Ss. CYRIL AND METHODIUS UNIVERSITY IN SKOPJE FACULTY OF VETERINARY MEDICINE - SKOPJE


Joint meeting
$5^{\text {th }}$ International Vet-Istanbul Group Congress
\&
$8^{\text {th }}$ International Scientific Meeting Days of veterinary medicine - 2018

# BOOK OF ABSTRACTS 

23-27 September 2018 Ohrid, R. of Macedonia

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Published by:
Faculty of Veterinary Medicine - Skopje, Lazar Pop Trajkov 5/7, 1000 Skopje
Tel: ++38923240700 Fax: ++ 38923114619
www. fvm.ukim.edu.mk


CIP - Каталогизација во публикација
Национална и универзитетска библиотека "Св. Климент Охридски", Скопје 636.09(062)(048.3)

INTERNATIONAL scientific meeting ( $8 ; 2018$; Ohrid)
Book of abstracts : join meeting : 5 th International Vet-Istanbul group congress \& 8 th International scientific meeting days of veterinary medicine - 2018, Ohrid, 2018 Republic of Macedonia / [editors Florina P. Percinic, Lazo Pendovski, Katerina Blagoevska]. - Skopje : Faculty of Veterinary medicine, 2018. - 200 стр. ; 23 см

Регистар
ISBN 978-9989-774-31-7
a) Ветеринарна медицина - Зборници - Апстракти COBISS.MK-ID 108195850

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It is my grea
$5^{\text {th }}$ International Days of veterinary $23^{\text {th }}$ to $27^{\text {th }}$ Septem

This year, in University, we'll b academic cooperat (VEE) partners, by the public good.

The Faculty together outstandin experts in the field researchers, veterina doctoral students, off in the veterinary edu following topics: anin and food safety.

The program for retain its usual popular student papers and pos authors, whose researc of the International Sc electing 117 papers fro followed by two poster invited speakers for the talks.

A successful confer place, where ideas abou debated, which will ope enjoyment.

On the behalf of the in Macedonia.

We look forward to w

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# DETERMINATION OF $\beta$-AGONIST RESIDUES IN LIVER USING LC-MS/MS METHOD 

Risto Uzunov ${ }^{1 *}$, Zehra Hajrulai-Musliu ${ }^{1}$, Velimir Stojkovski², Elizabeta DimitrieskaStojkovic ${ }^{1}$, Biljana Stojanovska-Dimzoska ${ }^{1}$, Stefan Jovanov ${ }^{1}$, Dean Jankuloski ${ }^{1}$
${ }^{l}$ Food Institute, Faculty of Veterinary Medicine, Ss. Cyril and Methodius University in Skopje, Lazar Pop Trajkov 5-7, 1000 Skopje, Macedonia ${ }^{2}$ Institute of Reproduction and Biomedicine, Faculty of Veterinary Medicine, Ss. Cyril and Methodius University in Skopje, Lazar Pop Trajkov 5-7, 1000 Skopje, Macedonia
$\beta$-agonists in human and veterinary medicine are mainly used for treatment of asthma and broncho-pulmonary diseases. Moreover, these substances are used illegally as growth promoters in animal husbandry, because improve feed conversion, increase protein synthesis, increase muscle mass and decrease fat deposition in farm animals. After administration as growth promoters in farm animals, the residues from these substances are accumulated in animal tissues. The residues from $\beta$-agonists may be fatal after acute consumption. Several acute intoxication in human after consumption of liver or muscle with these substances are documented. Because of negative effect on human health the European Commission banned the use of this drug in animal feeding. According to the Council Directive 96/22/EC the use of these substances in animal production should been monitored. The aim of this study was determination of $10 \beta$-agonists (clenbuterol, salbutamol, brombuterol, mabuterol, zilpaterol, cimbuterol, terbutaline, ractopamine, isoxsuprine and clenpenterol) in liver from bovine, pigs and chickens using LC-MS/MS method. The method was validated according criteria stipulated under the Commission Decision 2002/657/EC and the following parameters were determined: linearity of the method, decision limit ( $\mathrm{CC} \alpha$ ), detection capability ( $\mathrm{CC} \beta$ ), accuracy, precisionand reproducibility of the method. The accuracy, precision and reproducibility were determined at three different levels ( $0.25,0.5$ and $0.75 \mathrm{ng} / \mathrm{g}$ ) by spiking the blank liver samples.A totals of 101 liver samples were analyzed ( 25 bovine, 29 pigs and 47 chicken liver samples). The sample preparation was performed according to the confirmatory method from the reference laboratory for $\beta$-agonists from Berlin, Germany, and the analyses were carried out on the LC-MS/MS. The coefficient of correlation ( $\mathrm{r}^{2}$ ) ranged from 0.9850 to 0.9992 . The results from $C C \alpha$ varied from $0.160 \mathrm{ng} / \mathrm{g}$ to $0.315 \mathrm{ng} / \mathrm{g}$ and the results for $\mathrm{CC} \beta$ ranged from $0.115 \mathrm{ng} / \mathrm{g}$ to $0.344 \mathrm{ng} / \mathrm{g}$. The accuracy of the method for three concentration level ranged from $75.20-107.07 \%$. The precision and reproducibility were demonstrated by CV, \% (coefficient of variation, \%). The CV for precision varied from $3.430 \%$ to $15.565 \%$, while for reproducibility CV varied from $5.231 \%$ to $17.082 \%$.The results from validation study are in accordance with criteria from 2002/657/EC and the method is linear, accurate
rabeta Dimitrieskaean Jankuloski
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rary Medicine, rajkov 5-7,
ed for treatment of es are used illegally conversion, increase farm animals. After om these substances be fatal after acute n of liver or muscle on human health the g. According to the 1 production should gonists (clenbuterol, aline, ractopamine, s using LC-MS/MS Her the Commission mined: linearity of uracy, precisionand lity were determined ver samples.A totals ken liver samples). ty method from the alyses were carried n 0.9850 to 0.9992 . ults for $\mathrm{CC} \beta$ ranged concentration level emonstrated by CV, $130 \%$ to $15.565 \%$, ults from validation d is linear, accurate
and precise. In the liver samples the $\beta$-agonists were not detected. In summary, the LC-MS/ MS method for detection of ten $\beta$-agonists in liver was validated. The validation results showed that this method is applicable for routine analysis of bovine, pigs and chickens liver. Therefore, it is still necessary to monitor $\beta$-agonists in the future because the results of this study do not exclude the possibility of misuse of $\beta$-agonists.

Key words: $\beta$-agonists, liver, validation, LC-MS/MS

