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

PROCEEDINGS

DAYS OF VETERINARY MEDICINE 2013



The 4th International Scientific Meeting

06-08 September 2013
Struga, Republic of Macedonia

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4th International Scientific Meeting

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FOREWORD

Dear Colleagues,

This year the 4th International Scientific Meeting "DAYS OF VETERINARY MEDICINE-2013", is held from 6-8 September in Struga, Macedonia. The organizer is the Faculty of Veterinary Medicine at the University of Ss. "Cyril and Methodius" in Skopje.

The program offers unique opportunity for plenary lectures, scientific presentations and discussions regarding the following topics: animal health, food safety, public health, animal welfare and animal reproduction.

We believe that this meeting is an excellent occasion for renewal of old and making new contacts between scientists, veterinary practitioners and official veterinarian. Moreover it will be an open platform for dissemination the knowledge in the field of veterinary medicine.

Veterinary science has undergone tremendous development in all fields of research and gained ever-increasing importance in the management of many diseases. Faculty of Veterinary Medicine in Skopje continuously plays a fundamental role in these processes in our country and the region. These activities have generated significant results in various areas of the veterinary science and contributed greatly to the incorporation of science into every day veterinary practice.

The city of Struga is lying on the shore of Lake Ohrid, which is indeed the cultural heritage of the Republic of Macedonia in miniature and is ready to welcome all participants offering unique experience through a blend of beautiful lake, museums, old churches and above all traditional Macedonian food.

We kindly welcome you in Struga for this unique and stimulating event!

The Organizing Committee!

PROGRAMME
DAYS OF THE VETERINARY MEDICINE
2013

Day 1
(6 September 2013)

07:00-09:00 **Registration of Participants**

Session I: ANIMAL HEALTH
***(Moderators:** Mrenoshki Slavcho and Gilles P. Dupré)*

09:00-09:30 **Opening Addresses:**

Prof. Dr. Dine Mitrov, Dean of the Faculty of Veterinary Medicine-Skopje

Prof. Dr. Velimir Stojkovski, Rector of the University "Ss. Cyril and Methodius"

Mr. Dejan Runtevski, Director of the Food and Veterinary Agency

Plenary Lecture

09:30-10:10 **Success in Oncological Surgery**
Gilles P. Dupré

Oral Presentations

10:10-10:20 **Study Of The Effects Of Breed On Some Innate Immunity Parameters In Rams**
Krasimira Genova, Dimitrova Ivona, Stancheva Nevyana, Angelov Geno, Nakev Jivko, Mehmedov Tandju, Georgieva Svetlana

10:20-10:30 **Biochemical Characterization Of Paenibacillus Larvae Genotypes In Bulgaria**
Rusenova Velizarova Nikolina, Rusinov Parvanov Parvan

10:30-10:40 **Biliary Clearance Of Bromosulfophthalein In Healthy And Ketotic Holstein Cows**
Kirovski Danijela, Sladojević Željko, Šamanc Horea

10:40-11:00 ***Discussion***

11:00-11:30 ***Coffee Break***

11:30-11:40 **Investigations Of Some Haematological And Blood Biochemical Parameters In Cows With Spontaneous Bovine Leukemia Viral Infection**
Georgiev Sandev Nikolay, Stoycheva Zapryanova Dimitrinka, Stoycheva Zhekova Ivanka, Velizarova Rusenova Nikolina, Mircheva Georgieva Teodora

11:40-11:50 **Diagnostic Importance Of Liver Percutan Biopsy In Dairy Cows**
Janevski Aleksandar, Celeska Irena, Dzadzovski Igor, Ulcar Igor, Mitrov Dine

11:50-12:00 **Detection Of Recessive Mutations (BLAD And CVM) In HF Cattle Population In Republic Of Macedonia**
Adamov Nikola, Dovc Peter, Mitrov Dine, Esmerov Igor

12:00-12:10 **Constructing Of Two ShRNA Plasmid Specific Transcripts Of The BCL-2 Genes**
Esmerov Igor, Panov Sasho, Stojkovski Velimir, Atanasov Branko, Adamov Nikola, Mickov Ljubcho, Blagoevska Katerina

12:10-12:20 ***Discussion***

12:20-13:00 **ZOETIS – Commercial Presentation**

13:00-14:00 ***Lunch***

Session II: FOOD SAFETY AND VETERINARY PUBLIC HEALTH
*(**Moderators:** Jankuloski Dean and Ali Ajdin)*

Plenary Lecture

14:00-14:30 **Resistance Mechanisms Against Antimicrobial Agents Of Foodborne Staphylococcus Aureus**
Ali Ajdin, Mert Sudagidan

Oral Presentations

14:30-14:45 **Detection Of Hepatitis E Virus In Faeces And Liver Of Pigs Collected At Two Slovenian Slaughterhouses**
Lainšček Raspor Petra, Toplak Ivan, Kirbiš Andrej

14:45-15:00 **Aflatoxin M_1 In Milk Samples**
Breda Jakovac Strajn, Ksenija Šinigoj *Gačnik, Andrej Kirbiš, Gabrijela Tavčar Kalcher*

15:00-15:15 **Characterisation Of Lactococcus Garvieae Isolated From Raw Milk In Kosovo**
Mehmeti Ibrahim, Muji Skender, Diep B. Dzung, Nes F. Ingolf

15:15-15:30 ***Coffee Break***

15:30-15:45 **Novel Alternatives To Antibiotics In Veterinary Medicine**
Velev Romel, Ćupic Vitomir, Krleska-Veleva Natasa

15:45-16:00 **Antibiotic Resistance Of Enterococci Isolated From Dairy Products**
Mojsova Sandra, Sekulovski Pavle, Jankuloski Dean, Ratkova Marija, Prodanov Mirko, Angelovski Ljupco, Gavrilova Jovanka

16:00-16:15 **Study On Factors (Ph, Water Activitity, Salt Content) Affecting The Growth Of Listeria Monocytogenes In Raw Dried Cured Sausages**
Daskalov Hristo, Fejzullah Fejzulla, Stoyanchev Todor

16:15-16:30 **Dairy Sector In Republic Of Macedonia – Yesterday, Today, Tomorrow**
Sekovska Blagica, Todorovska Marina, Nikolova Krasimira

16:30-17:30 ***Poster Session***

20:00 ***GALA DINNER***

Day 2
(7 September 2013)

Session III: ANIMAL REPRODUCTION
*(**Moderators:** Dovenski Toni and Opsomer Geert)*

Plenary Lecture

09:00-09:40 **High Yielding Dairy Cows: To Produce Or To Reproduce And What Practitioners Should Know About This To Help Their Clients**
Opsomer Geert

Oral Presentations

09:40-10:10 **Highlights On Artificial Insemination (AI) Technology In The Pigs**
Khalifa Tarek, Kousenidis Kostas, Lymberopoulos Aristotelis

10:10-10:30 **First Results From Insemination With Sex-Sorted Semen In Macedonia**
Kochoski Ljupce, Filipov Zoran, Joshevski Ilcho, Ilievski Stevche, Davkov Filip

10:30-10:50 **Comparison Of Microbiological And Endoscopy Diagnostic Of Uterine Disease In Infertility Of Dairy Cattle**
Pavlovic Milosh, Pavlović Ivan, Vakanjac Slobodanka, Jakić - Dimić Dobrila, Pavlović Voislav, Djuric Mile, Radojičić Marina, Nedić Svetlana, Maletić Milan

10:50-11:00 ***Discussion***

11:00-11:30 ***Coffee Break***

11:30-11:50 **Kinetic Parameters Of Cryopreserved Yorkshire Boar Semen**
Mickov Ljupcho, Atanasov Branko, Blagoevska Katerina, Esmerov Igor, Nikolovski Martin, Dejanoski Toni, Dovenski Toni

11:50-12:10 **An analysis Of Sow's (RE) Productive Performances Associated With Management Practices In Commercial Swine Farms In R. Macedonia**
Angjelovski Branko, Mrenoshki Slavcho, Djurovski Ivica, Dejanoski Toni, Dovenski Toni

12:10-12:30 **Effect Of Reduced Glutatione On Kinematic Parameters Of Ram Spermatozoa**
Nikolovski Martin, Mickov Ljupco, Dovenska Monika, Petkov Vladimir, Atanasov Branko, Dovenski Toni

12:30-12:50 **Prediction Of Positive Pregnancy Diagnosis In Dairy Cows On Day 21 After Artificial Insemination – On Farm Report**
Atanasov Branko, Mickov Ljupco, Nikolovski Martin, Ilievska Ksenija, Paunkov Ilija, Burmuzoska Angelina, Dovenski Toni

12:50-13:00 ***Discussion***

13:00-14:00 ***Lunch***

Session IV: ANIMAL WELFARE
*(**Moderators:** Ilieski Vlatko and Munksgaard Lene)*

Plenary Lecture

14:00-14:30 **Can Animal Welfare Be Measured Automatically?**
Munksgaard Lene

Oral Presentations

14:30-14:45 **Animal Welfare In Slaughterhauses**
Novaković Zorica Jovana, Gerić Tamara Lazara, Turubatović Radoslav Lazara, Vesković-Moračanin Slavica Mirka, Turubatović Lazar Radoslava

14:45-15:00 **Usage Of Tri-Axial Acceleration Of The Hind Leg For Recognizing Sheep Behavior**
Radeski Miroslav, Janevski Aleksandar, Nikolovski Martin, Pendovski Lazo, Ilieski Vlatko

15:00-15:15 **Welfare Quality Protocol Provides An Insight In The Welfare Problems Of The Laying Hens In Republic Of Macedonia**
Prodanov Mirko, Ilieski Vlatko

15:15-15:30 **Possibilities Of IR Thermography For Early Diagnosis Of Lameness In Dairy Cattle**
Ilievska Ksenija, Atanasov Branko, Petkov Vladimir, Dovenski Toni, Trojacanec Plamen

15:30-15:45 **Fin Damage Of Farmed Rainbow Trout In Republic Of Macedonia**
Cvetkovikj Aleksandar, Radeski Miroslav, Blazhekovikj-Dimovska Dijana, Kostov Vasil, Stevanovski Vangjel

15:45-16:00 **Evaluation Of The Antiparasitic Effect Of The Button Mushroom Agaricus Bisporus**
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17:00-20:00 **Guided Ohrid Tour**

20:00 **DINNER at National Restaurant**

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Session I
ANIMAL HEALTH

SUCCESS IN ONCOLOGICAL SURGERY

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ABSTRACT

Surgery is the oldest modality of treating cancer and more patients are cured by surgery alone as by any other mean of cancer treatment. Nowadays, thanks to adjuvant therapy, aggressive treatments resulting in mutilations and amputations can be avoided leading nevertheless to better results. Some ethical aspects should be reminded and previous authors should be quoted

- Aegypt (3000 y BC) "Surgeon should contend with tumors that might be cured by surgery".

- Hippocrates (450 bc) "Surgeon should avoid treating terminal patients whose quality of life would be better without surgery".

Oncologic surgery embraces many aspects : Biopsies, surgical excision, cytoreduction/Palliative surgery, prophylactic surgery, emergencies, supportive surgeries and treatment of metastasis. We will concentrate ourselves on the most common indication for oncologic surgery, i.e tumor removal and will analyse the key for successfull surgeries.

Any of us before operating a tumor should be able to answer these questions:

- Do I know enough about this tumor?
- Do I know how to treat it?
- Can I operate it adequately?

Pre-operative work-up

The Pre-operative work-up should include a patient as well as a tumor evaluation. If the tumor can be palpated, tis could give some information but it should not be manipulated to avoid tumor cell spreading. Specific imaging techniques are often necessary to provide information about local invasiveness. Cytological and histological diagnosis is mandatory before any further recommendation can be made either medical or surgical. Local and focal lymph nodes as well as any distant metastatic possibility should be evaluated. Newest diagnostic modalities and experimented people are necessary in order to minimize the risk of operating patients with severe metastatic disease. Thanks to Ultra-sound or CT any suspicious tissue, even distant from the primary tumor itself, can be aspirated.

When cytological examination is not reliable or when more information is needed, tissue biopsies should be performed. Specific guidelines for biopsies

have been edicted :

- Choose the right sites at the junction between pathologic and healthy tissue
- Prefer multiple directions
- Avoid necrotic centers
- Avoid crossing healthy tissue
- Use Mini-invasive surgery for abdominal biopsies when fine-needle aspirates give negative results

Once enough information has been collected from the patient and its tumor (staging and grading), the veterinarian should wonder if he knows how to treat it. In this regard the most current literature should be consulted in up to dated textbooks. It is usually pretty sad when owners know from internet better than the practitioner himself…Once the need for surgery has been confirmed, the next question is: Can I operate it adequately ?

The pre-operative preparation include patient's preparation and surgeon's preparation, i.e preoperative planning. Many of the cancer patients suffer of poor nutritional status, mild to severe pain as well as an altered physical status. These should be evaluated and compensated before surgery. If needed a feeding tube is to be placed in order to restore an appropriate nutritional status before and after surgery. Paraneoplastic syndromes should also be corrected. As a surgeon the veterinarian should then plan tumor removal and reconstruction. Should closure difficulties be anticipated, the patient should be referred to an oncologic surgeon.

Peroperative rules

From the preparation room to the post-operative bandage, the surgeon should aim at preventing tumor cells from seeding. Agressive surgical scrubbing should be avoided and adequate excision has to be chosen.

The subtotal excision is basically intra-tumoral. It carries no indication except if the tumor is radiosensitive, chemosensitive or if only palliative removal is attempted.

In a local excision (intra-capsular), millions of fresh tumor cells are left in place. The capsule is nothing else but the outer limit of a tumor, where the most active tumor cells grow. Tumor cells are usually encountered in more than 50% of the « adhesions ».

In the wide excision a border of 2 to 3 cm of healthy tissue has to be taken away with the tumor istself. In the depth, at least one healthy tissue layer not in contact with the tumor base should be removed.

In radical excision the organ supporting the tumor is being removed (Amputation, mandibulectomy, hemi-pelvectomy…).

What to do with lymph nodes ? They should be aspirated when palpable or visible (Ultra-sound). Any positive, abnormal or tumor-associated lymph node should be removed with the tumor itself.

In surgery the oncologic surgeons should respect basic rules :

- No dissection, en-bloc excision
- Primary ligation of major tumor blood supply
- Control of local bleeding
- Prevention of dead space
- Closure after changing instruments, gloves and drapes

Surgery is simply not worth it if samples from the tumor and tissue edges are not submitted. After surgery, the surgeon should help the pathologist and specify his (her) needs : Type, grade, proliferation or mitotic index, specific staining, margins…

Post-operative

During the post-operative period, promoting factors for infection and pain should be evaluated, controlled and treated. Before and after surgery the need for further adjuvant therapies should be mentioned to the owner. At reception of the pathology results, these needs shall be further discussed.

Conclusion : Who is responsible for recurrence ?

The patient and its tumor…The fate…Ourself… ?

Key words: cancer, dog, cat, surgery, oncologic surgery guidelines, tumor seeding

STUDY OF THE EFFECTS OF BREED ON SOME INNATE IMMUNITY PARAMETERS IN RAMS

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ABSTRACT

Investigations were carried out on 27 rams from the breeds Karakachanian and Copper Red Shoumen. The non-specific immune parameters, phagocytic activity of leukocytes, bactericidal activity of phagocytes systems (oxygen-dependent and oxygen independent), inhibition of leukocyte migration and total plasma protein level were evaluated. Phagocytic response was evaluated against S. aureus 209-P with a certain percentage of active phagocytes (phagocytic index) and the number of absorbed particles per one phagocytic cells (phagocyte number). Phagocytosis completion index was defined as the percentage of the microbial cells that have been destroyed by phagocytes after incubation. State of the oxygen-dependent bactericidal systems of phagocytes was assessed in vitro using the NBT test, which reflects the ability of superoxide restore NBT in diphormazane. NBT test was evaluated by the degree of reduction in spontaneous and stimulated reactions, taking into account the intracellular deposits diphormazane.

Our studies and results shows that the rams from the two local Bulgarian breeds have a high activity of innate immune parameters and that's may be useful and important in the breeding programs as an indicator of resistance and highly tolerance to oxidative stress.

Key words: sheep, Karakachanian and Copper Red Shoumen breeds, non-specific immune parameters

BIOCHEMICAL CHARACTERIZATION OF *PAENIBACILLUS LARVAE* GENOTYPES IN BULGARIA

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ABSTRACT

Introduction

Paenibacillus larvae is the causative agent of American foulbrood, the most virulent bacterial disease of honey bees. Different genotypes of *Paenibacillus larvae* have been described in the last years. A recent study has shown that differences in genotype might correlate with the biochemical phenotype. Nothing is known about the biochemical profile of *Paenibacillus larvae* isolates in Bulgaria. Hence, the aim of this study was to characterize genotypes of the agent determined in Bulgarian apiaries with commercial identification system.

Materials and Methods

A total of 103 *Paenibacillus larvae* isolates (genotype AB, n=21; genotype ab, n=82 determined by rep-PCR using primers BOX A1R and MBO REP1) and a reference strain NBIMCC 8478 were analyzed using identification system BioLog Gen III microplates. In addition nitrate reducing ability was tested. Plates were processed following the manufacturer's instructions. To characterize the strains only the substrates providing carbon source (n=71) were taken into consideration.

Results

All tested strains were identified by the BioLog system. Genotype AB и ab isolates were able to metabolize D-trehalose, N-acetyl-D-glucosamine and N-acetyl-β-D-mannosamine. Hundred percent of the AB isolates used D-fructose and D-mannitol as carbon sources, while for the ab isolates the results were 5 % and 18 %, respectively. Glucose was utilized by 100 % of ab and 90 % of *Paenibacillus larvae* AB. All ab isolates used glycerol as carbon source, positive reactions for AB genotype were 10 %. Zero percent of genotype AB metabolized D-turanose, L-alanine and L-lactic acid. None of the tested ab isolates used D-melibiose as for the AB values were also low - 5 %. Considering the nitrate reducing ability, mannitol and salicin utilization, the strains were divided into biotypes I, III, IV and VIII. The reference strain belonged to biotype V. The majority of *Paenibacillus larvae* AB clustered into biotype III while ab were grouped mainly into biotype I. Biotypes I and IV were not found among the tested AB strains.

Conclusion

This study has shown the obvious link between genotype and biotype of Bulgarian *Paenibacillus larvae* strains. The provided information can be useful in epidemiologic situations to trace the source of infection.

Key words: Paenibacillus larvae, genotypes, biochemical characterization

BILIARY CLEARANCE OF BROMOSULFOPHTHALEIN IN HEALTHY AND KETOTIC HOLSTEIN COWS

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ABSTRACT

Introduction

Dairy cows are highly susceptible after parturition to developing ketosis. Ketosis is a metabolic disorder closely associated with liver lipidosis. Numerous tests have been developed to detect hepatic dysfunction in dairy cows. Among others, bromosulfophthalein (BSP) clearance is established as sensitive index of hepatic function. The objective of this study was to examine the difference of biliary excretion of BSP between the ketotic and healthy Holstein cows and to correlate this excretion with some other indicators of liver dysfunction.

Material and Methods

Twenty puerperal Holstein cows divided in two groups per 10 cows were involved in the study. First group of cows included healthy animals and the second group included cows with clinical symptoms of ketosis. Blood samples were taken at day 10 after parturition and concentrations of total protein, albumin, total bilirubin, Ca, P, total lipids, urea and glucose were determined. Immediately after blood sampling, BSP (bromosulfophthalein) test was performed. After BSP injection, blood samples were taken at minutes 5 and 45, and percent of retained color in sample obtained at minute 45 compared to minute 5 was calculated.

Results

Blood albumin and glucose concentrations were significantly higher in healthy compared to ketotic cows ($p < 0.05$ and $p < 0.001$, respectively) while total bilirubin concentration was significantly higher in ketotic compared to healthy cows ($p < 0.01$). Additionaly, BSP excretion was significantly higher in ketotic compared to healthy cows ($p < 0.001$). There was significant positive correlation between BSP values and total bilirubin concentrartions in both healthy ($r = 0.76$; $p < 0.01$) and ketotic cows ($r = 0.80$; $p < 0.01$) and significant

negative correlation between BSP values and glucose concentrartions in both healthy ($r = -0.816$; $p < 0.01$) and ketotic cows ($r = -0.74$; $p < 0.01$).

Conclusion

Based on obtained results it may be concluded that biliary clearance of bromosulfophthalein may be used as reliable method for detection of hepatic disfunction associated with clinical symptoms of ketosis in dairy cows. Additionally, total bilirubin and glucose concentrations may be good indicators of impaired function of liver in puerperal cows.

Key words: holstein cows, BSP test, ketosis

Acknowledgements: This work was supported by Ministry of Science and Technology, Republic of Serbia, Project Grant No 46002.

INVESTIGATIONS OF SOME HAEMATOLOGICAL AND BLOOD BIOCHEMICAL PARAMETERS IN COWS WITH SPONTANEOUS BOVINE LEUKEMIA VIRAL INFECTION

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ABSTRACT

Introduction

Enzootic bovine leukosis (EBL) is a chronic viral infection characterized by hyperplastic proliferations in haematopoietic organs accompanied by quantitative and qualitative changes in peripheral blood leukocytes.

The purpose of the present study was to monitor the changes in some haematological, whole blood or serum biochemical indices in cows, spontaneously infected with bovine leukaemia virus.

Materials and Methods

The experiment included 76 cows at various age and body weight. Serological leukosis tests were done by agar-gel immunodiffusion test with a commercial kit of Synbiotics – France, containing standardised gp 51 antigen and positive serum approved by the EU.

Blood samples were assayed on an automated analyser BC-2008 Vet to determine the total red blood cell counts (RBC), total white blood cell counts (WBC), mean corpuscular volume (MCV), haematocrit (HCT) and haemoglobin (HGB). The differential white blood cell counts were determined using routine analytical methods.

On the basis of haematological results, the cows were divided into three groups: first group – EBL-seropositive with normal haemogramme; second group – EBL seropositive with altered haemogramme and third group – controls.

The blood serum concentrations of calcium, inorganic phosphorus, triglycerides and cholesterol were assayed on an automated biochemical analyser BS – 3000 P (Sinnowa LTD) with Gisse Diagnostics (Italy) kits. The blood activities of transaminases (ASAT, ALAT) and total protein concentrations were determined with kits produced by Human (Germany). Alkaline phosphatase concentrations were analyzed with a kit of Chema Diagnostica (Italy). Gamma glutamyl transferase activity was analyzed with a kit produced by Hospitex

Diagnostics (Italy). The statistical processing of data was done with the Student's t test.

Results

In cows from the first and the second group, a statistically significantly increased blood cell counts was established compared to healthy controls. The total WBC were increased in the second group (leukocytosis) up to 33.21×10^9/l vs reference range of 5-10 $\times$ 10^9/l as well as lymphocyte percentages (lymphocytosis) – 81.89 % (reference 40–63 %). A reduction in the proportion of neutrophils to 12.78 % (relative neutropenia) vs the reference range of 22-49 % and monocytes (monocytopenia) to 1.78 % (reference range 2–6 %) was observed. A statistically significant reduction in Ca concentrations (4.41 mg/dl) and higher inorganic phosphate levels (5.28 mg/dl) were established in cows from the second group. Also, ASAT activity was considerably lower – 47.03 U/l, while alkaline phosphatase increased slightly within the reference range up to 167.68 U/l and 165.81 U/l in groups one and two, respectively.

Conclusion

The present haematological and whole blood/serum biochemical results in cows spontaneously infected with EBL virus could be used as prognostic markers of the course of the disease, to distinguish the stages of infection.

Key words: enzootic bovine leukosis, cows, haematological and blood biochemical assays

DIAGNOSTIC IMPORTANCE OF LIVER PERCUTAN BIOPSY IN DAIRY COWS

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ABSTRACT

Liver failure is the most common metabolic disorder in the high yielding dairy cows during the transition period. Diagnostic approach of the liver has the most significance importance in accurate diagnosis. The period of parturition is the period of overloading anabolic and catabolic biochemistry process in the liver. Liver is the one and only organ for gluconeogenesis from glucogenic precursors and maintaining normal concentration of glucose. The most common metabolic disease in high yielding dairy cows during the transition period is fatty liver and ketosis. Liver biopsy is very important for assessment liver function. Liver biopsy samples from phosphate buffered 10% formalin solution can be used for preparing patohistological slides and microscopic examination for stereometric and morfometric determination of fats in the hepatocytes for defining the degree of hepatic lipidosis. Also, the biopsy samples can be used for determination total concentration of genomic RNA and DNA with molecular methods and absolute and relative concentration of cell receptors. Percutaneous needle liver biopsy is made with clipping and shaving the area in the 11 intercostal space at the right side of torax. Local subcoutaneous infiltrative anesthesia of 2% lidocaine hydrochorid is used. The site of biopsy is 15 cm below the transverse process in the 11 intercostal space at the right side. The site is also defined by imaginary lines from the wing if the ileum to the point of elbow and the point of shoulder. The side is the area of the 11th intercostals space enclosed by these lines. Percutaneous and subcutaneous punctuate incision is made with scalpel blade, and sterile biopser perforate intercostals muscle, peritoneum and capsula fibrosa Glissoni. Biopser is applied in the liver parenchyma and the inner part of biopser is withdrown. The outer part of the biopser is switched deeply in the liver parenchyma, in order to cut a piece of the tissue. Applying the negative pressure with 20 ml syringe, the peace of liver is suction in the lumen of biopser. The tissue sample is used for further scientific examination, for histopathology examination and estimation of cell receptors quantification.

Key words: liver biopsy, dairy cows, biopser

DETECTION OF RECESSIVE MUTATIONS (BLAD AND CVM) IN HOLSTEIN-FRIESIAN CATTLE POPULATION IN REPUBLIC OF MACEDONIA

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ABSTRACT

Introduction

Bovine leukocyte adhesion deficiency (BLAD) and Complex vertebral malformation (CVM) are Holstein-specific autosomal recessive disorders which have had significant economic impact on dairy cattle breeding worldwide. The increased used of artificial insemination and the worldwide use of several bulls with superior proofs have led in the past to significant dissemination of these recessive alleles on global scale. This study was conducted with purpose to obtain a preliminary picture about the existence of these recessive alleles in R. Macedonia.

Matherials and Methods

Blood samples were obtained from 84 cattle of the HF breed reared on several different farms in Republic of Macedonia. Genomic DNA was extracted with DNeasy blood and tissue kit (Qiagen). This kit was also used for extraction of DNA from deeply frozen sperm doses from 6 different HF bulls that are property of the Faculty of veterinary medicine in Skopje. The target sequences harboring the recessive mutations were amplified by PCR. Identification of BLAD carriers was performed by digestion of the amplified products with *TaqI* and *HaeIII* restriction endonucleases and identification of the CVM carriers was done by digestion with *PstI* endonuclease. All heterozygote carriers were confirmed by capillary electrophoresis sequencing on ABI Prism 310 Genetic Analyser (Applied Biosystems).

Results

Among 90 HF cattle tested two cows were identified as carriers (+/-) of the BLAD allele and one was carrier of the CVM allele. The mutant allele frequencies were calculated as 0.011 and 0.006 for BLAD and CVM respectively with corresponding carrier prevalence of 1.22 % (BLAD) and 1.11 % (CVM).

Conclusions

This study demonstrates that carriers of BLAD and CVM are present in the Macedonian HF population, although at low frequency. PCR-RFLP is an efficient and inexpensive method for detection of these recessive alleles and should be used as a laboratory test in eradication programs. In order to establish a more accurate picture of the prevalence of these disorders, it is necessary to screen a larger number of animals. Also it is of great importance to test all the bulls intended for artificial insemination to prevent accidental transmission of these unwanted mutations to a large number of offspring.

Key words: BLAD, CVM, bovine autosomal recessive mutations

CONSTRUCTING OF TWO SH RNA PLASMID SPECIFIC TRANSCRIPTS OF THE BCL-2 GENES

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ABSTRACT

Prostate cancer is a malignancy of the largest gland in men, including the testes and seminal vesicles. The prostate cancer cells are generally not lethal by themselves, however if the tumor grows some of the cells begin to disintegrate and disseminate to distant parts of the body through the lymph and blood vessels. The most common target of the prostate metastases are the lymph nodes, seminal vesicles, lungs and bones.

The prostate cancer is a common disease since the beginning of this century, and it is diagnosed in a number of men in the Western Hemisphere. This type of cancer is slow growing, but can be very aggressive in the young population.

The techniques of genetic engineering and the general molecular biological procedures are carried out in accordance with appropriate laboratory manuals (Sambrook et al., 1989) as well as with the established methods in Molecular Biology Laboratory within the Institute (Panov, 2003; Panov, 2005). When using commercial sets and products, guidelines and protocols were used from the manufacturers as well as other recommended literature available online.

It was constructed two shRNA plasmid-specific transcripts of bcl-2 gene. The plasmid is designed according to previously published sequence for interferer RNA under the appropriate reference, using appropriate software (siRNA Wizard, InvivoGen). The designed single-strand DNA oligonucleotides were ordered with the highest degree of purity (PAGE) from appropriate manufacturer (SIGMA-Genosys).

The bacterial transformations are executed with the E strain GT116. coli (InvivoGen), which is deletion mutant of SbcCD allele for which the manufacturer claims is suitable for amplification of plasmids containing hairpin-s. If necessary, it will also be used the standard bacterial strain JM109 (SIGMA) which results in a high plasmid yield. Given that the cloning places for shRNA

passages are flanked by Lac-Z cassette alpha peptide, it is removed during cloning, so the discrimination of the transformed bacteria with successfully recombinant plasmids psiRNA is performed with white / blue screening (loss of alpha complementation) of strong RNA LB-based substrate with galactosidase inducer IPTG and substrate X-Gal. Antibiotic selection is conducted with antibiotic Zeocyn. Five colonies from each bacterial culture are tested by polymerase chain reaction (PCR) with an amplification in thermocycler (Perkin Elmer GeneAmp PCR System 2400) with primers specific for the plasmid (InvivoGen). Three isolates of each colony were verified by sequencing both strands with fluorescent kit Sanger dideoksiterminating sequencing (Applied Biosystems) with appropriate equipment for sequencing (Applied Biosystems Genetic Analyzer 310).

The verified bacterial colonies, each from a different shRNA-encoding recombinant plasmid and from the control plasmid psiRNAScr are to be cultivated on a large volume of liquid medium TB (Terrific broth, InvivoGen).

The plasmid DNA was cleansed from each bacterial culture with Endotoxin-free GeneElute Maxi-Prep Kit (SIGMA). DNA concentrations in each of the four psiRNA plasmid constructs and in the psiRNAScr control plasmid will be determined by UV spectrophotometry, and the integrity and the size of the plasmid DNA will be determined by the agarose gel electrophoresis.

Key words: bcl-2, PC-3, RNA interference, apoptosis, cell culture, plasmid construction, transfection

Session II
FOOD SAFETY
AND
VETERINARY PUBLIC HEALTH

RESISTANCE MECHANISMS AGAINST ANTIMICROBIAL AGENTS OF FOODBORNE *STAPHYLOCOCCUS AUREUS*

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ABSTRACT

Staphylococcus aureus is one of the important pathogens due to toxin production and development of resistance against antimicrobials and disinfectants. The use of antimicrobials in food animals and foods creates an important source of resistant bacteria that can spread to humans through the food supply. Therefore, the presence of resistant *S. aureus* strains in food can cause important infectious risks for public health. In the last decade, foodborne *S. aureus* strains have shown a considerable increase in resistance against most antibiotics in all over the world. Especially, the prevalence of vancomycin, methicillin and clindamycin resistant *S. aureus* strains was monitored carefully worldwide. The presence of resistance genes such as *van*A, *mec*A, *mec*C, *erm*A, *erm*B and *erm*C genes in *S. aureus* strains are also very important for the development of resistance mechanism. Additionally, the frequent usage of disinfectants in food industry lead to increase resistant bacteria against disinfectants and the number of resistant bacteria can be selected and grow rapidly in the food environment. The disinfectant resistance mechanism in *S. aureus* strains was established by *qac* genes especially *qacA*/*B*, *qacC*, *qacG*, *qacH* and *qacJ* encoded on plasmids. *qacC* and *smr* genes are responsible for resistance to one of the most important disinfectants quaternary ammonium compounds. Moreover, there is growing interest in the use of biopreservation methods that employ natural antimicrobial compounds. Nisin and lysozyme are commonly used in the biopreservation of food and these antimicrobials categorized as GRAS (In General Can Be Considered Safe) status in USA and Canada. In addition, nisin are numbered as European Food Additives list in 1983. The studies have shown that there are also nisin and lysozyme resistant *S. aureus* strains. Lysozyme did not inhibit commonly foodborne *S. aureus* strains but only an appropriately high concentrated nisin are effective against foodborne *S. aureus* strains.

Previous studies showed that the antimicrobial resistant *S. aureus* strains spread very quickly all of the world. In order to prevent growth of antimicrobial resistant bacteria, these antimicrobials should be use appropriate dosage in the environment include animals or food.

Key words: antimicrobial resistance, antibiotics, antimicrobials, S. aureus, public health

DETECTION OF HEPATITIS E VIRUS IN FAECES AND LIVER OF PIGS COLLECTED AT TWO SLOVENIAN SLAUGHTERHOUSES

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ABSTRACT

In recent years there have been numerous reports from different parts of the world describing hepatitis E virus (HEV) as a zoonotic agent, but the clinical cases in humans are still reported only sporadically.Domestic pigs represent the main reservoir of the HEV. Until recently it was believed that the HEV was transmitted only by faecal-oral route, but it has been proved that eating raw or undercooked pork meat and offal can cause acute HEV infection in human. This has triggered the alarm and many developed countries have already done a few studies to assess the percentage of infectedpigs.

In this study the situation regarding the risk factor of HEV among pigs that enter the food chain in Slovenia was evaluated. At two different slaughterhouses 87 faeces and liver samples were collected from pigs within two age groups. 32 faeces and liver samples were collected from 3 months old pigs and 55 faeces and liver samples from 6 months old pigs. Animals were brought to the slaughterhouse from different farms located at the north eastern part of Slovenia, where the majority of pig population is located. Collected samples were analysed with real-time RT-PCR method. Nucleic acids of HEV was found in 6 faeces samples from the younger age group (3 months of age), which represents 19% of examined samples. All liver samples from 3 months old pigs were negative. All samples of faeces and liver from 6 month old pigs were negative. The results were comparable with those from other European countries, where 7-30% of swine faeces samples were found HEV positive.

Key words: hepatitis E virus, pigs, slaughterhouse, undercooked pork meat, zoonosis

AFLATOXIN M_1 IN MILK SAMPLES

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ABSTRACT

Aflatoxins are secondary metabolites of moulds, contaminating mostly grain food and feed materials. In consideration of the carcinogenic properties of aflatoxin B_1, human exposure should be reduced to levels as low as reasonable achievable. Current EU legislation prescribed the maximum limit in various susceptible plant-derived food and feed commodities as well as in milk. Milk is contaminated with the hydroxy-metabolite aflatoxin M_1, following exposure of lactating animals to aflatoxin B_1 present in feedstuffs. As aflatoxin M_1 has toxicological properties comparable to those of aflatoxin B_1, albeit a lower carcinogenic potency, maximum levels for aflatoxin M_1 have been set for consumable milk at 0.05 μg/kg, and 0.025 μg/kg for infant formulae.

As it was generally acknowledged that the risk for aflatoxin B_1 contamination is only high in geographical regions with a tropical or sub-tropical climate, the monitoring of feed ingredients for the presence of aflatoxin B_1 focused, as of yet, on imported feeds from these regions. However, in 2003, Italy had to report for the first time an increase in the number of milk samples exceeding the statutory limit. Contamination rates above the legal limit amounted to 6% and 7.8%, respectively, in certain regions. These higher contamination rates were linked to a high contamination of locally grown maize that was used as animal feed.

In the last two years, among countries reported the presence of aflatoxins in milk samples were Slovenia, Croatia, Serbia and Macedonia. In these time, approximately 1000 milk samples were analysed in our laboratory. In a big part of samples, the concentration of aflatoxin M_1 was above the permitted levels; in some samples it was up to fifty times higher then the maximum permitted concentration. The general observations was, that when the source of contamination was found and removed, the concentration of aflatoxin M_1 in milk rapidly dropped to permitted level within a few days.

The impact of climate change has been identified as an emerging issue for food and feed safety. In particular, aflatoxins which are frequent in tropical and sub-tropical areas may become a concern in the Europe. *Aspergillus flavus* and *A. parasiticus*, the main aflatoxins producers, are xerophilic fungi. With climate

changes and expected higher temperatures and less rain, these fungi may find conditions that are more suitable for their development.

Key words: aflatoxins, aflatoxin B_1, aflatoxin M_1, milk samples, climate changes

CHARACTERISATION OF *LACTOCOCCUS GARVIEAE* ISOLATED FROM RAW MILK IN KOSOVO

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ABSTRACT

Lactococcus garvieae is an emerging pathogen in certain animals including farmed fish, cows and buffalos (mastitis), and represents a potential human pathogen. Based on our knowledge, only few reports have examined the presence of *L. garvieae* in milk and dairy products. In the present study we investigated the presence of *L. garvieae* and other pathogens as well as non-pathogens in raw milk from Kosovo. Milk samples from 221 farms were collected during the period November 2011 to June 2012. A total number of 2100 isolates were isolated on selective growth conditions (e.g., MRS and GM17) and/or by morpho-physiological and biochemical tests. Six hundreds of isolates were so far genotyped by 16S rRNA gene sequencing. The result showed that the diversity of lactic acid bacteria was very large with *L. garvieae* being relatively high in some locations. From total 221 farms, 43 (corresponding to 19% of the farms) showed high frequencies of *L. garvieae*, ranging from 4.4 to 6.17 $\log_{10}$ CFU ml^{-1} raw milk. The presence of *L. garvieae* in these locations appeared to be persistent as this organism was found with significant frequencies in two independent samplings separated by approximately 2 months. Interestingly, in most of the farms that showed a high frequency of *L. garvieae* in milk also disclosed a high incidence of staphylococci. The high frequencies of this bacterium and other food pathogens likely represent a potential health hazard for consumers of non-pasteurized milk from these regions. This study underlines a need for guidelines on how to achieve better hygiene on these farms.

Key words: Lactococcus garvieae, lactic acid bacteria, raw milk

NOVEL ALTERNATIVES TO ANTIBIOTICS IN VETERINARY MEDICINE

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ABSTRACT

Conventional antibiotics are used in the treatment and control of many types of infectious diseases in a wide variety of animal species. But in recent years an alarming increase in resistance of bacteria to antibiotics has been recognized worldwide by the WHO. The extended and uncontrolled use of conventional antibiotics representing the main risk factor for the development of bacterial resistance. The development of resistance can be minimised provided that a number of measures are observed to prolong the useful life of all antibiotics in both human and veterinary medicine. Yet insufficient attention has been given to the scientific breakthroughs and novel technologies that provide alternatives to antibiotics. Committee for Veterinary Medicinal Products encouraged an increased levels of innovation on treatment alternatives for infectious diseases. New insights into the physiology of bacterial populations and the use of new classes of compounds which impair bacterial communication and invasiveness offer the possibility of significantly reducing the amount of antibiotics used in veterinary medicine. The purpose of this paper is to present these new insights. Extensive research has been conducted on the development of seven groups of naturally occurring antimicrobials as novel alternatives to antibiotics: antimicrobial peptides, bacterial cell wall hydrolases, bacteriophages, probiotics and prebiotics, agents that interrupt bacterial communication, phytobiotics and agents which control antimicrobial resistance.

Key words: antibiotics, bacterial resistance, alternatives to antibiotics

ANTIBIOTIC RESISTANCE OF ENTEROCOCCI ISOLATED FROM DAIRY PRODUCTS

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ABSTRACT

Introduction

The growing interest in artisanal dairy products is partly due to the uniqueness of such food, in which specialized microorganisms derived from raw milk or natural starters can grow and contribute to the organoleptic and quality characteristics of cheeses. Enterococci are recognized as an essential part of natural microflora of many dairy products and, in some cheeses, they dominate over lactobacilli and lactococci (Clarrk and Reinbold 1966; Litopoulou-Tzanetaki et al., 1992). The reason for the prevalence of enterococci in dairy products has long been considered as a result of unhygienic conditions during collection and processing of milk, together with their resistance to pasteurization temperatures and their adaptability to different substrates and growth conditions (Giraffa et al., 1997).

However, the presence of enterococci in cheese and other fermented foods is a matter of debate, since some enterococcal species are involved in clinical infections such as endocarditis, bacteriaemia, urinary tract infections and neonatal sepsis (Schaberg et al., 1991).

The use of antimicrobials in animal feed as growth promoters has created large reservoirs of transferable antibiotic resistance genes and consequently a possible route of transmission of resistant *Enterococcus* spp. via food chain.

The aim of this work was to evaluate the antibiotic susceptibility of enterococci isolated from traditional and industrial dairy products.

Matherials and Methods

Enterococci originating from our own strain collection isolated from dairy products mainly artisanal cheeses in 2012-2013 and stored at -25 °C in 20 % glycerol medium were used in this study. Antibiotic sensitivity of the isolates was determined using VITEK 2 system (bioMerieux) according to the manufacturer's recommendation. The strains of enterococci were cultured on plate count agar and after 24 hours incubation were suspended in sterilized physiological saline to 0,5 McFarland standards. The bacterial suspension wasused to fill the AST P580 card, which was then inserted into the incubator-reader of the VITEK 2 system.

Results

The isolates that were choosen for this study were *E. faecalis* (16 isolates), followed by *E. faecium* and with *E. durans* (12 isolates), *E. hirae* (2 isolates), *E. casseliflavus* (1 isolate). All enterococcal isolates were susceptible to levofloxacin, linezolid, tiecoplanin, vancomycin, tigecycline. From the tested isolate 39 were resistant to Trimethoprim/ Sulfamethoxazole, 15 isolates were resistant to tetracycline, and 24 isolates were resistant to clindamycin.

Conclusion

Resistance to multiple antibiotics was observed. Most of the isolates were resistant to 2 or 3 antibiotics. The wide use of tetracycline, clindamycin and Trimetoprim/Sulfamethoxazole in animal husbandry is a possible explanation for the resistance frequently found among enterococcal isolates. None of the tested isolates was resistant to vancomycin.

Key words: enterococci, dairy products, antibiotic resistance, VITEK 2

STUDY ON FACTORS (pH, WATER ACTIVITITY, SALT CONTENT) AFFECTING THE GROWTH OF *LISTERIA MONOCYTOGENES* IN RAW DRIED CURED SAUSAGES

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ABSTRACT

Commission Regulation (EC) 2073:2005 considers the factors which can support or inhibit the growth of *L.monocytogenes* in ready-to-eat foods. The aim of the experiments was to examine the values of pH, water activity (a_w), salt content and level of contamination with *Listeria monocytogenes* of some popular in Bulgaria raw dried cured vacuum packed sausages, produced from June 2006 till May 2008. 81 vacuum-packed samples were taken from 5 different meat producing plants during the period of study. Average water activity level of the tested sausages was 0,87 ± 0,035; pH level - 5,61 ± 0,59 and salt content - 4,12 ± 1,11%. Four specimens contained *Listeria* spp. (two samples *L.monocytogenes*, one *L.welshimeri* and one *L.innocua*). All contaminated raw dried cured sausages had a_w bellow ≤ 0,92 and pH ≥ 4,4 or pH ≥ 5. After 3 months of storage of the same contaminated samples at 4^0C, in three of them *Listeria* spp. (two *L.monocitogenes* and *L.welshimeri*) survived and was detected. Salt content of the samples varied from 2,46 to 6,28% and was not able to affect the growth of *L.monocytogenes*.

Data showed that the detected levels of a_w could support the growth of *L.monocytogenes* in only 6 (7,4%) of the tested samples. pH values lower than 5 were presented in three samples and only the combination with low a_w was able to inhibit the growth of *L.monocytogenes*. The detected levels of salt content did not affect the presence and growth of *L.monocytogenes*. 'Microbiological criterion' set in COMMISSION REGULATION (EC) No 2073/2005 for ready-to-eat foods unable to support the growth of *L. monocytogenes* can be applied to 75 (92,6%) of the tested sausages.

Key words: L.monocytogenes, raw dried cured sausage, water activity, pH, salt

DAIRY SECTOR IN REPUBLIC OF MACEDONIA – YESTERDAY, TODAY, TOMORROW

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ABSTRACT

The aim of this study was to follow the changes in dairy sector in Republic of Macedonia. This study is an attempt to address the various important aspects related to dairy sector in Macedonia like source of milk production, average unit productivity, cost of milk production and milk supply channels. It may also provide an understanding of the opportunities and problems associated with the dairy enterprises in Macedonia. The findings of the study may help in ensuring development of country's dairy sector because the research based decisions of policy makers may have real impact on welfare of farmers and progress of all the stakeholders of the sector. The above mentioned objectives of study are achieved through surveying the farmers, statistical data, direct interviews with representatives of government institutions.

Key words: milk production, dairy farmers, dairy industry

Session III
ANIMAL REPRODUCTION

HIGH YIELDING DAIRY COWS: TO PRODUCE OR TO REPRODUCE AND WHAT PRACTITIONERS SHOULD KNOW ABOUT THIS TO HELP THEIR CLIENTS

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ABSTRACT

The present article aims to 'translate' the current – mostly theoretical – knowledge on fertility disorders in modern high yielding dairy cows, towards the actual situation in the stable with a main emphasis on the resumption of the ovarian activity after calving. While some detailed research has recently been done at our department to elucidate the association between a high level of milk production and the reproductive performance of the current dairy cow, the next challenge is to 'translate' this knowledge into practice and to offer possibilities and strategies to minimize the effects of the decrease in fertility. As the negative energy balance and general health status after calving are known to be paramount factors hampering fertility, it is apparent that avoiding both is among the most important preventive measures to be taken. Improvement of the energy status by achieving a high dry matter intake and the provision of optimal and well balanced nutrition during the transition period as well as during early lactation are key goals in this effort. To achieve these goals, we should not only calculate the rations on paper, but should also check in the stable to determine whether the calculated amount is really being consumed by the cows. Furthermore, veterinarians should use their 'clinical eyes' as well as other diagnostic tools to assess the general health status of the cows and to assess at which aspect of the process things are going wrong and need to be adjusted. Besides the control of the negative energy balance and health status, other management factors that need to be maximized include heat detection, cow comfort, insemination technique, time of insemination during estrus and sperm quality. Only if management is on a very high level can high milk production and good fertility be a feasible combination!

Key words: dairy cow, milk production, reproductive disorders, management

HIGHLIGHTS ON ARTIFICIAL INSEMINATION (AI) TECHNOLOGY IN THE PIGS

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Over the past decade, there has been a tremendous increase in the development of field AI services in the majority of countries concerned with pig production. The objective of this paper is to review: (1) the current status of swine AI in the world, (2) significance and limitation of AI with liquid and frozen semen, (3) the biological traits of porcine semen in relation to in-vitro sperm storage, (4) the criteria used for selection of a boar stud as a semen supplier, (5) how to process boar semen for liquid and frozen storage in the commercial settings, (6) how to improve fertility and prolificacy of boar semen and (7) the recent disciplines in evaluation of fresh and stored semen. More than 99% of the inseminations conducted worldwide are made with liquid-stored semen. AI with frozen semen is used only for upgrading the genetic base in a particular country or herd. Determining the initial quality of semen ejaculate coupled with selection of the optimum storage extender has a profound effect on the quality and fertility of AI doses. Different procedures have been used for improving fertility of preserved spermatozoa including colloidal centrifugation of the semen, deep intrauterine insemination and modulation of the uterine defense mechanism after AI. Pre-selecting the sex of offspring is an important technology for the pig industry. Despite the number of the available sex-sorted spermatozoa is too low for the practical use in pork production, laparoscopic insemination into the oviduct is a promising procedure for achieving this purpose.

Key words: boar, semen storage, artificial insemination, fertility

FIRST RESULTS FROM INSEMINATION WITH SEX-SORTED SEMEN IN MACEDONIA

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ABSTRACT

Long time the science was searching for the reliable method for controlling the sex of mammalian offspring. Recently, application of certain modern cellular methodologies has led to development of a flow cytometric system capable of differentiating and separating living X- and Y-chromosome-bearing sperm in amounts suitable for AI and therefore, commercialization of this sexing technology.

The aim of our study was to introduce the bovineAI with sex sorted semen, for the first time in Macedonia.

Insemination was done at two dairy farms in ZKPelagonija, with the sex-semen imported from USA. For the insemination total of 74 heifers (Holstein Friesian breed) were used. All of the heifers were previously synchronized for estrus and timed insemination, with the OvSynch protocol. For the purpose of insemination total of 107 heifers were prepared, but only 87 of them were put in OvSynch protocol, and 74 of them showed clear signs of estrus and were inseminated. Insemination was done into the ipsilateraluterine horn to the ovary where follicle larger than 1.8 cm was detected, by the means of ultrasound examination. Pregnancy was checked by ultrasound on day 30 after the insemination.

Reasonable pregnancy rates have beenachieved with low-dose sexed, cryopreserved sperm. Overall pregnancy rate for both farms was 43,24% (40,54% and 45,95%, respectively for farm 1 and farm 2). The sex of first 5 born calves was as predictedwith the sorted semen.

The results of our study are with the accordance of other authors (Seideland Garner, 2002; Weigel, 2004; Garner and Seidel, 2008; Otava 2010).

In conclusion, as first results from insemination with sex-sorted semen in Macedonia, they are very promising and will have much benefit in dairy sector. Average pregnancy rate is not much lowered as was expected regarding the low dose number of spermatozoa. Due this fact we can recommend insemination

only by experienced technicians within the TAI protocol and ultrasound examinations of the ovaries prior insemination, in order to lower the cost of insemination with the increasing of conception rate.

Key words: sex-sorted semen, cow, artificial insemination

COMPARISON OF MICROBIOLOGICAL AND ENDOSCOPY DIAGNOSTIC OF UTERINE DISEASE IN INFERTILITY OF DAIRY CATTLE

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ABSTRACT

Bacterial and fungal contamination of uterine lumen during pregnancy can persist and after parturition be a key cause of infertility. Dystocia, injuries and retention of fetal membranes are some of main risk factors, casing an ideal open door for infection. Once occur in uterus, infection takes long time of persisting. Such kind of changes which may occur in uterus, dramatically reduces the economic impact. Usually service period is extended and insemination's are repeated. Various methods helps in the diagnosis of infertility, sampling of uterine secretions, detection of changes in macroscopic appearance of the endometrium, etc. Endoscopy is used to determine the amount, nature and localization of secretion and microbiological examinations identifying type of infection. Application of endoscopy is equally important in the diagnosis of manifested and non-manifested endometritis. Postpartum uterine diseases are known to be the leading cases of reproductive inefficiency in dairy cows in veterinary literature. The examination of intrauterine fluid with bacteriological findings with respect to the involution of the uterus shows the nature of the inflammatory process. Polymorphonuclear cells as the predominant finding in intrauterine fluid and their relationship to the findings may indicate the reproductive performance of cows in the postpartum period. Lesions of the cervical mucus, combined with infection causes early embryonic death. Partial endometritis and changes in caruncules are not able to be palpated in routine method of examination. So it is the same with localized metrorraghia and presence of pathological-fermented liquid.

The remains of the fetal membranes around caruncula with necrotic and hemorrhagic content is usually place of infection location. It enables visibility of endometrium. Lysis of adhesion's, removal of localized changes in the endometrium, residual parts of the placenta, the fermented content, diagnosis and treatment of endometritis can be performed by endoscopy. Endoscopic review should be applied when abnormal uterus cannot be detected

by rectal palpation but sterile microbiological sampling is recommended. Other abnormalities in the uterus can be detected by ultrasound or combining these techniques. 30 Holstein cows were used that had an extended service period after calving, which amounted to 100 - 200 days. The lining of uterine horns often hides the identity of the present pathological changes, especially in terms of subclinical endometritis and state carancula. It is known that placentitis is a common reason for abortion or embryonic death . Existing of serious stable infection according to presence of E. Colli and other bacteria, also combined with fungal infection Aspergilus fumigatus, can persist during first pregnancy and latter be a cause of early embryonic death in repeated insemination.

KINETIC PARAMETERS OF CRYOPRESERVED YORKSHIRE BOAR SEMEN

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ABSTRACT

Despite the widespread of use of cryopreserved semen in cattle, its utilization in swine is limited, mostly as a result of the poor cryo-survival of boar spermatozoa. In order to overcome the mentioned problem, most of the work so far was focused on designing a suitable cryo-protocol for boar semen. Ejaculates obtained by 8 different Yorkshire boars, were cryopreserved using 2 different cryopreservation protocols. The samples were assessed by CASA before freezing, and after thawing. The objective of the study was to estimate the difference of the kinetic parameters (VAP, VSL, VCL, ALH, BCF, STR, LIN) of the spermatozoa in liquid semen, and the spermatozoa cryopreserved by two different protocols. The samples taken by glowed hand technique, and were extended to ratio 1:1 to be delivered to the laboratory for CASA evaluation, and further processing. The first group of ejaculates was cryopreserved according the procedure described by Westendorf et al. (1974), modified by Thurnston et al. (1975) (TCP Semen). In the second group, only P1 of SRF was cryopreserved according the procedure by Rodriguez-Martinez and Wallgren (2011) (XCP Semen). The analysis was performed by CASA (HTM IVOS V.12, Hamilton Thorne Research, USA) using the standard set-up for boar semen. The VAP values were 70.61 ± 1.20, 41.35 ± 0.76 and 46.62 ± 1.69 μm/s in liquid, TCP and XCP Semen respectively, with highly significant differences between the liquid and cryopreserved semen ($p < 0,00005$). The levels of VSL were 46.66 ± 0.52; 21.44 ± 0.75 and 30.25 ± 1.48 μm/s in liquid, TCP and XCP Semen respectively, and there was high significance between the all studied groups ($p < 0,00005$). VCL ranged from 162.26 ± 1.25 μm/s in liquid semen, 129.05 ± 1.93 μm/s in TCP semen and 120.63 ± 3.52 μm/s in XCP Semen; and there was high significance between liquid and cryoreserved semen ($p < 0,00005$). The ALH values were 7.74 ± 0.05, 7.73 ± 0.08 and 7.12 ± 0.14 μm in liquid, TCP and XCP Semen respectively, and there was high significance in the differences between the cryopreservation procedures ($p < 0,005$). BCF levels were 30.56 ± 0.20, 29.17 ± 0.30 and 29.12 ± 0.40 Hz in liquid, TCP and XCP Semen respectively, and there was high significance between liquid and TCP Semen ($p < 0,001$).

Key words: boar semen, CASA, cryopreservation, kinetic parameters.

AN ANALYSIS OF SOW'S (RE)PRODUCTIVE PERFORMANCES ASSOCIATED WITH MANAGEMENT PRACTICES IN COMMERCIAL SWINE FARMS IN R. MACEDONIA

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ABSTRACT

Our objectives in this survey were to determinate sow's performances in commercial farms in R. Macedonia, to investigate interrelationships between key measurements for productivity in the herds and to identify significant management factors associated with breeding-herd productivity.

One year (2012) retrospective study of sow's productivity data from large scale (400-1000 sows) and small scale (80-200 sows) commercial farms (n=9) was performed. The questionnaire that enclosed issues concerning general herd's characteristics, preventive measures, hygiene routine, farrowing management, feeding and water supply practices, was prepared before the study. Reproductive data based on mean results was also recorded. All data from questionnaire were obtained by the same author, through interviews with the pig farmers and correspondent veterinarians. Data were statistically analyzed by Statistica 7.0 software.

Great difference regarding preweaning mortality (PWM) 7.09 vs.12.0, average parity (AP) 3.3 vs. 6.0, pigs/weaned/sow/year (PWSY) 19.4 vs. 26.4, litters per mated female per year (LMFY) 2.0 vs.2.32 and non-productive days (NPD) 35.56 vs.69.0 was found between nine farms. Obtained results show significant differences in five production measures between two groups of farms. Small farms had elder sows 5.38±0.48 than large farms 3.76±0.38 ($p<0.05$). Also, small farms had higher litter size 13.77±1.28 vs. 11.73 ± 0.50 ($p<0.05$). Replacement rate was significantly higher ($p<0.05$) in large scale farms 45.85 % ±5.68, compared to 20.00 %±4.08 small farms. Average weight of the pigs at weaning in small farms was higher 8.6 kg±1.10 than 7 kg±0.35 in large farms ($p<0.05$). Mortality rate in sows was higher ($p<0.01$) in large farms (5.79±3.17 vs.1.33±0.26 in small ones). Small farms had also more frequent ($p<0.05$) lactation meals 3.25±0.50 than 2.2±0.45 in large farms. Large farms had more frequently individual than group sow gestation housing system in

comparison to small farms. Split weaning technique was used more frequently by small farms. Lactation duration (LD) of 29.0±2.00 and farrowing interval (FI) of 149.25±1.89 in small farms, were negatively correlated ($p<0.05$) with (LMFY) 2.15±0.10. In both groups of farms NPD was highly negative correlated (46.02±8.83 vs.55.32±9.13) with LMFY ($p<0.01$). LD ($p<0.01$) and FI ($p<0.05$) in small farms was positively correlated with LMFY. Also positive correlation ($p<0.05$) in small farms was found between pigs/born/alive (PBA, 11.98±1.35) and pig weaned/sow (PWS, 10.83±1.08). In large farms positive correlation ($p<0.05$) was found between PWS (9.78±0.61) and (PWSY, 21.9±1.45) as well as between LMFY (2.24±0.06) and PWM (10.05±1.74), respectively. NPD was negatively correlated with PWM in large farms ($p<0.05$).

The study demonstrates significant differences in breeding-herd data in commercial swine farms. Those data were strongly correlated with management practices during pregnancy, lactation and weaning that lead to a conclusion that those practices have significance influence in variation of sow's (re)productivity performances.

Key words: sow, management practices, productive performances

EFFECT OF REDUCED GLUTATHIONE ON KINEMATIC PARAMETERS OF RAM SPERMATOZOA

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ABSTRACT

Motility patterns of spermatozoa are an indicator of their fertilization capabilities during the migration process which takes place in the female genital tract. Reduced glutathione (GSH) has been reported as antioxidant additive in the semen extenders which has positive effect on biological post-thaw quality of the spermatozoa. The aim of this trial was to investigate the effects of GSH additive in the soy-bean based semen extender on the following kinematic parameters of the post-thawed ram spermatozoa: continuous line velocity (VSL), average path velocity (VAP), curvilinear velocity (VCL), amplitude of lateral (ALH), linearity (LIN), straightness (STR) and beat frequency (BCF). VCL and VAP have been reported to have positive correlation with the ability of post-thawed spermatozoa to successfully migrate in sheep cervical mucus. The methodology of this trial included analysis of 48 frozen ejaculates in liquid nitrogen. The ejaculates were previously collected from two rams with artificial vagina method, and were classified in two groups: Group 1 (with addition of GSH, n=24) and Group 2 (without addition of GSH, n=24). Each ram contributed with 12 semen samples in each group, collected on the same date (January - May, 2013). Before freezing they have been diluted with commercial soy-bean extender, and frozen in liquid nitrogen on -196°C degrees in programmable freezer. For the assessment procedure, samples have been thawed at 37°C degrees for 30 seconds in water bath, and additionally diluted with the commercial soy-bean extender until reaching final spermatozoa concentration of 20 million/ml. Kinematic parameters were assessed using CASA system (TOX IVOS, Hamilton Thorne Research). Results showed that only VSL and BCF have considerable statistical significance between Group1 and Group 2 (102.98±15.13 vs. 88.47±20.63, $t=2.77$, $p<0.01$ and 32.01±2.68 vs. 29.47±2.92, $t=3.13$, $p<0.01$, respectively). The summary of the investigation has highlighted these kinematic parameters as indicators that confer the positive effect of GSH as additive in semen extenders. This conclusion opens straightforward methodology for future semen-quality and GSH related scientific investigations, eliminating the necessity of including other kinematic parameters in the calculations.

Key words: ram, spermatozoa, reduced glutathione, kinematic parameters, CASA

PREDICTION OF POSITIVE PREGNANCY DIAGNOSIS IN DAIRY COWS ON DAY 21 AFTER ARTIFICIAL INSEMINATION – ON FARM REPORT

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ABSTRACT

Early and accurate pregnancy diagnosis has great importance in modern dairy farming. The aim of this on farm trial was to investigate the possibility to predict early pregnancy in dairy cows, by scanning the ovaries on day 21 after A.I. using a ultrasound. Total of 32 Holstein Frisian cows from one dairy farm with regular estrus cycles were assessed. The ultrasonic scanning was performed by B-mode "real time" ultrasound scanner equipped with 7.5 MHz rectal linear probe. Detection of CL at least 25 mm in diameter and present follicle/s, with diameters beyond 13 mm determined the **"**positive pregnancy" diagnosis, whilst an absence of functional CL or presence of CL with diameter ≤ 21 mm with concomitant follicle ≥ 15 mm determined the negative pregnancy diagnosis. Re-examination for pregnancy diagnosis was carried out on day 40 after A.I. The statistical analysis has shown high general accuracy of the test (75%), which rendering it to satisfactory level for early pregnancy diagnosis. Maximal accuracy (100 %) was observed in cows diagnosed as non- pregnant. Sensitivity (SE) of the test was 100 % with possibility to predict positive pregnancy (PPV) of 68%. Specificity (SP) of the test was 46.6 %. Previous high recorded embryonic mortality on the examined farm, were considered detrimental for the specificity and positive predicative value of the test. In conclusion, scanning the ovaries on day 21 after AI can be suitable and applicable method on the farm to predict pregnancy in dairy cows without using any laboratory tests.

Key words: ovaries, cows, ultrasound, CL, follicles

Session IV
ANIMAL WELFARE

CAN ANIMAL WELFARE BE MEASURED AUTOMATICALLY?

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ABSTRACT

The EU project Welfare Quality identified a number of important welfare indicators, and animal based welfare indicators are in focus in the current EU welfare strategy. Furthermore, the structural development in dairy farming has led to increased farm size, where each co-worker is responsible for monitoring an increasing number of animals. Thus there is a need for new tools for monitoring and troubleshooting at dairy farms. Welfare is related to the animals' experience of positive and negative emotional states such as fear or pain. However, emotional states cannot be measured directly; therefore measures of physiology, behaviour and pathological conditions etc. are used to access the level of welfare. It is a challenge to identify the indicators that lead to both a reliable assessment of the welfare and at the same time can be measured by methods or technology within limits of expenses such that the systems are beneficial to the farmer. However, during the last years new technology for automatic recording of dairy cow behaviour has become available, and this is a promising tool for assessment of different aspects of animal welfare.

Fulfilment of behavioural needs is an important aspect of animal welfare. A number of studies have shown that the time budget of dairy cows can vary considerably under commercial conditions. Changes in the time budget may reflect adaptation to a specific environment without negative consequences. However, if there are constraints on high priority behaviours, it may have negative consequences for the welfare. There is evidence that lying behaviour has very high priority and that thwarting of lying behaviour can induce both behavioural and physiological stress responses. Results from studies with focus on the need for lying behaviour and consequences of restriction of lying behaviour will be discussed.

Furthermore, the first symptom of a disease is very often a change in behavioural patterns. The most well-known changes in behaviour in response to disease are reduced activity; decreased feed intake and increased time spend lying. Development of devices for automatic recording of behaviour can contribute considerably to on-farm assessment of animal welfare as well as a tool for consultancy, and can be used as documentation for a given standard of animal welfare. Accuracy in recording of different activities of cows will be discussed as well as the value of the recordings of various activities of cows in relation to assessment of the status of the animal.

ANIMAL WELFARE IN SLAUGHTERHAUSES

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ABSTRACT

This paper provides an overview of the scientific and technical literature and different experiences in the field of animal welfare in our country and the world.

This work aimed to analyze the different experiences of the world and in us and through review of regulations in the field of animal welfare during slaughter in Serbia appears to estimate the value of educating consumers and producers of meat.

From the point of view of animal welfare, the paper discusses procedures with animals in slaughterhouses, after receiving in boxes, its keeping, movement and guiding, a pre-slaughter stunning, ending with the slaughter and bleeding. Specifies how to use the equipment, in order to prevent stress of animals.

In addition to the existence of adequate legislation, there must be a clear strategy and training for producers of meat, which are also an important element in the fight for animal welfare. Only due to raising consumer awareness and systematic training of meat producers will be able to reach the European level in the field of animal welfare.

Key words: animal welfare, slaughter conditions, education

USAGE OF TRI-AXIAL ACCELERATION OF THE HIND LEG FOR RECOGNIZING SHEEP BEHAVIOR

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ABSTRACT

Measuring the leg position and its motion could indicate strong distinction between sheep's lying, standing and gait types which could define sheep behavior and their welfare state. Tri-axial accelerometer simultaneously records acceleration and inclination through measurement of an analogue signal in each of its three axes (X, Y and Z) converting it to gravity units on different time logging interval. The objective of this study was to analyze the position and locomotion of the sheep's hind leg using attached accelerometer and to interpret the gathered data from the accelerometer for differentiation of lying, standing and various gait types.

Six sheep (3 rams and 3 ewes) divided in two groups (gender division) were used in this experiment. HOBO Pendant G tri-axial acceleration data logger was attached on the lateral side of the metatarsal region on the left hind leg. The logger was set for measuring acceleration and inclination of x, y and z axis, on fast mode logging interval (0.03sec.). Each sheep and the appropriate left hind leg were recorded with video camera.

For the standing position the acceleration of the x axis was in range 0.8 and 1 g ($\overline{x}$ =0.937±0.035) and the sum vector was from 0.85-1.3g ($\overline{x}$ =0.988±0.006) with the time interval longer than 0.7 seconds. For the lying position the z axis acceleration was considered as a most reliable axis ≈ 1 or -1g depending of the lying side. The single step analysis of walking and running showed distinctive patterns of the x-axis acceleration during stance and swing phase. The frequencies of acceleration values of x-axis and sum vector showed significant differences in the category 0-1g for walking (x-axis, $\overline{x}$ =65.56±8.37 and sum vector, $\overline{x}$ =37.74±5.92), trotting ($\overline{x}$ =35.87±7.07; $\overline{x}$ =16.41±6.16) and running ($\overline{x}$ =22.24±4.86; $\overline{x}$ =4.91±3.03). The value category 3-4g for walking ($\overline{x}$=3.39±1.5; $\overline{x}$ =4.97±2.15), trotting ($\overline{x}$ =9.99±4.15; $\overline{x}$ =16.31±4.95) and running ($\overline{x}$ =19.65±4.05; $\overline{x}$ =30.09±2.19). Average time duration of the steps during walking, trotting and running was 0.47±0.07; 0.31±0.02 and 0.25±0.02 seconds, respectively.

Accelerometers provide detailed real time analysis of the leg position in sheep's natural environment. Defining thresholds based on the frequencies of

acceleration values are the most reliable approach for acceleration analysis. The defined thresholds for sheep can be used for behavioral research and sheep locomotion, as well as a tool for herd and sheep behavior and animal welfare assessment.

Key words: accelerometers, behavior, animal welfare, sheep, gait

WELFARE QUALITY PROTOCOL PROVIDES AN INSIGHTIN THE WELFARE PROBLEMS OF THE LAYING HENS IN REPUBLIC OF MACEDONIA

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ABSTRACT

The Welfare Quality project developed protocols of assessing animal welfare and a standardized way of integrating this information to enable farms and slaughterhouses to be assigned to one of four categories. The welfare of laying hens in Macedonia is a challenge, mainly because all the laying hens are still kept in conventional cages.

The aim of this survey was to use the Welfare Quality protocol in order to access the welfare of laying hens.

The survey was carried out on 15 flocks laying hens. All the flocks were housed in conventional cages. The performed welfare assessment was designed in accordance with the established welfare principles, criteria and measures by the Welfare Quality Project, used twelve define welfare criteria.

The initial data showed that although three of the flocks declared to have enriched cages, the cages were modified in such way so they could house more birds and thus making them no different from conventional cages. Furthermore in all the flocks the available space per hen was well below the regulative minimum. The results showed relatively good scores in the criteria good health, but showed low scores on the criteria good feeding, good housing and appropriate behavior.

The low scores obtained in this survey were due to the lack of interest of the farmers to invest in alternative housing systems, because this will increase the cost of the production thus increasing the cost of the eggs. The Welfare Qualit® protocol is time consuming, but it gives is good ground for assessing the animal welfare of laying hens. The results obtained from this protocol are standardized and can easily provide information which segment needs to be improved in order to prove the welfare of the hens.

Key words: welfare quality, animal welfare, laying hens, conventional cages

POSSIBILITIES OF IR THERMOGRAPHY FOR EARLY DIAGNOSIS OF LAMENESS IN DAIRY CATTLE

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ABSTRACT

Infrared thermography is a non-invasive diagnostic technique that can transform skin surface temperature into a color images. Thermal camera is used to detect emitted infrared radiation that is directly correlated to the blood flow, and convert into the electrical impulses that can be visualized in different colors on a video screen. The ability of infrared thermography is to detect the location and the degree of local inflammation or injury through increased temperature and/or a change of skin surface temperature distribution. Since thermography detects abnormal thermal patterns, way before appearance of the clinical signs, it represents a very useful tool for early detection of orthopedic disease in horses, prediction of changes in udder temperature in dairy cattle, animal welfare, stress detection, inspection of testical function and to detection of inflammation associated with hot-iron and freeze branding in cattle. The aim of the study was to assess the ability of infrared thermography as non-invasive, on-farm tool, for early detection of different aseptic and septic conditions on the foot, especially subclinical laminitis in dairy cattle. Non-contact thermography was performed on the hind claws of total 80 lame and non-lame lactating Holstein-Friesian cows in a tie-stall farm using the hand-held portable thermal camera (Testo 880-3 Thermal imager, Testo AG, Germany). In order to determine the surface temperature of the coronary band (CB) and skin as control area (CA) above the coronary band and the temperature differences (ΔT) between the CB and CA, measuring was performed on the dorsal and palmar surface of the claws. The images were taken in the barn, at the same distance between 1-1.5 m from the subject, after removing any debris and moisture from the dorsal and palmar surface of the claws were free. The results have shown difference in the surface temperature between lateral and medial claws. Significantly elevated temperature was detected in cows with detected hyperemia of the coronary band and the surrounding skin, associated with acute laminitis. Infrared thermography have a potential as a non-invasive diagnostic tool for early detection of lameness

in dairy cattle, however, further investigations are needed for validation of the method and use in field conditions.

Key words: infrared thermography, laminitis, dairy cow, coronary band

FIN DAMAGE OF FARMED RAINBOW TROUT IN REPUBLIC OF MACEDONIA

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ABSTRACT

The aims of this study were to determine the prevalence of fin damage in farmed rainbow trout and to see whether the level of damage differed between different fish categories and farms. The study was field based and was conducted in two parts. The first part was a survey with the owner of the farm or the responsible technologist and covered the major technological operations in the farming process. The second part included the fin damage analysis and clinical description of the damaged fins. Fins were analyzed in two categories of fish (weight below 30g (min. 5g) and over 100g (max. 250g)). Thirthy fish per category were randomly selected, netted and each rayed fin was assessed and photographed (total of 5880 fins were analyzed in 840 fish from seven rainbow trout farms). The prevalence of fin damage was 100% and there was a large range in the level of damage which was mainly characterized by surface abrasions. Worst affected fins in both fish categories were dorsal and pectoral fins. Fin damage was present in lesser degree in the smaller categories, but there was fin damage in the smallest fish examined. Pattern of damage was Dorsal>Pectoral >Abdominal>Anal>Tail fin. Differences in fin damage in all surveyed farms indicate that some factor or group of factors specific to each farm influence the extent of damage. Future research should identify and explore the impact of the factors affecting fin damage and propose management practices that can minimise the level of fin damage.

Key words: rainbow trout, fin damage, fish welfare, welfare indicator

EVALUATION OF THE ANTIPARASITIC EFFECT OF THE BUTTON MUSHROOM *AGARICUS BISPORUS*

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ABSTRACT

Introduction

Farm animal welfare is the physical and psychological well-being of animals. The owner shall take all necessary measures to ensure the protection of animals and to ensure that animals are not caused unnecessary pain, injury, suffering and diseases. Gastrointestinal parasitism has been classified as a major health and welfare problem for ruminants. Parasitism, especially by helminth parasites, impairs health by causing inappetance, diarrhoea, anaemia and, in severe cases, death. In addition to compromising health and welfare, parasitism impairs productivity and results in poor growth and reproduction. Nutritional content of an animal feed is influenced not only by nutrient content but also by many other aspects such as, feed presentation, hygiene, digestibility, and effect on intestinal health; through coccidiostatic or histomonostatic effect. Mushrooms are known to have effective substances for antifungal, antiinflammatory, antiviral, antibacterial, hepatoprotective, antidiabetic, hypolipedemic, antithrombotic and hypotensive activities. In accordance with Article 17 of Regulation (EC) No 1831/2003 on additives for use in animal nutrition, the Commission has established a Community Register of feed additive, according to which the recommended natural animal feed supplement is Agaricus bisporus, and its extract CoE 543. Affirmed nutritional and immuno-modulatory effects of supplement from the button mushroom, *Agaricus bisporus* have indicated justification of blending that fungi in animal food as a natural alternative to

growth promotion antibiotics. The purpose of this research was to evaluate the antiparasitic effects of the button mushroom *Agaricus bisporus*.

Materials and Methods

One-year-old sheep, breed Lika Pramenka were used in this experiment.

The dietary groups were controled, one group feed by basal diet and the other group with diets containing 1.5% dry supplement of *Agaricus bisporus*, during the 6 weeks of winter feeding period. Upon completion of the experime nt, coprological examination of the faeces of sheep was done.

Results

The number of strongiloides eggs and oocysts Eimeria sp., were significantly decreased in sheep treated with *Agaricus bisporus* supplement compared to pair-fed controls.

Conclusions

The data provided by this study illustrate the antiparasitic potential of button mushroom *Agaricus bisporus* and supports the recommendation of *Agaricus bisporus* as a dietary component for farm animals.

Key words: welfare, *Agaricus bisporus,* sheep breed Lika Pramenka

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POSTER PRESENTATIONS

P1 ALTERATIONS IN SOME GLOMERULAR MARKERS IN DOGS WITH PYOMETRA

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ABSTRACT

Introduction

Impairment of renal function consequently to bacterial infection is of significant importance to human and veterinary medicine. The main post infection cause of renal dysfunction in dogs is the bacterial uterine infection in sexually mature nulliparous bitches. The aim of the present study was to investigate the diagnostic value of urine CRP/Cr and ALB/Cr ratios as glomerular markers in dogs with pyometra.

Materials and Methods

The study was performed on 18 dogs with pyometra and positive result for *E. coli* from the intrauterine bacterial culture, without any concomitant diseases (group P, n=18). Sixteen clinically healthy dogs were used as a control group (group H, n=16). Twelve dogs from the initially diagnoses 18 pyometra patients (experimental group P^{1}, n=12) were submitted to another physical examination, blood and urine analysis six months after the ovariohysterectomy (experimental group P^{11}, n=12). The other 6 patients by that time were ineligible due to fatal outcome. Urine samples were obtained by ultrasound-guided urinary bladder puncture. The analysis was performed within 30 min after sampling on an automated biochemical analyzer and commercial kits for determination of urinary creatinine (uCr) and urinary albumin (uALB. The analysis of CRP was performed at the University of Milan, Italy, on a microplate spectrophotometric reader with species-specific commercial ELISA kits.

Results

Although serum creatinine and urea concentrations in all groups were within the reference range for the species, the levels of uAlb and uCRP, as well as uAlb/Cr and uCRP/Cr ratios were considerably higher in dogs with pyometra than in healthy controls. A statistically significant difference was established for uCRP/Cr ($p<0.05$) and uAlb/Cr ($p<0.001$) in experimental groups P and P^{1}. In dogs from group P^{11} the values returned to normal and were similar to those of control dogs.

Conclusion

The diagnostic value of glomerular (uALB/Cr, uCRP/Cr etc.) markers, as compared to routine tests (sCr, BUN) was good with regard to the extent and localization of the renal damage. The glomerular markers utilized in the present study, are promising non-invasive tests in the diagnostics of renal damage in dogs with pyometra.

Key words: glomerular marker, dog, pyometra

P2 THE EFFECT OF ACUTE INFLAMMATION ON TOTAL ALKALINE PHOSPHATASE ACTIVITY IN DOGS

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ABSTRACT

Introduction

The main purpose of this study was to investigate the effect of acute inflammation on total alkaline phosphatase (ALP) activity in dogs. In this study total ALP activity were determined in dogs with experimentally induced acute inflammation in order to determine their potential value in this condition.

Materials and Methods

ALP concentrations were determined in plasmas from 9 mongrel male dogs (in experimental group) and 6 mongrel male dogs (in control group) at the age of 2 years and body weight 12-15 kg. The inflammation was reproduced by inoculation of 2 ml turpentine oil subcutaneously in lumbar region and same quantity saline in control dogs. Blood samples were collected into heparinized tubes before inoculation then at hours 6, 24, 48, 72 and on days 7, 14, 21. The total ALP concentrations were determined with commercial kits (Human-GmbH, Germany) on an automatic biochemical analyzer (BS-3000 P, Sinnowa, LTD Nanjing China). The statistical analysis of the data was performed using one way analysis of variance (ANOVA), Statistica v.6.1 (StatSoft Inc., 2002).

Results

In this experiment ALP activity began to increase compared to baselines at 72th h after injection and remained so high up to on day 14, although they were within the normal ranges. *S*tatistically significant difference was not found between the groups as well as within them.

Conclusion

In conclusion we can say that the total ALP activity was not significantly affected in dogs with experimentally induced acute inflammation.

Key words: alkaline phosphatase activity, acute inflammation, dogs

P3 COMPARISON OF THE EFFECTS OF SPONTANEOUS AND MECHANICAL VENTILATION ON BLOOD GASES DURING GENERAL ANESTHESIA IN DOGS

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ABSTRACT

Spontaneous ventilation during general anaesthesia leads to respiratory depression and atelectasis. Mechanical ventilation increases tidal volume and eliminates atelectasis. The study material consisted of a total of 20 dogs of different breed, age and gender. Dogs were divided into two groups, consisted of 10 dogs. The first group was established as the spontaneous ventilation (SV) group, while the second group was the mechanical ventilation (MV) group. For induction of anaesthesia, propofol was administered to both groups via intravenous injection at a dose of 6 mg/kg. Blood samples were collected from all dogs in 5 minutes after propofol administration. This period was determined as Minute 0 (T0). In both groups, inhalation anaesthesia was continued with isoflurane. Venous blood samples were collected from dogs in the SV and MV groups at 15 (T15), 30 (T30) and 60 (T60) minutes. Heart rate, respiratory rate, SpO2, body temperature and blood gases were monitored. Statistical evaluation of the study was carried out using the Repeated Measures Analysis of Variance method. The results obtained showed that there was no statistically significant difference between the SV and MV groups regarding the examined parameters. However, in the assessments within the group, results obtained from the dogs in the MV group were more reliable from the point of view of the patients remaining stable through out anaesthesia.

Key words: spontaneous ventilation, mechanical ventilation, blood gases, dog

P4 BACTERIAL SPECIES ISOLATED FROM CATS WITH LOWER URINARY TRACT INFECTION AND THEIR SUSCEPTIBILITIES TO CEFOVECIN

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ABSTRACT

The aim of this study was to determine the clinical findings, the bacterial species and in vitro activity of cefovecin against to the isolates from cats with one or more clinical signs (stranguria, haematuria pollakiuria, inappropriate urination, excessive licking of the genital area and frequent and/or prolonged attempts to urinate), which are suggestive of lower urinary tract infection. After physical examination of cats, urine samples were obtained via ultrasound-guided cystocentesis. The urine samples that had ≥ 5-6 leucocytes were cultured using standard bacteriologic techniques. The isolates were identified by conventional microbiological methods and tested for in vitro susceptibility by Kirby-Bauer disc diffusion method according to Clinical Laboratory Standards Institute. Bacterial growth was observed in 16 of 61 urine samples from the cats with ≥ 5-6 leucocytes. The following pathogens were isolated: *Escherichia* coli (n=4), Coagulase negative *Staphylococcus* spp. (n=3), *Enterococcus faecalis* (n=2), *S. intermedius* (n=2), *S. aureus* (n=1), *S. epidermidis* (n=1), *Corynebacterium renale* (n=1), *Enterococcus avium* (n=1), *Enterobacter cloacea* (n=1). Results of the antibacterial susceptibility tests showed that 81.25% of the isolates were susceptible to cefovecin.

Key words: cat, urine, urine culture, antimicrobial susceptibility, cefovecin

P5

BREED-RELATED DIFFERENCES IN THE LYMPHOCYTE TRANSFORMATION IN RAMS

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ABSTRACT

The development of cell-mediated immunity in rams was analyzed by the breed-dependent patterns of the responsiveness of peripheral blood lymphocytes to common mitogens (phytohemagglutinin, PHA; concanavalin A, ConA; poke-weed mitogen, PWM), in a mixed lymphocyte culture (MLC), in order to establish the basic immune parameters in clinically normal rams. Investigations were carried out on 27 rams from the breeds Karakachanian and Copper Red Shoumen. It was established that there are close differences between proliferative activities of lymphocytes of the two groups of animals. The immune parameters obtained for the two local sheep breeds may serve as a basis for further research on lamb systemic immune responsiveness regarding differences among breeds.

Key words: sheep, indigenous breeds

P6 SEROLOGICAL RESPONSE TO ACTINOBACILLUS PLEUROPNEUMONIAE IN GILTS AND SOWS ORIGINATED FROM FIVE COMMERCIAL FARMS

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ABSTRACT

Introduction

Porcine pleuropneumonia caused by *Actinobacillus pleuropneumoniae* is one of the most important respiratory disease of pigs and is widely spread among pig-keeping countries.

The clinical course of the disease can vary widely, ranging from the acute forms with severe clinical signs and a high mortality to the more chronic forms with few or even without any clinical symptoms.

There are two biotypes of *A.pleuropneumoniae* differentiated on the basis of their requirement for NAD.There are 13 serotypes of biotype I (1-12, 15) and 2 serotypes of biotype II (13,14), based on surface polysaccharide antigens. The four major RTX toxins ApxI, ApxII, ApxIII and ApxIV are secreted by the different serotypes in various combinations.

Materials and Methods

For *A.pleuropneumoniae* monitoring purposes, the detection of ApxIV antibodies in the serum is currently the most frequently used serological method. That is why we determined as the objectives of our work: Investigations of gilts and sows blood sera for the presence of antibodies against *A. pleuropneumoniae* using the iELISA test. For the investigations, samples were taken of the blood of gilts and sows originating from 5 swine farms. Investigations were carried out using the method of indirect ELISA with the following diagnostic kits: Chekit APP-Apx IV: *A. pleuropneumoniae* (App) antibody test Kit.

Results

A total of 465 blood sera samples of gilts (144) and sows(321) were examined. Antibodies against *A. pleuropneumoniae* were detected in 105 (22,58%) sera. Antibodies were present in 22 (15,27%) sera of gilts and in 83 (25,58%) sera of sows. Percentage of positive sera differed among the farms, ranging in gilts from 0-23,52 % and in sows 6-36,20%.

Conclusion

The results of the investigations have shown that infection with *A.*

pleuropneumoniae is present in sows on all 6 examined farms and on 5 farms in gilts. The successful control of *A. pleuropneumoniae* depends on the efficient prevention of the transfer of the infective agent, both between the farms and also between certain categories of animals on the same farm. Good results can be achieved using the strict implementation of reliable serological methods.

Key words: A.pleuropneumoniae, gilt, sow, antibodies, ELISA

P7 USE OF ULTRASONOGRAPHY IN TENDONS INJURY IN HORSE – INITIAL EXPERIENCE

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ABSTRACT

Injuries of tendons in horses, especially on the palmar region are common. Ultrasonography has been used for diagnosis over 25 years. Since then, technological advance of this imaging modality are more available in practice. The key issue for the clinician is to produce images of the highest quality to maximize diagnostic information obtained. It is now possible to identify subtle injuries and to image parts of the horse that before now could not be seen. Tendon partial injury is characterized by collagen fiber damage which is hypoehogenic on the ultrasound images. Once the injury is diagnosed, recheck ultrasound (usually at 60 days intervals) allow us to evaluate the healing and strength of the injury tendon or ligament. For this purpose controlled exercise, is essential for optimum healing of tendons and ligaments.

Key words: ultrasonography, horse, tendon

P8 SPLEEN HEMANGIOMA IN DOGS: CASE STUDY

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ABSTRACT

Hemangioma is a benign tumor of vascular endothelial origin. Hemangioma can occur in a different body parts, including skin, liver, spleen, kidneys, bone, tongue and heart. In this case, we focus on the appearance of hemangiomas changes in spleen in 12,5 old female French bulldog. This condition is very common at this age, the oldest animals, but the breed French bulldog is not prone for hemangioma of spleen. The first examination was done on 12.03.2013 year, when it was appearance of ascites. One of the suspect diagnose was sinus arrest, because the electrocardiography was changed with the progressive decrease of RR interval at lead II. The therapy was verapamil 2 mg/kg b.w., q-12h p.o., and furosemide-2 mg/kg b.w. q-12h.,p.o. One week after the therapy, ascites appeared once again. The punction liquid from the ascites was reddish. The microscopic examination has shown the presence of the red blood cells in different shapes and no appearances of tumorous cells. The routine complete blood count (CBC) panel was submitted by and at the Veterinary Center. CBC was performed on hematology counter BC 2800 Vet Mindry. First result was obtained on 28.03.2013. At 44 x 10 ^{9}/L, the number of thrombocytes was progressively decreased (reference interval (RI): 117-460 x 10^{9}/L). All other CBC and biochemistry parameters were unremarkable. Two more CBC panels were made. The second examination was performed on 01.04.2013, from which it was shown that the number of thrombocytes was additionally lower, 33 x 10^{9}/L (RI: 117-460 x 10^{9}/L). In addition, the third examination performed on 04.04.2013, showed that the number of thrombocytes was in low increase, 44 x 10 ^{9}/L (RI: 117-460 x 10^{9}/L). The team from the Veterinary Center made an ultrasound examination and there was a suspect of tumor near spleen. On insistence of the owner, with explanation of poor health of the patience, the team has agreed to perform a surgery. The surgical intervention lasted approximately for 1h and 10 min. The anesthesia was made with isoflurane and oxygen support. Incision was from linea alba till epigastrium. But unfortunately, till lasting operation led to the outcome, who was exitus letalis of the patience.

Percentage of occurrence of tumors in small animal practice is increasing, especially in animals over 10 years age. Although hemangioma is rare in French Bulldog, above mentioned report case, suggesting that it is not excluded in this breed. Therefore the need for regular routine controls in older individuals is an opportunity for early detection of tumor changes.

Key words: hemangioma, spleen, French bulldog, thrombocytes

P9 GRANULOMATOUS MENINGOENCEPHALOMYELITIS IN 8 MONTH MALE SAMOYED

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ABSTRACT

Granulomatous meningoencephalomyelitis (GME) is an idiopathic disease or condition of the central nervous system in dogs. There are three forms of GME: ocular, disseminated and focal. The etiology of disease is still unknown. The clinical signs of GME depend on which part of the brain is involved. The mean age for this disease range from 6 months to 8 years, but mostly between 4 and 6 years. The diagnosis is made based off the signalment and history, clinical signs, neurological examination findings, examining the cerebro-spinal fluid (CSF), complete blood count (CBC), biochemistry profile, radiographs, computed tomography or magnetic resonance. The treatment is based on the use of corticosteroids and antineoplastic agent, but in some idiopathic causes the use of antibiotics in required. The prognosis with this disease is poor, and the survival time ranges between 14 and to more than 1000 days. In this case is presented a 6 month male Samoyed, presented with acute ocular and post ocular disseminated granulomatous meningoencephalomyelitis, presented with dilated and unresponsive pupils, ataxia, circling, seizing, changed mental and behavior status, paresis and neck pain. The diagnosis was made based on the history, examination, CBC, biochemical profile and examination of the CF

Key words: granulomatous meningoencephalomyelitis, GME, encephalitis of unknown etiology, meningoencephalitis in dogs

P10 HISTOPATHOLOGICAL ALTERATIONS ON THE GILLS OF VARDAR CHUB (*SQUALIUS VARDARENSIS*) FROM THE MINING IMPACTED RIVERS IN THE NORTH-EASTERN MACEDONIA

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ABSTRACT

In spring 2012 (May/June), the effect of water contamination on bioindicator organism, Vardar chub (*Squalius vardarensis*), was compared for three rivers, two severely impacted by active Pb-Zn mines Zletovo and Toranica (Zletovska and Kriva River, respectively) and one moderately contaminated by agricultural waste (Bregalnica). The chemical analyses of the river water revealed increased concentrations of sulphates, chlorides, several macro (Ca, K, Mg, Na) and trace elements (Cd, Co, Cs, Cu, Li, Mn, Ni, Rb, Sn, Sr, Tl, and Zn) in the Zletovska River, whereas in the Kriva River prominent increase was observed for Cd and Pb. Since the exposure to chemicals could induce lesions and alterations in the fish organs, histological examination of the chub gills was applied as a tool to assess the effect of water contamination on chub health. The gills of five chub of similar size were examined for each river. Tissue samples were fixed in 4% formaldehyde, and processed using a standard histological technique: dehydration in the ethanol series, embedding in paraffin, and serially sectioning at 5 μm. Sections were stained with haematoxylin and eosin. In all fish, circulatory disturbances, regressive and progressive changes were found, whereas signs of inflammation and neoplasm were not observed. The frequency of the occurrence of these changes was comparable in all three rivers, but the severity of the changes differed. Based on the scores calculated

according to Bernet *et al*. (1999)*, the histopathological changes on the chub gills were the most severe at the Kriva River, whereas the mildest changes were observed at Bregalnica. The characteristic alteration on the gills of the chub from the Bregalnica River was hyperplasia of the gill epithelia revealed in a form of a fusion of the tips of the secondary lamellae, as well as a fusion of several secondary lamellae. More severe lesions were observed in the gills of the chub from two mining impacted rivers. Typical lesions on the Zletovica River included swelling and/or vacuolization of epithelial cells, congestion and oedema, usually on the tips of secondary lamellae. Acute necrosis of epithelium and calcified cartilage oedema was also seen. On the Kriva River, severe gill damage was found, such as severe degeneration and necrosis, exfoliation and delaminating of respiratory epithelium. Extreme telangiectasia was also found. Based on our results, the histopathological alterations on the chub gills could be used as a reliable, although nonspecific, indicator of the degree of water contamination.

Key words: active mines, gills, histopathology, river, Vardar chub

*Bernet, D., Schmidt, H., Meier, W., Burkhardt-Holm, P., Wahli, T. 1999. Histopathology in fish: proposal for a protocol to assess aquatic pollution. *Journal of Fish Diseases* **22**:25-34.

P11

THE ROLE OF FISH INTESTINAL PARASITES (ACANTHOCEPHALANS) IN METAL EXPOSURE ASSESSMENT AND PROTECTION OF FISH AGAINST METAL ACCUMULATION

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ABSTRACT

Metal contamination in aquatic systems is a critical environmental issue due to metal uptake, accumulation and possible toxicity in aquatic organisms. Although there are common and well established indicator organisms and tissues, as well as biomarkers of metal exposure, we are still in need of reliable and sensitive bioindicators to monitor metal pollution in the environment. In the past decades it was shown that fish intestinal parasites, acanthocephalans, can accumulate metals at concentrations that are orders of magnitude higher than those in the commonly used indicator organisms like fishes, bivalves or crustaceans. Accordingly, attempts have been made to utilize acanthocephalans as biological indicators of metal exposure in environmental risk assessment studies.

Parasites can cause mechanical (fusion of gill lamellae, tissue replacement), physiological (cell proliferation, immunomodulation, altered growth) and reproductive damage in fishes. Consequently, fishes under the stress of their parasite load are weakened and become more susceptible to other environmental stressors including contaminants. On the other hand, it was reported that parasites might alter metal uptake and accumulation in fish due to high metal accumulation capacity, resulting in reduced metal levels in tissues of infected compared to uninfected hosts.

In the present study selected essential (Cu, Fe, Mn, Zn) and non-essential (Ag, Cd, Pb) trace metals were analysed in acanthocephalan *Pomphorhynchus laevis* and gastrointestinal tissue of European chub (*Squalius cephalus* L.) from the Sava River in Croatia. The sampling was performed during April/May (chub spawning period) and September (post-spawning period) in order to assess if acanthocephalans can reflect seasonal variability in metal levels of their host. The mean intensity of infection with *P. laevis* was higher in April/May (4.2) than in September (3.0). Although all measured metal levels in chub gastrointestinal tissue were higher during chub spawning, such seasonal

variability was not reflected in *P. laevis*. The bioconcentration factors (the ratio of metal concentration in the parasites to that in host gastrointestinal tissue), which represent a measure of duration of environmental exposure, were also comparable between seasons and indicated low metal exposure of chubs in the Sava River. In both seasons, Cu and Cd levels were higher in uninfected than chub infected with *P. laevis*, indicating possible protective role of acanthocephalans against metal accumulation in fish.

Key words: european chub, Pomphorhynchus laevis, gastrointestinal tissue, essential/non-essential metals, spawning/postspawning period

P12 *AEROMONAS SALMONICIDA* INFECTIONS IN SALMONIDS

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ABSTRACT

Management of furunculosis remains a considerable clinical challenge, in part due to their ability to cause disease in the different salmonids, both in the culture systems and in the wild environment. *Aeromonas salmonicida* subsp. *salmonicida* is a rapidly spreading pathogen associated predominantly with skin infections in the salmonids with high mortality and the disease can cause major economical losses in the hatcheries. These characteristics provide a challenge to clinicians for the fast and successful diagnosis and therapy.

Here we report the isolation of *A. salmonicida* from different salmonids and evaluating identification methods and antimicrobial sensitivity of these isolates.

A. salmonicida isolates were isolated and characterized based on their morphology and host range, and evaluated for their antimicrobial sensitivity. Isolates were from Arctic charr (*Salvelinus alpinus*) and brown trout (*Salmo trutta*) obtained on the Tryptic Soy Agar plates (BD-BBL) after incubation for 24-48h at 22ºC. Phenotypical characteristics, Gram staining and oxidase production were determined for isolates and further characterization was carried out by using the API kits (Bio Mérieux). Isolates were also analyzed using MALDI TOF (Bruker) mass spestrometry with confirmation of isolated bacteria by PCR. The antimicrobial susceptibility of the isolated strains was determined with Kirby-Bauer disk diffusion method on Difco™ Mueller Hinton II agar (Becton, Dickinson and Company) using the discs obtained from the same manufacturer. The following antimicrobial discs were used in antibiogram: ampicillin (AM_{10}), chloramphenicol (C_{30}), enrofloxacin (ENO_5) erythromycin (E_{15}), flumequine (AR_{30}), nitrofurantion (F/M_{300}), norfloxacin (NOR_{10}), novobiocin (NB_5), oxytetracycline (T_{30}), penicillin (P_{10}), piperacillin (PIP_{100}), sulfamethoxazole/trimethoprim (SXT), tetracycline (TE_{30}) and trimethoprim (TMP_5).

Only one type of colony was recovered in pure culture on Tryptic soy agar media from all positive organs from examined fish in open waters and culture systems i.e. brown trout and Arctic charr. Gram staining revealed the presence of

gram-negative bacilli. The colony characteristic was brown diffusible pigment on the agar. The identification of bacterial isolates by API tests, MALDI TOF mass spectrometry and PCR analysis confirmed the presence of *A. salmonicida*. Resistance was recorded for ampicillin, enrofloxacin, erythromycin, norfloxacin, novobiocin, penicillin, piperacillin, tetracycline and trimethoprim

The results obtained in this study suggest that *A. salmonicida* exist in the fish from open waters as well as in the culture systems of salmonids. That fact might increase the possibilities of transfer of this pathogenic bacterium.

Key words: Aeromonas salmonicida, arctic charr, brown trout, MALDI TOF, PCR

P13 ACCUMULATION OF Pb AND Cd IN THE GILLS AND LIVER OF VARDAR CHUB (*SQUALIUS VARDARENSIS*) FROM THE MINING IMPACTED RIVERS IN THE NORTH-EASTERN MACEDONIA

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ABSTRACT

Active Pb-Zn mines in the north-eastern Macedonia, Zletovo and Toranica, are known to cause degradation of the environmental status of the regional rivers. As a consequence of the leaching from the mine tailings or direct input of the mine waste into the rivers, increased concentrations of several metals were found in the Zletovska and Kriva rivers compared to the reference area, downstream section of the Bregalnica River. Dissolved Cd was found increased in both Zletovska and Kriva rivers (0.27 μg L^{-1}), whereas dissolved Pb was found increased only in the Kriva River (1.85 μg L^{-1}). Both Pb and Cd are regarded as the priority toxic substances by the European Water Framework Directive, but only Cd has exceeded the European recommendations. The aim of this study was to assess if increased Pb and Cd concentrations in the river water have resulted with the increased bioaccumulation of these metals in two selected organs (gills and liver) of bioindicator organism, Vardar chub (*Squalius vardarensis*). In May/June 2012, 30 chub specimens were sampled in each river, and their gills and livers were dissected. Metal concentrations were measured in the soluble tissue fractions, i.e. in supernatants obtained after tissue homogenization and centrifugation at 50,000×g, by high resolution inductively coupled plasma mass spectrometry. The comparison of metal concentrations in

two organs indicated more pronounced Pb accumulation in the gills than liver, whereas higher accumulation in the liver was observed for Cd. Nevertheless, both organs revealed the same spatial pattern of metal bioaccumulation, which reflected metal contamination of the river water. Cytosolic Pb concentrations were the highest in the gills and the liver of the chub from the Kriva River, whereas cytosolic Cd concentrations were higher in the gills and the liver of the chub from both Kriva and Zletovska rivers compared to the least contaminated Bregalnica River. Furthermore, the association of metal bioaccumulation with several biometric parameters was analyzed, but no clear pattern was observed, except for the gender related differences. At higher level of metal exposure, such as observed in the Kriva and the Zletovska rivers, Pb and Cd were significantly higher in the liver of males than females, whereas opposite trend was observed for Cd in the gills. Our results pointed to possible use of cytosolic Pb and Cd concentrations in both gills and liver of Vardar chub as indicators of metal exposure in the river water.

Key words: active mines, cadmium, gills, lead, Vardar chub

P14 PARASITOFAUNA OF FARMED FISH: IMPACT ON HUMAN HEALTH

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ABSTRACT

Aquaculture is nowadays a fast growing food sector. However, along with the fast development of aquaculture in Mediterranean there is a great concern of the emerging fish diseases directly affecting food safety. European sea bass (*Dicentrarchus labrax*) and gilthead sea bream (*Sparus aurata*) are two most important fish species in Croatian aquaculture and together with bluefin tuna (*Thunnus thynnus*) farming constitute significant part of this industry in the Croatia. For the purpose of this study three sea bass and one tuna farm were investigated, all located in the East Adriatic Sea. Sampling sites were chosen in order to cover different regions: south, central and north Adriatic. Sampling was done during spring and autumn in the period from 2005 to 2010. A total of 333 randomly sampled sea bass and 10 tunas were analysed. Samples for ectoparasite analyses were collected from skin and gills, and were microscopically analysed *in situ*. Further macroscopic investigation of internal organs during necropsy was made for detection of the presence of endoparasites. A subsequent molecular analysis of sampled tuna parasites was made in the laboratory using PCR method. Five parasitic species have been found on sea bass and two on tuna. Although infection with *Diplectanum* sp. had high prevalence, economical loss wasn't detected. Obtained results suggested difference between investigated sampling seasons. In addition, bacteriological and virological analyses of fish were also done together with physicochemical analysis of the collected seawater. Detected parasites could present threat for aquaculture, by inducing mortality or suppressing fish growth they lead to the great economical losses while on the other side they pose direct risk to the human health. Results of this investigation, together with previous knowledge, should be the basis for future monitoring of sea bass and tuna farm impacts elsewhere.

Key words: aquaculture, parasites, sea bass, tuna

P15 FAST SIMPLE CHROMATOGRAPHIC METHOD FOR DETERMINATION OF B-COMPLEX VITAMINS IN VETERINARY FOOD PREMIXES

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ABSTRACT

Introduction

We have performed extended research for optimal chromatographic method, considering mobile phase content, DAD signal selecting, buffer characteristics and choice for most suitable column, for successful and satisfying separation of at least 6 vitamins from the group of B-complex mixtures and vitamin C as eventual additional constituent in animal food premixes. The main aim of our analytical research was to create, develop and validate fast, simple and accurate HPLC method for simultaneous determination of 7 hydro-soluble vitamins in premixes for veterinary food supplements. Main idea was creating an isocratic elution chromatographic method with measuring UV signal generated by PDA-detector.

Materials and Methods

All chemicals used, acetonitrile, methanol, KH_2PO_4, diethylamine, were PhEur grade, purchased from Merck Darmstadt, Germany. The reversed phase columns C8 and C18 used for testing were products of Merck, and Supelco, USA. The chromatographic system was Varian ternary gradient ProStar model with DAD 330, controlled by Varian Star version 6.2. The vitamin standards substances were purchased by BASF, Germany.

Results

The main target was development of strictly isocratic separation of all vitamins, without use of an ion-pair reagents, high temperatures, high flow rates, with generating high selectivity, sensitivity, reproducibility and accuracy of the quantification. Gradient elution was avoided as time consuming procedure, especially with long 250 mm chromatographic columns, which limits large number of analysis per day and method validation at all.

We tested 7 different HPLC reversed phase columns, C8 and C18 with different chemical and physical characteristics, The best choice in our set of tested column was LiChrospher RP Select B 250 x 4mm, with 5um particles, used with flow rate 1 ml/min, mobile phase: methanol, acetonitrile and phosphate

buffer with pH=2.7, and signal selection with DAD at 205nm and 277nm with different reasons, even method can operate accurately and successfully with UV monitoring at 205nm only, with run time about 12 minutes or less.

Conclusions

Baseline separation was achieved for 7 vitamins, which make this method very applicative for their quantification in many different vitamin premixes, formulations and any type of food supplement or pharmaceutical dosage forms in veterinary and human medicine. The method can be easily redesigned, readapted for increasing separation factor, sensitivity, selectivity and applicability of it for additional component in premixes. The method can also be used for bromatological studies of hydro-soluble vitamins, their qualification and quantification in different matrixes.

P16 OCURENCE OF PATULIN IN APPLE-BASED JUICES AND CONCENTRATES

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ABSTRACT

Introduction

Patulin is a toxic secondary metabolite produced by a wide range of fungal species of the *Penicillium, Aspergillus* and *Byssochlamys* species growing on fruit, including apples, pears, grapes and other fruit. The principal risk arises when the unfit fruit is used for the production of juices and other processed fruit. Apples and apple products are excellent substrates for *Penicillium expansum*, the casual agent of "blue mould rot", to produce the patulin. According to the International Agency for Research on Cancer (IARC) it is classified in Group 3, and therefore the European Commission (EC 1881/2006) has set the maximum permitted level (MRL) of patulin for apple juice and reconstituted apple juice concentrate at 50 μg/kg.

Materials and Methods

Eighteen commercially available apple juice and twelve concentrated apple juices have been purchased from the market, or supplied from local producers and importers. The samples were stored in refrigerator at +4 ^{o}C prior to analysis. Sample preparation has been performed utilizing molecularly imprinted - solid-phase extraction (MIP-SPE) polymer cartridges, a product by R-Biopharm (Darmstadt, Germany). For detection and quantification a High-Performance Liquid chromatography (HPLC) with Diode Array Detector (DAD) at 276 nm has been applied. Gradient separation was carried out on reverse-phase C18 analytical column (GL-Science, Torrance, CA, USA), with 1 ml/min flow, in duration of 30 minutes.

Results

Applying the chromatographic conditions described above, a good separation of the patulin peak with no apparent interferences has been achieved. Linearity has been established from 10 μg/L to 500 μg/L, with a correlation coefficient (R^2) of 0.9998, and average precision less than 1 %. The estimated limits of detection (LOD) and quantification (LOQ) were 1.8 μg/L and 5.3 μg/L, respectively. Method accuracy has been tested at 25 μg/kg and 50 μg/kg, and

the determined recoveries were > 80 %. 23.3 % of the samples being tested contained patulin less than the LOQ of the applied method. Patulin has been detected and confirmed in 23 analysis, comparing the ultraviolet spectra of standards and test samples. Concentration levels in the positive samples ranged from15.0 to 87.8 μg/kg; 6.7 % of the tested samples have been confirmed to be non-compliant.

Conclusion

The performances of the HPLC-DAD method have confirmed its fitness as precise and accurate analytical technique for detection and quantification of patulin in apple juices and concentrates. The performed analysis revealed patulin's presence in over 76 % of the tested samples.

Key words: patulin, apple juice, molecuraly imprinted polymers, HPLC-DAD

P17 SPECTROPHOTOMETRIC DETERMINATION OF TRIMETHOPRIM WITH BROMOCRESOL GREEN IN DIFFERENT COMMERCIAL FORMULATIONS

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ABSTRACT

Introduction

Trimethopim is a bacteriostatic antibiotic known as dihidrofolate reductase inhibitor. Combinations of a sulfamethoxazole with trimethoprim in a fixed ratio in commercial preparations very often are used for chemotherapeutic practice in veterinary medicine, as well as in human medicine. Veterinary formulations with this combination of active drugs ingredients, which are commonly termed "potentiated sulfonamide", must be of good quality, safe and effective. Quality control of trimethoprim is very important because these potentiated drugs are frequently used as main choice for treatment of urinary infections at humans and animals.

Materials and Methods

A standard stock solution with concentration 1 mg mL^{-1} was prepared with Trimethoprim (TMP) purchased from Sigma-Aldrich. An adequate mass portion in methanol (Sigma-Aldrich) (equal to concentration of 100 μg mL^{-1} TMP) of the different commercial products: Hemosul-P, Trimetosul, Bactrim and Lidaprim, were subjected to the assay procedure. Dilution was assisted by sonification in ultrasonic bath for 45 minutes. In the following step, to the aliquot of the sample, Bromocresol Green (0.001 mol/L in methanol) solution was added. The pH value of the media was adjusted adding a suitable amount of base.

The UV spectra of the standard and sample solutions, and appropriate blanks were recorded on a Varian Carry 50 UV/Visible Spectrophotometer, at room temperature in 1 cm quartz cell. The wavelength range was from 190 to 800 nm, with resolution 0.5 nm and scan rate of 300 nm/min.

Results

At the beginning of the experiments extraction with ethanol and methanol was tested. Methanol was chosen as more appropriate solvent, since better precision and accuracy were achieved. The method revealed good linearity (r=0.9953) in the concentration range 1–35 μg⁄ml, with maximum absorbance at 618 nm.

It was found out that the method has satisfactory accuracy and precision for human drugs (Bactrim and Lidaprim), but for veterinary products (Hemosul-P and Trimetosul) method performances have been rather poor. To improve the performances for veterinary compilations, 10 % *N,N*-dimetilacetamide in methanol as solvent has been used. The obtained linearity was satisfactory (r=0.9961) with significantly improved accuracy and precision. With some slight modifications the method has been successfully applied for different commercial formulations.

Conclusion

Statistical analysis showed the method is sufficiently accurate and precise, and may be successfully applied for quantitative determination of TMP in different formulations, when performing the official quality control of commercial drugs.

Key words*:* trimethoprim, bromocresol green, UV spectroscopy, drugs, determination

P18 ANTIMICROBIAL EFFECT OF WHITE WINE AND LEMON JUICE MARINADE AGAINST*SALMONELLA ENTERITIDIS* AND *LISTERIA MONOCYTOGES* IN CHICKEN MEAT

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ABSTRACT

Introduction

In this study, we evaluated the antimicrobial effect of white wine and lemon juice marinade (in combination with or without salt) on chicken breasts which were contaminated with *Salmonella enteritidis* and *Listeria monocytogenes.* The raw chicken meat can easily be contaminated with food borne pathogens, and it is well known that every day we are looking for better and more effective ways of eliminating them.

Materials and Methods

Raw chicken meat, was cut in 10 g. pieces and they were contaminated with *S. enteritidis* (NCBB 100284) and *L. monocytogenes* (NCTC 11994) reference strains in bacterial suspensions of 8 log CFU/ml. After 30 min. the pieces of meat were put in four combinations of marinade (in duplicates): white wine (Tikvesh, R. Macedonia), white wine and salt, lemon juice (100 % lemon juice) and lemon juice and salt. The marinades were placed in refrigerator at 4 ºC, and viable counts of marinated meat samples were made after 1h, 12h and 24h. Serial dilutions were made from the samples and they were spread plated on XLD (Oxoid, England) for samples with *S. enteritidis* and on Ottaviani Agosti Agar (Oxoid, England) for *L. monocytogenes*.

Results

The results of the analyses showed that after 1h, *S. entertidis* counts were lowered for the white wine, white wine and salt, lemon juice and lemon juice and salt as following : 7.71 log CFU/g; 6.71 log CFU/g; 5.69 log CFU/g and 5.04 log CFU/g respectively. After 12h and 24h, the viable counts for the chicken meat in the white wine marinade lowered from 5.60 to 5.30 log CFU/g., and for the white wine and salt from 5.47 to 5.30 log CFU/g respectively. The *S. enteritidis* was entirely eliminated after 12h and 24h in the chicken meat in the lemon juice and lemon juice and salt marinade.

The viable counts for *L. monocytogenes* examined after 1h, 12h, and 24h were as folowing: white wine marinade (6.90 log CFU/g; 5.78 log CFU/g and 5.60 log CFU/g respectively), for the white wine and salt marinade (6.67 log CFU/g; 5.20 log CFU/g and 5log CFU/g respectively), for the lemon juice marinade (6.44 log CFU/g; 4.60 log CFU/g and 4.30 log CFU/g respectively) and for the lemon juice and salt marinade (6.28 log CFU/g; 4.20 log CFU/g and 4.15 log CFU/g respectively).

Conclusions

The results from this study showed that marinades of lemon juice with and without salt were effective against *S. enteritidis* (8 log CFU/ml) in chicken meat after 12h and 24h. There was a synergistic effect of the wine and lemon with salt. The viable counts for *S. enteritidis* were lowered up to 2.70 log CFU/g in white wine marinade and white wine and salt marinade after 24h.The viable counts for *L. monocytogenes* were lowered up to 2.40 log CFU/g in white wine marinade, 3 log CFU/g in white wine and salt marinade, 3.70 log CFU/g in lemon juice marinade and 3.85 log CFU/g in lemon juice and salt marinade after 24h. This gave us a picture that all four combinations of marinades could be very effective if the pathogens are in smaller concentrations, and could be used for chicken meat preparations with a benefit of their antimicrobial and better flavouring effect.

Key words: food borne pathogens, *S. enteritidis, L. monocytogenes*, white wine, lemon juice, salt marinade, chicken meat

P19 PRESENCE OF TOTAL AFLATOXINS IN CORN FLOUR AND POLENTA

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ABSTRACT

Introduction

Aflatoxins are a group of naturally occurring, extremely toxic chemicals, mainly produced by two moulds, *Aspergillus flavus* and *Aspergillus parasiticus*. Under favorable conditions of temperature and humidity, these fungi grow on certain foods and feeds (corn and corn products, tree nuts, cottonseed, spices and milk). The major aflatoxins of concern are designated AFB_1, AFB_2, AFG_1 and AFG_2; however, aflatoxin AFB_1 is usually predominant and is the most toxic one. International Agency for Research on Cancer made classification of AB_1 as carcinogenic to humans (Group 1). Total aflatoxin content in food is regulated by legislation worldwide. The maximum residual level (MRL) for total aflatoxins content is set on 4,0 μg/kg for cereals and their products. The HPLC-FLD method with immunoaffinity column clean-up is the most used method for determination of aflatoxins due to its efficiency, specificity, accuracy and sensitivity.

Materials and Methods

Total of 35 corn flour and 17 polenta samples were bring to our laboratory by border health inspectors or from the food operators itself. The extraction and purification of samples for aflatoxins analysis was done according to AOAC Official method 991.31. HPLC-FLD procedure was performed according to ISO 16050:2003. For clean-up IAC Aflaprep from R-Biopharm Rhône were used. The validation procedure was performed according to Decision 2002/657/EC and Regulation 401/2006/EC.

Results

The validation procedure provides satisfactory values for all performing criteria for the method. Calibration curves were linear in the proposed concentration range for all four aflatoxins with satisfactory coefficient of correlation (R^2) in the range of 0,9993-0,9999. Limit of detection (LOD) and limit of quantification (LOQ) ranged 0,003-0,005 μg/kg and 0,009-0,023 μg/kg, respectively, were acceptable. Method accuracy estimated by recovery has been

tested and the mean recovery for total aflatoxins was 88,21%. The results for the repeatability of the method (RSD_r) are in the range 0,171 – 2,626%. RSD_R values (within laboratory reproducibility), show good correlation between two days (4,93-11,87%). 13 corn flour samples (37,14%) show total aflatoxins content below the LOD and 6 samples (17,14%) are over the MRL and contain aflatoxins in the range of 5,71-92,77 μg/kg. 4 polenta samples (23,52%) are with aflatoxins content less then LOD and only one sample was positive with total aflatoxins content of 7,46 μg/kg.

Conclusions

It was confirmed, through the validation procedure that employed HPLC-FLD method is suitable for total aflatoxins analysis in corn flour and polenta samples and it can be implemented for routine analysis in the laboratories.

Key words*:* total aflatoxins, HPLC-FLD, validation, corn flour, polenta

P20 CHEMICAL COMPOSITION OF COW MILK IN THE EARLY STAGES OF LACTATION IN TIED AND FREE STALL SYSTEM OF HOLDING

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ABSTRACT

The milk obtained from dairy cows represents the main final product in the cattle industry. With its high biological value is an important part in the human diet whether in the form of processed milk or in the form of dairy products. The yield and the quality of the milk are influenced by many factors (cattle diet, system of holding, lactation stage, air temperature, relative air humidity) whose values change during the entire year and as result the yield and quality of the milk change accordingly. The variability and the changes in the milk's chemical composition are also influenced by the way of holding and housing.

In this study 38 cows were included during the early stages of lactation (in the first 40 days after calving). From the free stall system 20 cows were included and from the tied stall system 18 cows were included. Milk samples were taken for analysis (Milkoscan 4000, Foss Electric) during the morning and the evening milking.

Results from the testing of the chemical composition of milk from dairy cows in the early stages of lactation indicate higher content of fat and dry matter in the samples taken from the free stall system.

Higher values of the ingredients in the chemical composition in the free stall system is due to the method and system of milking, milking procedure, higher hygiene of living spaces, milking machines, compared to the tied system of holding. From the results we can conclude that the free stall system with automatic milking has a positive effect on the chemical composition of the milk, compared to the bound system of holding and manual milking.

Key words: milk quality, dairy cows, stall system, milkoscan

P21 VALIDATION OF ELISA SCREENING METHOD AND DETECTION OF BOLDENONE IN CATTLE URINE

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ABSTRACT

Introduction

17 β Boldenone (androsta-1,4-dien-17-ol-3-one) is a synthetic androgenic steroid hormone and synthetic derivatives of the male hormone testosterone. The anabolic effects are considered to be those promoting erythropoiesis, protein synthesis and muscle growth. Because of this effects boldenone are used to improve the growth and food conversion of cattle and therefore can be abused for more efficient meat production. Boldenone, like the other anabolics, are banned substances in the European Union but might still be illegally applied as growth promoters. The aim of this study was validation of screening ELISA method and detection of boldenone in cattle urine.

Materials and Methods

For detection of boldenone we used ELISA kit for boldenon from Tecna. For our study 34 urine samples from cattle were obtained and they were kept frozen until use. The ELISA method for boldenone was previously validated and for determination of limit of detection we used 20 blank bovine urine samples. Detection capabilities (CCβ) was evaluated by analyzing 20 spiked bovine urine on ½ of MRPL level and recovery was determined at three levels by spiking on blank urine on 0,5; 1 and 1,5 ng/ml.

Results

The results of this study showed that the mean recovery for detection of boldenon with ELISA method was 84,32%. The limit of detection was 0,27 ng/ml and detection capability was 0,67 ng/ml. In the analyzed 34 cattle urine samples the concentration of boldenone was less than detection capability.

Conclusion

In our study screening ELISA method was used for monitoring of boldenone, anabolic steroid, in cattle urine. The method was validated and the detection capability was satisfactory. Also the recovery of the method was good and boldenon wasn't detected in cattle urine. Due to good recovery and satisfactory CCβ, our method is applicable in laboratories involved in official routine analysis for monitoring the illegal use of anabolic steroids.

Key words: boldenone, ELISA, validation, cattle urine, anabolic steroid

P22 VALIDATION OF ELISA METHOD FOR DETECTION OF TRENBOLONE IN BOVINE URINE

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ABSTRACT

Introduction

ELISA method has been developed for the determination of trenbolone residues in bovine urine. Trenbolone acetate is a synthetic anabolic hormone which is used as a feed supplement to promote the growth of bovine. Trenbolone is rapidly hydrolyzed to its metabolite 17*β*-trenbolone. Twenty-four hours after application, 80% of the 17*β*-trenbolone will be converted to 17α-trenbolone and then excreted by urine. The use of trenbolone in food producing animals is prohibited in most countries of the EU. For this reason in our study validation of ELISA method for determination of trenbolone in bovine urine is described.

Materials and Methods

The analyte was extracted with sodium acetate buffer and cleaned by C18 solid phase extraction cartridge. For determination of limit of detection we used 20 blank bovine urine samples. The method recovery was determined at three levels by spiking on blank urine (1; 1,5 and 2 ng/ml). Detection capabilities (CCβ) was evaluated by analyzing 20 spiked bovine urine on ½ of MRPL level. Precision was expressed as the Coefficient of variation (CV) of the calculated standards and sample concentrations.

Results

Detection limit for trenbolone in bovine urine was 0,20 ng/ml and CCβ was 1,08 ng/ml The overall recovery varies from 85% to 97,1% at the three target concentration. The precision (CV%) in trenbolone standards ranged from 2.0% to 8.9%. The precision in spiked cattle urine samples ranged from 2.8% to 9.2 %.

Conclusion

The method was reliable, sensitive and reproducibility, its performance can meet the requirements of the domestic and international legislation. Because of good recovery and precision, and satisfactory CCβ, it is applicable in official control laboratories as a screening method for determination of trenbolone in bovine urine. But in the case when the target analyte is clearly identified above

CCβ the sample is considered as non compliant and we must confirm the results with GC/MS, LC/MS or another confirmatory method.

Key words: trenbolone, anabolic steroid, urine, ELISA

P23 ISOLATION AND IDENTIFICATION OF LACTIC ACID BACTERIA IN KASHAR CHEESE

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ABSTRACT

Introduction

The aim of this study is to identify the lactic acid bacteria (LAB) in fresh and ripened kashar cheese samples (37 fresh and 13 ripened kashar samples) retailed in Erzurum.

Materials and Methods

A total of 38 strains of LAB from fresh kashar samples and 10 strains of LAB from ripened kashar samples were isolated and identified using API 50CHL (Biomerieux, France) methods.

Results

In the fresh kashar samples: One (2,6%) of the LAB was *Lactobacillus delbrucki* ssp. *lactis*, 5 (13,2%) of them were *Leuconostoc lactis*, 8 (21,1%) of them were *Lactobacillus curvatus* ssp. *curvatus*, 3(7,9 %) of them were *Lactococcus lactis* ssp. *lactis*, 3 (7,9 %) of them were *Lactobacillus fermentum*, 4 (10,5%) of them were *Lactobacillus paracasei* ssp. *paracasei*, 5 (13,2%) of them were *Lactobacillus brevis*, 1 (2,6%) of them was *Weissiella viridescens*, 5 (13.2%) of them were *Lactobacillus plantarum*, 1 (2,6%) of them was *Leuconostoc mesenteroides* ssp. *Mesenteroides dextranicum*, 1 (2,6%)of them was *Lactobacillus rhamnosus*. Only one isolate was identified as *Pediococcus pentosaceus* (2,6%). In the ripened kashar samples; *Lactobacillus curvatus* 4 (40%), *Lactococcus lactis* ssp. *lactis*, 1 (10 %) *Lactobacillus paracasei* ssp. *paracasei* 4(40%), *Lactobacillus brevis* 1(10%) were the main isolates.

The LAB, total aerobic–mesophilic, *Staphylococcus–Micrococcus*, yeast and mould counts were in fresh and ripened kashar were 6,13±1,21-6,26±1,54 log kob/g, 6,57±1,01-5,99±1,25 log kob/g, 4,46±1,64-3,94±0,91 and 3,53±1,94-4,46±1,48 log kob/g, respectively. *Enterobacteriaceae* and *Salmonella-Shigella* (SS) were not detected in any sample.

Conclusions

It can be concluded from the sedata that the dominant flora in kashar is *Lactobacillus* species.

Key words: lactic acid bacteria (LAB), API 50 CHL, kashar cheese

P24 MASTITIS IN DAIRY GOATS: ISOLATED MICROORGANISMS AND ANTIMICROBIAL RESISTANCE

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ABSTRACT

Mastitis is an inflammation of the mammary gland (udder) that causes a chemical and physical reaction in milk produced by goats. It is more frequent in dairy and meat goats raised under intensive and semi-intensive management practices. Depending on the severity of the disease, mastitis could result in decreased revenues for producers.

Mastitis is generally associated with poor hygienic practices and caused by the bruising of mammary tissue or teats from traumas, nursing, fly bites, or other wounds to the skin that provide an important barrier to infection. Mastitis is also associated with viral, bacterial or fungi and their toxins. Under stressful conditions such as extreme temperatures, muddy and wet living conditions, or a sudden change in diet, a doe's immune system is compromised and has a difficult time fighting off the invasion of foreign bodies that cause diseases like mastitis.

In this study 66 samples were analyzed. The samples were collected from goats with high somatic cell count (>500.000 scc/ml). The testing was done using standard microbiological methods and the antimicrobial resistance was determined using Kirby Bauer test.

The results present the fact that coagulase negative staphylococci were the most often detected 60.6% (40 samples), *Streptococcus* spp. was next with prevalence of 16.6% (11 samples) and the lowest prevalence was detected for *Staphylococcus aureus* 13.6% (9 samples).

The disc diffusion method for antimicrobial resistance showed the following results: the coagulase negative staphylococci were highly resistant to tetracycline (50%), nalidixic acid (55%) and penicillin (62.5%), the strains of *Streptococcus* spp. were highly resistant to tetracycline (45.5%) and enrofloxacin (63.6%). *Staphylococcus aureus* strains showed the highest resistance towards penicillin (66.7%), nalidixic acid (55.6%) and neomycin (77.8%).

While mastitis cannot be totally eliminated from the goat herd, incidence can be held to a minimum. Key elements in control are good husbandry practices and sanitation. Major techniques for the control of goat mastitis are: post-milking

teat dipping and therapy at dry off (end of the lactating period). Intra-mammary therapy at dry off has the two functions: elimination of existing infections and prevention of the establishment of new infections.

Key words: mastitis, goats, somatic cells, antimicrobials

P25 EFFECTS OF PRODUCTION SYSTEM AND SLAUGHTER WEIGHT ON FINISHING PERFORMANCE AND CARCASS QUALITY IN KIVIRCIK LAMBS

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ABSTRACT

Fifty three Kivircik lambs were used to investigate the effects of production system (intensive or semi-intensive) and slaughter weight (Low=24-27 kg; Medium=29-32 kg; High=34-37 kg) on lamb growth, slaughter characteristics and carcass quality. Intensive lambs (n=27) were weaned at 75 days of age, and then placed into the finishing pen. These lambs were fed *ad-libitum* with good quality alfalfa hay and concentrate feed until slaughter. Semi-intensive lambs (n=26) were allocated in a pen built for them and their mothers in the sheepfold until 75 days of age. Then, these lambs and their mother were grazed on pasture in the day-time, while they had free access to good quality alfalfa hay during the night in the sheepfold until slaughter age. Lambs of both production systems were divided into three subgroups (Low, Medium and High weight groups) according to the target slaughter weight.

Average daily gain (ADG) until slaughter age was higher ($P<0.001$) in intensive system (200.3 g) then semi-intensive system (154.9 g). Lambs from Low, Medium and High slaughter weight groups had similar ADG during finishing period. Intensive lambs had higher hot carcass weight, hot dressing percentage and omental and mesenteric fat weight compared with semi-intensive lambs ($P<0.001$). Omental and mesenteric fat weight of Low group was lower than those of Medium and High groups. Intensive production system yielded higher weights of ribs and pelvic limb cuts than semi-intensive system ($P<0.001$). On the other hand,lambs from intensive system had also higher kidney knob channel fat (KKCF) weight ($P<0.001$), and subcutan ($P<0.001$) and intermuscular ($P<0.01$) fat weight in the dissection of pelvic limb. Lambs from High and Medium groups had similar fatness levels (KKCF, subcutan and intermuscular fat weights), which were alsohigher than those of Low group. Lamb carcasses from semi-intensive system had higher fat lightness (L*) value measured from tail root than intensive carcasses, however production system had no significant influence on fat yellowness (b*) value.

In conclusion, intensive production system yielded higher levels ofgrowth

rate, carcass weight, carcass dressing, weights of high value carcass cuts, but also fattier carcasses compared with semi-intensive system. If increasing consumer demands for less fattier carcasses is taken into consideration, slaughtering of semi-intensive Kivircik lambs at 24-27 kg weight might be suggested to the sheep farmers as leanest carcasses were obtained from this production model.

Key words: lamb, intensive system, semi-intensive system, growth performance, carcass quality.

P26 MEAT QUALITY CHARACTERISTICS OF LAMBS REARED UNDER EXTENSIVE, INTENSIVE AND ORGANIC PRODUCTION SYSTEMS

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ABSTRACT

This study aimed to investigate meat quality of male, single born, crossbred lambs (Chios x OstFriz x Kivircik) reared under extensive (n=10), intensive (n=10) and organic (n=9) production systems. The differences among production systems in terms of carcass pH, drip loss, cooking loss and Warner-Bratzler shear force values were not significant (P>0.05). The effect of production system on meat colour characteristics was also not significant, except meat lightness value measured at 24 h after cutting (L^{24h}). Meat samples obtained from extensive lambs had higher L^{24h} value (42.30) than that of intensive (39.43) and organic (39.00) lambs.

According to the sensory panel evaluation results, meat samples of three production systems had similar lamb odour intensity and juiciness. Panellists gave lower scores to the meat of organic lambs for flavour intensity and flavour quality. Panellists evaluated the meat samples of intensive lambs being more tender than organic lambs. Meat samples obtained from organic lambs had lower overall acceptability score (4.82) than that of extensive lambs (5.36).

Results of the study indicated that although the differences among production systems in terms of instrumental meat quality characteristics were not significant, meat of extensive system were evaluated more acceptable by panellists in sensory evaluation.

Key words: lamb, production system, meat quality, sensory evaluation

P27 ACTIVITY OF ^{137}Cs AND ^{40}K IN SHEEP MILK

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ABSTRACT

The human does not exclude himself from the environment, thereby the changes in the radiologic contamination of the environment (water, air, soil, food products) also condition the level of radioactive contamination of the human organism.

In the tissues of animal organisms in European countries, the maximal value of radioactive contamination has been achieved in the period from 1963 to 1964 after the nuclear trial performed in China and after the accident of the nuclear power plant in Chernobil in 1986.

After this incident, the sheep were the most endangered species of domestic animals since they are bred in the mountain regions and they are fed from pastures, so for this reason, consumption of milk and meat was forbidden in Europe for a long time period.

The sheep milk is one of the links in the chains of circulation of radioactive substances, which through the nutrition reach the human and affect human's health. For this reason, radiation control of the content of natural radionuclides should be performed, including ^{137}Cs. Hence, the objective of this research was to determine the presence of ^{40}K and ^{137}Cs in samples of sheep milk, from different regions of Republic of Macedonia. 25 samples of sheep milk were analysed, taken from different regions in the Republic of Macedonia. While taking the samples, it has been confirmed that the animals are not fed up and they were kept exceptionally eating from pastures. The samples were analysed with clean semi-conductor gamma spectrometer Canberra Packard, with efficiency of the detector of 30% measured at ^{60}Co Na. On the basis of the obtained results, it has been determined that ^{137}Cs is present in all regions of the Republic and its activity is within the range from 0,22 Bq/kg to 0.68 Bq/kg, while the activity of ^{40}K is from 19,30 to 37,20Bq/kg. So the level of ^{137}Cs in sheep milk is not even close to the maximal allowed limits of pollution of 370Bq/kg. From the above indicated it follows that at the current level of radioactive contamination for the food products with plant origin similarly to agricultural products, animal body tissues and the food products of animal origin, there would be no need for taking measures in regard to reduction of the radioactive pollution, taking into consideration that the values from the radioactive pollution are lower, compared

to the level of radiological contamination from natural originAfter the performed comparison with the Rulebook for maximal allowed quantity of radionuclides in food, it has been shown that all samples fulfill the criteria given in the same.

However, continuous control of ^{137}Cs in sheep milk is required, because it represents an indicator of contamination of the environment

Key words: radionuclides, milk, analysis, gamma spectrometry, results

P28 REPLACEMENT OF FISH MEAL BY RIBOTRCIN IN DIETS OF CARP (*CYPRINUS CARPIO*)

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ABSTRACT

Introduction

Waste nonstandard fish combine with wheat bran filler in ratio 60:40 were used for production of new nontraditional protein component – Ribotricin. This study was conducted to investigate the effects of new protein component Ribotricin on growth performance, feed utilization and feed ingredients digestibility of carp (Cyprinus carpio L., 1758) fish.

Materials and Methods

The experiment (45 days) involved 160 carp fish rear in eight concrete tanks with an initial average weight of 0,046 ± 8,04 g in (RT) tank, and fish with an initial average weight of 0,047 ± 6,52 g in (FM) tanks.

Results

In the experiment, a total of carp fish were equally divided into two groups of 80 (four replicates of 20 carp each). At the end of the experiment, there were no significant differences among the groups in Body Weight (BW), Feed Conversion Ratio (FCR) Specific Growth Rate (SGR), Protein Efficiency Ratio (PER) and apparent digestibility ($p<0.05$).

Conclusions

It is concluded that Ribotricin (RT) can be substitution fish meal in the diets of carp fish without adverse effects on the measured parameters.

Key words: carp, fish meal, feed conversion ratio, specific growth rate

P29 GAME MEAT SAUSAGES AS MYCOTOXICOLOGICAL RISK TO CONSUMERS

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ABSTRACT

Introduction

Game, as a representative of wildlife, is considered to be a suitable bio-indicator of environmental pollution and also subject to contamination with mycotoxins. Game has a freedom to choose on what to feed, but the feeding thereby being dependent on seasonal availability of certain types of food. The presence of mycotoxins in food and feed depends on many environmental factors, such as the region of origin, season, humidity and temperature, as well as on the conditions under which the crops are harvested, stored and processed. When not appropriately controlled, these toxins can be transferred onto animals and humans through the ingestion of contaminated feed and food.

Material and Methods

For the sake of aflatoxin B_1 (AFB_1) and ochratoxin A **(**OTA) determination, competitive enzyme-linked immunosorbent assay (ELISA) was implemented and validated, whereas the concentration of citrinin (CIT) was determined using high performance liquid chromatography with fluorescence detection (HPLC-FLD).

Results

OTA was detected in 86.73% of game sausage samples (n=15), with its concentration ranging from <0.05 to 3.07 μg/kg. In addition to OTA, CIT and AFB_1 were found in one sample of wild boar sausage in concentrations of 1.0 μg/kg and 1.5 μg/kg, respectively. OTA in the mean concentration of 2.84 μg/kg was found in wild boar sausages, while the samples of rabbit sausages harbored OTA in the mean concentration of 2.27 μg/kg. Mixed sausages made from wild boar, deer and domestic pigs were shown to contain OTA in the mean concentration of 2.16 μg/kg. Deer and roe deer sausages were contaminated with OTA in concentrations less than 2.00 μg/kg. ANOVA showed significant differences in OTA concentrations across various types of game sausages (P<0.05). In the samples of game meat sausages, moulds in the outer and inner part were not proven.

Conclusions

As hunters and members of their households frequently consume game meat, and because of the possible *carry over* effect entering additional amounts of mycotoxins in humans, in order to protect the public health, the occurrence of these contaminants in game sausages should be constantly monitored.

Key words: game meat sausages, aflatoxin B1, ochratoxin A, citrinin

P30 CONSUMERS' SENSORY ASSESSMENT IN RELATION TO FREE FATTY ACIDS CONTENT THROUGHOUT RIPENING OF CHEESE IN A SACK

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ABSTRACT

Introdution

Cheese in a sack (local name: sir iz mišine) is one of Croatia's traditional cheeses, produced on family farms in central Dalmatia. The main specificity of this cheese is ripening in a sack made of lamb skin for a period of 2-3 months. The leading process during cheese ripening in an animal skin, which is responsible for the cheese unique strong and piquant flavour and aroma, is lipolysis. Therefore, the aim of this paper was to study sensory properties throughout ripening of cheese in a sack in relation to free fatty acids (FFA) content.

Material and Methods

The production and ripening of 25 batches of raw ovine milk cheeses ripened in lamb sack were observed at five family sheep farms. Samples of cheese at 30, 45 and 60 days of ripening were taken for lipolysis analysis as well as for the sensory evaluation by a group of local consumers (n=29). The relationship between sensory scores (odour, taste and total) and free fatty acids content was done by calculation of Pearson correlation coefficients.

Results

The concentration of fatty acids C 14:0, C 14:1, C 17:0, C 18:0, C 18:2n-6 and C23:0 in 45 days matured cheese resulted in significantly positive correlation ($p<0.05$) with cheese odour. After 60 days of ripening, as a result of the ripening time, accumulation of C 4:0, C 6:0 and C 8:0 fatty acids, which directly affect the taste and aroma of cheese, have led to a negative correlation between C 4:0 and odour ($p <0.05$), C 6:0 and taste ($p <0.05$) and C 8:0 and taste, aroma and total sensory scores ($p <0.01$, $p <0.01$, 0.05). Accumulation of polyunsaturated FFA (> C18), with the exception of C 18:2*tt*, C 20:2 and C 22:6 n-3, had a positive impact on the sensory properties of cheese. As unsaturated fatty acids are precursors for synthesis of acids and alcohols, it

is possible that the larger amounts of the above three fatty acids adversely affect the sensory properties of cheese, indirectly through reactions of catabolism.

Conclusion

Extensive lipolysis led to a fact that the fully ripened cheese (60 days) was not well accepted by consumers. In terms of sensory quality the optimal ripening time of cheese in a sack was determined to be 45 days. The study clearly showed that cheese ripening for too long, commonly practiced by cheesemakers, is not recommended.

Key words: cheese in a sack, free fatty acids, sensory assessment

P31

THE EFFECTS OF USE THE LITTER OF VERMICULITE ON SOME PHYSICOCHEMICAL ANDMICROBIOLOGICAL PARAMETERS IN BROILER MEATS

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ABSTRACT

Introduction

This study was conducted to determining the effects of different litter materials on the meat quality of broiler meat.

Materials and Methods

Throughout the experiment, 195 day-old chickens were used. The chicks were placed in different partitions which three different litter material (wood shavings=WS, mixture of wood shavings-vermiculite=WSV and vermiculite=V) were used. The experiment has been carried on for 42 days. At the end of this period the breast and drumstick meats derived from slaughtered broilers were packed and stored at 2°C 24 hours and then microbiological and physicochemical analyzes were made.

Results

In this study, only the difference between the groups in terms of aw and pH values were determined on the breasts ($p<0,01$). The L* values of both breast ($p<0,01$) and drumstick ($p<0,05$) meats were important differences between the groups. For the lowest value of L* in breast and drum stick samples were detected on V group. For the values of a* and b* the difference was important between the groups for the breasts and the drumstick. For the highest value of a* and b* in breasts and drumstick samples was observed on V group.

At the end of the microbiological experiment, it has been ascertained that the litter material have no significant ($P>0.05$) difference on the count of total mesophilic aerobic bacteria (TMAB), total psychrotrophic aerobic bacteria (TPAB), enterobacteria, coliform bacteria, *Lactobacillus spp*. And *Pseudomonas*

spp., Enterococcus spp. and *S. aureus* in both breast and drumstick samples except for the count of *Staphylococcus*/*Micrococcus* in the breasts ($p<0,05$).

Conclusions

As a result, it was observed that use of different litter type has effect on some physicochemical properties of broiler meat, while the effect on the microbiological properties of the meat has been only the count of *Staphylococcus*/*Micrococcus* in the breasts.

Key words: broiler, litter material, vermiculite

P32 A STUDY ON DETERMINATION OF SOME CHEMICAL SUBSTANCES INJECTED INTO THE BEEF AND POULTRY MEAT

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ABSTRACT

Introduction

The commercial preparations consist of the organic acids such as sodium acetate, trisodium citrate and ascorbate with dry glucose, flavor and salt that applied to meat by injection are used in recent years, increasingly because of their abilities of preventing weight loss of 25-30 % levels during baking process, providing cooking at low temperatures, decreasing the pH to 5,2-5,5 levels, providing ripening through activating cathepsin and the other proteolytic enzymes, having positive effects on color, aroma, texture and carrying antioxidant properties.

The aim of this study were not only to prevent carcinogenic effects of such applications, but also to obviate unjust enrichment due to inhibition of loss of water and to achieve consumption of fresh red meat and poultry meat suitable for food codex.

Materials and Methods

In this study, the totally 150 samples (75 fresh red meat and 75 fresh poultry meat) were randomly obtained from the hotels operating for tourism, catering companies and the restaurants in connection with these catering companies in Provincies of Bursa, Balikesir and Canakkale located in the South Marmara Region of Turkey. The presence of sodium acetate, trisodium citrate and ascorbate in the samples were investigated by using HPLC device under some modifications in the method described by Shui et al. 2002.

Results

According to the analyses results, ascorbic acid in 27 (12,7%) (17 fresh red meat and 10 fresh poultry meat) out of 150 samples, citric acid in 8 (5,3%) (3 fresh red meat and 5 fresh poultry meat) out of 150 samples and acetic acid in 8 (5,3%) (3 fresh red meat and 5 fresh poultry meat) out of 150 samples were detected.

Conclusions

It was concluded that the presences of these chemicals in the meat samples could be threated to public healthy and the checking mechanisms intended for this problem should be activated for obviating unjust enrichments of producers and purveyors, made these adulterations and for achieving the consumption of fresh red meat and poultry meat suitable for food codex.

Key words: red meat, poultry meat, sodium acetate, sodium citrate, ascorbate

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P33 CHARACTERISTIC OF BACTERIOCINS PRODUCED BY LACTIC ACID BACTERIA

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ABSTRACT

In recent years, consumers prefer natural foods produced without any addition of chemical preservatives. Thus natural preservatives have attracted the food industry's attention. One of the alternatives applied in the food industry is bacteriocins. Bacteriocins are natural antimicrobial substances of protein structure produced by a large of bacteria including lactic acid bacteria. They demonstrate strong and wide-spectrum inhibitory activite against closely related and some spoilage and pathogens (etc. *Listeria monocytogenes, Staphylococcus aureus, Bacillus cereus*, *Clostridium botulinum)*. Bacteriocins are usually produced by *Lactococcus, Pediococcus and Leuconostoc.* The important ones of bacteriocins from lactic acid bacteria are nisin, diplococcin, acidophilin, bulgarican, helveticins, lactacins, and plantaricins. Because of similar activity, in many sources are confused bacteriocins wiht antibiotics. However, there are some differences between bacteriocins and antibiotics. While bacteriocins are ribosomally synthesized, antibiotics gain active forms through enzymatic processing. Bacteriocins are primary metabolites produced during the growth phase, though antibiotics are usually secondary metabolites produced in the stationary phase. There are four main classes of bacteriocins: the lantibiotics (Class I); the small (<10 kDa) heat-stable bacteriocins (Class II), the large (>30 kDa) heat-labile bacteriocins (Class III) and the complex bacteriocins (Class IV). Class II may be further subdivided in the pediocin-like and anti-*Listeria* bacteriocins (subclass IIa), the two-peptide bacteriocins (subclass IIb), and the cyclic peptides (subclass IIc) and the non-pediocin one-peptide linear bacteriocins (subclass IId). This paper gives the general knowledge about bacteriocins produced by lactic acid bacteria.

Key words: bacteriocin, lactic acid bacteria, classification

P34 LC-MS/MS METHOD FOR THYREOSTATIC AGENTS IN URINE AND MUSCLE

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ABSTRACT

It is known that „the use of thyreostatic substances is prohibited in food producing animals and no residues of these substances should be present in animal products imported or produced within the EC (Directive 96/23/EC)". Generally thyreostats are divided in two groups: thyiouracil analogues as thyouracil, methylthiouracil, propylthyouracil, phenylthyouracil; mercaptoimidazole analogue and its analogue tapazole. According 96/23/EC thyreostats belong to A5 group.

Present an analytical method for determination of thyreostats in urine and muscle by LC-MS/MS detection after extraction and by derivatization of the analytes with 3-iodobenzilbromide. The analytes were separated on Poroshell 120 EC-C18 chromatographic column with easy eluting program using aqueous formic acid and methanol as a mobile phases. The recoverys at corresponding concentration to MRPL-level are between 48 and 113%. Linear range, repeatability, reproducibility were also determinate. The mas-spectrometry detection was used to ensure the qualitatively identification of derivatized substances. The method was validated according Commission Decision 2002/657/EC criteria and was approved as confirmatory method at the Bulgarian Reference Laboratory.

Key words: LC-MS/MS, derivatization, thyreostats, extraction

P35 REQUENCY OF CONTAMINATION WITH *LISTERIA MONOCYTOGENES* OF RAW DRIED CURED VACUUM PACKED SAUSAGES

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ABSTRACT

The aim of this study was to collect actual data concerning the frequency of contamination with *Listeria monocytogenes* of some very popular in Bulgaria raw dried cured vacuum packed sausages, produced from Oct. 2004 till May 2008. 148 vacuum-packed samples were taken from 9 different food business operators during all seasons of the year. The samples were analyzed according to USDA method for meat foods, described by Ryser and Donelly (15). Ten specimens were positive for presence of *Listeria monocytogenes* equal to 6,75% of all tested samples. In two other raw dried cured sausages *L.welshimeri* and *L.innocua* were found, but these species are not pathogenic for consumers. In the period before the official implementation of HACCP system (01.01.2006) in Bulgaria, 52 samples were examined and 5 *Listeria monocytogenes* isolates were found (~10%). 2,5 years after the HACCP implementation, 96 specimens from the same meat factories were tested and 5 *Listeria monocytogenes* isolates (5,2%) were detected. Samples taken from lots, produced in winter time were contaminated with *Listeria monocytogenes* more often (7 of all 10) than specimens taken during other seasons. Data were discussed through the point of view of the effectiveness of hygienic practices and HACCP system application. Also, application of 'microbiological criterion' set in COMMISSION REGULATION (EC) No 2073/2005 for ready-to-eat foods unable to support the growth of *L. monocytogenes* was considered.

Key words: L.monocytogenes, raw dried sausage, contamination, HACCP

P36

POSSIBILITIES OF IMPLEMENTING PROGESTERONE EIA TEST IN THE CONTROL OF REPRODUCTION IN DAIRY COWS

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ABSTRACT

The aim of this work was to implement progesterone EIA test developed in our laboratory by using anti-progesterone antibody (Yamaguchi University, Japan) in order to determine the optimal moment for insemination and to detect pregnancy in Holstein-Friesian cows according to the progesterone concentration in the whole milk. Also, the influence of β-carotene, applied at the day of insemination and human chorionic gonadotrophin applied on day 7 after AI on the progesterone level and pregnancy rate were evaluated.

For the accuracy of estrus detection, the milk samples from 70 cows were collected on the day of insemination. Milk samples from 148 cows were collected 19-22 days following insemination for pregnancy check.

After naturally occurring oestrus (day 0) and AI, cows were divided in the following groups: group A (n = 19) was treated with 200 mg β-carotene (20 ml Carofertin® i.m.), group B (n = 17) was treated with 1 500 IU hCG i.m. and control (non treated) group C (n = 18). The milk samples for EIA progesterone concentration analysis were collected on the day of AI, the 14th and the 20th day of oestrus cycle.

Oestrus detection errors and inappropriate moment of insemination according to the progesterone concentration were detected in 22.86% animals (16/70). The test accuracy for non-pregnant cows was 90.48% (76/84). The accuracy of progesterone test in pregnant cows was 75% (48/64). False positive results (high progesterone level but the cows were not pregnant) was detected in 25% of cows (16/64) as a result of prolonged oestrus cycles, embrional mortality and endomethritis (10/16 cases).

The treatment of cows with 1 500 IU of hCG, on the day 7 of oestrus cycle, resulted in statistically significant increase of progesterone concentration in diestrus ($P < 0.01$). The most successful insemination was in the group of cows that was treated with hCG (47.05%; 8/17), then in the control group (38.88%; 7/18) and, at the least, in Carofertin group - 36.84% (7/19). These differences were only numerical ($p>0.05$).

We may thus conclude, that our progesterone EIA test could be used for accurate progesterone testing in the whole milk.

Key words: progesterone, b-karotene, cows, EIA test, hCG, milk

P37 SEXUAL BEHAVIORS OF KIVIRCIK EWES HAVING DIFFERENT LEVEL OF RECEPTIVITY INDEX

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ABSTRACT

Reproduction is important for sheep breeding since the greatest part of the income in sheep farming is supplied through lamb production. One of the most important determinants of successful reproduction is the willingness and ability of animals to display adequate levels of sexual behaviour. The present study was performed to investigate thealterations in the frequencies of some sexual behaviorsdisplayed around the time of estrus by Kivircik ewes having Low, Medium and High level of receptivity index (RI). The effects of different levels of RI on attractivity, proceptivity and some estrus parameters were also investigated.

During the breeding season, 43 Kivircik ewes were synchronized with intravaginal spongesimpregnated with 30 mg FGA left for 12 days plus an injection of 600 IU PMSG at the time of sponge removal.Behavioral observations were performed at 8 hours intervals starting at 16 h following sponge removal. During the observation periods ewes were introduced to rams one by one, and sexual behaviour of eachewe was video-recorded for 3 min. After the evaluation of video-records the ewes were divided into three subgroups (Low, RI<50%; Medium, RI=50-80%, and High, RI>80%) according to their RI level.

The differences among groups in terms of estrus onset, estrus duration, average attractivity and average proceptivity were not significant statistically ($P>0.05$). Frequencies of urination and vocalisation behaviours during the receptive period were lower than those recorded at 8 h before, and 8 h after the receptive period. The frequency of non firm standing behaviour was similar both at the receptive period and at the previous observation period.

It is concluded that the level of receptivity index does not have any significant effect on attractivity, proceptivity, and some other sexual behaviours in Kivircik ewes. A ewe with low receptivity index display adequate level of sexual behaviour as a ewe with high receptivity index does.

Key words: Kivircik ewe, sexual behavior, receptivity index, attractivity, proceptivity

P38

INFLUENCE OF GEOGRAPHICAL LOCATION CHANGE ON ONSET OF SEXUAL ACTIVITY IN INDIGENOUS PRAMENKA OF LIKA SHEEP

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ABSTRACT

Introduction

Pramenka of Lika is an indigenous breed of sheep which is raised in the mountain areas of Lika and Gorski Kotar in Croatia and it is the most typical strain of Pramenka sheep, with strong physical condition and resistant to illnesses. It is the source of income for the rural population of Lika and Gorski Kotar and the production of cheese and lamb meat depends on the seasonal breeding and specific period of lambing of sheep. For this reason, management of herd's reproduction at family households is of great importance. The aim of this study was to determine the onset of sexual activity and environmental influence in two consecutive breeding seasons, by measuring serum progesterone concentration (P_4).

Materials and Methods

In the study, 8 sheep at age from 1.5 to 5.0 were brought at the Clinic for Reproduction and Obstetrics at the Veterinary Faculty Zagreb from a family household situated in Lika. Blood samples were taken in the period from 10.06.2011 until 05.09.2012, once a week and serum was extracted by centrifugation within 15 minutes from sampling. They were packed, labeled and stored at -20 °C in 2ml cuvettes until assessment. The P_4 testing was made by Immulite®One automatic system.

Results

In the first year of study, the start of the breeding season was first recorded in one sheep in July, 23rd, 32 days after the summer solstice, which is considered to be the trigger of sexual activity in sheep. In August, 26th, cyclic activity was noted in another one. During September, serum P_4 levels were elevated at 5th, 13th and 29th in one, three and one sheep, respectively. One sheep entered the breeding season as late as the 10th of October, 2011. Next year, ovarian cyclic activity was registered in 6 sheep in August, at 10th in one sheep, at 17th in two more, as well as the 24th and in one sheep at the very end of the month, 31st.

Serum P_4 levels showed cyclic activity of the ovaries in one sheep at 5th of September, 2012 and only one sheep did not establish ovarian cyclic activity.

Conclusion

Based on the results, we can conclude that the breeding season in the first year of study started two weeks later than the year after (~ September, 8th vs. ~August, 25th). Although this difference is not statistically significant, it points out that the change of geographical location could postpone the start of sexual activity in Pramenka of Lika sheep.

Key words: breeding season, environment, progesterone, Pramenka of Lika, sheep

P39 HISTOPATHOLOGICAL CHANGES OF THE LIVER IN T-2 MYCOTOXICOSIS IN BROILERS

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ABSTRACT

Introduction

T-2 toxin is a trichothecene mycotoxin produced by some Fusarium fungi. The harmful consequences of ingestion of dietary T-2 toxin have been examined in many species after its prolonged administration, but there are very few records of its toxicity after short term administration. Therefore, the aim of this study was to examine the effect of T-2 mycotoxicosis on the histopathological changes in the broiler's liver after short term (3 day) administration.

Materials and Methods

One day old broilers were divided in two groups, both given feed and water ad libitum. T-2 mycotoxin was dissolved in water and given to the experimental group with daily oral gavages in doses of 0.250 mg per bird for 3 consecutive days. Histopathological analyses of the liver were made 24 hours after the last application. Liver was fixed in buffered 10% formalin, embedded in paraffin and 5μm thick sections were stained with haematoxylin and eosin.

Results

Compared to the control group, the body weight and the absolute liver weight of the broilers in experimental group were significantly reduced by 27,26 % and 19,19%, respectively. The relative liver weight was significantly increased by 5,30% compared to the control chicks. Histopatological analyses showed areas with destruction of the normal liver lobular histology, accompanied with cell necrosis. Mononuclear cell infiltration was evidenced in Kiernani's interlobular spaces. In all experimental animals, fat degeneration of hepatocytes was noticed in a form of small lipid vacuoles present in their cytoplasm. Fat degeneration was observed in the central parts of the lobules (around vena centralis), as well as in the ventral and periportal parts of the liver lobes. Dilated sinusoids were also registered.

Conclusion

The obtained results showed that three day application of T-2 mycotoxin provokes damage of the histological architecture of the liver with distinguish fat degeneration of the hepatocytes.

Key words: T-2 mycotoxin, liver, histopathology, broilers

P40 T-2 TOXIN AND IT'S EFFECT UPON BROILER'S TISSUE ANTIOXIDANT STATUS

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ABSTRACT

Mycotoxins are secondary metabolites of various fungal species and are unavoidable contaminants of food and feed, because their formation is weather dependant and effective prevention is impossible. From all Fusarium mycotoxins, trichotecenes, especially T-2 toxin has an enormous influence on poultry health and performance. Their presence in high concentrations in poultry feed may induce genotoxic, cytotoxic, cancerogenic and teratogenic effects. They are fast acting potent inhibitors of protein and nucleic acid synthesis, inhibition of mitochondrial function, effects on cell division, membrane effects and apoptosis induction. The changes that T-2 provokes in the biochemical status vary greatly, however, lipid peroxidation is considered as one of the most important. Inducing lipid peroxidation, trichotecenes influence on cell membrane integrity, resulting in different disorders in animals, such as malabsorption syndrome. The pro-oxidant effect of T-2 in many cases may be mediated via influence on the glutathione synthesis and the antioxidant enzymes. For that purpose, 40 day-old broilers were divided in two groups, both given feed and water *ad libitum*. T-2 mycotoxin was dissolved in water and given to the experimental group with daily oral gavages in doses of 0.250 mg per bird for 3 consecutive days. On day four, 5 chicks from each group were sacrificed after total ether anesthesia and liver tissue samples were taken for further analyses. Total gluthatione and superoxide dismutase levels were determined in liver homogenates, as well as TBARS concentration. The results of this study demonstrate that trichothecenes stimulate lipid peroxidation with consequent decrease of GSH content and superoxide dismutase, and increase the level of TBARS in tissue liver homogenate. These data demonstrate that T-2 can adversely affect broiler health. The effects of this toxin may be exacerbated by other factors when under field conditions; hence, the potential detrimental effects of the toxin cannot be dismissed.

Key words: T-2 toxin, lipid peroxidation, liver homogenate, broilers

P41

EFFECTS OF COLD STRESS ON THYROID GLAND IN RATS

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ABSTRACT

Introduction

In rats, thyroid gland is located in the caudal part of larynx bilaterally. The gland, releases hormones for growth and reproduction of the body. The hormones released from thyroid are tetraiyodotironin (T_4) and triiyodotronin (T_3).

Materials and Methods

In this study 24 weeks-old 200-220 gr 48 male and female Sprague-Dawley race rat was used. Rats separated into two groups each of which consists of 24 rats. First group was structured as 12 male and 12 female control group, and the other group was cold stress applied group in the same number male and female. Stress was provided by applying of 4-6 degree cold to animals in the stress applied group, while no process applied to animals in the control group.

Findings obtained at the end of the study were investigated both morphologically and statistical analysis was performed by identifying triiyodotironin (T_3) and tetraiyodotironin (T_4) blood serum hormone levels which were obtained from each group and by identifying follicle cell dimension.

Results

In the study, it was observed that; in male materials of the stress group colloid quantity declined from place to place in some areas, borders of epithelial cells becoming indistinct while colloid completely disappearing in some of the follicles, and nucleus increase in size and forms of these epithelial cells deformed. It was also observed that females have more colloid quantity respect to males in the same group, and their nucleus size slightly decreased.

In the assessments performed on follicle cells it was determined that diameter of cell decreased depending on stress in a significant level respect to control group, and it was also determined that this result is statistically significant ($P<0,01$) in both male and female stress applied groupsStatistically there was no significant change in T_3 level related to stress though cold stress causes an increase in T_3 level in stressed group ($P>0,05$). T_4 levels decreased respect to control group and this decrease was statistically significant ($P<0,05$). Furthermore, it was determined that T_3 values of females higher than of males and T_4 values of males higher than of females in the stressed group.

It was determined that in males of stressed group there was an increase in T_3 and T_4 levels compared to control group while there was an decrease both T_3 and T_4 levels of female of stressed group compared to control group.

In conclusion**,** although in general present findings are accord with literature there were also some certain differences. These differences were possibly originated from factors such as age, sex, environment and husbandry of animals.

Key words: rat, thyroid, cold stress, triiyodotironin (T_3), tetraiyodotironin (T_4)

P42 INFLUENCE OF MICROCLIMATE CONDITIONS ON THE PRODUCED QUANTITY AND QUALITY OF MILK IN THE EARLY LACTATION STAGE IN TIED AND FREE STALL HOUSING SYSTEM

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ABSTRACT

The living conditions, the diet of dairy cows, including the microclimate conditions of the environment influences the quality and quantity of the milk.

The goal of this study was to evaluate the influence of air temperature and relative air humidity on the produced daily milk quantity and its quality (number of bacteria, number of somatic cells) in tied and free stall systems of housing. In this research 40 dairy cows were included for 40 days (April-June, 2013) in the early lactation stage after calving. The milk samples were taken during morning and evening milking with automatic milking in the free system of holding and with manual milking in tied system of holding in the total amount of 50 ml including the preservative.

The average air temperature was 16°C in the tied system of holding with 95% relative air humidity and 20°C in the free system of holding with 96% relative air humidity. The average produced quantity of milk during the morning milking was 15.0 kg for free holding systems, 13.5 kg in tied system of holding. In the evening milking the average quantity of milk was 11.5 kg for free holding system and 11.0 kg for tied system of holding.

The total produced quantity of milk from the two milkings was 26.5 in the free holding system and 24.5 kg in the tied system of holding. There is an insignificant difference of 2 kg between the two types of holding systems. The average number of bacteria in milk at the free system of holding was 609.000/ml and the number of bacteria in milk in the tied system of holding was 154.000/ml. The number of somatic cells in the milk was 25.000/ml for the free holding system and 78.000/ml for tied system of holding.

The microclimate condition had a positive and beneficial influence in daily produced quantity of milk during the morning milking in the free stall system. For the evening milking the quantity of produced milk is the same from both systems. The total daily quantity of milk from the two milkings was higher in the free stall system in the quantity of 2.0 kg.

Key words: milk quality, somatic cells, microorganisms, lactation

P43

THE KAP 1.1, KAP 1.3 AND K33 GENES POLYMORPHISMS IN CHIOS, KIVIRCIK AND AWASSI SHEEPS

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ABSTRACT

Since the improvement of linkage maps, many researchers have managed identifying QTL affecting economically important traits. At the beginning investigations focused on identifying QTL affecting production traits, however traditional selection ways have been effectual in development production traits in sheep without the DNA marker information.

The aim of the study was to determine to investigate the effects of KAP 1.1, KAP 1.3 and K33 genes which play important roles in determining phenotypes for the wool fibre quality and productive traits in Chios, Kivircik, Awassi sheep. In the study unrelated 50 Chios, 50 Kivircik and 50 Awassi were used. The genomic DNA was isolated from whole blood using standard salt-out protocol. Blood samples were taken into 2 ml sterilized tubes with EDTA from V. Jugularis for having genomic DNA samples. Regions were amplified for the determination of KAP 1.1, KAP 1.3 and K33 polymorphisms in DNA samples by using PCR. Gene polymorphisms and chi-square test was used to determine whether the populations are in Hardy-Weinberg equilibrium using by POPGENE32 software.

Key words: KAP 1.1, KAP 1.3 and K33 genes, sheep, polymorphism

P44 EVALUATION OF THE ANTIMICROBIAL POTENTIAL OF THE BUTTON MUSHROOM *AGARICUS BISPORUS*

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ABSTRACT

Introduction

Farming methods have changed dramatically in recent decades. Production has become increasingly industrialised, with larger numbers of animals stocked at higher densities, coupled with breeding and feeding strategies aimed at maximising production. These changes have a huge impact on the welfare of farmed animals we rear for food and can increase the risks to people and animals from some zoonotic diseases.The arising awareness of the relationship between diet and diseases has evolved the concept of "functional foods". A food may be considered to be functional if it contains a food component (whether a nutrient or not) which affects one or more identified functions in the body in a positive manner, which are in different name forms, e.g. dietary supplements, nutraceuticals, medicinal foods and foods for specific health uses.Fears from possible risk for human health because of use and/or misuse of antibiotics growth promotors in food for animals for consumption, led to a ban of their use in the EU (Regulation EC No. 1831/2003). The scientific community, in searching for new therapeutic alternatives, has studied many kinds of mushrooms and has found variable therapeutic activities such as anticarcinogenic, anti-inflammatory, immuno-suppressor and antibiotic among others. In accordance with Article 17 of Regulation (EC) No 1831/2003 on additives for use in animal nutrition, the Commission has established a Community Register of feed additive, according to which the recommended natural animal feed supplement is *Agaricus*

bisporus, and its extract CoE 543. The purpose of this research was to evaluate the antimicrobial potential of the button mushroom *Agaricus bisporus*.

Materials and Methods

Experiment were conducted on 60 Yorkshire x Landras crossbred pigs, after weaning. The animals were divided in two dietary groups, one group feed by basal diet and the other group with diets containing 1% dry supplement of *Agaricus bisporus*, during the 5 weeks. Upon completion of the experiment, microbiological examinations of jejunal aspirates was done.

Results

At the beginning of the experiment colony-forming unit in jejunal aspirates of all animal was approximately equal, 10^7CFU/ml. Upon completion of the experimentCFU were significantly decreased (10^5CFU/ml) in animals treated with *Agaricus bisporus* supplement compared to pair-fed controls (10^8CFU/ml).

Conclusions

Weaning is a traumatic event for piglets regardless of their age. The change in nutrition from a largely milk based diet to a pelleted ration affects gut local immune status and gut microflora. In addition, changing the accommodation and mixing piglets can all have consequences on the piglet physical, nutritional, immunological, and behavioral status. The data provided by this study illustrate the antimicrobial potential ofbutton mushroom *Agaricus bisporus* as well asbetter production results in animals treated with *Agaricus bisporus* supplement.

Key words: welfare, Agaricus bisporus, pig, weaning

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P45 ADDITIONAL PART OF M. SARTORIUS IN THE DOG AND ITS IMPORTANCE IN THE FEMORAL HERNIA SURGERY

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M. sartorius is superficial muscle that lies at the cranio medial surface of the thigh, and in the dog is composed of the cranial and caudal part. Pars caudalisof the M. Sartorius does not cover the femoral canal but represents its caudal edge which allows palpation of the heart rate. Anatomical section of the pelvic limb for student education, established an additional part of this muscle in the dog. Since the femoral hernia in dog cures by resection of part or the integral parscaudalis of M. sartorius, we believe that finding anomalie in the structure of these muscle have clinical significance and contribution.

Key words: dog, m. sartorius, femoral hernia

RABIES IN A COW IN TURKEY

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A 5-year-old female Holstein cow was admitted to Kirikkale University Veterinary Faculty, Department of Internal Medicine clinics with complaints of anorexia and thursty. Upon physical examination; abdominal pain, excess salivation, tenesmus, strabismus, kyphosis, stranguria, dysphagia, hydrophobia, and high rate pulsation were detected. Upon the hematological examination all parameters were not in physical ranges. Glucose were detected in urine by the using of speed test strip. Cow was died on the 8th day under quarantine period. The aim of this case was to rabies should be taken into account in cattle showing anorexia and apathy.

Key words: cow, rabies, anorexia, thursty

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1. Cracknell V and Flogdell K 1988, Proceedings of the XV WBC pp505-508
2. Bradley AJ *et al*, Survey of the incidence and aetiology of mastitis on dairy farms in England and Wales, Vet. Rec. 2007 Feb 24;160(8): pp253-7



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