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PREFACE

The Proceedings contains 38 papers presented at XI International Symposium on Agricultural Sciences "AgroReS 2022" in Trebinje, Bosnia and Herzegovina, from 26 to 28 May, 2022. In the Proceedings are published only papers for which their authors choose that way of publishing

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Seed germination of *Calendula officinalis* L. under influence of different light conditions

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Abstract

The aim of this study was to examine germination rate and morphological characteristics of English marigold seedlings under influence of different light conditions. Seeds of *Calendula officinalis* L. were collected from the natural population of the Botanical garden of the University of Banja Luka. Experiment was set up in four replicates for each light treatment. Petri dishes with seeds were placed in growth chamber under artificial white (FLUO) and blue, red, and combination of blue/red (LED) light with 16h/8h photoperiod. Germination energy was tested after 7 days and germination of the seeds was tested after 14 days. Results showed significant difference in germination energy, germination rate, hypocotyl height, root length, and fresh weight. The highest average values of the germination energy and germination rate of the *Calendula officinalis* L. were recorded under red LED light (32%; 47%) while the lowest values were recorded under blue LED light (1%; 23%). The highest average values of hypocotyl height, root length and fresh weight were recorded also under red LED light (3,70 cm; 6,33 cm; 0,97 g) while the lowest values were recorded under combination of blue/red LED light (1,95 cm; 2,52 cm; 0,28 g). It can be concluded that the use of red LED light is recommended in the seed germination phase, not only for better germination but also for better morphological development.

Key words: germination, *Calendula officinalis* L., morphological parameters, LED light

Introduction

The key factors for plant growth and development are light and temperature. Plants have

photoreceptors that respond to the wavelength and light intensity (Zhang and Folta, 2012). The effect of light during plant growth and fertility process is undeniable. Among the various naturally occurring abiotic factors regulating plant development, light plays an important role in photosynthesis and photoperiodism, and only wavelengths ranging 400 to 700 nm can be used (Teklić, 2012).

Many of light sources which are used to enhance photosynthetic levels have very low energy use efficiency for growing plants. Solar light consists of electromagnetic radiation with wavelengths ranging 400 to 700 nm (violet, blue, green, yellow, orange and red). The changes in the light quality (wavelength) influences seed germination, seedling growth, photosynthesis, and flowering depending on the species and developmental stage (He et al., 2017).

In contrast to other light sources, lightemitting diode (LED) lighting system have various advantages, including the ability to set the desired spectral combination, permanence, specific wavelength, low heating and the electrical input (Lin et al., 2013). These advantages make them suitable light source for growing plants. The LEDs give wavelengths that can be matched with plant photoreceptors to provide optimal production and influence morphology and metabolism of plant (Morrow, 2008). Experimentations on impact of light variables on plant growth and development is attracting attention of plant scientists. Changes in different light attributes like intensity and duration, can influence all aspect of plant growth and physiology especially morphology and photosynthetic responses of the plants. Red and blue lights have great effects on plant growth because they contain the two main light spectra for photosynthetic CO₂ fixation in plants (Kasajima et al., 2008). It has been reported that red light is the most important light spectrum for growth and phytochrome responses in plants (Wang et al., 2016).

Red light is important for the development of the photosynthetic apparatus of plants and may increase starch accumulation in several plant species by inhibiting the translocation of photosynthates out of leaves (Paradiso et al., 2011). In contrast, blue light is important in the formation of chlorophyll, chloroplast development, stomatal opening, enzyme synthesis, activation of the photosynthesis, and photomorphogenesis (Kang et al. 2008; Demarsy and Frankhauser 2009).

Green light, in the process of seed germination of *Arabidopsis*, stimulates the early elongation of the stems, antagonizing the growth inhibition by light whereas the white and red light, in ferns can delay the chlorophyll loss due to senescence (Burescu et al., 2015). It has been reported that light is key factor regulating the seed germination in numerous plant

species. Jala (2011) found that seeds of *Nepenthes mirabilis* first germinated under white and red light, and the last germinated under green light, and the highest average speed of emergence was also recorded under red light.

Lal and Sachan (2017) investigated the effect of different colours of light (natural, red, blue, yellow, and green) on seed germination, hypocotyl growth, biomass production in *Vigna unguiculata* L. Walp., an important crops for grain and fodder purposes. Red light showed maximum % of germination (98%) at 84 hours while, green light showed almost no germination even after 96 hours. Blue light and yellow light caused significant reduction in % of germination to 71 and 56, respectively, at 84 hours. Root and shoot growth were highest in red light and the order of biomass production was red > yellow > natural > blue > green.

English marigold (*Calendula officinalis*) is an annual or biennial plant species that belongs to the Asteraceae family. It is native to North Africa and Southern Europe and represents as one of the most used ornamental plants in the gardens and green spaces. English marigold has different applications in horticulture industry: as a pot plant, garden plant, cut flower, food and medicinal plant. The aim of this study was to examine germination rate and morphological characteristics of English marigold seedlings under influence of different light conditions.

Material and Methods

Investigation was conducted in laboratory condition at the Faculty of Agriculture, University of Banja Luka. Seeds of *Calendula officinalis* L. were collected from the natural population of the Botanical garden of the University of Banja Luka. The experiment consisted of four light treatments: three different colors of light (blue, red, and combination of blue/red LEDs) as treatments, and white FLUO light, as control. The experimental design was completely randomized, with four replications of 25 seeds per experimental unit for germination test. Petri dishes were sterilized with 96% ethanol, and lined with moistened filter paper with 3 mL of purified water. One hundred surface sterilized seeds of *Calendula officinalis* L were placed in a series of five petri dishes for each colors of light.

These petri dishes were placed on shelves, exposed to different wavelength (white, blue, red, and combination of blue/red light) with 16h/8h photoperiod. Temperature during the research was constant (20±1°C). 2 ml of water per day was added to each petri dish. Seeds were kept under these conditions for 14 days. Germination energy was tested after 7 days and germination of the seeds was tested after 14 days. Both values are expressed as percentage (%). Also, after 14 days hypocotyl height, root length, and fresh weight were obtained. The

obtained data were statistically analysed (LSD, F-test, t-test) using standard computer programs and VV-Stat paket (Vukadinović, 2017). Means comparison were performed using low significant differences procedure (LSD), with a significance level of 5% ($P < 0.05$).

Results and Discussion

Statistically analyzed obtained results of germination energy, seed germination, number of cotyledons, hypocotyl height, root length, and fresh weight of *Calendula officinalis* L. under influence of different light conditions are presented in Table 1. and Table 2.

Treatment T₁-red LED light had the best result in all of three parameters. Germination energy and number of cotyledons were under very significant influence ($p=0.01$), while germination rate was under significant influence ($p=0.05$) of different light conditions.

The highest average values of germination energy of the English marigold was recorded in the treatment T₁ (32%), while the lowest value was recorded in the treatment T₂-blue LEDs (1%). Also, average value of germination rate was recorded in the treatment T₁ (47%), while the lowest value was recorded in the treatment T₂ (23%). The highest average development of hypocotyls were recorded in the treatment T₁ (11.50) and the lowest in the treatment T₂ (4.5).

Table 1. Influence of different light conditions on germination energy, seed germination, number of cotyledons of English marigold - *Calendula officinalis* L. seeds

Treatment variant	germination energy %	germination rate %	number of cotyledons
Control K	12 ^b	38 ^b	9.5 ^b
Treatmen T ₁	32 ^a	47 ^a	11.50 ^a
Treatmen T ₂	1 ^c	23 ^c	4.5 ^c
Treatmen T ₃	10 ^b	26 ^c	5.75 ^c
Average	13.75	33.5	7.81
Analyses of variance - F	10.73822**	4.032787*	7.062718**
LSD	germination energy %	germination rate %	number of cotyledons
0.05	12.2941	17.0185	3.7676
0.01	17.2366	ns	5.2822

(K-white FLUO light; T₁-red LED light; T₂-blue LED light; T₃-combination of red/blue light); means marked with different letters ^{a,b,c} significantly differ at $p=0.05$

ns=not significant

Data shown in Table 2. indicates very significant difference ($p=0.01$) between the average values of hypocotyl height, root length, and fresh weight of *Calendula officinalis* L. seeds under influence of different light conditions. Treatment T₁ - red LEDs, had the best result on all of three parameters. The highest average values of hypocotyl height and root length of

English marigold were in the treatment T₁ (3.70 cm; 6.33 cm), while the lowest average values of hypocotyl height and root length of English marigold were in the treatment T₂ (1.95cm; 2.52 cm). The same ratio was in fresh weight with the highest average values in the treatment T₁ -red LED light (0.97 g), than in control - white FLUO light (0.70 g). Lower average values were recorded in the treatment T₃ - combination red/blue LEDs (0.34 g) while the lowest average values were recorded in the treatment T₂ - blue LEDs (0.28 g) (Table 2.).

Table 2. Influence of different light conditions on hypocotyl height, root length, and fresh weight of English marigold - *Calendula officinalis* L. seeds

Treatment variant	hypocotyl (cm)	height	root length (cm)	fresh weight (g)
Control K	3.00 ^b		5.18 ^b	0.70 ^b
Treatmen T ₁	3.70 ^a		6.33 ^a	0.97 ^a
Treatmen T ₂	1.95 ^c		2.52 ^c	0.28 ^c
Treatmen T ₃	2.08 ^c		2.81 ^c	0.34 ^c
Average	2.68		4.21	0.57
Analyses of variance - F	11.43219**		18.816**	8.599304**
LSD	hypocotyl (cm)	height	root length (cm)	fresh weight (g)
0.05	0.7519		1.3111	0.3397
0.01	1.0541		1.8382	0.4762

(K-white FLUO light; T₁-red LED light; T₂-blue LED light; T₃-combination of red/blue light); means marked with different letters ^{a,b,c} significantly differ at p=0.05

ns=not significant

Higher light intensities during growth of soybean - *Glycine max* (L.) Merrill resulted in increases in photosynthesis rate, light saturation intensity, and specific leaf weight (Bowes et al., 1972). Blažević (2016) confirmed that blue LED light increased the germination energy of *Calendula officinalis* L. and *Tagetes patula* L. but energy rate was not under different light conditions. Astolfi et al. (2012) investigated used of LED lamps which emitted a continuous spectrum thanks to a mixture of blue, green, red and far-red LEDs. Their results showed that plant response to light quality seems to be related to the plant species. In *Fagus sylvatica* L. seedlings fresh and dry weight, shoot height and leaf area were greatest when plants were cultured under LED light, and lowest under fluorescent lamps. Also, they found that LED-induced reduction of chlorophyll contents in *Fagus sylvatica* L. and *Quercus ilex* L. leaves resulted in an increase of the carboxylase capacity in the same plant species suggesting an improvement of light use efficiency in these plants.

Behzadi et al. (2012) confirmed that exposed basil seeds with LED light with wavelength of 620-625 nm before sowing, induced plant growth density parameters, increasing of the internal energy on seeds and change increasing growth rate, chlorophyll content, fertility, and biological parameters.

Fraszczak (2013) exposed *Anethum graveolens* L. seeds with short term of red and blue light. Results showed that the values of plant fresh mass, area and height parameters were the highest for plants treated with red light.

Conclusion

The results of this research showed positive effects on germination energy, germination rate, and all morphological investigated parameters (hypocotyl height, root length, and fresh weight) in English marigold (*Calendula officinalis* L.) seeds exposed to red LED light, where the following conclusions were obtained:

- the highest percentage of germination was in the treatment with red light (T₁) of 47%, with an increase of 104% over the treatment with blue light T₂ (23%);
- the highest value for the average hypocotyl height was 3.70 cm on red light (T₁) in compare with the lowest on blue light (T₂) - 1.95 cm;
- the highest value for the average root length was 6.33 cm on red light (T₁) in compare with the lowest on blue light (T₂) - 2.52 cm;

Based on the obtained results we can conclude that the treatment with red LED light can enhance seed vigor and plant productivity of English marigold (*Calendula officinalis* L.). Blue light and combination of red/blue light had a similar results in average values of hypocotyl height and root length, and significantly reduced plant growth, in compare with red light. Also, we can conclude that the effects obtained may be greater if the seeds are preventively treated with a fungicide.

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