weight. Grouping of the examined options was made by a hierarchical cluster analysis. The method of intergroup combining was used (Ward, 1963, Dyuran, B., P. Odelly, 1977). The results were presented in a table with the steps of combining the clusters and intergroup distances, as well as graphically, through dendrograms. The dotted horizontal line of the dendrograms showed a rescale distance, at which the clusters were formed.

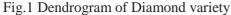
	Di	iamond		Enola				
Steps	Cluster Co	ombined	Coefficients	Steps	Cluster Co	ombined	Coefficients	
1	2	3	3.398	1	2	8	2.661	
2	9	10	3.826	2	2	6	3.698	
3	7	8	4.983	3	1	9	6.530	
4	6	7	7.221	4	4	7	7.454	
5	2	5	9.486	5	3	10	7.713	
6	1	4	9.640	6	4	5	9.679	
7	1	6	10.981	7	1	3	10.294	
8	2	9	13.339	8	1	2	13.713	
9	1	2	17.153	9	1	4	17.769	

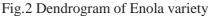
Table1. Combining clusters and inter-group distances of varieties "Diamond" and "Enola"

Results from the cluster analysis are the following:

*Diamond Variety:* Ten treatment options were put to a cluster analysis. The dendrogram shows that the options were separated in three cluster groups (Fig. 1). In the first cluster group, options 2 and 3 were with very close results. The second group included options 9 and 10, the third one – options 7 and 8, and option 6 joined later. There was an obvious resemblance in the second cluster group. It was due to the effective control of the broad-leaved weeds, which was performed by the separate application of Logran pesticide. Option 7 was close to option 8. The small Euclidean distance between them could be explained with the treatment with a same preparation. The preparation was first in the experiment scheme. The separate insertion of the first two herbicides contributed to the formation of plants with similar biometrical indicators. The impact of the imported anti-wheat products was weaker (Axial and Tracsos). The control option, in reference to all observed indicators, was closest to option 4 with a coefficient of 9.640, and it was most distant to option 2 with a coefficient of 17.153 (Table1).





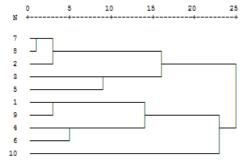


*Enola Variety:* The sort's dendrogram (Fig.2) shows that the options formed three cluster groups. The first group was with very close results and it included options 2, 8 and 6. They were at a comparatively small distance 2.661- 3.698. The reported similarity and the small Euclidean distances were due to the measured close biometric indicators. The second cluster group included the other options 1, 9, 3, 10 which acted equally. They combined with the first group at the next stage. They were least similar and with least Euclidean distance between them. The third cluster group included 3 options: 4, 7 and 5. It was due to the high degree of similarity of the examined indicators. In reference to all observed indicators, the control option was closest to option 9 with a coefficient of 6.530, and it was most distant to option 2 gives a high assessment on the treatment with the combined herbicide Axial one. The complex assessment of the examined indicators is a result from the treatment's impact upon annual wheat and broad-leaf weeds.

Apolon Variety: The sort's results show that the options were divided into three clusters. It can be seen by the dendrogram (Fig. 3) that the options were grouped as follows: I-st cluster – options 7, 8 and 2 were grouped, close to other indicators; II-nd cluster – options 3 and 5; III-rd group – options 4 and 6. The first cluster included options 7 and 8 which were very close in distance, and later option 2 joined. The treatment was performed with the same anti-broad-leaf herbicide (Lintur). Subsequently, the conditions for the crop's growth were made equal. The later joining of option 2 was due to the effect of florasulam, which was one of the active substances in the combined treatment Axial one. There was closer distance for options 3 and 5. It could be explained with the application of Lintur, although it was a tank mixture. The third cluster group options were characterized with close indicators: number of wheat-ears, number of grains in an ear, etc. It was a result of the equal activity and the positive effect of the imported herbicide mixtures, as one of the treatments was Logran. In reference to all indicators, the control was most distant from options 2 and 10. The applied treatments Axial one, and Logran and Tracsos, which were imported separately, showed high herbicide activity and removed the rival weeds. Thus, they were favorable to the plants growth, as well as obtaining higher structural indicators with coefficients of 17.153 and 15.472, respectively. Option 9 was closest to the control with a coefficient of 4.067.

		Apolon		Bologna				
Steps	Cluster Combined		Coefficients	Steps	Cluster Co	ombined	Coefficients	
1	7	8	2.690	1	4	6	2.417	
2	2	7	3.911	2	2	3	3.893	
3	1	9	4.067	3	4	8	5.061	
4	4	6	5.402	4	1	9	7.124	
5	3	5	7.324	5	2	5	9.009	
6	1	4	10.345	6	4	10	10.237	
7	2	3	11.765	7	2	4	12.671	
8	1	10	15.472	8	2	7	15.658	
9	1	2	17.153	9	1	2	18.503	

Table 2. Combing clusters and inter-group distances of "Apolon" and "Bologna" varieties



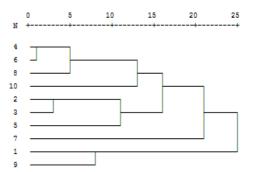


Fig.3.Dendrogram of Apolon variety

Fig.4. Dendrogram of Bologna variety

*Bologna Variety:* Ten treatment options were put under a comparative analysis. The dendrogram (Fig. 4) shows that there was high similarity between options 4 and 6, 2 and 3. Grouping the first two options was due to the anti-broad-leaf treatment Logran. Options 2 and 3 were very similar by the crop's examined structural elements. Results were due to the high effectiveness of the applied Axial 1 and the tank mixture of anti-broad-leaf Lintur and anti-wheat Tracsos. The control option, in reference to all others, was most distant from option 2, and closest to option 9. The Euclidean distance for this variety was once more greatest between the control option and option 2. The biometric indicators were lowest for the weeded control. While, after treatment with Axial 1, there were high values of the biometric indicators.

*Illico Variety*: Ten treatment options were put under a comparative analysis. The dendrogram (Fig. 5) shows that the options were divided one more time into two main cluster groups. The first cluster group, with very close distances in it, included two subclusters. One of the sub-clusters had options 3, 5, and 4, and later option 2 joined (Fig. 3). The second cluster included one main cluster with options 8, 6, 9, and 1. The presence of small group distances between options 3 and 5 in the first cluster group was due to the effectiveness of the applied anti-broad-leaf Lintur, although it was in a mixture. Option 4, and later, option 2, showed similarity of indicators. When defining the biometrical indicators, small differences were reported: number of grains in an ear (33.05-33.89), mass of 1000 grains (72.9-73.7g). The applied treatments showed a high degree of effectiveness in respect of the crop's weeding.

		Illico		Ingenio				
Steps	Cluster Co	Cluster Combined		Steps	Cluster Co	Cluster Combined		
1	3	5	2.711	1	6	9	3.356	
2	3	4	3.421	2	7	8	3.830	
3	2	3	6.607	3	3	6	4.614	
4	6	9	6.619	4	3	4	6.525	
5	2	8	8.920	5	2	3	7.804	
6	2	6	9.435	6	1	2	10.183	
7	1	2	10.977	7	7	10	11.079	
8	1	10	16.689	8	1	5	11.677	
9	1	7	28.180	9	1	7	19.984	

Table 3. Combing clusters and inter-group distances of "Illico" and "Ingenio" varieties

The dendrogram shows that for Iliko variety, the control of all indicators was most distant from options 7, 10, 2, and it was closest to option 2 (Table 3). Close by other indicators were options 3, 5, 4; 2 and 3; 6 and 9. Practically, they could be taken as equal. Inter-group distances between the given options showed that they, selfsame, differed by complex evaluation of the examined indicators.

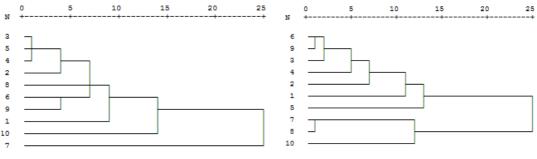


Fig.5. Dendrogram of Illico variety

Fig.6. Dendrogram of Ingenio variety

*Ingenio Variety:* The dendrogram (fig. 6) shows that options were grouped into two clusters. The first cluster included 4 sub-clusters. Options 6, 9, and 3 were in the first sub-cluster. They were characterized with a high similarity. Later, option 4 joined, which formed the second cluster. Next, option 2 joined the second cluster, followed by options 1 and 5. The control option was most distant from options 5 and 7 with coefficients 11.677 and 19.984, and it was closest to option 2 with a coefficient of 10.183. For Ingenio variety, the given results showed a greatest effectiveness from the application of broad-leaf Lintur, as a tank mixture and separately. The mixtures and their herbicide activity eliminated the weeds' tormenting effect and gave conditions for wheat's normal growth and structural elements formation of high potential. The performed classification and grouping of options through a hierarchical cluster analysis allowed high objectivity in evaluation the

complex impact of treatment options upon the biometrical indicators of the examined wheat varieties.

# Conclusions

Option 9 was closest to the control option with a coefficient of 4.067 for Apolon variety, followed by Enola variety with a coefficient of 6.530 and Bologna variety with 7.124. It can be explained ecologically that the treatment can go without it. For Illico and Ingenio varieties, the closest option to the control was option 2 with coefficients of 10.977 and 10.183. The effectiveness was weakly expressed, so applying herbicides and herbicide mixtures with higher effectiveness could be offered.

In reference to all indicators, the control option was most distant from:

- Option 7, with a coefficient of 28.180 for Illico variety, followed by Ingenio variety with a coefficient of 19.984;
- Option 4, with a coefficient of 17.769 for Enola variety, followed by Apolon and Diamond varieties, with coefficients 10.345 and 9.640 respectively;
- Option 2, as it was most strongly expressed for Bologna variety with a coefficient of 18.503, followed by Diamond and Apolon varieties with an equal coefficient of 17.153.

The options `classification and their grouping through a hierarchical cluster analysis allow higher objectivity in evaluating the complex impact of the treatment options upon the structural elements of the examined wheat varieties.

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# HEAD RICE YIELD AND WHITE RICE YIELD OF ARPA AND ONICE -TWO NEWLY INTRODUCED RICE VARIETIES (Oryza sativa L.) IN THE REPUBLIC OF MACEDONIA

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## Abstract

This paper presents the results of investigation of the head rice yield, white rice yield and one thousand grain weight of Arpa and Onice, two newly introduced Italian rice varieties, grown in Macedonia. Two standard varieties were used for comparison: Prima riska (Macedonian variety) and San Andrea (domesticated Italian variety). Field experiments (randomized blocks) were set-up during vegetation years 2013 and 2014. The head rice yield (the weight of whole white rice grains remaining after milling, as a percentage of the total weight of the paddy) was determined by laboratory milling, conducted on a paddy quality testing machine. The results showed the highest head rice yield in the variety Arpa (62.32%), and the lowest in the standard variety San Andrea (56.54%) during the two years of investigation. The highest average white rice yield per hectare was obtained in the standard variety *Prima riska* (5783.53 kg ha<sup>-1</sup>), while in the other standard (*San Andrea*) the average white rice yield was the lowest (4977.82 kg ha<sup>-1</sup>), compared to all the varieties in the investigation. For the same characteristic, the introduced varieties reached the values of 5177.79 kg ha<sup>-1</sup> (Arpa) and 5497.68 kg ha<sup>-1</sup> (Onice). Regarding the thousand grain weight, the standard Prima riska obtained the highest results both for paddy (40.00g) and also for white rice (30.40 g), while in opposite, the lowest thousand grain weight was found in Arpa (26.6 g for paddy rice, 18.80 g for white rice).

Key words: rice, varieties, head rice yield, yield, one thousand grain weight.

# Introduction

After the harvest of the rice crop, the obtained paddy grain cannot be directly used for human consumption. The next step for obtaining an edible, white rice kernel is the postproduction, carried out in the milling factories. The basic objective of the milling is to remove the husk (hulls), embryo and bran layers. The milling process of the paddy grain results in the following fractions: milled grains, paddy (non-milled) grains, brokens, husks and bran or flour. During the initial step of milling, or dehusking the paddy, the straw to brown-coloured grain is obtained, named also as brown rice (cargo rice or husked rice). By polishing cargo rice, the rice brans, aleuron layer and small part of the endosperm are being removed in order to produce white rice. White rice (or milled rice) represents milled

and polished rice without the embryo and bran layers. The post-harvest processing of the paddy rice results in different percent of whole grains of milled rice (head rice yield). The head rice yield is genetically determined in each variety or genotype, but it is also highly dependent on the applied production technology as well as on the environmental conditions (Kunze, 1985; Sürek and Beşer, 1998; Andov et al., 2003; Andreevska et al., 2005/2006 and 2007; Ilieva et al., 2000, 2007 2008 and 2009). The head rice yield is also influenced by the time of harvesting and the grain moisture content. According to Ali et al. (1993), high head rice yield was reached when the harvesting was done 36 to 39 days after flowering, with the grain moisture content between 20% and 30%. In order to increase the yield and improve the quality of white rice, the use of high-yielding and good quality varieties is of particular importance, beside the climatic conditions and applied production technology. Andov et al. (2003, 2008/2009, 2010) sugest the selection of the most suitable rice varieties adapted to the climatic and soil conditions of the specific geografic region which will provide full expression of the yield potential and quality properties. In parallel with breeding new rice varieties, there is also need for introduction, acclimatization and investigation of high yielding and good quality varieties, with the aim to enrich the spectrum of varieties available to the producers. The aim of this investigation was to assess the head rice yield, the yield of white rice and a thousand grain weight of the paddy rice and white rice on a two newly introduced Italian rice varieties, grown under the environmental conditions of Kochani region in the Republic of Macedonia.

# Material and methods

Investigations were done during 2013 and 2014 by conducting field trials in the experimental field of the Institute of Agriculture – Skopje, Department for rice in Kochani (locality "Mishjak"). As a research material, two newly introduced Italian rice varieties (*Arpa* and *Onice*) were used by comparing them with two standard varieties: *Prima riska* (Macedonian rice variety, registered since 2004) and *San Andrea* (Italian rice variety, domesticated and widespread under Macedonian rice growing conditions). The field trials were randomized blocks in three repetitions. Standard production technologu was applied. The head rice yield – whole grains and by-products (brokens, hulls and bran) were determined on a paddy quality testing machine by milling three average samples per treatment (each weighing 100 g).The time of milling was 1.40 min. The obtained results were statistically performed (ANOVA and LSD testing). One thousand grain weight was assessed on a paddy and on white rice (two samples x 500 grains per treatment).

# Soil and climatic conditions

The soil type where the trials were set-up was alluvium (Tab. 1), carbonate-free at the examined depths (Ubavić et al., 2001). According to American classification, presented by Filipovski (1984), the pH of the soil in water solution was very strong acid at 0-20 cm and slight acid at 20-40 cm depth, while in nKCL solution pH was acid at 0-20 cm and slight acid at 20-40 cm depth. The soil samples were medium supplied with easy available potassium and phosphorus, according to applied AL method (Džamić et al., 1996).

Tuble 1. Some enemieur properties of the son nom rocurty "Misijuk								
Donth [am]	CaCO <sub>3</sub> %	pH		Available mg/100 g of soil				
Depth [cm]		H <sub>2</sub> O nKCl		$P_2O_5$	K <sub>2</sub> O			
0-20	-	5.9	5.1	16.88	14.03			
20-40	-	6.2	5.6	12.66	11.15			

Table 1 So	me chemical	properties	of the soil	from local	ity "Mishjak"
1 abic 1. 50	ine enemicai	properties	of the son	nom local	ty misijak

During the vegetation of the rice crop (April-October), the average monthly air temperature in 2013/2014 was 19.4°C, maximum temperature was 26.4°C, while minimum temperature was 12.6°C (Tab. 2). The average monthly air temperature in the first year of investigation (20.4°C) was higher than the second year (18.4°C). The total (average) sum of rainfall for both years of investigation during the vegetation was 433.8 mm (Tab. 2). It should be pointed out that in the second year the sum of rainfall (551.0 mm) was larger than in the first year (316.5 mm). During the first year of rice vegetation (April-October), the largest monthly sum of rainfall was assessed in June (130.5 mm), while the least was in August (11.2 mm). During the second year, Appril was the month with the largest sum of rainfall (121.0 mm), and August with the least monthly sum of rainfall (31.0 mm).

Year	Months							Average	
Average monthly temperature (C°)									Veg.
2013	15.3	20.1	21.8	23.9	26.1	19.8	15.7	14.7	20.4
2014	12.4	16.8	20.8	23.2	23.8	18.3	13.8	13.8	18.4
Average	13.9	18.5	21.3	23.6	25.0	19.1	14.8	14.3	19.4
Average monthly max. temperature (C <sup>o</sup> )									
2013	21.7	26.5	28.2	30.8	33.3	27.2	23.1	20.6	27.3
2014	18.0	23.0	28.1	30.9	31.8	25.0	20.7	20.1	25.4
Average	19.9	24.8	28.2	30.9	32.6	26.1	21.9	20.4	26.4
	Average monthly min. temperature (C <sup>o</sup> )								
2013	7.9	13.0	15.4	16.2	17.5	11.2	7.3	8.4	12.6
2014	7.4	10.8	14.0	16.5	16.6	13.4	8.7	8.5	12.5
Average	7.7	11.9	14.7	16.4	17.1	12.3	8.0	8.5	12.6
	Sum								
2013	39.0	45.0	130.5	32.0	11.0	29.0	30.0	559.5	316.5
2014	121.0	92.0	116.0	65.0	31.0	89.0	37.0	794.0	551.0
Average	80.0	68.5	123.3	48.5	21.0	59.0	33.5	676.8	433.8

Table 2. Data on meteorological elements of the rice vegetation period in Kocani

# **Results and discussion**

# Head rice yield

The results of investigation of the head rice yield – whole grains and by-products are presented in the Tab. 3. The highest average head rice yield was obtained in the newly introduced variety *Arpa* (62.32 %), while the lowest average head rice yield was assessed in the standard variety *San Andrea* (56.54 %). In 2013, highest head rice yield was assessed in the newly introduced variety *Onice* (57.17%) which was significantly higher than standard *Prima riska* at probability level of 0.05 and significantly higher than standard *San Andrea* for both levels of probability (0.05 and 0.01). The lowest head rice yield in

2013 was obtained in the standard variety San Andrea (43.20 %). In 2014, the highest values for the head rice yield was achieved in the standard variety San Andrea (69.87 %), significantly higher than standard Prima riska (62.13 %) at both levels of probability (0.05 and 0.01) and significantly higher compared to the newly introduced variety Onice (65.47 %) at probability level of 0.05. According to Tab. 3, the highest percent of husks was found in San Andrea (21.00 %) and the lowest in Oniche (17.61 %). Also, during the laboratory milling, the highest percent of bran was generated in Prima riska (14.3 %) and the lowest percent of bran was in Onice (9.51 %). Regarding the fraction of chalky grains, the highest value was found in Arpa (4.19 %) and the lowest in Onice (1.17 %). The high percent of chalky grains in the post-harvest procedure indicates to insufficiently matured paddy grain that is related to the vegetation length of each of the varieties. According to investigations of Andov et al. (2003), the amount of fractions generated through the milling process (whole grains, brokens, hulls and bran) varied in different varieties, depending on the genotype, vegetation year and cropping systems (first or second crop). Ilieva et al. (2009) explored the optimal time for harvesting rice crop applying five different terms of harvesting on five rice varieties (Monticelli, Biser-2, San Andrea, R-76/6 and Prima riska). The authores assessed the highest head rice yield of the paddy harvested at the grain moisture content between 18 % and 20% (the third term of harvesting).

## Yield of white rice

The results for yield of white rice are presented in Table 4. The highest average yield of white rice after milling the paddy grain was obtained in *Prima riska* (5783.53 kg ha<sup>-1</sup>); in opposite, the lowest average yield of white rice was assessed in *San Andrea* (4977.82 kg ha<sup>-1</sup>). The yield of white rice in the newly introduced varieties was between the values of the standard varieties, achieving 5177.79 kg ha<sup>-1</sup> (in *Arpa*) and 5497.68 kg ha<sup>-1</sup> (in *Onice*). In the vegetation year 2013, the highest yield of white rice was determined in the standard *Prima riska* (5552.22 kg ha<sup>-1</sup>), significantly higher than all other varieties, for both levels of probability. The lowest yield of white rice was assessed in *San Andrea* (4105.56 kg ha<sup>-1</sup>). In the vegetation year 2014, the standard variety *Prima riska* was also best yielding for white rice among all the examined varieties (6014.84 kg ha<sup>-1</sup>), but the differences among means were not significant. By exploring several rice varieties (*Monticelli, Osogovka, M-101, Onda, Lido* and *Radon*) in different cropping systems, Andov et al. (2003) assesed higher yield of white rice in the variant when rice was grown as first crop, compared to the variant when rice was grown as second crop.

Varieties		Head	Brokens		Total	Chalky	Rice	Husks	
	Year	rice	1/3	2/3	Total	Milled	grains	bran	
		yield	1/5	2/5	Total	Rice yield			
Prima	2013	51.23	2.20	9.23	11.43	62.66	0.50	17.33	19.51
riska (st.)	2014	62.13	2.50	5.10	7.60	69.73	1.87	10.93	17.47
	2013/14	56.68	2.35	7.17	9.52	66.20	1.19	14.13	18.49
San	2013	43.20	3.03	14.00	17.03	60.23	5.37	9.97	24.43
Andrea	2014	69.87	1.00	1.40	2.40	72.27	0.77	9.39	17.57
<i>(st)</i>	2013/14	56.54	2.02	7.70	9.72	66.26	3.07	9.68	21.00
Arpa	2013	56.87	1.0	2.30	3.30	60.17	5.98	12.57	21.28
	2014	67.77	-	0.30	0.30	68.07	2.40	9.23	20.30
	2013/14	62.32	0.50	1.30	1.80	64.12	4.19	10.90	20.79
Onice	2013	57.17	2.90	9.80	12.70	69.87	0.97	11.15	18.01
	2014	65.47	1.90	6.20	8.10	73.57	1.37	7.86	17.20
	2013/14	61.32	2.40	8.00	10.40	71.72	1.17	9.51	17.61
Year	2013	2014							
LSD 0,05	5.34	3.96							
LSD 0,01	8.09	6.00							

Table 3. Head rice yield - %

# Table 4. Yield of white rice (kg ha<sup>-1</sup>)

Varieties	Y	ear		Index from		
varieties	2013 2014		Average	Prima riska	San andrea	
Prima riska(st.)	5552.22	6014.84	5783.53	0	+16.19	
San andrea (st)	4105.56	5850.08	4977.82	-13.93	0	
Arpa	4527.09	5828.48	5177.79	-10.47	+4.02	
Onice	4929.81	6065.55	5497.68	-4.94	+10.44	
Average	4778.67	5939.74	5359.21			
LSD 0,05	405.23	374.96				
LSD 0,01	614.48	568.58				

# The thousand grain weight

The thousand grain weight is genetically determined characteristic in different species and varieties. In rice it varies in large extents, depending on maturity degree of the grain, grain origin, location of the grain on the mother plant, environmental conditions, applied technology etc. Since the grain moisture has an important impact on the thousand grain weight, the thousand grain weight will be expressed as related to grain dry matter content. In Table 5, the results for thousand grain weight on a paddy and on white rice are presented. According to the results obtained, the newly introduced Italian rice varieties are short-grained, having in general lower thousand grain weight in comparison with long-grained standards *Prima riska* and *San Andrea*. The standard variety *Prima riska* characterized with the highest thousand grain weight (paddy – 40.00g and white rice 30.40 g). The lowest thousand grain weight was found in newly introduced variety *Arpa* (paddy – 26.24 g and white rice 18.80 g). Also the other Italian variety *Onice* had lower thousand grain weight of the other standard *San Andrea* was 35.36 g (paddy) and 27.90 g (white rice). The investigated Italian varieties *Arpa* and *Onice* reach higher thousand grain

weight under Italian rice growing condition then under Macedonian rice growing conditions.

		Paddy		White rice				
Varieties	Ye	ear	A	Ye	ear	A		
	2013	2014	Average	2013	2014	Average		
Prima riska(st.)	41.40	38.60	40.00	30.60	30.20	30.40		
San Andrea (st)	34.93	36.33	35.63	28.80	27.00	27.90		
Arpa	26.80	25.67	26.24	19.07	18.53	18.80		
Onice	33.20	32.33	32.77	23.80	24.60	24.20		

Table 5. The thousand grain weight (g)

## Conclusions

Based on the presented results on the investigation of the two newly introduced Italian rice varieties (*Arpa* and *Onice*) compared to two standard varieties *Prima riska* and *San Andrea*, the following conclusions can be done:

- The highest average head rice yield was obtained in the newly introduced variety *Arpa* (62.32 %), while the lowest average head rice yield was assessed in the standard variety *San Andrea* (56.54 %).

- The highest percent of husks was found in standard *San Andrea* (21.00 %) and the lowest in *Oniche* (17.61 %).

- The highest percent of bran was generated in the standard *Prima riska* (14.3 %) and the lowest percent of bran was in *Onice* (9.51 %).

- The highest value of the fraction of chalky grains was found in Arpa (4.19 %) and the lowest in *Onice* (1.17 %).

- The highest average yield of white rice after milling the paddy grain was obtained in *Prima riska* (5783.53 kg ha<sup>-1</sup>); the lowest average yield of white rice was assessed in *San Andrea* (4977.82 kg ha<sup>-1</sup>).

- The yield of white rice in the newly introduced varieties was between the values of the standard varieties, achieving 5177.79 kg ha<sup>-1</sup> (in *Arpa*) and 5497.68 kg ha<sup>-1</sup> (in *Onice*).

- The standard variety *Prima riska* characterized with the highest thousand grain weight (paddy -40.00g and white rice 30.40 g). The lowest thousand grain weight was found in newly introduced variety *Arpa* (paddy -26.24 g and white rice 18.80 g)

- The milling quality characteristics of rice (the head rice yield and the yield of white rice) are due to many factors as genetic constitution of the varieties, different vegetation years and the environmental conditions of growing.

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### YIELD AND SOME MORPHOLOGICAL CHARACTERISTICS OF THE NEWLY INTRODUCED RICE VARIETIES (*Oryza sativa* L.) IN THE REPUBLIC OF MACEDONIA

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#### Abstract

Two newly introduced Italian rice varieties Arpa and Onice were investigated under the growing conditions of Kochani region, Republic of Macedonia, in comparison with two standard varieties Prima riska (Macedonian variety) and San Andrea (domesticated Italian variety). The investigations were conducted during 2013 and 2014 by setting-up field trials (randomized blocks). During the two vegetation years, the highest results for average paddy rice yield and biological yield among all the varieties were obtained by the standard variety Prima riska (10,257.00 kg ha<sup>-1</sup> and 27,422.50 kg ha<sup>-1</sup> respectively). The lowest paddy rice yield was found in the variety Arpa (8,286.50 kg ha<sup>-1</sup>), while the lowest biological yield in the variety Onice (21,027 kg ha<sup>-1</sup>). The obtained differences in the yield were significant. The newly introduced varieties were characterized with shorter stem (Arpa – 84.97 cm and Onice – 80.80 cm), compared to standard varieties (Prima riska – 107.54 cm and San Andrea 106.22 cm). Also, the panicle length was, in average, shorter in the Italian varieties (Arpa - 14.55 cm and Onice - 12.60 cm) than in standard varieties in the investigation (Prima riska – 21.85 cm and San Andrea 17.85 cm). The highest average number of productive tillers per  $m^2$  was assessed in the variety Arpa (602.23), and the lowest in San Andrea (484.67).

Key words: rice, variety, yield, stem, panicle, productive tillers.

### Introduction

In the last couple of years, the rice production is spread to a surface area of 4,500 to 5,140 ha within the overall agricultural plant production of the Republic of Macedonia. In our country, rice is mainly grown along the valley of the Bregalnica river, in the regions around Kochani, Shtip, Vinica, Blatec, and on the smaller areas in the regions around Veles and Probishtip. The average rice yields for the period 2011-2014 are 5,612 kg ha<sup>-1</sup> of paddy rice, or 2,806 kg ha<sup>-1</sup> of white rice, calculated at 50% head rice yield (according to the Statistical Yearbooks of the Republic of Macedonia 2011-2014). Important factors in rice production are soil and climatic conditions together with applied production technology, but for boosting yields and improving quality of rice, the use of high quality rice varieties is of particular importance. The right selection of varieties for each particular

complex of environmental conditions would provide full expression of varieties' productive and quality properties. The rice varieties in Macedonian rice production (*R*-76/6, San Andrea, Prima riska etc.) are mainly characterized with plant height over 100 cm. These varieties are susceptible to lodging, thus unsuitable for intensive production technology and for high doses of mineral (especially nitrogen) nutrients. In the past few years with unpleasant environmental conditions during the harvesting, serious failure in rice yield and deterioration in paddy quality were recorded (Andreevska et al., 2007, 2009). Therefore, while introducing new varieties, it is of special interest to focus on high yielding, stable and good quality varieties, suitable for intensive production technology. The use of new short-stem rice varieties (semi-dwarf or intermediate) would improve in the entire rice production, but also it would provide enrichment of the germplasm in rice breeding programs for creating new rice varieties. The aim of this investigation was to assess the yield of paddy rice, the biological yield and certain morphological and productive traits (plant height, panicle length and number of productive tillers) in two newly introduced rice varieties.

#### Material and methods

Field trials were set-up during 2013 and 2014 in the experimental field of the Institute of Agriculture – Skopje, Department for rice in Kochani (locality "Mishjak"). Two newly introduced Italian rice varieties (*Arpa* and *Onice*) were used as a research material and compared with two standard varieties: *Prima riska* (Macedonian rice variety) and *San Andrea* (Italian rice variety, domesticated and widespread under Macedonian rice growing conditions). The field trials were randomized blocks in three repetitions. Standard production technology was applied. The following traits were investigated: yield of paddy rice, the biological yield, plant height, panicle length and number of productive tillers per m<sup>2</sup>. The obtained results were statistically performed (ANOVA and LSD testing).

#### Soil and climatic conditions

The soil type where the trials were set-up was alluvium (Tab. 1), carbonate-free at the examined depths (Ubavić et al., 2001). According to American classification, presented by Filipovski (1984), the pH of the soil in water solution was very strong acid at 0-20 cm and slight acid at 20-40 cm depth, while in nKCL solution pH was acid at 0-20 cm and slight acid at 20-40 cm depth. The soil samples were medium supplied with easy available potassium and phosphorus, according to applied AL method (Džamić et al., 1996).

	chemical properties of the soft from focality withingak										
Depth [cm]	CaCO <sub>3</sub> %	p	Н	Available mg/100 g of soil							
Depui [ciii]		H <sub>2</sub> O	nKCl	$P_2O_5$	$\underline{K}_2 \underline{O}$						
0-20	-	5.9	5.1	16.88	14.03						
20-40	-	6.2	5.6	12.66	11.15						

Table 1 Some chemical properties of the soil from locality "Mishjal
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During the vegetation of the rice crop (April-October), the average monthly air temperature in 2013/2014 was  $19.4^{\circ}$  C, maximum temperature was  $26.4^{\circ}$ C, while minimum temperature was  $12.6^{\circ}$ C (Tab. 2). The average monthly air temperature in the

first year of investigation  $(20.4^{\circ}C)$  was higher than the second year  $(18.4^{\circ}C)$ . The total (average) sum of rainfall for both years of investigation during the vegetation was 433.8 mm (Tab. 2). It should be pointed out that in the second year the sum of rainfall (551.0 mm) was larger than in the first year (316.5 mm). During the first year of rice vegetation (April-October), the largest monthly sum of rainfall was assessed in June (130.5 mm), while the least was in August (11.2 mm). During the second year, April was the month with the largest sum of rainfall (121.0 mm), and August with the least monthly sum of rainfall (31.0 mm).

Year				Months		•		Ave	rage		
		Average	monthly te	emperature	e (C°)			Years	Veg.		
2013	15.3	20.1	21.8	23.9	14.7	20.4					
2014	12.4	16.8	20.8	23.2	23.8	18.3	13.8	13.8	18.4		
Average	13.9	18.5	21.3	23.6	25.0	19.1	14.8	14.3	19.4		
	Average monthly max. temperature (C <sup>o</sup> )										
2013	013 21.7 26.5 28.2 30.8 33.3 27.2 23.1										
2014	18.0	23.0	28.1	30.9	31.8	25.0	20.7	20.1	25.4		
Average	19.9	24.8	28.2	30.9	32.6	26.1	21.9	20.4	26.4		
	Α	verage mo	onthly min	. temperat	ure (C°)						
2013	7.9	13.0	15.4	16.2	17.5	11.2	7.3	8.4	12.6		
2014	7.4	10.8	14.0	16.5	16.6	13.4	8.7	8.5	12.5		
Average	7.7	11.9	14.7	16.4	17.1	12.3	8.0	8.5	12.6		
		Mo	nthly rainf	falls (mm)				Su	m		
2013	39.0	45.0	130.5	32.0	11.0	29.0	30.0	559.5	316.5		
2014	121.0 92.0 116.0 65.0 31.0 89.0 37.0							794.0	551.0		
Average	80.0	68.5	123.3	48.5	21.0	59.0	33.5	676.8	433.8		

Table 2. Data on meteorological elements of the rice vegetation period in Kocani

#### **Results and discussion**

#### Yield of paddy rice

The results for the yield of paddy rice are presented in Table 3. The highest average yield of paddy rice from the two years of investigation was achieved by the standard variety *Prima riska* (10,257.00 kg ha<sup>-1</sup>), while the lowest was assessed in the newly introduced variety *Arpa* (8,286.50 kg ha<sup>-1</sup>). In the vegetation year 2013, the yield of paddy rice in both Italian varieties *Arpa* and *Onice* was significantly lower for both levels of probability, compared to two standard varieties. In 2014, the yield of paddy rice in *Arpa* and *Onice* was significantly lower for both levels of probability, compared to the standard *Prima riska*. But, in comparison with the other standard *San Andrea*, Italian varieties were better regarding the yield of paddy rice, *Arpa* gained significantly better yield for the level of 0.05, and *Onice* was significantly higher yielding for both levels of probability.

Table 3. Yield of paddy rice (kg ha<sup>-1</sup>)

#### SECTION 9. FIELD CROP PRODUCTION

	Y	ear		Index from					
Varieties	2013	2014	Average	Prima riska	San andrea				
Prima riska(st.)	10.834	9.680	10.257	0	+14.70				
San Andrea (st)	9.512	8.373	8.942.50	-12.82	0				
Arpa	7,973	8.600	8.286.50	-19.21	-7.34				
Onice	8.624	9.267	8.945.50	-12.79	+0.03				
Average	9.235.75	8.980.00	9.107.88						
LSD 0,05	455	157.15							
LSD 0,01	68.96	238.30							

## Biological yield

According to the results for biological yield (grain + straw), presented in Table 4., the highest average biological yield in the two years of investigation was found in the standard variety *Prima riska* (27,422.50 kg ha<sup>-1</sup>), while the lowest biological yield was in the variety *Onice* (21,027.00 kg ha<sup>-1</sup>). In 2013, significantly higher biological yield for both levels of probability was determined in the standard variety *Prima riska* (28,178 kg ha<sup>-1</sup>), compared to all other varieties in this research – *San Andrea* (24,278 kg ha<sup>-1</sup>), *Arpa* (25,284 kg ha<sup>-1</sup>) *and Onice* (18,254 kg ha<sup>-1</sup>). In the vegetation year 2014, the biological yield of the standard variety *Prima riska* (26,667 kg ha<sup>-1</sup>) was significantly higher for both levels of probability compared to *San Andrea* (25,120 kg ha<sup>-1</sup>) and *Onice* (23,800 kg ha<sup>-1</sup>), while compared to *Arpa* (26,373 kg ha<sup>-1</sup>), differences between means were no significant.

Varieties	Ye	ear	Augrage	Index from				
varieties	2013	2014	Average	Prima riska	San Andrea			
Prima riska(st.)	28.178	26.667	27.422.50	0	+11.03			
San Andrea(st)	24.278	25.120	24.699.00	-9.93	0			
Arpa	25.284	26.373	25.828.50	-5.81	+4.57			
Onice	18.254	23.800	21.027.00	-23.32	-14.87			
Average	23.998.50	25.490.00	2444.25					
LSD 0.05	165.06	463.90						
LSD 0.01	250.29	703.46						

Table 4. Biological yield (straw + grain) kg ha<sup>-1</sup>

### Stem height

In general, the investigated Italian rice varieties (*Arpa* 84.97 cm and *Onice* 80.80 cm) had shorter stem than standards (*Prima riska* 107.54 cm and *San Andrea* 106.22 cm), according to the results presented in Table 5. In 2013 as well as in 2014, *Arpa* and *Onice* had significantly lower stem height for both levels of probability, in comparison with two standards *Prima riska* and *San Andrea*.

1 doie 5. Stelli liei							
Varieties	Year	Х	$\sigma(S)$	Sx	CV %	min	max
Prima riska(st.)	2013	103.00	5.86	1.07	5.69	90.00	114.00
	2014	112.07	4.51	0.82	4.03	104.00	119.00
	average	107.54	5.19	41.54	4.86	97.00	116.50
San Andrea(st.)	2013	98.27	5.15	0.94	5.25	86.00	106.00
	2014	114.17	8.12	1.48	7.11	102.00	129.00
	average	106.22	6.64	1.21	6.18	94.00	117.50
Arpa	2013	80.97	2.79	0.50	3.44	77.00	86.00
	2014	88.97	4.62	0.84	5.19	79.00	97.00
	average	84.97	3.71	0.67	4.32	78.00	91.50
Onice	2013	77.90	4.44	0.81	5.71	70.00	87.00
	2014	83.70	2.73	0.49	3.26	78.00	91.00
	average	80.80	3.59	0.65	4.49	74.00	89.00
Year	2013	2014					
LSD 0.05	5.37	8.28					
LSD 0.01	8.14	12.56					

Table 5. Stem height (cm)

### Panicle length

The average panicle length (Tab. 6) in the newly introduced Italian varieties *Arpa* (14.55 cm) and *Onice* (12.60 cm) was shorter compared to standard varieties *Prima riska* (21.85 cm) and *San Andrea* (17.85 cm). In both years of this study (2013 and 2014), the average values for the panicle length in *Arpa, Onice* and *San Andrea* were significantly lower for both levels of probability, in comparison with the standard *Prima riska*.

Varieties	Year	Х	σ (S)	Sx	CV %	min	max
Prima riska(st.)	2013	22.50	1.48	0.27	6.58	20.00	26.00
	2014	21.20	2.09	0.38	9.86	17.00	25.00
	average	21.85	1.79	0.33	8.22	18.50	25.50
San Andrea(st)	2013	18.27	1.80	0.33	9.84	15.00	22.00
	2014	17.43	1.73	0.32	9.96	13.00	20.00
	average	17.85	1.77	0.33	9.90	14.00	21.00
Arpa	2013	14.83	1.09	0.20	7.32	13.00	17.00
	2014	14.27	1.23	0.22	8.62	11.00	17.00
	average	14.55	1.16	0.21	7.97	12.00	17.00
Onice	2013	12.37	0.96	0.18	7.80	11.00	14.00
	2014	12.83	1.32	0.24	10.25	10.00	16.00
	average	12.60	1.14	0.21	9.03	10.50	15.00
Year	2013	2014					
LSD 0,05	0.95	0.74					
LSD 0,01	1.43	1.13					

Table 6. Panicle lenght (cm)

### Number of productive tillers

According to Tab. 7, the variety Arpa generated the highest average number of productive tillers per m<sup>2</sup> (602.23); in opposite, *San Andrea* had the lowest number of productive tillers (484.67). In 2013, *Arpa* characterized with the highest average number of productive

tillers per m<sup>2</sup> (708.33), significantly higher for both levels of probability, compared to all other examined varieties. In 2014, the highest average number of productive tillers per m<sup>2</sup> was assessed in *Onice* (648.33), significantly higher for both levels of probability, compared to all other examined varieties.

	Y	lear		Index from				
Varieties	2013	2014	Average	Prima riska	San Andrea			
Prima riska(st.)	587.33	463.33	525.33	0	+7.74			
San Andrea(st)	602.67	366.67	484.67	-7.74	0			
Arpa	708.33	496.33	602.33	+14.66	+24.28			
Onice	594.33	648.33	621.33	+18.27	+28.13			
Average	623.17	493.67	558.42					
LSD 0.05	11.88	28.94						
LSD 0.01	18.02	43.88						

Table 7. Number of productive tillers per  $m^2$ 

### Conclusions

From the two-year study of the newly introduced rice varieties *Arpa* and *Onice* and two standard varieties *Prima riska* and *San Andrea* based on the results presented above, the following conclusions have been done:

- The highest average yield of paddy rice from the two years of investigation was achieved by the standard variety *Prima riska* (10,257.00 kg ha<sup>-1</sup>), while the lowest was assessed in the newly introduced variety *Arpa* (8,286.50 kg ha<sup>-1</sup>).

- The highest average biological yield in the two years of investigation was found in the standard variety *Prima riska* (27,422.50 kg ha<sup>-1</sup>), while the lowest biological yield was in the variety *Onice* (21,027.00 kg ha<sup>-1</sup>).

- The investigated Italian rice varieties (*Arpa* 84.97 cm and *Onice* 80.80 cm) characterized with shorter stem than standards (*Prima riska* 107.54 cm and *San Andrea* 106.22 cm)

- The average panicle length in the newly introduced Italian varieties *Arpa* (14.55 cm) and *Onice* (12.60 cm) was shorter compared to standard varieties *Prima riska* (21.85 cm) and *San Andrea* (17.85 cm).

- The variety Arpa generated the highest average number of productive tillers per m<sup>2</sup> (602.23); in opposite, *San Andrea* had the lowest number of productive tillers (484.67).

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### ESSENTIAL OIL COMPOSITION OF OCIMUM BASILICUM L. IN SEMI-ARID CONDITIONS OF TURKEY

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#### Abstract

Sweet basil (Ocimum basilicum L.) is known locally as "feslegen or reyhan", belonging to the Lamiaceae family and is widely grown as an ornamental, medicinal and spice crops in the Mediterranean countries, including Turkey. Basil (O. basilicum) is a important medicinal plant bearing essential oil. The used plant part is leaves and flowers. In Divarbakır region, basil is cultivated in small areas like gardens. However this plant must be produced in the shape of field farming for a new crop. Cultivar, cropping season, plant ontogeny and plant part had significant effects on the yield and quality of the essential oil of O. basilicum as withotheressential oilplants. The present work was undertaken with the main objective to investigate the essential oil rate isolated from the aerial parts of O. basilicumcultivated in Southeast Anatolia Region of Turkey as affected by different development stage and the constitutents of essential oil at flowering stages. The constituents of essential oils isolated by hydrodistillation of the overground parts of O. basilicum L. from Divarbakir province were examined by GC-MS. The yield of essential oil from different development stages varied between 1.2-1.6%, and we found that the main components of O. basilicum L.essential oil are linalool and eugenol in flowering stages.

Key words: Ocimum basilicum, essential oil, composition.

### Introduction

The genus *Ocimum* L. (Lamiaceae) consists of 30-160 species, spreading throughout the tropical regions of Asia, Africa and Central and South America (Beatovićet al., 2015). In Turkey, sweet basil (*O* . *basilicum* L.) is known locally as "fesleğen or reyhan", is widely grown as an ornamental or spice crops. Basil (*Ocimum basilicum* L.) leaves are used fresh and dried as flavorings or spices in sauces, salad dressings, vegetables, vinegar and confectionery products (Vieira ve Simon, 2006). *Ocimum* is largely used for culinary and medical properties such as for headaches, coughs, diarrhea, worms and kidney disorders (Telci *et al.*, 2006). Extracted essential oil of the plant is of various biological activities which are related to their various interesting applications as antimicrobial, antioxidant, repellent, insecticidal, larvicidal, nematicidal and therapeutic (anti-inflammatory,

antinociceptive, antipyretic, antiulcer, analgesic, anthelmintic, anticarcinogenic, skin permeation enhancer, immunomodulatory, cardio-protective, antilipidemic) agents (Pandey et. al., 2014). Several of the factors may influence commercially important crops in respect ofoptimizing the cultivation conditions and time of harvest and to obtain higher yields of high-quality essential oils according to consumer requests. Knowledge of the factors that determine the chemical variability and yield for each species are thus very important (Figueiredo etal., 2008). Saharkhiz et al., (2009) stated that medicinal and aromatic plants have great variability with regard to chemical and biological aspects depending on climatic condition, location, genetic diversification, different plant part, different ontogenetic and harvesting time. Beltrame et al., (2014) stated that Ocimum basilicum L. (basil) essential oil is chemical composition was already largely studied, due to the useful properties of the plant as essential oil-producing crop, culinary herb, medicinal plant and insect-controlling substances. The main chemotypes found are monoterpenoids, sesquiterpenoids and phenylpropanoids, being linalool, methyl chavicol, eugenol, methyl eugenol and geraniol the main reported substances for O. basilicum essential oil. This plant species has attracted interest in the area due to the large amount of linalool present in the essential oil, an important component in many formulations of perfumes with high market acceptance. Thereforeit is important to know the chemical structure of the plantgrown in the region. The objective of this research was to determine chemical composition in flowering stages and yield of essential oil of Ocimum basilicum at different development stages in Southeast Anatolia of Turkey.

### **Material and Methods**

### Plant Material and Growth Conditions

This experiment was conducted in the experimental area of the Department of Field Crops, Faculty of Agriculture, Dicle University, Diyarbakir, Turkey (latitude 37°53'N, longitude 40°16'E, altitude 680 m above sea level) with dominant semi-arid characteristics during 2009 and 2010 years. The climate in Diyarbakir is dry and hot in summer and cold in winter. Precipitation regime is irregular during winter. Southeastern Anatolia Regionhas acontinental climatefeatures. Diyarbakirharshcontinental climateprevails inthe semi-aridhighland climate. The summers arevery hot, dryand long, cold wintersand a littlerainy. Seeds of *Ocimum basilicum* L. were purchased from local bazaar in Netherland and planted in the greenhouse in March2011, in polystyrene containers with 40 holes, In May 2011, nursery plants were transplanted into the collection garden of Dicle University, Agriculture Faculty, Field Crops Department. Harvesting of the plants was performed at preflowering, flowering and post-flowering stage in 07.07. 2015, 17.07.2015 and 30.07.2011, respectively for essential oil rate but at flowering stages for determining essential oil constitutent. Plants were air-dried in the shade, then stored.

### Essential oil extraction

Essential oil of 20 g dried plant samples were subjected to hydro-distillation for 2.5 hours under continuous steam using a Clevenger-type apparatus (v/w).

The essential oils were stored in glass bottles at 4 °C until analysis. *Gas chromatography-mass spectrometry (GC/MS) analysis* GC-MS/FID analysis were conducted in the Plant

Physiology Laboratory in Biology Dept. of Kahramanmaras Sutcu Imam University. Qualification of the oil was analyzed on an Agilent 5975C Mass Spectrometer coupled with a Agilent GC-6890II series. The GC was equipped with HP-88 capillary column (100 m x 250  $\Box$  m x 0.20  $\Box$  m film thickness) and He was used carrier gas with flow rate of 1.0 mL/min. The GC oven temperature was programmed as follows: 70 oC (1 min), 230 oC at of 10 oC/min and then kept at 230 oC at 20 min. The injector temperature was 250 oC. The mass spectrometer was operating in EI mode at 70 eV. Split ratio was 20:1. Mass range 35-400m/z; scan speed (amu/s): 1000. 10  $\Box$ L of the oil was mixed 500 µl diethyl ether and 1  $\Box$ L of the concentrations injected into the column. Agillent two-way microfluid splitter (G3180B) allowed for simultaneous data acquisition using two different detectors MS and FID. The components of the oil were identified by mass spectra with those of pure authentic samples and NIST08, Willey7n.1 and HPCH1607 libraries reference compounds.

### **Results and discussion**

The plants cultivated in Southeast Anatolia Region of Turkey have essential oils varied between 1.20-1.60% according to development stages. The highest essential oil was obtained from flowering stages, followed by pre-flowering stages and the lowest essential oil content was obtained post-flowering stages (Table 1). Literature data show differences in essential oil content of Ocimum species. Vieira and Simon (2006) found that the yield of essential oils from the aerial parts of O. basilicum varied from 0.57% to 1.12%, while basil grown in Egypt had an essential oil yield of 1.7% (Ismail, 2006). Hussain et al. reported the hydro-distilled essential oils content ranged from 0.5% to 0.8%. Wesołowska(2012) found that in three sweet basils average of the essential oil yields was 0.44%. Furthermore, Hadipanah et al (2015) stated that the essential oil yields were obtained from the aerial of O. basilicum, 0.68, 0.71 and 0.96 ml / 100 g dry matter identified in Baghe-Bahadoran, Shahreza and Falavarjan province, respectively, cultivated in Iran.An earlier report by Sajjadi (2006) indicated the oil yield of the aerial parts of O. basilicum cv. purple and O. basilicum cv. green collected at full flowering from the same region were 0.2% and 0.5% (v/w) respectively. The research was agree with our study said that herb and oil yield were maximum at complete flowering (Randhawaand Gill, 1995). Great variations in the essential oil content of O. basilicum across geographic regions might be attributed to variable agroclimatic conditions and different agronomic techniques for cultivating. Given these facts, our results are in accordance with previously published reports on basil oils (Beatović et al., 2015). The mainconstituents of studied oil, their relative percentage of thetotal chromatogram area and retentiontimes are summarized in Table 1.

No	RT	Compound	Concentration (%)
1	12.86	limonene	0.33
2	13.42	gamma terpinene	0.78
3	13.79	terpinolene	0.14
4	13.96	eucalyptol	7.03
5	16.26	α-copaane	0.18
6	17.03	<i>n</i> -octyl acetate	0.31
7	17.35	α- cis bergamotone	1.01
8	17.77	linalool	39.58
9	17.94	beta elemene	7.05
10	18.25	e- caryophyllene cis	1.34
11	18.87	sabinene hydrate	0.27
12	19.04	βlongipenene	0.16
13	19.26	α- guaiene	1.95
14	19.44	terpinen-4-ol	8.60
15	19.82	γ- muurolene	1.58
16	20.12	germacrene d	2.21
17	20.44	camphor	1.42
18	20.53	3-cyclohexane	3.28
19	21.00	borneol	0.98
20	21.49	cis calamene	0.09
21	23.14	z- nerolidol	0.17
22	23.72	z- ocimene	0.04
23	24.44	cubenol	0.31
25	24.76	α- gurjunene	0.31
26	25.51	epi-alpha-cadinol	1.74
27	25.77	spathulenol	0.77
28	26.28	carvacrol	0.12
29	26.94	eugenol	16.23
30	28.52	valerenol	0.14
Total			98.12
Essential oil rate %	Pre-flowering stage: 1.2	%; Flowering stages: 1.4%; F	Ost-flowering stage: 1.6%

Table 1.Chemical composition of *Ocimum basilicum* L. oil at flowering stages and essential oil rate (%) according to development stages.

In the leaf essential oil in the flowering stages, 30 compounds were identified, representing 98.12% of the total oil. The major components were linalool (39.58%) and eugenol (16.23%). Other components present in appreciable contents were:, terpinen-4-ol (8.60%), beta elemene (7.05%), eucalyptol (7.03%), 3-cyclohexane (3.28%), germacrene-d (2.21%),  $\alpha$ -guaiene (1.95%), epi-alpha-cadinol (1.74%),  $\gamma$ - muurolene (1.58%), camphor (1.42%), e-caryophyllene cis (1.34%),  $\alpha$ -cis bergamotone (1.01%) (Table 1).

Various basil species and cultivars provide essential oil with different compositions and aroma. The chemo-taxonomical range of sweet basil is very wide (Zheljazkov *et al.*, 2008). The chemical composition of essential oils of *Ocimum* species has been studied previously. As prevalent components many basil essential oils contained monotherpene derivatives (camphor, limonene, 1,8-cineole, linalool, geraniol) and phenylpropanoid derivatives (eugenol, methyleugenol, chavicol, estragole, methyl-cinnamate). Different

chemotypes of basil have been recognised based on the predominant essential oil constituents (e.g. linalool, methyl chavicol, methyl cinnamate, methyl eugenol, eugenol). Basil essential oils contain a broad array of chemical compounds depending on variations in chemotypes, flower and leaf colours, aroma and particularly the origin of the plant (Beatovićet al., 2015). A significant differences in chemical composition of a particular species is found, which may be due to their occurrence in different eco-climatic zones and changes in edaphic factors (Pandey et. al., 2014).Da Costa et al. (2015) reported several studies assessing the chemical composition of 18 basil essential oils observed that the samples distributed into seven distinct types, each one presenting as the major volatile compound among the following: linalool, methyl cinnamate, methyl cinnamate/linalool, methyl eugenol, citral, methyl chavicol (estragole), and methylchavicol/citral.Pirbalouti Ghasemi (2014) also identified in total, 38 and 28 components were determined in essential oils of Purple and Green landraces of basil, respectively. The major constituents of the essential oil from the aerial of Purple landrace were methyl chavicol or estragol  $(63.32\pm6.0\%)$  and linalool  $(7.96\pm0.1\%)$ . The main compositions of the essential oil from the aerial of Green landrace were methyl chavicol (31.82±5.4%), geranial (24.60±1.1%) and neral (22.65±0.7%). Mohamed H. M. Abd El-Azim et al (2015) notified that the most predominant compounds were Linalool (33.9%), Eugenol (8.31%) and 2,6-Dimethyl-6-(4methyl-3-pentenyl)-bicyclo [3.1.1] hept-2-ene (8.04%) of the oil constituents.Reported by Telci et al. (2006) identified seven different chemotypes in 18 basil landraces from Turkey and majority were characterized by high linalool contents. According to results of Keita et al. (2000) that the oil of O. basilicum contained linalool (69%), eugenol (10%), (E)- $\alpha$ bergamotene (3%) and thymol (2%). Marotti et al. (1996) reported the composition of essential oils, all characterized by a high content of linalool, included three chemotypes: "linalool," "linalool and methylchavicol," and "linalool and eugenol" from Italian cultivars. Similarly, Sajjadi (2006) identified 20 constituents in the volatile oil of O. basilicum cv. purple collected at full flowering which the main constituents were methyl chavicol (52.4%), linalool (20.1%), epi- $\alpha$ -cadinol (5.9%) and trans- $\alpha$ -bergamotene (5.2%) and 12 components in the volatile oil of O. basilicum cv. green collected at full flowering which methyl chavicol (40.5%), geranial (27.6%), neral (18.5%) and caryophyllene oxide (5.4%) were the major components. In O. basilicum from Pakistan, Hussain et al. (2008), The essential oils consisted of linalool as the most abundant component (56.7-60.6%), followed by epi-a-cadinol (8.6-11.4%), a-bergamotene (7.4-9.2%) and c-cadinene (3.2-5.4%).Nurzyńska-Wierdak et al. (2012) found that the concentration of linalool was from 55.4% to 69.8%, depending on the cultivar and plant growth stage. The oil extracted at the flower bud stage was characterized by the highest proportion of linalool (in both cultivars) and of 1,8-cineole (only in the cultivar 'Kasia'). Bouzouita et al. (2003) found that the O. basilicum oil contains high proportions of methylchavicol and linalool (34.0% and 32.5%, respectively) and lower levels of 1,8-cineole (8.7%), (E)-methylcinnamate (4.2%) and eugenol (1.7%).Linalool (44.18%), 1,8-cineol (13.65%), eugenol (8.59%), methyl cinnamate (4.26%), iso caryophyllene (3.10%), and a-cubebene (4.97%) as the main components of the essential oils of O. basilicum cultivated in Egypt (Ismail, 2006). Hadipanah et al. (2015), The major constituents of the oil Baghe-Bahadoran were methyl chavicol (58.35%), neral (11.64%), linalool (9.34%) and 1,8-cineole (4.13%). The major constituents of the oil Shahreza were methyl chavicol (45.82%), linalool (17.31%), neral (13.21%), 1,8-cineole (5.73%), geranial (5.84%) and the major constituents of the oil Falavarjan were methyl chavicol (62.69%), geranial (7.36%), linalool (6.91%),  $\beta$ -caryophyllene (4.68%) and neral (4.67%).

Our results show that the majority of the basil cultivars growing in Southeast Anatolia Region of Turkey belonged to the "linalool chemotype".

#### Conclusions

In conclusion, the results obtained in our study indicated that the major components of the oil of *O. basilicum*harvested at flowering stage werelinalool, eugenol and terpinen-4-ol. Differences in the essential oils of studied *O. basilicum* could be attributed to genetic, chemotype, different location/zone and climatic conditions.

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#### EFFECTS OF DIFFERENT SUN PEST DAMAGE RATIOS ON BREAD WHEAT QUALITY

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#### Abstract

The research was conducted with Nina, Krasunia, Pehlivan, Flamura 85, Guadalupe and Gelibolu bread wheat varieties in 2010 and 2011. Extensograph extensograph, alveograph analyses were made to determine the effects of different sun pest damage ratio on the quality characteristics of bread wheat. In crops which have different damage of sun pest with Nina, Krasunia, Pehlivan, Flamura 85, Guadalupe and Gelibolu samples reduced considerably depending on the Rmak and extendibility from extensograph properties; water absorption ratio and softening value from farinograph properties; resistance, extension from alveograph properties. The effects of sun pest damage on quality characteristics of bread wheat were changed according to varieties. While %1 sun pest damage in some bread wheat varieties reduced considerably on quality characteristics, decreasing of quality characteristics in some bread wheat varieties was very low levels. **Key words**: Bread wheat, sun pest damage, quality, farinograph, extensograph, alveograph.

### Introduction

It is reported that sun pest, which is also called as "suni bug", "cereal bug", "stink bug", "sun pest", or "wheat bug" in literature, is encountered in almost all wheat cultivated areas in Turkey (Sivri 1998) and leads to economic loss. Sun pest is known to have 15 varieties in the world, 7 in Turkey, the most significant of which are *Eurygaster integriceps Put.*, *Eurygaster maura L.* ve *Eurygaster austriacus Schr.*, and *Eurygaster integriceps Put.* is the most common in our region. In Thrace region, there are a great number of various bread wheat types that have various yield and quality properties depending on ecological conditions. These varieties yield much more (approximately twice amount) than the average of the country. However, change in the quality properties of the wheat types due to various factors (climate, variety, sun pest damage, etc.) lead to a number of problems for the producers. One of the factors that brings about a negative change in the quality properties of the bread wheat is sun pest (*Eurygaster integriceps Put.*). Sun pest leads to deterioration in the crop quality by damaging kernel during a specific development stage of

wheat (milk stage of wheat). Sun pest damages the kernel by absorbing the protein part of the crop in the milk stage wheat. The types of wheat which are in the milk stage during the period when sun pest start to cause damage are particularly affected in an adverse manner. (Gültekin 1990, Talay 1997, Matsoukos and Morrison 1990, Aja et al. (2004), Tayyar 2008, Kütük et al. 2010, Aktar 2011). There are not enough number of studies on the relationship between sun pest damage and phenological properties of wheat types in the region. Moreover, this study aims at providing valuable information for local producers and plant breeders who may want to embark on resistance studies by revealing the relationship between sun pest damage ratio and crop yield; and the effect of sun pest damage ratio on quality characteristics of bread wheat.

#### **Material and Methods**

The study was carried out with five bread wheat varieties in the experimental area of Department of Field Crops of Tekirdağ Agricultural Faculty in 2010 and 2011 growing seasons. In this study, grains of Gelibolu, Flamura 85, Guadralupe, Krasunya and Nina bread wheat varieties was used experimental material. Variation of quality characters in the grains of bread wheat varieties affected by different sun pest damage was examined. The change in quality properties of the kernels was determined with the help of extensograph, farinograph, and alveograph analyses

#### **Results and Discussion**

In today's world, extensograph measures the stretching properties, the resistance to extension, and gas retention of dough. Dough's resistance to extension and its extensibility are central parameters that affect baking properties of flour. Dough's resistance to extension is (Rm), height of the curve (cm), Brabender units (BU), Extensograph unit (EU). Dough's extensibility (E) is floor length of the curve and is related to dough's elasticity which is represented in mm or cm. Significant changes were observed in terms of their extensograph properties when sun pest damaged and undamaged bread wheat kernel samples were analyzed. Gelibolu variety with 0.2% sun pest damage was found to show 447-560 (Rm) resistance to extension in 45 minutes in both years while its resistance was found 715-834 BU in 90 minutes and 763-998 BU in 135 minutes. Dough's extensibility (E), which is directly proportional with its dough workability, was found 148-163 mm in 45 minutes, 122-142 mm in 90 minutes, and 105-140 mm in 135 minutes. When sun pest damage was 3.8% with Gelibolu variety, its resistance to extension declined dramatically to 266 BU in 45 minutes, 244 BU in 90 minutes, and 191 BU in 135 minutes. However, its extensibility increased to 172 mm in 45 minutes, 156 mm in 90 minutes, and 151 mm in 135 minutes. One of the pioneering researchers in Turkey on sun pest, Tekeli (1964) and Lodos (1980) pointed out that technological property of wheat is lost when sun pest damage ratio on wheat commodity is 2%. With 2% sun pest damaged Flamura 85 variety in both years; R max value was found 521-473 BU, extensibility value 170-179 mm, and ratio 3.1-2.6 in 45 minutes while R max value was 866-740 BU, extensibility 146-145 mm, and ratio 5.9-5.1 in 90 minutes. In 135 minutes, R max value was found 887-685 BU, extensibility 133-133 mm, and ratio 6.7-5.2. When sun pest damage ratio was 4.5, there

was a noteworthy fall in these values. In 45 minutes, R max value shrank five times to 109 BU, extensibility 149 mm, and ratio 0.7 while R max value was found 91 BU, extensibility 125 mm, and ratio 0.7. These values could not be measured in 135 minutes. Even with Flamura 85 variety which has favorable values in terms of quality in the region, 4.5% sun pest damage ratio led to a remarkable fall in extensograph quality properties. With 0.2 sun pest damaged Guadralup bread wheat variety kernel samples in both years; R max value was found 456-475 BU, extensibility 157-165 mm, and ratio 2.9 while R max value was found 577-697 BU, extensibility 174-143 mm, and ratio 3.3-4.9 in 90 minutes. In 135 minutes, R max value was found 579-776 BU, extensibility 156-134 mm, and ratio 3.7-5.8. When sun pest damage ratio was 6.0 %; in 45 minutes, R max value fell dramatically to 201 BU, extensibility 139 mm, and ratio 1.5. in 90 minutes, R max value was found 152 BU, extensibility 134 mm, and ratio 1.1 while R max was found 119 BU, extensibility 121 mm, and ratio 1.0 in 135 minutes. When these figures are analyzed, it is found out that increscent sun pest damage leads to a substantial fall in R max value and a considerable decline in extensibility values with Guadralupe variety. With no sun pest damage or 0.2 damaged Krasunya variety, which has proper values in terms of quality, R max value was found 470-634 BU, extensibility 177-158 mm, and ratio 2.6-4.1 in 45 minutes. While in 90 minutes, R max value was found 701-911 BU, extensibility 139-149 mm, and ratio 5.0-6.1; R max value was found 835-998 BU, extensibility 146-135 mm, and ratio 5.7-7.4 in 135 minutes. According the figures attained; for Flamura 85 variety, there was a substantial fall in quality properties with highly sun pest damaged kernels compared to undamaged crop samples. With no sun pest damage and 0.4 damaged Nina bread wheat variety samples; in both years, R max value was found to be 381-453 BU, extensibility 146-176 mm, and ratio 2.6 in both years. While R max value was found 498-538 BU, extensibility 142-178 mm, and ratio 3.5-3.0 in 90 minutes, R max was found to be 545-587 BU, extensibility value 132-170 mm, and ratio 4.1-3.4 in 135 minutes. Crops with 6.0% sun pest damage presented a fall in values in 45 minutes R max as 153 BU, extensibility as 175 mm, and ratio as 0.9. R max was found to be 140, extensibility 161, and ratio 0.9 in 90 minutes while R max value was found 115, extensibility 157, and ratio 0.7 in 135 minutes. Taking these figures into consideration, it was found out that there appears a significant

Taking these figures into consideration, it was found out that there appears a significant fall in R max, extensibility, and ratio values when we compare undamaged or slightly sun pest damaged crop samples with highly sun pest damaged variety samples. It shows us that sun pest damage leads to a substantial resistance deterioration in bread wheat crop varieties. In their study, Köse et al. (1997) prepared 0% (undamaged), 4%, and 6% sun pest damaged crop samples by blending 4 different wheat varieties. They indicated that quality properties of each wheat variety are different from others due to their genetic and variety characteristics even though they are sun pest damaged to the same degree. Sun pest damage caused a fall or an increase to different degrees in extensibility value depending on the wheat variety. On the other hand, Rm/extensibility ratios displayed a considerable change with high sun pest damage ratios. These figures indicate that even 1% sun pest damage ratio causes a notable plummet in extensograph properties, especially in resistance value. Water absorption ratio is the amount of water required for the optimum dough consistency. Kitterman and Rubenthaler (1971) defined flour with water absorption ratio less than 60% as soft wheat flour. After flour is made into dough, farinograph is used to

determine flour's bread properties and kneading properties of dough. With the help of farinograph, information is gathered about dough properties of gluten proteins by measuring dough's resistance to kneader paddles during kneading process, flour's water absorption, and rheological properties (development time, stability, and softening degree) during kneading process. It has been found out that making bread with sun pest damaged wheat is harder depending on the degree of damage; and in most cases, there has been a decrease in processing properties of flour in this condition in the sense that dough from the flour tends to have a fluid quality; and there has been a deterioration in bread characteristics, volume of bread in particular (Elgün and Ertugay 1997). Flours that will be used for bread production should have high water absorption and should not have long kneading time. Long kneading time is an undesirable trait for bakers in the sense that it causes waste of time and energy. However, flours which have short kneading time generally have low bread quality. Water absorption is the amount of water that flour absorbs for suitable dough consistency (500 BU) for high quality bread production. It is expressed on a percentage basis of flour weight. Humidity content of flour plays a crucial role in water absorption. Furthermore, gluten quantity and quality, and the amount of damaged starch also affect water absorption. Development time is between the start of kneading process and the point where curve centers 500 consistency line and starts to fall. Development time is expressed in minutes. Stability is between the points where curve reaches and leaves 500 consistency line and is expressed in minutes. Softening is the distance between curves mid-point after 20 minutes and 500 consistency line.

		s	Softening Degree (BU)	29	45	52	65	61	97	34	76	42	113	67	92	54	86	14	46	105	116	28	72
		Propertie	Stability (dk)	5,7	11,1	5,5	9,2	5,2	4,1	14	2,1	2,2	9,2	3,1	4,7	4,6	3,7	17,0	17,0	2,4	2,7	16	2,5
		Farinograph Properties	time (dk) Development	2,2	2,3	2,3	2	2,5	2,4	2	1,5	1,7	3	1,9	1,9	1,8	2,2	3,2	2,2	1,5	1,5	1,8	1,5
		Fai	Water absorption %	58,90	56,30	56,20	53,4	56,80	58,10	55,5	55,2	56,50	60,50	58,0	53,8	57,70	56,70	59,40	55,00	51,40	52,20	53,50	48,9
			Area (cm <sup>2</sup> )	96	45	173	97	146		116	116	119	104	129	20	151	62	175	121	91	16	126	28
		nutes	сія£Я (Ялах/Е)	7,3	1,3	7,1	4,9	6,7	be drawn	5,2	5,2	3,7	7,5	5,8	1,0	5,7	2,0	7,4	3,5	4,1	6'0	3,4	0,7
s		135. minutes	Extensibility (E) (mm)	105	151	140	124	133	Could not be drawn	133	133	156	107	134	121	146	146	135	163	132	114	170	157
age ratio	es		لا 300) الا 301	763	191	866	601	887	C	685	685	579	798	776	119	835	293	866	567	545	86	587	115
ı pest dan	Extensograph Properties	s	oitsF (HxanF)	5,9	1,6	5,9	4,6	5,9	0,7	5,1	5,1	3,3	6,05	4,9	1,1	5,0	2,1	6,1	3,2	3,5	6'0	3,0	6'0
ferent sur	ensograp	90. minutes	Extensibility (E) (mm)	122	156	142	141	146	125	145	145	174	122	143	134	139	139	149	182	142	126	178	161
with dif	Ext	6	(BU) R max	715	244	834	654	866	91	740	740	577	738	697	152	701	286	911	586	498	112	538	140
t varieties		s	Сайо (Ктах/Е)	3	1,5	3,4	3,1	3,1	0,7	2,6	2,6	2,9	2,91	2,9	2,1	2,6	1,8	4,1	2,3	2,6	1,0	2,6	6'0
ead whea		45. minutes	Extensibility (E) (mm)	148	172	163	151	170	149	179	179	157	158	165	139	177	166	158	193	146	140	176	175
lues of br		4	(BU) R max	447	266	560	463	521	109	473	473	456	460	475	201	470	299	634	453	381	142	453	153
Table 1. Quality analysis values of bread wheat varieties with different sunpest damage ratios	Sun	pest	damage ratio %		3.8	0.2	1.2	0.2	4.5	0.2	2.5	0.2	1.3	0.2	6.0	0	1.0	0.2	1.3	0	2.5	0.4	6.0
Quality a	Year			0100	0107	100	1107	0100	0107	1100	1107	0100	0107		1107	0100	0107	100	1107	0100	0107	1100	
Table 1. (	Variety Name			Colibelu.	מבווססות			20000000	Comment				ouamanup			A	Masuriya			Ning	PIIIA		

#### SECTION 9. FIELD CROP PRODUCTION

Alveograph is the process of measuring dough's resistance to extension (inflation) after dough is produced as a mixture of flour, salt, and water under fixed conditions and a certain piece of it is cut apart and put into a specific shape which is let sit for a while for inflation (Özkaya and Kahveci 1989). In alveograph test, P is tenacity, L: extensibility, P/L: configuration ratio, G: swelling, W: energy index.

It is desired that resistance value is high to a certain extent; and extensibility capacity, swelling value, and energy value are high. If P/L ratio of wheat is 1 or approximately 1, it shows us that resistance and elasticity are well proportioned and the quality of dough is high.

Farinograph and alveograph properties of wheat varieties were analyzed and it was found out that 0.2 sun pest damaged Gelibolu variety had 58.9% water absorption ratio while 3.8 sun pest damaged samples had 56.3% water absorption ratio. The development time of 0.2 sun pest damaged crop samples in both years was 2.2-2.3, stability 5.7-5.5, softening degree 45-65, resistance 138.6-89, extensibility 50.79-70.00, P/L 2.7-1.27, swelling 15.68-16.3, and energy value 281-245. With 3.8% sun pest damaged crop samples, there was a significant deviation from these figures. The development time of 3.8 sun pest damaged kernels was 2.3, stability 11.1, softening degree 29, resistance 88.66, extensibility 46.99, P/L 1.89, swelling 16.52, and energy value 268. It has been found out that sun pest damaged crops at different levels are physically downgraded and flour output is adversely affected. Protein amount is not affected but sedimentation value is reduced because of sun pest damage. Farinogram development time, stability time, mixing tolerance index, and softening degree; and alveogram W, P/L, P, S, and L values are reduced by sun pest damage. If sun pest damage ratio is higher than a specific percent (15%), it is impossible to gluten wash and make bread using that wheat (Atlı et al. 1988).

In 2010-2011 harvest periods, 0.2% sun pest damaged Flamura 85 variety had 2.5-2.0 development time, softening degree 61-34, resistance 110-115, extensibility 67.31-59, P/L 1.6-1.78, swelling 18.05-17.10, and energy value 323-241. The development time of 4.5% sun pest damaged crops was 2.4, stability 4.1, softening degree (climbed to) 97, resistance 76.34, extensibility 33.95, P/L 2.22, and swelling 12.82. Energy value, on the contrary, was found to be much lower due to sun pest damage, 96. These figures show us that sun pest damage around 4-5% results in a deterioration alveograph and farinograph values for Flamura 85 variety, which is regarded as high quality bread wheat.

0.2% sun pest damaged Guadralup variety had 1.7-1.9 development time, 2.2-3.1 stability, softening degree 113-92, resistance 110-102, extensibility 53-58, P/L 2.08-1.76, swelling 16.96-17.0, and energy value 330-231. However, 6% sun pest damaged crop samples demonstrated an undesirable change for these values. The development time of 6% sun pest damaged Guadralup was 1.9, stability 4.7, softening degree 67, resistance 61, extensibility 71, P/L 0.86, swelling 18.8, and energy value 132.

	Alveograph Properties											
Variety Name	Year	Sun pest damage ratio %	P Resistance (mm)	L Extensibility (mm)	P/L	swelling (G) (cm <sup>3</sup> )	W Energy (joule)					
	2010	0.2	138.60	50.79	2.7	15.68	281					
Gelibolu	2010	3.8	88.66	46.99	1.89	16.52	268					
Genbolu	2011	0.2	89.00	70	1.27	16.3	245					
	2011	1.2	48.00	101	0.48	22.4	155					
	2010	0.2	110.00	67.31	1.6	18.05	323					
Flamura85	2010	4.5	76.34	33.95	2.22	12.82	96					
Fiamuraos	2011	0.2	105.00	59	1.78	17.1	241					
		2.5	74.00	45	1.64	14.9	126					
	2010	0.2	110.00	53.00	2.08	16.96	330.9					
Guadralup		1.3	105.00	49.43	2.12	16,04	322					
Guadralup	2011	0.2	102.00	58	1.76	17	231					
	2011	6.0	61.00	71	0.86	18.8	132					
	2010	0	129.00	77.00	1.6	19.32	406.8					
Vaccumuc	2010	1.0	92.95	69.19	1.3	18.30	246					
Krasunya	2011	0.2	146.00	76	1.92	19.4	393					
	2011	1.3	109.00	73	1.49	19.0	319					
	2010	0	59.18	68.30	0.8	18.16	162					
Nina	2010	2.5										
inina	2011	0.4	71.00	67	1.06	18.6	178					
	2011	6.0	69.00	31	2.23	12.4	92					

Table 2. Quality analysis values of bread wheat varieties with different sun pest damage ratios

The development time of undamaged or 0.2% sun pest damaged crop samples was 1.8-3.2, stability 4.6-17, and softening degree 54-14. However; with 1.3% sun pest damaged crop samples, the development time was 2.2, stability 17.0, and softening degree (climbed to) 46. Alveograph values show us that undamaged or 0.2% sun pest damaged samples had highly favorable values such as 129-146 resistance, 77-76 extensibility, 1.6-1.92 P/L, 19.32-19.40 swelling, and 406.8-393 energy value. However, 1.3% sun pest damaged crop samples had 109 resistance, 73 extensibility, 1.49 P/L, swelling 19, and energy value (reduced to) 319.Undamaged Nina bread wheat variety samples had 1.5 development time, 2.4 stability, 105 softening degree, 59.18 resistances, 68.30 extensibility, 0.8 P/L, 18.16 swelling, and 162 energy value. 6% sun pest damaged crop samples had 1.5 development time, 2.5 stability, 72 softening degree, 69 resistance, 31 extensibility, 2.23 P/L, 12.4 swelling value. Energy value which constitutes one of the most important criterions for wheat quality reduced about 40% to 92. When farinograph and alveograph properties are analyzed, it has been found out that high sun pest damage results in significant decline in quality properties even if the figures may alter from one variety to another. Particularly, softening degree appears to rise in various properties while energy values tend to fall significantly. With some of the varieties, these falls appear to be smaller due to sun pest damage. However; with others, even 1% sun pest damage leads to a notable decrease in quality properties of wheat.

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## EFFECT OF PHYTOREMEDIATION ON ABSORPTION OF HEAVY METALS IN THE VELES REGION

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#### Abstract

The paper is based on phytoremediation as a biological measure to improve the overall productivity and revitalization of degraded soils as a potential farmland. This measure is also a way to implement a sustainable system of agricultural production. In the Veles region is established a soil pollution with toxic metals as lead, cadmium, zinc and others thus release of these elements from lead and zinc smelter located in the city of Veles. On the basis of these findings, the research has been performed including four plant species: oilseed rape (*Brassica napus* D.C.), white clover (*Trifolium repens* L.), corn (*Zea mays* L.), and alfalfa (*Medicago sativa* L). Soil samples were taken at the beginning and the end of the assessment for laboratory analysis. The paper presents results of above mentioned species yield and their ability for toxic heavy metals extraction. Analyses show that the highest yield is determined in oilseed rape which shows statistical significant impact (P<0.01) compared with white clover, corn and alfalfa. In terms of heavy metals tested, regardless of plant species and the year of research, the biggest absorption was observed in zinc (Zn). White clover and alfalfa showed the biggest heavy metal extraction in the third trial year, from all trial years.

Keywords: phytoremediation, plant species, heavy metals, Pb, Cd, Zn

### Introduction

Environmental pollution long time ago is the subject of extensive scientific research of the modern society. One method for the decontamination of polluted soils with heavy metals is the method by cultivating plants that perform hyper accumulation of heavy metals. This method or measure in the literature is called phytoremediation (Sekulic, 2003). Those plants that have a high coefficient of bioaccumulation of heavy metals are well known as hyper accumulators. By their growing, there is possibility to be reduced the presence of heavy metals in contaminated soils for several years. In R. Macedonia has been identified 16 hot spots (Environmental statistic, 2007), where the soil is contaminated with heavy metals. "Local Agenda 21 for the Municipality of Veles" presents alarming data, caused by the presence of heavy metals in soils, especially Pb and Zn which provides conditions for receiving and accumulation in the aerial parts of the crops that are grown in Veles region.

So far in the Republic of Macedonia, research methods of decontamination of soils contaminated with heavy metals are not applied, where cultivated plants will be tested in order to establish which of them is right for phytoremediation. This research provides an opportunity to start solving one of the biggest problems of the citizens of Veles, towards obtaining healthy living and production environment.

#### Material and methods

The experiment was set up on the productive areas of Public Communal Enterprise PCE "Derven" in Veles, according to the randomized block system experimental design with four test plants (oilseed rape - Brassica napus Oleifera D.C., white clover - Trifolium repens L., alfalfa - Medicago sativa L., and corn - Zea mays L.) in four repetitions within a period of three years (2008-2010). Before the experiment, average soil samples were taken from 0-20 cm and 20-40 cm for determination of the total and soluble forms of heavy metals (lead, cadmium and zinc) in the soil. Preparation of soil samples was performed in accordance to the ISO 11464:2006 standard. Total soil concentrations of metals were extracted by the Aqua Regia (HCl-HNO<sub>3</sub>, 3:1) method (ISO 11466:1995), their soluble forms were extracted by EDTA (Damić et al., 1996), whereas the plant material was digested in a nitric acid (HNO<sub>3</sub>), (Jones and Case, 1990) and in all extracts observed metals were detected by Atomic Absorption Spectrophotometry (AAS): Varian AA 220 with graphite furnace. During the vegetation, the following parameters were observed: phenophases development of plant, plant height, number of plants per  $m^2$  (vegetative composition) and the yield of green mass. The examinations include generally accepted methods for filed and laboratory examination of soils and the vegetative material (Mitrikeski and Mitkova, 2006, Damić, 1996), and the obtained data is presented in tables.

#### **Results and discussion**

The latest studies of environmental pollution show that the population living near smelters for lead and zinc contain 50% more lead in their bodies than the population of other areas. In conditions of high lead contamination, received lead (Pb) in the tissues of humans and animals via the respiratory or digestive organs, accumulates in the lungs, kidneys, bones and other organs which causes many health disorders, even death. The experiment was made on the right side of the bank of the Vardar River, formed with the sedimentation of the suspended alluvium from the river water. The fixation/mobilization potential of the soil with respect to heavy metals is dependent on the pH, organic matter and clay content of the soil. Table 1 and 2 shows the mechanical composition and chemical properties of the soil.

-												
	Fine soil separates in %											
Ν	Coarse sand Fine sand		sand	Total sand		Silt		Clay		Silt+clay		
IN	<sup>N</sup> 0.2-2mm		0.02-0	.2mm	m 0.02-2mm		0.002-0.02mm		<0.002mm		<0.02mm	
2	$\frac{1}{x}$ %	SD	$\frac{1}{x}$ %	SD	$\frac{1}{x}$ %	SD	$\frac{-}{x}$ %	SD	$\frac{-}{x}$ %	SD	$\frac{1}{x}$ %	SD
_	23.10	2.69	55.10	3.68	78.20	6.36	11.60	1.84	10.20	4.53	21.80	6.36

Table 1. Texture of the soil used for the experiment

x % - average; SD – standard deviation

In soil separates, the fine sand fraction (55.10%) dominates and together with the coarse sand fraction amount to 78.20%, the silt fraction is third (11.60%) and the clay (10.20%) is the last abundant fraction. The soils are characterized by favorable mechanical composition which is confirmed with the classification of the fluvisols in texture classes (fine sandy loam) according to Scheffer and Schachtschabel, (Mitrikeski and Mitkova, 2006).

The results of the research of the chemical properties of the alluvial soil (Table 2) show that the examined soil had little (1.38%) humus contents, medium contents of carbonates (5.95%), (Penkov, 1996) and low alkaline (pH = 7.68) reaction.

	Chemical properties													
Ν	Hu	mus	N	V	pH in	n H <sub>2</sub> O	pH in	n KCl	CaC	$CO_3$	$P_2O_5$		<b>K</b> <sub>2</sub>	0
	0	%	%				%		mg/100g		mg/100g			
2	$\frac{-}{x}$	SD	$\frac{-}{x}$	SD	$\frac{-}{x}$	SD	$\frac{1}{x}$	SD	$\frac{1}{x}$	SD	$\frac{-}{x}$	SD	$\frac{1}{x}$	SD
	1.38	0.33	0.08	0.02	7.68	0.11	6.98	0.04	5.95	1.06	2.86	1.56	11.26	1.20

Table 2. Chemical properties of tested soil used for experiment

x - average; SD – standard deviation

The neutral and low alkaline reaction of the soil solution as well as the organic and mineral colloids, limit the heavy metal mobility, including their easily available forms. In the Republic of Macedonia we still refer to the EU standards on maximally permitted concentration of heavy metals in the soil. Maximum Allowable Concentrations (MAC) of trace elements in agricultural soils from EU standards should not be more than: Pb - 100 ppm, Zn (150-300 ppm) and Cd 1-3 ppm (Kabata-Pendias et al., 2000). In our country (Филиповски, 2003) suggests the following MAC: Pb - 100 ppm, Zn - 200 ppm and Cd - 3 ppm. The results on the total contents of heavy metals in the soil have indicated that all three elements are present over the maximally permitted concentrations (Table 3) which means that the soils are not safe for healthy food production.

Total (mg kg <sup>-1</sup> )							EDTA-extractable (mg kg <sup>-1</sup> )						
Ν	Pb	)	Cd		Zn		Pb		Cd		Zn		
2	$\frac{1}{x}$	SD	$\frac{-}{x}$	SD	$\frac{1}{x}$	SD	$\frac{1}{x}$	SD	$\frac{1}{x}$	SD	$\frac{1}{x}$	SD	
2	176.80	1.18	5.58	0.21	330,34	12.03	81.60	10.47	3.46	0.25	64.27	2.04	

Table 3. Content of heavy metals in the soil used for the experiment

x - average; SD – standard deviation

This condition of soil contamination is suitable for implementation of the phytoremediation measure which is not very efficient when it comes to greater contents of heavy metals in the soil. Table 3 shows the total and soluble forms of heavy metals in the soil. Soluble forms of the lead are (46%) of the total amount, cadmium (62%) and zinc (19.45%) of total forms of these heavy metals in the soil. The survey results (Table 4) showed that the yield of hay (dry matter) for perennial crops (alfalfa and white clover) is the highest in the second year and for annual (maize and oilseed rape) in the first year. Average yields of hay (dry matter) showed that oilseed rape gave the highest yields – 32.041 kg/ha from all plant species in the three year study, compared to alfalfa – 13.409,83

kg/ha, maize – 13.305,6 kg/ha and white clover – 83.83,4 kg/ha (Table 4). White clover in the three year study showed the greatest power of extracting zinc (118,50 mg/kg) compared with the power of extracting lead (17,34 mg/kg) and cadmium (1,17 mg/kg). Its power to extract heavy metals is highest in the second year, with the exception of cadmium, whose extraction is greatest in the first year. Alfalfa in the three year study showed highest total power of extracting zinc (112,93 mg/kg), then the lead (23,88 mg/kg) and the lowest in cadmium (0,70 mg/kg). The greatest extraction of all tested heavy metals was noticed in the second year. Maize in the three-year period of research showed the greatest power of extracting zinc (39,69 mg/kg), then the lead (16,78 mg/kg) and at least cadmium (0,49 mg/kg). Extraction of zinc and cadmium is highest in the first year of the study, unlike the lead whose extraction is greatest in the second year of the study.

Oilseed rape during the study showed the greatest power in the zinc extraction (18,31 mg/kg), then the lead (4,83 mg/kg) and the cadmium (0,43 mg/kg). Its power to extract heavy metals is strongest in the second year, with the exception of cadmium, whose extraction is greatest in the first year. The analysis of the power of extracting heavy metals per year for each of the tested crops showed that the highest absorption of lead showed alfalfa (23,88 mg/kg) in the examined period, while the smallest oilseed rape (4,83 mg/kg). The greatest power of extracting cadmium is established in white clover (1,17 mg/kg), The greatest power of extracting zinc is determined in white clover (118,50 mg/kg), while the smallest is determined in oilseed rape (18,31 mg/kg).

		Hay yield (kg/ha)		
Year	White clover	Alfalfa	Maize	Oilseed rape
2009	5465.70	12633.30	20416.7	41208.0
2010	11320.50	14890.70	12583.4	28917.0
2011	8364.00	12705.5	6916.7	25998.0
Average	8383.40	13409.83	13305.60	32041.00
		Pb (mg/kg)		
Year	White clover	Alfalfa	Maize	Oilseed rape
2009	5.58	8.68	6.75	1.25
2010	41.00	58.00	43.50	13.00
2011	5.43	4.95	0.10	0.25
Average	17.34	23.88	16.78	4.83
		Cd (mg/kg)		
Year	White clover	Alfalfa	Maize	Oilseed rape
2009	1.23	0.14	1.00	0.53
2010	1.20	1.70	0.40	0.50
2011	1.08	0.26	0.06	0.26
Average	1.17	0.70	0.49	0.43
		Zn (mg/kg)		
Year	White clover	Alfalfa	Maize	Oilseed rape
2009	73.34	48.26	106.33	16.41
2010	195.10	275.00	4.50	32.10
2011	87.06	15.53	8.25	6.42
Average	118.50	112.93	39.69	18.31

Table 4. Hay yield (dry matter) and heavy metal content in the tested crops

Besides mathematical obtained values and established average crop yields as well as their power of extracting heavy metals from the soil over a period of three years, the statistical analysis of the results were obtained (Table 5).

Hay yield (kg/ha)								
Year	White clover	Alfalfa	Maize	Oilseed rape				
2009	5465.70	12633.30	20416.7	41208.0				
2010	11320.50	14890.70	12583.4	28917.0				
2011	8364.00	12705.5	6916.7	25998.0				
Просек	8383.40	13409.83	13305.60	32041.00				

Table 5. Hay yield of trial crops for a period of 3 years

LSD <sub>0,05</sub> = 10334

LSD <sub>0,01</sub> = 15654

The analysis of the hay (dry matter) yield showed that there are statistically significant differences among the tested crops at the level of 0.01% of probability. Differences arise between oilseed rapes compared to all other crops.

	White clover	Alfalfa	Maize	Oilseed rape				
mg/kg								
Pb	17.34	23.88	16.78	4.83				
Cd	1.17	0.70	0.49	0.43				
Zn	118.50	112.93	39.69	18.31				

Table 6. Heavy metals content in assessed crops (mg/kg)

LSD <sub>0,05</sub> = 19,32

LSD  $_{0,01} = 26,04$ 

In terms of heavy metals assessed, regardless of plant species and the year of research, the biggest absorption was observed in zinc (Zn). White clover and alfalfa showed the biggest heavy metal extraction in the third trial year, from all trial years at the level of 0,05% - LSD  $_{0,05} = 2,51$  and LSD  $_{0,01} = 3,67$ .

### Conclusion

White clover in the three year study showed the greatest power of extracting zinc. Its power to extract heavy metals is highest in the second year, with the exception of cadmium, whose extraction is greatest in the first year. Alfalfa in the three year study showed highest power of extracting zinc. The greatest extraction of all assessed heavy metals was noticed in the second year. Maize in the three-year period of research showed the greatest power of extracting zinc. Extraction of zinc and cadmium is highest in the first year of the study, unlike the lead whose extraction is greatest in the second year of the study. Alfalfa and oilseed rape have shown the greatest absorption of zinc in the period under study. The biggest draw of the lead in the examined period showed alfalfa (23,88 mg/kg) and the lowest oilseed rape (4,83 mg/kg). The greatest power of extracting zinc is determined in white clover (118,50 mg/kg), while the smallest is determined in oilseed

rape (18,31 mg/kg). There are statistically significant differences among the tested crops at the level of 0.01%. Differences arise between oilseed rapes compared to all other crops.

In terms of heavy metals assessed, regardless of plant species and the year of research, the biggest absorption was observed in zinc (Zn). White clover and alfalfa showed the biggest heavy metal extraction in the third trial year, from all trial years at the level of 0,05%. High yield of oilseed rape does not affect the extraction of zinc. The biggest draw is observed in perennial crops alfalfa and white clover.

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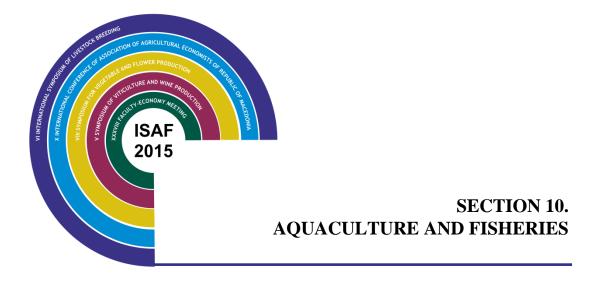
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### DETERMINATION OF RNA/DNA RATIO IN WHITE MUSCLE SAMPLES OF KOI CARP USING DIFFERENT TECHNIQUES

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#### Abstract

The RNA/DNA (R/D) ratio is an indicator of muscle growth capacity in fish. The amount of DNA in a cell is constant; while the amount of RNA indicates how actively the cell is synthesizing proteins and growing. An increase or decrease in the R/D ratio at a given temperature would indicate a change in growth rate. Some studies have advocated caution in the use of the RNA/DNA ratio because of the lack of sensitivity of some techniques. In this study we estimated the RNA/DNA ratio in 24 samples of Koi carp divided in 2 groups (small and large size) and three temperature subgroups 5°C, 25°C and 30°C. All fish were grown under the same feeding conditions for 6 weeks. Two different techniques (Triazol™ and Qiagene<sup>TM</sup>) were used to isolate and purify RNA and DNA. The Triazol<sup>TM</sup> method was slightly modified, while the commercial Qiagene method was used for simultaneous isolation of RNA and DNA according to the manufacturer's recommendation. The results showed that the average values for the R/D ratio in large fish adapted to 25°C was the highest in both cases (Qiagene<sup>TM</sup>3.3 and Triazol<sup>TM</sup>4.23), while the difference was shown in the samples from fish adapted to 5°C and 30°C. The results obtained using the Triazol<sup>™</sup> method are in agreement with the expected values where the fish adapted to 5°C were higher compared with those from the fish adapted to 30°C (P=0.027). The results from the Qiagene<sup>™</sup> isolation were opposite (P=0.085). The values in all samples are higher using Triazol<sup>™</sup> method by approximately 25% (P=0.000). Based on these results we propose that the Triazol<sup>™</sup> method is a more reliable method in determining the RNA/DNA ratio in fish muscle samples.

Key words: koi carp, RNA/DNA ratio, Triazol<sup>TM</sup>, Qiagene<sup>TM</sup>, temperature, growth

### Introduction

The commonly used tools to measure recent growth and nutritional condition of fish species are based on DNA and RNA concentration ( $\mu$ g/mg tissue), RNA/mg tissue, ratio of

RNA to DNA etc. The amount of DNA in a cell is constant while the amount of RNA indicates how actively the cell is in synthesizing proteins (Chicharo, L. M. Z., Chicharo, M. A. A. T. 1995). RNA comprises much of the cell's machinery for protein biosynthesis and its content fluctuates in response to food availability in the natural food habitats (Rooker, J. R., Holt, G. J. 1996). When properly calibrated with laboratory experiments, the RNA/DNA (R/D) ratio can also be used to estimate instantaneous growth rates. Changes in protein synthesis directly correlate with R/D, although this pattern depends on tissue type and the extent of laboratory treatment (Lowery, MS, Somero GN. 1990). Some studies have advocated caution in the use of R/D ratios on the basis of the lack of sensitivity of some techniques and the influence of the analytical protocol and standard used (Calderone, E et. al. 2006). It has also been pointed out that the R/D ratio may be influenced by temperature. Quantification of R/D ratio can be done using spectrophotometric, fluorometric and HPLC methods (Triant, D.A., Whitehead, A. 2009). In this study we were comparing the R/D ratio isolated from the white muscle samples from different sizes of koi carp grown at different temperatures using two spectrophotometrical techniques.

### Material and methods

*Material*. Twenty four individuals divided in two groups (small and large) and in three subgroups based on the growing temperatures (5, 25 and 30°C) were investigated during the study. Technically, 4 fish of "small" and "large" group (in total 8) were grown in 3 tanks of 50 L on the three mentioned temperatures for 6 weeks. After that, all fish have been harvested and stored at -80 °C. The sample data are given in a Table 1.

Temperature ( <sup>0</sup> C)	Size	Weight (g)	Temperature ( <sup>0</sup> C)	Size	Weight (g)	Temperature ( <sup>0</sup> C)	Size	Weight (g)
	Small	16,8 13,6 16,4 16,1	25	Small	52,69 66,37 59,01 60,18	20	Small	48,7 56,71 56,35 67,54
5	Large	57,6 56,8 56,9 65,4	25	Large	182,58146,71171,95214,02	30	Large	160,72 132,1 129,87 132,2

Table 1. Fish data

*Methods*. Simultaneous extraction of RNA and DNA was done using TRIzol and Qiagene method.

**TRiazol** method for simultaneous extraction of RNA and DNA subsequently was performed according to the manufacturer's protocol (9) with some modification as follow. Approximately 50 mg of white muscle was scraped from -80 °C frozen fish, precisely measured ( $\pm$  0.0001 g) and immersed in 1.5 mL of TRIzol reagent in Petri dish for *RNA extraction*. The tissue was minced using scalpel blade and transferred to cryovial. The muscle tissue was than homogenized by sonification at maximum speed for 30 seconds. The sample was incubated for 10 minutes at room temperature and after the incubation, 0.2 ml of chloroform was added and shaked for 30 sec It was followed by incubation for 5 min

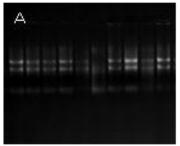
at room temperature and transfer to 1.5 ml microfuge tube. The solution was centrifuged in a microfuge at 12.000 g/ 10' at room temperature. The aqueous layer very carefully was transferred in a new microfuge tube avoiding any contact with interphase and organic layer. In order to extract total RNA, we performed the new step of rinsing the interphase and organic layer with 0.2 ml HPLC grade water. The sample was again centrifuged at 12.000 g/ 10' at room temperature. The rest of aqueous phase was merged with the previous amount, 0.5 ml isopropanol were added and incubated at room temperature for 10 minutes. The solution was spined at 12.000 g for 15' at room temperature. The supernatant was discarded and the pellet was washed with 1 ml 75% ethanol. The solution was vortexed briefly and spin at 12.000 g for 5 minutes at room temperature. Supernatant was discarded by decanting and the tube was placed upside down on wipe to dry for 15 minutes. The pellet was resuspended as RNA extract in 50% formamide/50% DEPC water and stored at - 20°C. The interphase and organic layer from the procedure for RNA isolation were used for DNA extraction following the protocol from TRiazol method. They were twice rinsed with 200 µl HPLC grade H<sub>2</sub>O and centrifuged at 12,000 x g for 5 min at room temperature. In the aqueous phase were added 450 µl of absolute ethanol. The content was mixed by inverting the tube few times. Than it was incubated for 5 minutes and centrifuged at 2,000 x g for 5 min. The supernatant was removed and DNA pellet was washed with 1 ml 0.1 M sodium citrate and 10% ethanol solution (pH 8.5). Again it was incubated for 30 min and centrifuge at 2,000 x g for 5 min. In the tube was added 1.5 ml 75% ethanol and incubated for 15 min with occasional gentle inversion. It was followed by centrifugation at 2,000 x g for 15 min. DNA pellet was dried for 10 min and re-suspended in 100 µl of 8 mM NaOH. The solution was centrifuged at 12,000 x g for 10 min. The insoluble material was removed and keep DNA solution was stored at 4°C.

**Oiagene** protocol (6) was performed according to the manufacturer manual. Briefly, 30 mg of white muscle were precisely measured, disrupted and homogenized in 0,6 mL buffer RLT Plus. The lysate was pipetted directly into a QIA shredder spin column placed in a 2 ml collection tube, and centrifuged for 3 min at maximum speed. The supernatant was carefully removed by pipetting, transferred it to the AllPrep DNA spin column placed in a 2 ml collection tube (supplied) and centrifuged for 30 s at 8000 x g. AllPrep DNA spin column was put in a new 2 ml collection tube and store at 4°C for later DNA purification. The flow-through solution was used for RNA purification. One/one volume (approximately 600 µl) of 70% ethanol was added to the flow-through solution and mix well by pipetting. The sample including any precipitate that may have formed (about 700 ul) was transferred to an RNeasy spin column previously placed in a 2 ml collection tube (supplied). The column was centrifuged for 15 s at 8000 x g. The flow-through was discarded and 700 µl of buffer RW1 were added to the RNeasy spin column. The centrifugation for 15 s at 8000 x g was done to wash the spin column membrane. The flowthrough was discarded, 500 µl of buffer RPE were added to the RNeasy spin column and centrifuged for 15 s at 8000 x g to wash the spin column membrane. The flow-through was discarded and 500 µl of buffer RPE were added to the RNeasy spin column and centrifuged for 2 min at 8000 x g to wash the spin column membrane. The RNeasy spin column was placed in a new 1.5 ml collection tube, 30-50 µl RNase-free water was added directly to the spin column membrane and centrifuged for 1 min at 8000 x g to elute the RNA. The genomic DNA purification was continued by adding of 500 µl Buffer AW1 to the AllPrep DNA spin column and centrifuged for 15 s at 8000 x g. The flow-through was discarded, 500 µl buffer AW2 was added to the AllPrep DNA spin column and centrifuged

for 2 min at full speed to wash the spin column membrane. The AllPrep DNA spin column was placed in a new 1.5 ml collection tube, 100  $\mu$ l Buffer EB were added directly to the spin column membrane, incubated at room temperature for 1 min and centrifuged for 1 min at 8000 x g. RNA and DNA were simultaneously isolated from 24 samples. All extractions were done in two replicates. The quantification of extracted nucleic acids was performed spectrophotometrically using Nanodrop technique. The integrity of isolated NA was checked on 0.9% agarose TBE gel.

### **Results and discussion**

Simultaneous and subsequent isolation of RNA and DNA from the same initial material and obtained RNA/DNA ratio can serve as an indicator of the growing potential in fish white muscle. But, there are some obstacles which may compromise the interpretation of RNA/DNA ratio (Dellanno A., et al 1998). The main problem is fragility of RNA molecule during the extraction process and the sureness of a quantitative collection of DNA after the previous RNA isolation from the same sample. That's why we approach the extraction of NA from white muscle of *Cyprinus carpio haematopterus* (Koi carp) using two different techniques, Qiagene and TRIzol. The 0.9% Agarose gel electrophoresis on the Figure 1 demonstrated 18S and 28S RNA bands indicating intact RNA. It can be seen that the isolates of RNA obtained using Qiagene were not uniform in the quantity compared to those obtained with TRIzol, although the same quantity of starting material was used and same amount of extracted RNA was loaded on the gel.



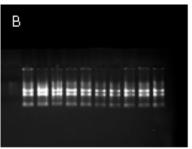
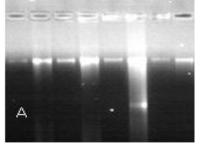


Figure 1. 0.9% AGE of RNA isolates. A) RNA isolates using Qiagene. B) RNA isolates using TRIzol

The extracted DNA isolates obtained using both techniques showed high level of DNA integrity (Figure 2). It should be point out that having in mind the previous procedure of extraction of RNA, the extracted DNA very often can't be used for further functional analysis.



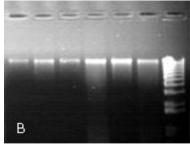
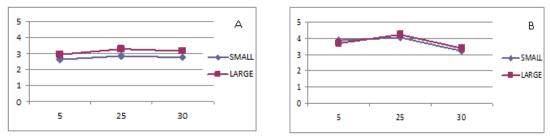


Figure 2. 0.9% AGE of DNA isolates A) DNA isolates using Qiagene. B) DNA isolates using **TRIzol** 

The quantification of extracted NA was done spectrophotometricaly using Nanodrop. The results were expressed as ng NA/ $\Box$ L and afterward based on the final volume were converted in absolute value of RNA and DNA, respectively.

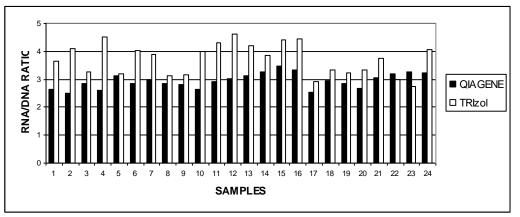
Based on the experimental design, we calculated mean values of RNA/DNA ratio in small and large fish acclimated on 5, 25 and 30°C, respectively. The results from these calculations are presented in Graphic 1.



Graphic 1 Average values of RNA/DNA ratio among different groups of fish grown at different temperatures determined using A) Qiagene kit, B) TRIzol techniques

The results showed that the average values for the R/D ratio in large fish adapted to 25°C was the highest in both cases (Qiagene<sup>TM</sup>3.3 and Triazol<sup>TM</sup>4.23), while the difference was shown in the samples from fish adapted to  $5^{\circ}$ C and  $30^{\circ}$ C. The results obtained using the TRIazol<sup>TM</sup> method are in accordance with the expected values where in the fish adapted to  $5^{\circ}$ C were higher compared with those from the fish adapted to  $30^{\circ}$ C (P=0.027) (Dellanno A., et al 1998). The results from the Qiagene<sup>™</sup> isolation were opposite (P=0.085).

The results of RNA/DNA ratio obtained using both techniques are shown in Graphic 2.



Graphic 2. Cumulative data for RNA/DNA ratio obtained with Qiagene and TRIzol technique

### Conclusion

The values in all samples are higher using Triazol<sup>™</sup> method compared to Qiagene by approximately 25% (P=0.000).

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# THE RATIO BETWEEN THE INTESTINE LENGTH AND THE BODY LENGTH IN SALMO OHRIDANUS STEINDACHNER FROM OHRID LAKE

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### Abstract

With the aim to investigate the relationship between the intestine length and the body length od Ohrid belvica (*Salmo ohridanus*, Steindachner, 1892), investigations were conducted on individuals captured in vegetative and reproductive development period of belvica, on three localities of Ohrid Lake: Kalista, Kaneo and Elesec, on depth from 45 to 70 m. Samples for researching have average body mass from 90 to 100 g and average total body length from 217 to 225 mm. It was found that this ratio varies from 0.73 to 0.98 for the three localities during the two developmental stages, or that is an average of 0.80 to 0.89. It has been established that the differences between the intestine length and the absolute body length, such as between localities and between the stages of development of Ohrid belvica are small and statistically insignificant. According to the results of the ratio between the intestine length and the body length, Ohrid belvica belongs to the group of zoophagous or carnivores.

Key words: Ohrid belvica (Salmo ohridanus); intestine length; body length.

### Introduction

The ratio determination between the intestine length and the body length of various fish species was being studied by many authors (Aganović M., Kapetanović, 1970; Aganović et al., 1976; Aganović & Vuković, 1966; Fryer & Iles, 1972; Rahman et al., 2012; Vuković, 1966; Vuković et al., 1972). Most of the authors concluded that in fish, as in many other taxa, the intestine length is often an indicator of diet. Species that eat only algae or higher plants (herbivores) tend to have longer intestines than species that eat both plant and animal material (omnivores), and this in turn tend to have longer intestines than species that species that eat only other animals (carnivores) (Fryer & Iles, 1972; Kapoor et al., 1975; Karachle & Stergiou, 2010a, 2010b; Kramer & Bryant, 1995a, 1995b). In fact the ratio between the intestine length and the body length, according to Al-Hussaini (1947) and Kapoor et al. (1975) ranges from 0.5 to 2.4 for carnivores, 0.8 to 5 for omnivores, and 2 to 21 for herbivores.

### Material and methods

Investigations were conducted on fish species of Ohrid belvica (*Salmo ohridanus*, Steindachner, 1892). The samples were captured in vegetative and reproductive development period of Ohrid belvica, on three localities of Ohrid Lake: Kalista, Kaneo and Elesec, on depth from 45 to 70 m. Tests were performed on 15 samples of each locality and each phase of development or a total of 90 fish. The body length was determined by using a caliper and a metal ruler with an accuracy of 1 mm. Measuring the total (absolute) length of the body is made under the scheme of Smitt, modified by Правдин (1966) and it is length from the top of the muzzle (snout) to the end of the middle rays of the caudal fin. For the purposes of measuring the intestine length it has been performed carefully extracting the digestive tract of the fish after cutting the esophagus and rectum. The ratio between the intestine length and the body length was calculated by the following formula (Ta\_ABEKM, 2003): Rt = intestine length / total body length. The data obtained with the measurements are processed by the method of the smallest squares, and the results consist of the arithmetic mean ( $\overline{H}$ ), standard deviation (SD), coefficient of variation (Cv) and differences in  $\overline{H}$ .

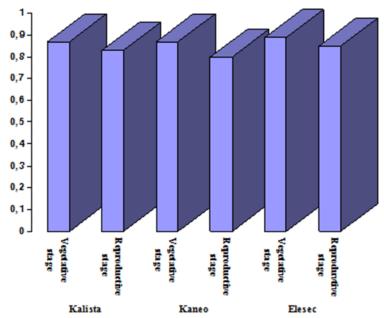
### **Results and discussion**

The average relation to the intestine length and the absolute body length of Ohrid belvica is shown in Table 1 and Graph 1. The value of the ratio of the individuals from the locality Kalista during the vegetative period, varies in the range of 0.77 to 0.95, while during the reproductive stage varies in the range of 0.74 to 0.90. At the individuals from the locality Kaneo, values are ranging from 0.76 to 0.94 in vegetative or from 0.73 to 0.82 in the reproductive stage. The Elesec locality values are ranging from 0.76 to 0.98.

Index	H	SD	CV	Differences in $\overline{H}$	
KALISTA					
Vegetative stage	0,87	0,06	6,90	0,04 <sup>NS</sup>	
Reproductive stage	0,83	0,05	6,02	0,04	
KANEO					
Vegetative stage	0,87	0,05	5,75	0,07 <sup>NS</sup>	
Reproductive stage	0,80	0,03	3,75	0,07	
ELESEC					
Vegetative stage	0,89	0,07	7,86	0,04 <sup>NS</sup>	
Reproductive stage	0,85	0,06	7,06	0,04	

Table 1. Ratio between the intestine length and the total body length in Ohrid belvica (n=15)

NS – not significant



Graph 1. Ratio between the intestine length and the total body length in Ohrid belvica

The differences in the ratio of the intestine length and the total body length, between localities as well as between the stages of development of Ohrid belvica, are small and statistically insignificant. It might be concluded that the intestine length of Ohrid belvica is slightly smaller than the body length, not taking consideration, understandably, the participation of pyloric ceca.

In the ichthyologic literature there can be found enough data about the ratio of the intestine length and the body length for the different fish species. Most authors have come to the conclusion that this morphological character is dependent on the species, especially the way of his diet, but on environmental conditions as well. It has been established that the ratio between the intestinal length and the body length in fish is gradually narrowing towards the phytophagous to zoophagous (Das & Moitra, 1958, quoted by Илиев & Цинева, 1974; Šorić, 1982). Most authors (Aganović & Kapetanović, 1970; Aganović et al. 1976; Aganović & Vuković, 1966; German & Horn, 2006; Horn, 1989; Илиев, 1971; Vuković, 1966, 1968; Vuković et al., 1972) concluded that the intestine length in fish depends, above all, on their diet, i.e. its qualitative and quantitative composition. Namely, the intestine length of a fish in which diet animal component prevails (carnivorous fish species) is less, apart from those in which plant foods prevail. According to Wootton (1998), the intestine length depends on the philosophy of digestion since the intestinal surface required for the since the absorption of nutrients is directly linked to digestive efficiency. Fish that consume food of high quality (e.g. other fish) can absorb nutrients by possessing guts shorter than their total body length. Conversely, fish that prey on food items, that are resistant to digestion (e.g. plant material and crustacean axoskeletons) seem to have a proportionally longer intestine compared to their body length.

The intestinal length in relation to the body length in zoophagous may be up to 50%. Fish species, characterized according to diet as zoophytophagous, occupy an

intermediate position. The shortest intestinal tract is at typical predatory fish, such as trout, zander and others, which is understandably considering the nutrition. Generally, in all types of fish that throughout their life are fed exclusively with animal foods, the ratio between the intestine length and body length is always less than 1, or reaches a maximum around 1 (ichthyophagous, zoobentophagous and zooplanktophagous). In those fish, which besides zooplankton diet, occasionally use plant components as well, the observed ratio varies from 1 to 3, and in those in which the plant component is prevalent in the diet, intestinal tract is over 3 times longer than their body length (Aganović & Kapetanović, 1970).

In predatory fish the intestine is short and straight, in omnivores it is with medium length and in herbivores and makroplanktophagous it is up to fifteen times longer than the body length (Јовановић-Кршњанин, 1989, quoted by Талевски, 2003). The length of the intestinal tract can be used as a systematic characteristic of the fishes (Aganović & Kapetanović, 1970; Šorić, 1982).

# Conclusions

Based on the results of the determination of the ratio between the intestine length and the total body length of Ohrid belvica from three localities of Lake Ohrid during the vegetative and reproductive stage, it might be concluded the following:

- The ratio between the intestinal length and the total body length of Ohrid belvica ranges from 0.73 to 0.89, or an average from 0.80 to 0.89.

- Differences between the intestine length and the total body length regarding localities and the stages of development of Ohrid belvica are small and statistically insignificant.

- According to the ratio between the intestinal length and the body length, Ohrid belvica belongs to the group of zoophagous or zooplanktophagous, zoobentophagous.

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# WATER QUALITY FOR FISH FARMING IN THE POND "MODRIC"

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### Abstract

Water quality for fish farming was examined in the pond "Modric". Pond is on the way for Modric village, about 400 m away from the main road Debar–Struga. It has an area of  $1200 \text{ m}^2$ . The pond is coldwater and rainbow trout (*Oncorhynchus mykiss* Walbaum) is grown in it. During the months: March, May, October and December, physical-chemical and microbiological testing of water were carried out at the entrance and the exit in pools where fish were raised. It was found that the quality of water, according to the physico-chemical and microbiological characteristics, is good for growing trout. It is recommended frequently cleaning the fish ponds, preferably twice a week, because healthy fish for the market are grown in clean water.

Key words: pond water, physico-chemical and microbiological characteristics.

### Introduction

Fish production in fish farms in our country has seen a trend of rapid development in recent decades. The number and the type of fish facilities are multiplied as well as the quantity of obtained fish and the intensity of cultivation. The large amount of water that is used for construction of fish ponds is varying in type and size. The quality of the water in the ponds is of great importance for success in the cultivation of fish and their quality. Fish health, productivity and profitability of the pond are directly dependent on the water. Chemical and microbiological characteristics and the purity of water are of primary importance for fish cultivation, as well (Gorlenko et al., 1983; Зибероски & Наумовски, 1993; Стевановски & Христова, 2010). When water pollution will exceed the allowed amount, a negative impact on the aquatic life and fish may be expected. This adverse effect is a consequence of lack of oxygen or due to the direct harmful effect and toxicity of certain chemical compounds which contaminated water. The morphological and biochemical characteristics of microorganisms in the water depend on the type of aquatic ecosystem, location, depth (Gorlenko et al., 1983; Perez, 1987; Rodina, 1972).

From a biological point of view, pure water is defined as water in which the fish might normally live, eat, grow, proliferate and generally perform all biological processes specific to them. From the physical-chemical aspect, wholesome and biologically pure water is the most important link in the chain of circulation of matter and energy in aquatic ecosystems. Each disrupting of the limits, in which the most important physical and chemical parameters of the water have to move, reflected on the wildlife in it, and consequently the fish, for which the water is environment (Наумовски & Беличовска, 2009). Therefore, studies of the water quality are of crucial importance for growing fish.

### Material and methods

Pond "Modric" is on the road to the village of Modric, about 400 m away from the main road Struga-Debar. It began working in 1996. There is an area of  $1200 \text{ m}^2$ , it is built of reinforced concrete and has 16 swimming pools set in two rows of 8 pools. One of the pools is used for breeding offspring, 9 pools are used as growing up pools for smaller fish and 6 fish ponds are used as growing up pools for commercial fish. A part of the water from Modric River is used for the purposes of the pond. The pond is coldwater and rainbow trout (*Oncorhynchus mykiss* Walbaum) is grown in it. The capacity of the pond is 30 000 kg fish per year. The pond is supplied with the spawn from fish ponds that produce raw materials in the country.

Tests were performed on samples of water taken during the months: March, May, October and December. Physical-chemical and microbiological testing of water were carried out at the entrance and the exit in pools where fish were raised. Common methods were used to determine the chemical composition of the water, and for the microbiological analysis standard methods for examination of various microorganisms, especially specific harmful bacteria were applied. Microbiological methods used for determination of certain groups of microorganisms are following:

- 1. For total bacteria count MPA method is used.
- 2. Brilliant green agar is used for Salmonella detection.
- 3. Chapman substrate for detection of Staphilococcus pyogenes.
- 4. SS substrate for detection of Proteus and Streptococcus.
- 5. Sulphite reducing bacteria are detected by sulphite agar.
- 6. Clostridia are detected by sulphite agar.
- 7. MPA agar for detecting of mesophilic bacteria.
- 8. Sabourald-agar for determination of yiests.
- 9. Czapek agar for detecting of moulds.
- 10. Suitable substrate is used for detecting of Pseudomonas aeruginosa.
- 11. Endo-agar for Escherichia coli.
- 12. Starch-ammonia agar and Andrade indicator for detecting the contaminants.

Tests were performed at the Faculty of Agricultural Sciences and Food in Skopje.

### **Results and discussion**

The results of chemical and microbiological analysis of the water taken by the pools of the pond "Modric" are shown in Tables 1–8.

Constituents of the water	MPQ	M-1	М-2	M-3	M-4	М-5	М-6	М-7
Organic Substances, %		0,3	0,4	0,5	0,7	0,8	0,9	0,5
pH	6,5 - 9	7,3	7,2	7,2	7,3	7,3	7,4	7,4
NH <sub>4</sub>	0,001	0	0	0	0	0,0001	0,0002	0,0001
H <sub>2</sub> S	0,005	0,0001	0	0	0	0,0002	0,0001	0,0001
Cyanide	0	0	0	0	0	0	0	0
Free Residual Chlorine	0,005	0	0	0	0	0	0	0
Chloride	0,005	0,0003	0,0004	0,0001	0,0001	0,0001	0,0001	0,0002
Phenols	0	0	0	0	0	0	0	0
Nitrites	0,005	0,004	0,0045	0,0039	0,002	0,003	0,0035	0,004
Nitrates	15,0	8,0	10,0	9,5	8,9	10,5	7,5	8,4
Carbonates	400	184	179	201	188	195	180	185
Sulphates	0,05	0,002	0,001	0,004	0,006	0,002	0,007	0,008
Phosphates	0,05	0,001	0,006	0,002	0,003	0,001	0,009	0,008
Lead	0,05	0,0001	0,0002	0,0001	0,0002	0,0002	0,0001	0,0001
Zink	5,0	0,0001	0,0001	0,0003	0,0001	0,0001	0,0003	0,0002
Mercury	0,001	0	0	0	0	0	0	0
Cadmium	-	0	0	0	0	0	0	0
Arsenic	-	0	0	0	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0	0	0	0
O <sub>2</sub> – Oxygen	> 5,0	11,0	8,5	17,5	12,5	10,5	12,5	11,6
$CO_2$ – Carbon dioxide	< 5,0	3,0	4,5	5,3	5,1	5,3	4,3	3,9
Electrical conductivity	500	420	390	380	410	400	420	430
Purity	pure	pure	pure	gary	gary	gary	gary	gary
Ca – Calcium	200	200	180	190	195	187	200	195
Mg – Magnesium	850	115	120	110	120	100	110	115
Fe – Iron	0,03	0,002	0,003	0,003	0,002	0,004	0,002	0,003
Pesticides	0	0,0001	0,0001	0	0	0	0,0001	0

Table 1. Chemical analysis of water on the 12<sup>th</sup> of March (ppm)

MPQ - maximum permissible quantity; M-1 - water pool with offspring;

M-2 – water from entering the incubator; M-3 – water from the entrance of the pool with offspring;

M-4 - water pool without fish; M-5 - water while leaving the incubator "spring water";

M-6 – water while leaving the pond; M-7 – water from entering the pond.

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the limits of allowed and even oxygen was quite present, and  $CO_2$  was not so amassed to cause a problem in fish. It is important that there were no contamination with heavy metals and pesticides. Deviations in separate pools is, apparently, due to inadequate flow of water in the pool, the higher amount of added food than necessary, or falling the deteriorating material from outside or untimely cleaning the pool, considering the fact that in the other pools situation is normal in terms of the individual parameter.

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			1	1	1	
M-1	M-2	M-3	M-4	M-5	М-6	M-7
0	0	0	0	0	0	0
0	0	0	0	0	0	0
0	0	0	0	2	1	0
0	0	0	0	0	0	0
0	0	0	0	0	0	0
0	0	0	0	0	0	0
0	0	0	0	0	0	0
0	0	0	0	0	0	0
0	0	0	2	0	2	2
0	0	0	0	0	0	0
0	0	0	0	0	0	0
8	12	18	22	6	15	12
650	640	750	840	490	690	710
0	0	0	0	2	3	3
0	0	0	0	0	0	0
0	0	0	0	0	0	0
	M-1 0 0 0 0 0 0 0 0 0 0 0 0 0 8 650 0 0 0	M-1         M-2           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           0         0           8         12           650         640           0         0           0         0	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$

Table 2. Microbiological analysis of water on the 12<sup>th</sup> of March

According to the microbiological analysis, water had some increased amounts of organic matters, due to which it was a little muddy. That was the reason for an increased number of bacteria, even a small number of coliform bacteria, which were probably a result of the food used for feeding the fish. However, this water may be used for fish farming, because the bacteria and other microorganisms were in the permissible limit under Regulation for Sanitary Safety of Drinking Water (Official Gazette 33/1987).

Constituents of the water	MPQ	М-1	M-2	M-3	М-4	М-5	М-6	М-7
Organic Substances, %		0,20	0,219	0,18	0,20	0,17	0,18	0,17
pН	6,5 - 9	6,8	6,9	6,9	6,8	6,8	6,8	6,8
NH <sub>4</sub>	0,1	0,0003	0,0011	0,0006	0,0005	0,0004	0,0005	0,0006
H <sub>2</sub> S	0	0,0003	0,0006	0,0007	0,0006	0,0006	0,0007	0,0005
Cyanide	0	0	0	0	0	0	0	0
Free Residual Chlorine	0,005	0	0	0	0	0	0	0
Chloride	0,005	0,0002	0,0002	0,0003	0,0002	0,0001	0,0002	0,0002
Phenols	0	0	0	0	0	0	0	0
Nitrites	0,005	0,005	0,0046	0,0039	0,0035	0,003	0,0047	0,004
Nitrates	15,0	5,0	5,0	6,5	6,0	5,9	6,2	4,9
Carbonates	400	250	250	295	250	270	285	267
Sulphates	0,05	0,008	0,007	0,005	0,006	0,004	0,006	0,004
Phosphates	0,05	0,006	0,004	0,008	0,006	0,003	0,008	0,006
Lead	0,05	0	0	0	0	0	0	0
Zink	5,0	0	0	0	0	0	0	0
Mercury	0,001	0	0	0	0	0	0	0
Cadmium	-	0	0	0	0	0	0	0
Arsenic	-	0	0	0	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0	0	0	0
O <sub>2</sub> – Oxygen	> 5,0	12,0	11,0	13,0	13,0	12,0	11,0	12,0
CO <sub>2</sub> – Carbon dioxide	< 5,0	2,7	2,8	2,88	2,2	2,1	2,3	2,2
Electrical conductivuty	500	400	320	325	380	330	350	340
Purity	pure	pure	pure	pure	pure	pure	pure	pure
Ca – Calcium	200	189	195	200	190	195	194	200
Mg – Magnesium	850	135	125	130	136	138	150	168
Fe – Iron	0,3	0,001	0,003	0,002	0,003	0,002	0,001	0,001
Pesticides	0	0	0	0	0	0	0	0

Table 3. Chemical analysis of water on the 19<sup>th</sup> of May (ppm)

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the allowed limit or slightly above it. Oxygen was fairly present and  $CO_2$  was not so present to cause problems in fish. There wasn't contamination with heavy metals and pesticides. According to the results, water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

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Microorganisms	M-1	М-2	M-3	M-4	M-5	М-6	M-7
Salmonella	0	0	0	0	0	0	0
Shigella	0	0	0	0	0	0	0
Coliform bacteria of fecal origin	0	0	0	1	1	2	0
Escherichia coli	0	0	0	0	0	0	0
Staphiloccoci of fecal origin	0	0	0	0	0	0	0
Strept. Pyogenes	0	0	0	0	0	0	0
Proteus	0	0	0	0	0	0	0
Pseudomonas aeruginosa	0	0	0	0	0	0	0
Intestinal protozoa	0	0	0	0	2	0	0
Intestinal helminths	0	0	0	0	0	0	0
Vibrioni	0	0	0	0	0	0	0
Algae that cause a change of smell	3	4	3	2	2	0	0
Aerobic mesophilic bacteria	654	590	490	630	880	920	510
Total coliform bacteria	0	0	0	2	4	3	0
Sulfite-reducing bacteria	0	0	0	0	0	0	0
Enterobacteriaceae	0	0	0	0	0	0	0

Table 4. Microbiological analysis of water on the 19<sup>th</sup> of May

According to the microbiological analysis, water was suitable for fish farming. There was some contamination with coliform bacteria, but it is probably from the food used for feeding the fish, which indicates that there was no danger for presence of coliform bacteria. The water was clear, which is a good basis for fish production. Bacteriologically, the water fits to the Regulation (Official Gazette 33/1987). The trout pools require frequent cleaning according to the results shown from the microbiological analysis.

Constituents of the water	MPQ	М-1	М-2	M-3	M-4	М-5	М-6	М-7
Organic Substances, %		0,14	0,16	0,16	0,20	0,14	0,19	0,12
pH	6,5 - 9	6,9	6,9	6,9	6,9	6,9	6,9	6,9
NH <sub>4</sub>	0,1	0,0003	0,0005	0,0006	0,0007	0,0006	0,0007	0,0006
H <sub>2</sub> S	0	0,0003	0,0005	0,0006	0,0007	0,0005	0,0007	0,0008
Cyanide	0	0	0	0	0	0	0	0
Free Residual Chlorine	0,005	0	0	0	0	0	0	0
Chloride	0,005	0,0005	0,0007	0,0008	0,0007	0,0008	0,0007	0,0007
Phenols	0	0	0	0	0	0	0	0
Nitrites	0,005	0,004	0,004	0,0045	0,003	0,0043	0,0037	0,005
Nitrates	15,0	6,0	7,0	6,9	7,3	7,4	8,0	7,6
Carbonates	400	370	375	376	380	385	372	381
Sulphates	0,05	0,008	0,008	0,009	0,007	0,006	0,008	0,008
Phosphates	0,05	0,006	0,008	0,009	0,008	0,006	0,008	0,008
Lead	0,05	0	0	0	0	0	0	0
Zink	5,0	0	0	0	0	0	0	0
Mercury	0,001	0	0	0	0	0	0	0
Cadmium	-	0	0	0	0	0	0	0
Arsenic	-	0	0	0	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0	0	0	0
O <sub>2</sub> – Oxygen	> 5,0	14,0	8,0	13,0	9,0	13,0	13,0	18,0
CO <sub>2</sub> – Carbon dioxide	< 5,0	3,0	4,9	3,5	5,0	3,5	4,8	3,9
Electrical conductivuty	500	400	380	375	385	388	392	345
Purity	pure	pure	pure	pure	pure	pure	pure	pure
Ca – Calcium	200	200	200	189	190	199	185	190
Mg – Magnesium	850	140	150	135	153	138	141	144
Fe – Iron	0,3	0,006	0,007	0,009	0,007	0,007	0,008	0,008
Pesticides	0	0	0	0	0	0	0	0

Table 5. Chemical analysis of water on the 15<sup>th</sup> of Oktober (ppm)

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the allowed limits. There wasn't contamination with heavy metals and pesticides. According to the results, the water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

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Microorganisms	M-1	M-2	M-3	M-4	M-5	М-6	M-7
Salmonella	0	0	0	0	0	0	0
Shigella	0	0	0	0	0	0	0
Coliform bacteria of fecal origin	0	1	0	3	0	4	2
Escherichia coli	0	3	0	2	0	0	0
Staphiloccoci of fecal origin	0	0	0	0	0	0	0
Strept. Pyogenes	0	0	0	0	0	0	0
Proteus	0	0	0	0	0	0	0
Pseudomonas aeruginosa	0	0	0	0	0	0	0
Intestinal protozoa	0	0	0	0	2	0	0
Intestinal helminths	0	0	0	0	0	0	0
Vibrioni	0	0	0	0	0	0	0
Algae that cause a change of smell	0	0	3	5	6	6	8
Aerobic mesophilic bacteria	480	329	535	650	606	800	970
Total coliform bacteria	0	1	0	3	0	4	5
Sulfite-reducing bacteria	0	0	0	0	0	0	0
Enterobacteriaceae	0	0	0	0	0	0	0

Table 6. Microbiological analysis of water on the 15<sup>th</sup> of Oktober

According to the microbiological analysis, water was suitable for fish farming There was some contamination with coliformic bacteria, but it is probably from the packaging and the protein part of the food used for feeding the fish, which indicates that there was no danger for presence of these bacteria when it comes to this number - a few. The water was clear, which is a good basis for fish production. Bacteriologically, the water fits to the Regulation (Official Gazette 33/1987). According to the shown results of microbiological analysis, the trout pools require frequent cleaning because coliform bacteria from type Escherichia coli were found in the digestive tract.

Constituents of the water	MPQ	M-1	M-2	M-3	М-5	М-6	М-7
Organic Substances, %		0,11	0,14	0,16	0,18	0,13	9,5
pH	6,5 - 9	6,9	6,9	6,9	6,9	6,9	6,9
NH <sub>4</sub>	0,1	0,0004	0,0006	0,0008	0,0009	0,0009	0,0007
$H_2S$	0,001	0,0002	0,0005	0,0006	0,0006	0,0008	0,0007
Cyanide	0	0	0	0	0	0	0
Free Residual Chlorine	0,005	0	0	0	0	0	0
Chloride	0,005	0,0002	0,0004	0,0003	0,0005	0,0005	0,0004
Phenols	0	0	0	0	0	0	0
Nitrites	0,005	0,005	0,004	0,0047	0,004	0,0035	0,003
Nitrates	15,0	6,0	6,5	7,0	7,8	8,0	8,0
Carbonates	400	300	300	330	310	320	315
Sulphates	0,05	0,003	0,003	0,003	0,003	0,003	0,004
Phosphates	0,05	0,004	0,006	0,005	0,005	0,006	0,007
Lead	0,05	0	0	0	0	0	0
Zink	5,0	0	0	0	0	0	0
Mercury	0,001	0	0	0	0	0	0
Cadmium	-	0	0	0	0	0	0
Arsenic	-	0	0	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0	0	0
O <sub>2</sub> – Oxygen	> 5,0	10,0	8,0	11,0	7,0	10,0	8,0
$CO_2$ – Carbon dioxide	< 5,0	4,0	3,5	3,0	3,5	4,0	4,5
Electrical conductivuty	500	370	390	385	375	360	300
Purity	pure	pure	pure	pure	pure	pure	pure
Ca – Calcium	200	200	195	200	185	180	190
Mg – Magnesium	850	185	170	165	170	160	175
Fe – Iron	0,3	0,0035	0,0039	0,0030	0,0032	0,0035	0,0034
Pesticides	0	0	0	0	0	0	0

Table 7. Chemical analysis of water on the 24<sup>th</sup> of December (ppm)

According to the analysis, the water wasn't contaminated with harmful substances. All parameters were in the allowed limits. Water was pure in all tested samples, pH value was 6.9. There wasn't contamination with heavy metals and pesticides. According to the results, the water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

### Microorganisms M-1М-2 M-3M-5 М-6 M-7Salmonella Shigella Coliform bacteria of fecal origin Escherichia coli Staphiloccoci of fecal origin Strept. Pyogenes Proteus Pseudomonas aeruginosa Intestinal protozoa Intestinal helminths Vibrioni Algae that cause a change of smell Aerobic mesophilic bacteria Total coliform bacteria Sulfite-reducing bacteria Enterobacteriaceae

### SECTION 10. AQUACULTURE AND FISHERIES

Table 8. Microbiological analysis of water on the 24<sup>th</sup> of December

According to the analysis, the water was suitable for fish farming. From this situation the conclusion arises that pollution comes from the fish feed or packaging which keeps food for fish. Therefore, if it is necessary, daily cleaning of the pools should be done. There is no danger for presence of these bacteria, when it comes to so little number of harmful bacteria. The water was clear, which is a good basis for fish production. Bacteriologically, the water fits to the Regulation (Official Gazette 33/1987).

### Conclusions

Based on the research results of the chemical and microbiological safety of the water from the pond "Modric", it may be concluded:

- 1. The water quality in the pond during the research period, according to their physicalchemical and microbiological characteristics fits to the Regulation for sanitary safety of drinking water.
- 2. Water is good for growing trout.
- 3. It is recommended frequently to clean the fish ponds, preferably twice a week, because healthy fish for the market are grown in clean water.

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# THE INFLUENCE OF SIZE AND LIVING TEMPERATURES ON THE MUSCLE PROTEIN PROFILE OF KOI CARP (*CYPRINUS CARPIO HAEMATOPTERUS*)

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### Abstract

The goal of this paper is to show the eventual variations in the muscle protein gene expression in Koi carp (*Cyprinus carpio haematopterus*) caused by the fish size and living temperature differences. SDS-PAGE methodology was used as standard procedure, where samples from 24 different fish were analyzed, separated into two groups, large and small fish, raised in three different temperatures (5,  $25 \times 30^{\circ}$ C). The results showed concentration differences in proteins located at the same region in the gels, present in some of the large fish raised at temperatures of  $5^{\circ}$ C and  $25^{\circ}$ C, which points to the possible influence of the fish size and living temperatures on the muscle protein profile of the Koi carp. These types of variations were not recognized in the remaining analyzed samples.

Key words: muscle proteins, Koi carp, size, temperature.

### Introduction

One of the oldest fish farmed by man is the common carp, and the intensive breeding and mixing with other species of carp over the years, has led to the creation of a new species of carp known as *Koi carp*. The fish muscle proteins can be divided into 3 groups: *myofibrillar proteins* - soluble in concentrated salt solutions (actin, myosin, actomyosin, tropomyosin etc.); *sarcoplasmic proteins* - soluble in water or soluble in diluted salt solutions (myogen, globulin, myoglobulin etc.); and *binding tissue proteins*- proteins insoluble in water or salt solutions (collagen). (Min D., McCormick R.J, 2009). Electrophoresis is a commonly used method in many scientific fields for separation of charged molecules (proteins and nucleic acids). The main factors that determine the migration rate of the molecules are the medium used for separation, the total charge of the molecules, the power of the electrical field as well as size and conformation of the molecules (Gersten D.M. 1996). Protein separation is commonly done using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). This kind of electrophoresis is named as denaturizing because of the fact that Sodium dodecyl sulfate

(SDS) is an anionic detergent that is applied to the protein samples in order to denature (linearize) proteins, to impart a negative charge on them, and to achieve only separation only by length of their polypeptide chains. The gels are formed as acrylamide monomer polymerizes into long chains that are linked together by bis molecules (N,N'-methylenebisacrylamide). To initiate the start of the polymerization ammonium persulfate (APS) and N,N,N',N'- tetramethylethylenediamine (TEMED) are added to form the free radicals. TEMED accelerates the decomposition of persulfate molecules into sulfate free radicals and these in turn, initiate the polymerization. (Laas, T. 1989).

### Material and methods

For this study 24 samples were prepared from 24 different fish (*Cyprinus carpio haematopterus*) separated into two groups by size – small and large, raised on three different temperatures of 5, 25 and  $30^{\circ}$ C. The samples were prepared by homogenizing 50 mg fish muscle in 8M urea, 2M thiourea, 10% SDS, 2mM DTT reducing buffer. After this, the homogenized mixture was centrifuged at 10,000 x g, for 10 minutes and the supernatant containing the whole muscle proteins was used for analysis. (Wick M. 2010). In the table 1 are presented recipes for the preparation of the reagents for SDS-PAGE.

4x Lower Tris	4x Lower Tris pH 8.8		4x Upper TRIS pH 6.8			ouffer(10X)	
Tris BASE	18.17 g	Tris BASE	6.06 g	Tris BASE		60 g	
10% SDS	4.0 mL	10% SDS	4.0 mL	G	lycine	288 g	
q.s. with H <sub>2</sub> O to	100 mL	q.s. with H <sub>2</sub> O to	100 mL	SDS	(add last)	20 g	
pH to 8.8 before ad	lding SDS	pH to 6.8 before a	q.s. w	ith H <sub>2</sub> O to	2.0 L		
Reducing buffe	r (2X)	Coo	ant Blue R-250 gel stain				
10% SDS	3.0 mL	Coomassie B	rilliant Blue	R-250		400 mg	
Upper Tris(4x)	1.25 mL	М	lethanol			400 Ml	
b-mercaptoetanol	500µ L	Glacia	l acetic acid			400mL	
Glycerol	1.0 mL		q.s. wit	h dH20	D to 1L		
Bromphenol Blue	Pinch	Destain			Protogel		
q.s. with H2O to	10.0 mL	10% acetic acid				0.8% bis	

Table 1. SDS-PAGE reagents

The gel used for electrophoresis was divided into an upper stacking gel with lower concentration and lower resolving gel with smaller pores. The stacking gel has a role to deposit the proteins at the top of the resolving gel as a narrow band. In the electrophoretic set two gels were casted at the same time. The preparation of two resolving gels is achieved using reagents listed in Table 2.

Table 2. SDS-PAGE Gel Formulae for resolving gel	l
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Reagents	Volume		
dH <sub>2</sub> O	5.6 mL		
4X Lower TRIS	3.4 mL		
Protogel	4.6 mL		
TEMED	20µL		
10% APS	80µL		

Preparation of 3% stacking gel was done using 1.6 mL dH<sub>2</sub>O, 0.62 mL 4X Upper TRIS, 0.3 mL protogel, 10  $\mu$ L TEMED, and 30  $\mu$ L APS. The electrophoresis was performed under voltage of 100 V for 80-100 min. The analysis of the gels was done by Phoretix software package.

### **Results and discussion**

Analysis of the fish white muscle proteins can be done separately on sarcoplasmatic and myofibrilar or on total muscle proteins. In this study we were analyzing the composition of total fish white muscle proteins. On Figure 1 a noticeable higher concentration difference of some protein is visible only in sample 8. Also, the software analysis results show that the largest number of proteins are identified in sample number 6 (23 proteins), and the lowest in sample 1 (16 proteins).

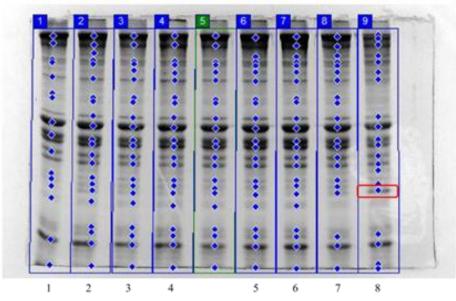


Figure 1.10% SDS-PAGE analysis: samples 1, 2, 3 and 4 were prepared from small fish raised at5<sup>o</sup>C; samples 5, 6, 7 and 8 were prepared from large fish raised at 5<sup>o</sup>C

On Figure 2 samples 13, 14 and 16 also have higher concentrations of a protein located in the same region as the one in sample 8 from Figure 1. All the samples that have a higher protein concentration belong in the large fish group. The largest number of proteins detected by the software is in samples 14 and 16 (16 proteins) and the lowest number in sample 9 probably due to the so called smiley effect (pattern).

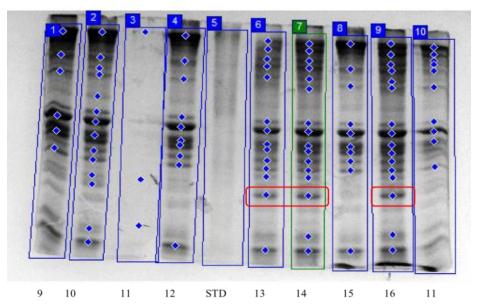


Figure 2.10% SDS-PAGE analysis: samples 9, 10, 11 and 12 were prepared from small fish raised at 25<sup>o</sup>C; samples 13, 14, 15 and 16 were prepared from large fish raised at 25<sup>o</sup>C

On Figure 3 the protein profiles look identical in terms of concentration, and the software analysis show that samples 18, 19, 20, 21, 22, 23 and 24 have 21 proteins, sample 17 has 20 proteins and samples 22 has 19 proteins detected.

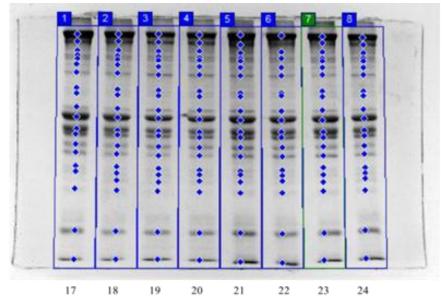


Figure 3.10% SDS-PAGE analysis: samples 17, 18, 19 and 20 were prepared from small fish raised at 25<sup>o</sup>C; samples 21, 22, 23 and 24 were prepared from large fish raised at 25<sup>o</sup>C

# Conclusions

Based on the results from the analysis, several conclusions can be drawn:

- There are visible differences of higher protein concentration present in the same region in some of the samples from the large fish group raised at temperatures of 5 and 25<sup>o</sup>C.
- The protein profiles of the fish raised at temperature of 30°C are almost identical (they have no significant visual difference).
- For a more detailed analysis the myofibrilar and sarcoplasmic proteins can be run separately, and if some specific protein fractions are identified that are present only in some group of fish they can be analyzed using a mass spectrometer.
- If this study continues, the determination of quantitative differences of the protein profiles should be approached by analysis of the gene expression using the mRNA isolated from the muscle tissue of the fish, that will be transcribed into complementary DNA and analyzed on Real Time PCR on which the differences in the expression of the genes active in the fish growth can be followed.

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# CONVENIENCE OF WATER FROM POND "CRESOVO" FOR FISH FARMING

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### Abstract

Quality of water that provides the pond is of especially importance for fish farming i.e. breeding of fish in artificial ponds. Fish health, productivity and profitability of the pond are directly dependent on the water. For this purpose, physico-chemical and microbial characteristics of water in pond "Cresovo" were examined. The pond is situated in Cresovo village, region of Skopje. It has an area of 600 m2. The pond is hotwater and pond carp (*Cyprinus carpio*, Linnaeus) is grown in it. During the months: April, May, August and October, physical-chemical and microbiological testing of water were carried out at the entrance, in the middle and exit in pools where the fish are raised, as well as in the hatchery. It was found that the quality of water, according to the physico-chemical and microbial characteristics, is good for growing carp.

Key words: pond water, physico-chemical and microbial characteristics.

### Introduction

The production of fish in the fishing farms in the Republic of Macedonia is established and developed over the last four decades. The condition of the fish production in the country from day to day changes in favor of farming fish.

Water's quality, which is used for fish farming in the pond, has particular importance for production of fish or breeding of fish in artificial ponds. More authors performed research on the quality of water in which fish are raised (Buras et al., 1985, 1987; Buras, 1990; Fattal et al., 1993; Hejkal et al., 1983). Water is a basic and unique environment that allows normal fish growth. It is an environment in which the fish are fully adapted for life. Water affects the fish and the progress of their normal life cycle with many factors, which means that the conditions that water quality provides can positively or negatively influence the fish. Therefore, studies of the physical-chemical and microbiological characteristics of water are essential for determining water quality and directly utilization towards fish farming.

### Material and methods

Pond "Cresovo" is built in the village of Cresovo, in the region of Kuklevica, Skopje, and began operations in 1998. It has an area of  $600 \text{ m}^2$  and the material which the farm is made from is land, using a nylon to cover the bottom. The farm consists of 1 large and 2 smaller pools, with individual area of 300, 200 or 100  $m^2$  and a total area of 600  $m^2$ . It is secured by a road, electricity and water supply. The source of water for breeding fish is a well deep 11,5 m, with a water column of 5,5 m, which means that the pools are filled with underground well water. Cresovo pond is a hotwater pond and provides appropriate conditions for growing of carp (Cyprinus carpio, Linnaeus). The capacity of the farm, according to the project, amounts to 10 tons of fish. The current offspring is passed from R. Bulgaria, from the farm "Krdzhali". Nutrition in the pond is done with grains (wheat), pelleted and extruded feed. The conditions of storing the food in the warehouse is at a temperature of 18°C. The food is complemented by a new one once a month. Tests were performed on samples of water taken during the months: April, May, August and October. Physical-chemical and microbiological testing of water were carried out at the entrance to the swimming pools, in the middle, at the exit in pools, where fish were raised, and in the hatchery. Common methods were used to determine the chemical composition of the water, and for the microbiological analysis standard methods for examination of various microorganisms, especially specific harmful bacteria were applied. Microbiological methods used for determination of certain groups of microorganisms are following:

- 13. For total bacteria count MPA method is used.
- 14. Brilliant green agar is used for Salmonella detection.
- 15. Chapman substrate for detection of Staphilococcus pyogenes.
- 16. SS substrate for detection of Proteus and Streptococcus.
- 17. Sulphite reducing bacteria are detected by sulphite agar.
- 18. Clostridia are detected by sulphite agar.
- 19. MPA agar for detecting of mesophilic bacteria.
- 20. Sabourald-agar for determination of yiests.
- 21. Czapek agar for detecting of moulds.
- 22. Suitable substrate is used for detecting of Pseudomonas aeruginosa.
- 23. Endo-agar for Escherichia coli.
- 24. Starch-ammonia agar and Andrade indicator for detecting the contaminants.

Tests were performed at the Faculty of Agricultural Sciences and Food in Skopje.

### **Results and discussion**

The results of chemical and microbiological analysis of the water taken by the pools of the pond "Cresovo" are shown in Tables 1–8.

Constituents of the water	MPQ	Entry	Middle	Exit	Hatchery
Organic substances, %		0,2	0,35	0,4	0,2
pH	6,5 - 9	7,2	7,2	7,3	7,2
NH <sub>4</sub>	0,1	0	0,0001	0,0005	0,0001
$H_2S$	0	0	0,0001	0,0021	0
Cyanide	0	0	0	0	0
Free Residual Chlorine	0,005	0	0	0,0001	0
Chloride	0,005	0,0001	0,0020	0,0030	0,0001
Phenols	0	0	0	0	0
Nitrites	0,005	0,004	0,0045	0,005	0,004
Nitrates	15,0	9,0	10,0	12,5	9,0
Carbonates	400	260	260	265	250
Sulfates	0,05	0,0020	0,0020	0,0025	0,0020
Phosphates	0,05	0,0030	0,0030	0,0032	0,0030
Lead	0,05	0,0001	0,0001	0,0001	0,0001
Zink	5,0	0,0003	0,0004	0,0005	0,0003
Mercury	0,001	0	0	0	0
Cadmium	-	0	0	0	0
Arsenic	-	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0
$O_2 - Oxygen$	> 4,0	11,0	10,0	9,0	12,5
$CO_2$ – Carbon dioxide	< 5,0	3,0	4,5	5,2	3,1
Electrical conductivity	500	450	450	420	410
Color	White	White	White	White	White
Ca – Calcium	200	190	200	195	180
Mg – Magnesium	850	140	130	140	140
Fe – Iron	0,3	0,003	0,003	0,003	0,003
Pesticides	0	0	0	0	0

Table 1. Chemical analysis of water on the 15<sup>th</sup> of April (ppm)

MPQ – maximum permissible quantity

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the limits of allowed and even oxygen was quite present, and  $CO_2$  was not so amassed to cause a problem in fish. It is important that there were no contamination with heavy metals and pesticides. According to the results, water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

Microorganisms	Entry	Middle	Exit	Hatchery
Salmonella	0	0	0	0
Shigella	0	0	0	0
Coliform bacteria of fecal origin	0	0	0	0
Escherichia coli	0	0	0	0
Staphiloccoci of fecal origin	0	0	0	0
Strept. Pyogenes	0	0	0	0
Proteus	0	0	0	0
Pseudomonas aeruginosa	0	0	0	0
Intestinal protozoa	0	0	3	0
Intestinal helminths	0	0	0	0
Vibrioni	0	0	0	0
Algae that cause a change of smell	2	7	10	1
Aerobic mesophilic bacteria	400	420	520	220
Total coliform bacteria	0	0	4	0
Sulfite-reducing bacteria	0	0	2	0
Enterobacteriaceae	0	0	0	0

Table 2. Microbial analysis of water on the 15<sup>th</sup> of April

According to the microbiological analysis, water was suitable for fish farming. The water was quite clear, which is a good basis for fish production. Bacteriologically, the water fits to the Regulation (Official Gazette 33/1987).

Constituents of the water	MPQ	Entry	Middle	Exit	Hatchery
Organic substances, %		0,25	0,27	0,22	0,20
pH	6,5 - 9	6,9	6,9	6,9	6,9
NH <sub>4</sub>	0,1	0,0008	0,0009	0,0009	0,0009
$H_2S$	0	0,0006	0,00059	0,0008	0,0005
Cyanide	0	0	0	0	0
Free Residual Chlorine	0,005	0,0003	0,0005	0,0004	0,0005
Chloride	0,005	0,0004	0,0007	0,0006	0,0002
Phenols	0	0	0	0	0
Nitrites	0,005	0,0045	0,0009	0,0042	0,0039
Nitrates	15,0	6,0	7,0	7,5	6,0
Carbonates	400	350	355	350	350
Sulfates	0,05	0,007	0,008	0,008	0,006
Phosphates	0,05	0,010	0,035	0,038	0,010
Lead	0,05	0	0	0	0
Zink	5,0	0,006	0,0067	0,0069	0,006
Mercury	0,001	0	0	0	0
Cadmium	-	0	0	0	0
Арсен – Arsenic	-	0	0	0	0
Hexavalent Chromium	0,05	0	0	0	0
Trivalent Chromium	0,1	0	0	0	0
O <sub>2</sub> – Oxygen	> 4,0	16,0	14,0	11,0	17,0
$CO_2$ – Carbon dioxide	< 5,0	2,9	2,9	3,0	2,3
Electrical conductivity	500	400	400	400	415
Color	White	White	White	White	White
Ca – Calcium	200	195	185	190	200
Mg – Magnesium	850	140	146	150	145
Fe – Iron	0,3	0,001	0,001	0,002	0,001
Pesticides	0	0	0	0	0

Table 3. Chemical analysis of water on the 19<sup>th</sup> of May (ppm)

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the allowed limit. Oxygen was quite present and  $CO_2$  was not so present to cause problems in fish. In all tested samples water was clean. There wasn't contamination with heavy metals and pesticides. According to the results, water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

Microorganisms	Entry	Middle	Exit	Hatchery
Salmonella	0	0	0	0
Shigella	0	0	0	0
Coliform bacteria of fecal origin	0	0	2	1
Escherichia coli	0	0	0	0
Staphiloccoci of fecal origin	0	0	0	0
Strept. Pyogenes	0	0	0	0
Proteus	0	0	0	0
Pseudomonas aeruginosa	0	0	0	0
Intestinal protozoa	0	0	3	0
Intestinal helminths	0	0	0	0
Vibrioni	0	0	0	0
Algae that cause a change of smell	4	5	4	0
Aerobic mesophilic bacteria	490	920	980	340
Total coliform bacteria	0	8	10	1
Sulfite-reducing bacteria	0	0	0	0
Enterobacteriaceae	0	0	0	0

Table 4. Microbial analysis of water on the 19<sup>th</sup> of May

According to the microbiological analysis, water was suitable for fish farming. There was some contamination with coliform bacteria, but it is probably from the food used for feeding the fish. The water was clear, which is a good basis for fish production. Bacteriologically, the water fits to the Regulation (Official Gazette 33/1987). More frequent cleaning the pools from sludge is required, according to the results shown from the microbiological analysis.

Constituents of the water	Entry	Middle	Exit	Hatchery
Organic substances, %	0,03	0,06	0,04	0,02
pH	6,9	6,9	6,9	6,9
NH <sub>4</sub>	0	0,0135	0,0111	0,0111
H <sub>2</sub> S	0,0001	0,0145	0,0112	0,0113
Cyanide	0	0	0	0
Phenols	0	0	0	0
Free Residual Chlorine	0	0	0	0
Chloride	0	0,0002	0,001	0
Nitrates	2,24	4,38	2,22	1,56
Nitrites	0,0002	0,0004	0,0003	0,0002
Carbonates	173	180	200	234
Sulfates	0,0002	0,0003	0,00048	0,00052
Phosphates	0,0002	0,0006	0,0004	0,0003
O <sub>2</sub> – Oxygen	12,0	6,0	9,0	13,0
CO <sub>2</sub> – Carbon dioxide	3,0	5,0	4,0	3,0
GCO – 5*	12	10	42	53
NCO in KMnO <sub>4</sub> **	8	14	32	53
Color	White-green	White-green	Green	White-green
Hardness	1,1646	1,1522	1,4342	1,3478
Electrical conductivity	525	480	490	500
Bicarbonates	144	146	152	163
Ca – Calcium	152	149	163	166
Mg – Magnesium	1200	114	122	134
Fe – Iron	0,0002	0,0005	0,0003	0,0002
Lead	0	0	0	0
Zink	0	0	0	0
Mercury	0	0	0	0
Cadmium	0	0	0	0
Arsenic	0	0	0	0
Hexavalent Chromium	0	0	0	0
Trivalent Chromium	0	0	0	0
	0	-		

Table 5. Chemical analysis of water on the 12<sup>th</sup> of August (ppm)

GCO - 5 = Gross consumption of oxygen

\*\*NCO in KMnO<sub>4</sub> = Net consumption of oxygen

According to the chemical analysis, the water wasn't contaminated with harmful compounds. All parameters were in the allowed limit. The pH value was 6,9. Oxygen was quite present and  $CO_2$  was not so present to cause problems in fish. It is important that there wasn't contamination with heavy metals and pesticides. According to the results, water fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

Microorganisms	Entry	Middle	Exit	Hatchery
Salmonella	0	0	0	0
Shigella	0	0	0	0
Coliform bacteria of fecal origin	0	2	1	0
Escherichia coli	0	0	0	2
Staphiloccoci of fecal origin	0	0	0	0
Proteus	0	0	0	0
Pseudomonas aeruginosa	0	0	0	0
Intestinal protozoa	0	0	0	0
Intestinal helminths	0	0	0	0
Vibrioni	0	0	0	0
Algae that cause a change of smell	4	12	6	2
Aerobic mesophilic bacteria	226	2590	1240	1100
Sulfite-reducing bacteria	0	8	412	0
Enterobacteriaceae	0	0	0	0

Table 6. Microbial analysis of water on the 12<sup>th</sup> of August

There wasn't microbiological contamination with presence of harmful bacteria in the water from entering the pond (from entrance) and in the hatchery. There were small amounts of coliform bacteria at the exit of the pools and in the middle, but certainly they are part of the protein or food packaging. However, algae are found quite especially at the exit of the pools, even in the hatchery, which is a sign of some increased oxygen consumption. However, the water from these samples was relatively clean, especially when it is used for fish farming, insofar the chemical analyzes show good results.

Constituents of the water	Entry	Middle	Exit	Hatchery
Organic substances, %	0,3	0,6	0,9	0,3
pH	6,9	6,9	6,9	6,9
NH <sub>4</sub>	0,0005	0,0009	0,0012	0,0004
H <sub>2</sub> S	0,0001	0,0005	0,0007	0,0001
Cyanide	0	0	0	0
Free Residual Chlorine	0	0	0	0
Chloride	0,0002	0,0005	0,0005	0,0002
Nitrites	0,0005	0,0008	0,0009	0,0004
Nitrates	6,0	6,0	6,0	5,0
Carbonates	180	180	180	180
Sulfates	0,0004	0,0005	0,0012	0,0004
Phosphates	0,0003	0,0004	0,0007	0,0003
Lead	0	0	0	0
Zink	0	0	0	0
Mercury	0	0	0	0
Cadmium	0	0	0	0
Arsenic	0	0	0	0
Hexavalent Chromium	0	0	0	0
Trivalent Chromium	0	0	0	0
O <sub>2</sub> – Oxygen	11,0	8,0	6,0	13,0
$CO_2$ – Carbon dioxide	2,0	3,0	4,0	2,0
GCO – 5*	10	10	10	9
NCO in KMnO <sub>4</sub> **	17	18	19	17
Hardness	1,1237	1,1234	1,1235	1,1234
Electrical conductivity	389	379	366	400
Color	Bright	White	Green	White
Ca – Calcium	144	143	144	142
Mg – Magnesium	120	122	122	122
Fe – Iron	0,0001	0,0001	0,0004	0,0001
Pesticides	0	0	0	0

Table 7. Chemical analysis of water on the 15<sup>th</sup> of Octomber (ppm)

\*GCO – 5 = Gross consumption of oxygen

\*\*NCO in KMnO<sub>4</sub> = Net consumption of oxygen

The chemical analysis shows that there wasn't presence of harmful compounds and elements in the water from all four samples, so it fits to the Regulation (Official Gazette 33/1987) for using in fish farming.

#### Microorganisms Entrv Middle Exit Hatchery Salmonella Shigella Coliform bacteria of fecal origin Escherichia coli Staphiloccoci of fecal origin Strept. Pyogenes Proteus Pseudomonas aeruginosa Intestinal protozoa Intestinal helminths Vibrioni Algae that cause a change of smell Aerobic mesophilic bacteria Total coliform bacteria Sulfite-reducing bacteria Enterobacteriaceae

#### SECTION 10. AQUACULTURE AND FISHERIES

Table 8. Microbial analysis of water on the 15<sup>th</sup> of October

According to the microbiological analysis, water taken from the pond, corresponds to the Regulation (Official Gazette 33/1987) for use as water for fish farming. There was some contamination with coliform bacteria, but they were most likely the result of rarely cleaning the pond, possible contamination from the food or food which shelf life was over.

#### Conclusions

Based on the research results from the chemical and microbiological safety of the water from the pond "Cresovo", it may be concluded:

1. The water quality in the pond, during the research period, according to their physicalchemical and microbiological characteristics fits to the Regulation for sanitary safety for drinking water.

2. Water is good for growing pond carp.

3. It is recommended frequently to clean the fish ponds, preferably twice a week, because healthy fish for the market are grown in clean water.

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#### SOME DATA ABOUT THE TREATMENT OF GYRODACTYLUS SPP. IN RAINBOW TROUT (ONCHORHYNCHUS MYKISS) IN ALBANIA

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#### Abstract

The monogenean Gyrodactylus spp. is an important parasite of farmed fish in Europe because of the heavy damages and losses that induce. Gyrodactylys spp. is susceptible to a range of commonly used reagents. After the analysis of the level of infestation of rainbow trout in some regions in Albania and the analyses of the implementation of the sanitary hygienic measures for the limitation of this infestation we have tested various therapeutic baths to prove the efficacy against this helminthes. Bath treatments included high salinity water, formaldehyde and chlorine/iodine based compounds. The performed baths showed a reduction of parasites in trouts mainly the exposure to formaldehyde exposure has reduced them significantly. We have also checked for the effects of the treatment on fish after the baths. The information of this study is very important because the Gyrodactylus is a very difficult parasites to be removed after established in fish farms.

Key words: parasites, Gyrodactylus, treatment, baths, effect.

#### Introduction

The invasions caused by monogenean trematodes bring high economical losses (Fridman, S et.al. 2014; Levy, G et.al. 2015). In particular Gyrodactylus sp. parasites affect a high number of fresh waters fish. To control the spreading of the disease are applied different methods, chemical treatments (Jero, J et.al 1986; Richards, et.al. 1996; Schelkle, B, et.al. 2011) use of the pharmaceutical preparation in food (Tojo, J.L., M.T Santamarina 1998), the application of bath treatments with anthelmintic preparation (Schmahl, G., Taraschewski, H. 1987; Santamarina, M.T et.al. 1991; Olafsdottir S.H., K. Buchmann 2004; Truong, D.H and Kim, V.V. 2014), the use of plant extraction (Ramadan, R.A.M et.al 2012; Tu, X et.al. 2013; Reverter, et.al. 2015). Formalin and green malachite used until now for the control of gyrodactylosis and other ectoparasites have not given the expected results because of their toxicity and the low specificity (Tojo, J et.al 1992), and some pharmaceutical preparations applied as bath cause negative impacts in the water environment (Jones, O.A.H et.al 2003). The frequent use of chemicals has developed in some parasites the resistance forms for this treatment and has damaged the environment. For this reason the need for new alternative chemical treatments more effective and sustainable is very required in the last years (Truong, D.H., Kim, V, V. 2014). The present work has as the main purpose to prove the anthelmintic effect of some chemicals and plant extractions applied as water baths in the cases of infestation with Gyrodactylus sp.

#### Material and methods

One year rainbow trouts naturally infested with Gyrodactylus sp. were obtained from the farms in Pogradec and Librazhd district in 2015. The average weigh of the fish was Wg = $9.725 \pm 0.517$ ; Var % = 5, 32. The fish transportation was done in plastic containers with 100 L of water and supplied with liquid O2. The tests are performed in glass tanks: 2 tanks with 0.34m<sup>3</sup> volumes, 2 tanks with 0.1m<sup>3</sup> volumes and 1 with 0.030 m<sup>3</sup> volume and 2 fiberglass tanks 3 m<sup>3</sup>. The fish divided in two groups were feed with a commercial diet (Coppens International). The tests are performed in 2 groups: the therapeutic and the control group, the pectoral gills are controlled with stereomicroscope to define the number of dead parasites. Before the therapeutic baths is tested the toxicity for two preparations praziquantel and bithinol in tanks with 30L water inside, with 5 fish in each tank. The test is performed 2 times in two high concentrations for 1-6 hours.

Table 1. The scheme used for the testing of the toxicity effect of Praziquantel and Bitnioi.					
Solution used	Doses (mg/L)	Exposure time (hours)			
Praziquantel	10/20	1, 3, 6			
Bithionol	20/60	1, 2, 3			

Table 2. The scheme used for the antienminite effect of mentioned solutions.					
Solution used for water bath Experiment doses (mg/L)		Exposure time (min)			
Praziquantel	10	180			
Sodium chloride	25 000	5			
Bithionol	5 and 20	180			
Water/garlic extract solution	12,5 and 8,5	60			

Table 2 The scheme used for the anthelmintic effect of mentioned solutions

The optimal anthelmintic concentration is considered the concentration that provided the highest level of dead parasites, but with absence of any toxicity signs in fish. The concentration of anthelmintic solutions use in this paper that caused the death of less than 20% of parasites is considered not efficient. The mortality of parasites for every treatment is calculated with Wang G.X.,et.al, 2010 formula:

M(%) = [(B-T)/B]\*100

Where: M- Mortality of Gyrodactylus sp. parasites (%), B – Mean number of survived parasites in the control group in interval "t"; T - Mean number of survived parasites in the treated group in interval "t"; "t" - duration of the exposure of the fish in the water bath. The results of the study are analyzed using the statistic data tool SPSS 16. The variance analyze (ANOVA) is performed to prove the reduction of the main values of invasion after the treatments with anti Gyrodactylus sp.preparations. The mortality of parasites after every treatment is calculated with Wang G.X. et.al. 2010 formula.

#### **Results and discussion**

The tables 3 and 4 shows the results obtained from the toxicity test for: praziguantel and bithionol applied in trouts in two concentrations and in three exposure times.

	Doses and time of exposure (mg/L; hours)					
Indicator	10 mg/L; 1h	10 mg/l-3 h	10 mg/l-6h	20 mg/l-1 h	20 mg/l-3 h	20 mg/l-6 h
General behavior	-	-	-	+	++	++
Clumsy swimming	-	-	++	+	++	++
Swimming in bottom	-	-	-	-	-	++
Increase of respiration	-	-	-	-	+	++
Death	-	_	-	-	-	+

Table 3. Results of toxicity test for two concentrations of Praziquantel used in water baths.

Notes :(+) one individual ;(++) 2-3 individuals ;(+++) 4-5 individuals

The test proved that the use of praziquantel in 10 mg/L; 1-6 hours does not cause visible changes and loss of any individual. The situation was different with the application of doses 20 mg/L that resulted in changes of physiological status of the trouts.

	Doses and time of exposure (mg/L; hours)					
Indicator	20 mg/L; 1	20 mg/l-3	20 mg/l-6	20 mg/l-1	20 mg/1-3	20 mg/l-6
	h	h	h	h	h	h
General						
behavior	-	-	-	-	+	+
Clumsy						
swimming	-	-	+	+	+	++
Swimming						
in bottom	-	-	-	-	-	+
Increase						
of	-	-	-	-	-	-
respiration						
Death	-	-	-	-	-	-

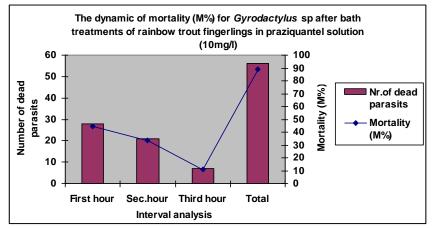
Notes :(+) one individual;(++) 2-3 individuals;(+++) 4-5 individuals

The toxicity test for bithionol is performed as with praziquantel in the concentrations 20mg/L and 60 mg/L in three intervals of exposure; 1, 2 and 3 hours. During the application of 20 mg/L concentrations of bithinol was observed the slowdown of the swimming. The tripling of the concentration was not accompanied with changes in swimming or behavior of trouts. During 3 hours exposure in 60 mg/L bithionol 50% of the trouts showed clumsy swimming and no changes in respiratory rates and no mortality.

#### The efficacy of praziquantel treatments (10 mg/L)

Since during the use of praziquantel solution in concentration 20 mg/L and exposure time 1-3 hours was not observed any death, there was not performed a anti parasitary test for this concentration. The increase of the respiratory rates may be a stress indicator as a result

of the extension exposure times 6 hours in 20 mg/L praziquantel. The anthelmintic efficacy test of praziquantel is performed applying 3 hours water baths in concentration 10 mg/L. In order to estimate the dynamics of the mortality rates (M %) 7 trouts are collected 1, 2 and 3 hours after the beginning of the test and is performed the microscopic control of the pectoral fins to count the dead parasites.

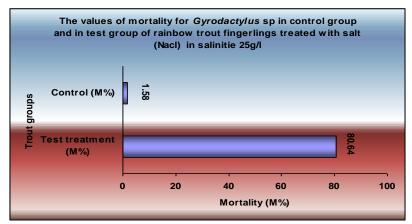


Graph.1 The dynamic of mortality values (M%) of Gyrodactylosis spp. after 10 % solution of Praziquantel bath.

Graph 1 show that the higher average value of mortality (M%=44.51±2.688) was obtained after the first hour (in average 4 dead parasites/ trout). The mortality rates of Gyrodactylus parasites after the second hour with 10% Praziquantel solution was reduced (average 3 dead parasites/trout; M%=33.32±1.941). The statistical analyses proved the difference between the mortality for the first and second hour of the treatment (t = 3:39; P <0:01). In the last hour of the test the mortality reduction was more pronounced. (M = 9.11 ± 0.861%). Taking into consideration the average value of invasion (9.75 ± 4418) and the average number of parasites that remain alive (1:08 ± 0:04) resulted that the efficiency of the praziquantel solution applied in the concentration 10 mg/L , to interval exposure of three hours was 88.87%.

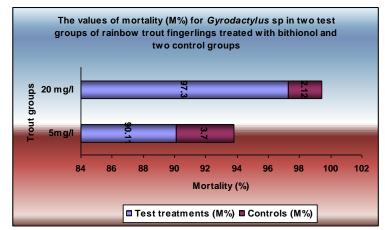
#### The efficacy of sodium chloride treatments (25 mg/L)

The anthelmintic efficiency of sodium chloride solution bath in concentration 25g/L for 5 minutes exposure interval was tested with 21 individuals of rainbow trout. Graph. 2 shows the change in the mortality values (M %) in the treated and in the control group, within 5 minutes. In the two groups we have calculated the mortality value of Gyrodactylus sp (M% = 80.64 for treated group and M% = 1:58 for the control group.



Graph.2 Mortality of Gyrodactylosis sp. after the tratment with 25 % solution of sodium chloride/5 min

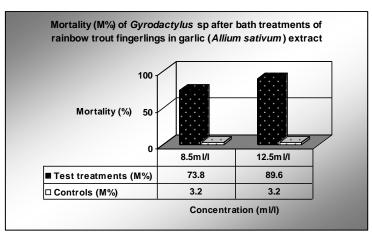
The efficacy of bithionol treatments (4 and 20 mg/L).



Graph 3. Mortality of Gyrodactylosis sp. after the treatment with bithinol solution 5 mg/L and 20 mg/L/180 min.

The comparing of the values in the Graph. No.3 prove that the increase by five times of the concentration of bithionol has improved with 1:08 times the efficiency against Gyrodactylus sp, in the 5mg/L bithionol solution for 180 min the mortality of the parasites resulted M% = 90.11, while in the concentration 20mg/L the helminthes mortality was M % = 97.3. In two control groups the mortality values resulted M% = 3.7 and 2.12. After the first hour of treatment with Bithionol the mortality value of Gyrodactylus sp was  $M\% = 33.52 \pm 1.747$  and in the end of the second hour was  $M\% = 44.24 \pm 2.352$ . The changes between these two values resulted significant (t = 3.66; P <00:01). During the third hour of the treatment the average mortality of the parasite was  $M\% = 1027 \pm 19:56$ . From the comparison of the values we have proved that the anthelmintic efficiency against the parasites was similar for the first two hours. The difference noted in the last hour was caused by the higher values of parasites mortality after the treatment with bithionol in concentration 20mg/L.

The efficacy of garlic extracts treatment (8.5 ml/L and 12.5 ml/L).



Graph.4 Results of garlic extract treatment against Gyrodactylus sp. (8.5 ml/L and 12.5 ml/L).

The garlic extract test was performed by applying two concentrations (8.5ml/L and 12.5ml/L) for 60 minutes. In the first concentration the mortality of the parasites resulted M % = 73.8. The use of water bath with garlic extract solution in 12.5 ml/L concentration for 60 minutes showed an increased mortality of the parasites (M = 89.6%). The increase by about 1.5 times of the concentration caused the improvement of effect by more than 1.2 times. Despite the positive effect the increase of garlic extract concentration was followed by some abnormal fish behavior. It is very important to give a particular importance to the European Union policy related to the reduction of the chemical substances in aquatic environments. The combination of the correct treatments with other health management strategy for the disease prevention, as vaccination, improvement of the water quality, alternative treatments can help to ensure successful development of this sector in the future (Athanassopoulou, F. et.al.2009). The toxicity test results proved that praziquantel 20mg/L and bithionol 60mg/L, for 3 hours, cause toxic effects to rainbow trout fingerlings and manifested by alterations in swimming behavior, frequent respiratory, tendency to swim at the end of aquariums and in one case the death of a individual. The bithionol showed higher activity compared with other chemicals involved in this study. For the praziquantel solution in 10mg/L the anti-Gyrodactylus sp. efficacy was about 88.9%, and for the garlic extract in 8, 5 mL/l the mortality rate resulted 73.8%, and the toxic effect in was found in the concentration 12.5 mL/l.

#### Conclusions

1. We have tested the anti parasite activity of praziquantel, bithionolit, sodium chloride and garlic extract, in rainbow trout (O.mykiss) infested with the monogenic parasites Gyrodactylus sp.

2. The highest value of helminthes mortality (M = 97.3%) was achieved during the use of bithionol solution in a concentration of 20mg/L for 3 hours. During the use of praziquantel in 10mg/l concentration for 3 hours the parasites mortality was M%=88.87%.

3. The use of garlic extract was implemented as an alternative solution to combat the Gyrodactylus sp. During the application of garlic solution in the concentration 8.5mL/l the helminthes mortality resulted in M% = 73.8. Keeping the fish for 60 minutes in garlic extract solution in concentration 12.5mL/l caused increased mortality values of the parasites (M = 89.6%) but was also accompanied by the occurrence of stress signs in trouts.

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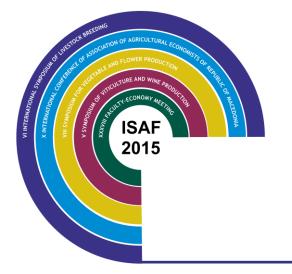
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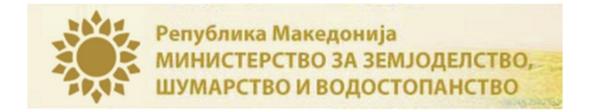
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