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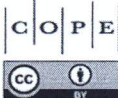
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didn't show big fluctuation during the years and are far below the proposed limit. All samples fulfil the regulations of the European Commission regarding maximal concentration of OTA in wine and other wine and/or grape must based beverages.

### P73

#### Validation of ultra-high performance liquid chromatography coupled with triple quadrupole mass spectrometry method for determination of thyreostats in bovine urine

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**Introduction:** Thyreostats are veterinary drugs that may be used in livestock production as growth promoter. They cause retention of water in the subcutaneous and muscular tissues as well as in the gastro-intestinal tract by inhibiting thyroid hormone production; thus administration of thyreostats results in increasing of body weight. On the other hand, residues of thyreostats in meat pose a risk for human health due to the teratogenic and carcinogenic effect. For this reason, in 1981 the European Union banned their use in animal production. The aim of this study was validation of the UHPLC MS/MS method for determination of thyreostats in bovine urine.

**Material and methods:** Our study included four thyreostats: thyouracil (TU), methylthiouacil (MTU), propylthiouacil (PTU) and tapazole (TA). Besides these thyreostats, dimethylthiouacil was used as an internal standard. Negative bovine urine was used for validation. Before extraction, the pH of the urine was adjusted to 1. Then derivatisation of the samples was performed with 3 iodobenzylbromide, 1h at 40°C and liquid/liquid extraction was performed with ethyl acetate. The method was validated according to 2002/657/EC. The decision limit (CC $\alpha$ ), detection capability (CC $\beta$ ), accuracy, precision and reproducibility were validation parameters which were obtained with validation procedures. The calibration curve for all standards was from 1.0 to 30  $\mu\text{g/kg}$ .

**Results:** The linearity of the method showed good correlation for all standards with  $r^2=0.9913$  for TA,  $r^2=0.9977$  for TU,  $r^2=0.9969$  MTU and for PTU  $r^2=0.9946$ . The accuracy was evaluated by determining the recovery of spiked urine samples on three concentration level at 5 $\mu\text{g/l}$ , 10 $\mu\text{g/l}$  and 15 $\mu\text{g/l}$ . The recovery for TA was from 98.22 to 123%, for TU was from 81.11 to 102.33%, for

MTU was from 92.89 to 93.67% and for PTU the recovery was from 92.33 to 106.33%. The precision of the method ranged from 4.58 to 17.45% and the reproducibility of the method was from 1.61 to 20.70%. The value from CC $\alpha$  were 2.26 $\mu\text{g/l}$  for TA, 10.71  $\mu\text{g/l}$  for TU, 2.79 $\mu\text{g/l}$  for MTU and 2.22 $\mu\text{g/l}$  for PTU. CC $\beta$  were found to be 2.93 $\mu\text{g/l}$  for TA, 16.75 $\mu\text{g/l}$  for TU, 3.60 $\mu\text{g/l}$  for MTU and 3.04 $\mu\text{g/l}$  for PTU.

**Conclusion:** The UHPLC-MS/MS method for detection and identification of four thyreostats compounds in urine was developed. Good linearity, good precision, recovery and reproducibility, make this method applicable for determination of thyreostats in bovine urine.

### P74

#### RP-HPLC determination of 5-hydroxymethylfurfural in honey from the Republic of Macedonia as a control parameter of its quality with some analytical aspects: a random collection study

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The decomposition of sugars produces the organic compound 5-hydroxymethylfurfural, also known as HMF. HMF affects the color and taste, thus manufacturers monitor its levels in some foods, including honey. HMF production increases with extended shelf life and/or heat exposure, and is used commonly as an indicator of heat and storage changes in honey. But, HMF is not a harmful substance in the levels present in food. According to the Codex Alimentarius Standard for Honey (1981), "HMF content of honey after processing and/or blending shall not be more than 40 mg/kg", and "in the case of honey of declared origin from countries or regions with tropical ambient temperatures, and blends of these honeys, the HMF content shall not be more than 80 mg/kg."

The aim of this work was to develop, test and implement simple, fast, accurate, rugged and robust chromatographic method for determination and quantification of 5-hydroxymethylfurfural in honey.

In this research we used HPLC system automated Varian ProStar with ternary high pressure mixing pump, Autosampler 410 with column oven and Diode Array Detector 330, controlled by software Varian-Star Version 6.31; the chemicals were Ph.Eur. grade, products of Merck Darmstadt, Germany. Several columns were tested: LiChrosphere C18 125 x 4 mm, 5mm; LiChroCart C18e 125 x 4mm, 5mm, protected with C18e pre-columns 4 x 4 mm; Eurosphere EC Knauer C18 250 x 4,6 mm, 5 mm and Perkin Elmer Brownlee C18 Pecosphere 33 x 4,6 mm with 3 mm particles.

The extraction procedure was simple, using demineralized water; the monitoring signal at 282 nm, column oven set