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LIVESTOCK PRODUCTION: RECENT TRENDS AND FUTURE PROSPECTS

Abstracts

A COMBINATION OF WHEAT CEREAL OUTER LAYER THERMO-MECHANICAL PROCESSING AND BIOCONVERSION FOR SUSTAINABLE VALUE-ADDED FEED STOCK PRODUCTION

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    Wheat (*Triticum* spp.) is one of the most popular crops worldwide; however, the most popular part of wheat is the endosperm, and, until now, other layers have been used as low nutritional value feed stock. However, it should be pointed out that most of the bioactive compounds in cereals are concentrated in the outermost tissues. On the other hand, undesired compounds (mycotoxins, etc.) also occur in these fractions. For all these reasons, pre-treatment technologies to improve the properties of wheat bran (WPBP) are being studied, of which fermentation with selected lactic acid bacteria (LAB) strains is the most popular for WPBP valorization. Also, possible strategies for WPBP functionalization include extrusion. Extrusion causes many structural, physicochemical, and microbial transformations of stock, e.g., depolymerization of starch, denaturation of proteins, oxidation of lipids, decontamination of bacteria, etc., and these changes are influenced by extrusion process parameters. Until now, no study on the combination of extrusion and fermentation, especially with LAB strains which possess antimicrobial properties, has been published in the literature to date. The aim of this study was to evaluate the influence of combining extrusion and fermentation (with *L. casei* and *L. paracasei* strains) processes on the chemical and biosafety of WPBP. An extrusion experiment was performed by testing two different temperatures (115°C and 130°C) and three different speeds of extruder screw (16 rpm, 20 rpm, and 25 rpm). Very prospective results were obtained, and it was concluded that the appropriate extrusion parameters and LAB strain selection lead to higher formation of L(+) isomers and lower WPBP microbial contamination. Extrusion, as well as extrusion in combination with fermentation, reduces total biogenic amine content (on average, 2 times). The lowest mycotoxin concentration was found in $W_{ex130/screwspeed20}$ and $W_{ex130/screwspeed25}$ samples fermented with both LAB strains. Finally, the combination of extrusion and fermentation with *L. casei* and *L. paracasei* strains can be confirmed as a prospective innovative pre-treatment for WPBP, potentially capable of enhancing its composition and safety characteristics.

    Keywords: wheat bran, extrusion, fermentation, mycotoxins, safety.

THE EFFECT OF DIFFERENT GENETICAL ARCHITECTURES FOR GENOMIC PREDICTION OF CARCASS FATNESS BY MACHINE LEARNING IN CATTLE

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Carcass fatness in cattle is an important phenotype in the consumer markets, and it plays a key role in metabolism of meat flavour properties including colour and tenderness. More recently, literature has emerged that offers findings about molecular markers (for instance, single nucleotide polymorphisms, SNPs,) in association with the phenotypes of interest termed as genomic prediction (GP) and genome wide association study (GWAS) [1]. This study set out to investigate the usefulness of various ML models for GWAS and GP of carcass fatness obtained by ultrasound technology. The dataset consisted of 1439 Nellore cows genotyped for 35237 single nucleotide polymorphism (SNP) for the phenotype of carcass fatness. Bayesian and machine learning (ML) analyses were based on the Bayesian ridge regression (BRR), Bayesian lasso (BL), Bayes A (BA), Bayes B (BB) and Bayes Cπ (BC), Bayesian least absolute shrinkage and selection operator (BL), reproductive kernel Hilbert space (RKHS), support vector machine (SVM), genomic best linear unbiased prediction (GBLUP) and elastic net (EN). EN, SVM and RF resulted in the lowest value of predictive ability of carcass fatness. The highest GP accuracy was obtained from GBLUP (0.9225). Linkage disequilibrium pruning resulted in a small variation on the accuracy results. Employment of pedigree and genomic kinship matrixes on the accuracies was also discussed.

Keywords: carcass fatness, cattle, genome wide association study, genomic prediction, machine learning.

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THE INVESTIGATION OF QUANTITATIVE GENETIC PARAMETERS OF CARCASS FATNESS IN *CATTLE*

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The main aim of this study was to investigate various genetic parameters for carcass fatness (CF) in cattle (n = 1439) located across three Brazilian states, born between 2009 and 2018 [1]. Ultrasound measurements of carcass fatness (corrected for age) were obtained by a private company.

Data were analyzed using Bayesian segregation analyses. Gibbs sampling was used to make statistical inferences on posterior distributions; inferences were based on single run of the Markov chain for each trait with 500 000 samples, with each 10th sample collected due to the high correlation among the samples [2]. Posterior mean (and SD) of major gene variance for CF was 8.7004 (1.5161), for additive gene effect, it was 3.7722 (0.0601), and for dominant effect, it was 7.0851 (0.2656). The highest posterior density regions for CF did not include zero which supported the evidence for a major gene.

Keywords: bayesian analyses, carcass fatness, major gene analyses, segregation analyses.

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THE INVOLVEMENT OF *IRS1* GENES IN MILK PROTEIN SYNTHESIS IN DAIRY GOAT MAMMARY CELL

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Goat milk has been consumed by humans since ancient times and it is an important source of nutrition for humans of all ages. The content of milk protein is the prodominant evaluation of milk qualitity. Milk protein synthesis is highly regulated by insulin and the gene of insulin receptor substrate 1 (*IRS1*) plays a central role in insulin signal transduction.

Material and methods. The gene expression of *IRS1* was reduced by RNA interference (RNAi) in goat mammary epithelial cells (GMECs) and evaluated by RT-qPCR. The content of casein was determined by ELISA. Comparative proteomic analysis was adopted to analyse the protein profile. Results. The content of casein in GMECs significantly changed when *IRS1* expression was reduced. Several differential expression proteins were identified.

Conclusion. The involvement of *IRS1* regulated the milk protein synthesis in GMECs.

THE EFFECT OF DAIRY COW STATURE AND LIVE WEIGHT ON THE PRODUCTIVITY AND LONGEVITY TRAITS

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Dairy cow potential longevity exceeds 15 years, but in modern dairy farming, due to various internal and environmental factors, cattle lifespan has been reduced to little under 3 closed standard lactations. Although it is possible to breed exceedingly bigger animals, it comes with numerous negative effects, one of which is a shorter lifespan. In modern dairy farming, it is not recommended to have Holstein Black and White breed cows taller than 150 cm, as increased stature usually leads to leg problems, increased treatment costs and possible forced culling from herd, which increases the costs per animal and increases the demand of replacement heifers. The aim of our paper was to determine the effect of dairy cows' stature and live weight at the beginning of the first lactation on cow longevity and lifetime milk productivity. Analysed data were collected from the Latvian Data Centre milk recording records about 272 633 Holstein Black and White, Holstein Red and White and Red breed group dairy cows born in timespan form year 2010–2019. For study purposes, we distributed dairy cows in 4 separate groups, depending on their live weight and stature in the first 100 days of the first lactation. To characterize total cow milk productivity, we used energy corrected milk (ECM). All analysed dairy cows during the first standard lactation produced 7179.7 ± 4.05 kg ECM and showed a tendency to increase the productivity level until the third lactation (8358.3 ± 7.34 kg ECM), but in later lactations, cows produced significantly ($P < 0.05$) less (accordingly 8180.4 ± 9.18). The average lifespan in the analysed cow population was 1925.4 ± 1.31 days (approximately 5.27 years) with the average lifetime productivity of 24 352.3 ± 26.67 kg ECM (average per 1 life day 11.9 ± 0.01 kg ECM). Cows born in the timespan 2010–2019 demonstrated a strong tendency to gradually increase live weight and stature not only in the firstᵗ, but also in the third lactation. In the meantime, the age at first calving decreased from 825.5 ± 0.80 days for cows born in the year 2010 to 766.3 ± 0.66 days for cows born in the year 2019. Different researchers explain this tendency with increased dairy cow growth potential, body capacity and metabolism. This combination of factors also gives a significant impact on the dairy cow milk productivity level [1]. Cows with live weight in the first lactation > 600 kg were characterized by a significantly ($P < 0.05$) shorter lifespan of 1758.8 ± 2.24 (approximately 4.8 years), but a higher lifetime milk productivity (24 757.9 ± 58.99 kg ECM) than in other studied groups, which could be explained by that fact that cows with higher live weight have a grater body capacity that can lead to increased dry matter intake and milk productivity. Cows with a bigger stature (above 151 cm) were characterized by a significantly shorter lifespan (1694.3 ± 2.40) and lifetime milk productivity (22 557.9 ± 61.34) that could be explained by the increased pressure on cow feet and claws, which leads to increased treatment costs and premature culling from herds.

Keywords: live weight, stature, lifespan, milk productivity.

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THE INFLUENCE OF THE BULL AND BULL LINE ON THE CONTENT OF FATTY ACIDS IN THE MILK OF BULLS' DAUGHTERS

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In recent years, increasing attention has been paid to improving the quality of dairy products. Therefore, the selection of animals according to the composition of milk fatty acids is of increasing interest. [1] The composition of milk fat has a significant influence on dairy products, shelf life and quality of milk processing. Unsaturated milk fatty acids have the greatest impact on human diet and health. [2] Myristic (C14:0) and palmitic acid (C16:0) are associated with cardiovascular disease due to increased blood cholesterol, and shorter chain saturated fatty acids (C4:0 C12:0) are associated with a positive health effect [3]. Milk producers are looking for methods to improve the physical and functional properties of cow's milk and to optimize the fat composition of milk for human health. Increasing knowledge of milk fatty acid synthesis, revealing the genetic background of milk fatty acids, may help to modify the fat composition of cow's milk [4].

The research was carried out in 2021–2022 at the Lithuanian University of Health Sciences, Department of Animal Breeding and Practical Training and Research Center of the Lithuanian University of Health Sciences. The milk samples of 263 cows of the Holstein breed bred in Lithuania were studied. The objective of this study was to focus on the genetic factors that affect the content of fatty acids in milk. In this study, cows were grouped according to the cow's father bull and the line of a bull. Statistical characteristics were calculated using statistical software SPSS 25.

There was no statistically significant impact on the fatty acids content in the milk of offspring's during the analysis of the influence of genetic factors on content of fatty acids, but the amount of fatty acids in the milk of the offspring of different bulls varies.

The biggest differences were detected in the content of unesterified fatty acids, preformed, saturated and unsaturated fatty acids in milk ($P > 0.05$). Meanwhile, the difference in the amount of fatty acids in the milk of the progeny of bulls of different lines was not significant. The most significant differences were in the amounts of non-esterified fatty acids, preformed, unsaturated fatty acids and mono-unsaturated fatty acids, and the lowest differences were found in the amounts of polyunsaturated and palmitic acids in milk ($P > 0.05$).

Keywords: cows, fatty acids, milk production, bulls.

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STUDY OF ANTIMICROBIAL EFFICACY OF TEAT DIP SOLUTIONS WITH POVIDONE IODINE AND CHLORHEXIDINE DIGLUCONATE AGAINST *P. ZOPFII* ALGAE ISOLATED FROM BOVINE MASTITIS MILK

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Prototheca zopfii is an environmental pathogen that causes chronic bovine mastitis. Reproduction of the pathogen in the mammary gland and regional lymph nodes causes granulomatous inflammation of the udder, damage to mammary parenchyma, atrophy of affected udder quarters, reduction of milk yield, and an increase in the count of somatic cells to 1 000 000/mL [1, 2]. *Prototheca* mastitis usually occurs due to contact with contaminated water sources or equipment [3]. There are no effective or approved treatment methods for *Prototheca* mastitis [4]. *Prototheca* survives pasteurization and is resistant to disinfection with chlorine compounds [5]. Frequent intramammary antibiotic infusions may contribute to the development of *Prototheca* infection in the udder [6]. It is difficult to eradicate the pathogen from the herd, infected cows are eliminated, and the only disease control measure is prevention.

The aim of this study was to determine in vitro sensitivity of *P. zopfii* isolated from bovine mastitis milk to post-milking teat dips *Profidip Iodine* and *Profidip Chlorhexidine.*

Research methods. The research was conducted in the Laboratory of Microbiological Research, Institute of Microbiology and Virology, Faculty of Veterinary Medicine, Lithuanian University of Health Sciences. *Prototheca zopfii* isolated from the milk of cows with chronic bovine mastitis was used. The research was carried out using the agar diffusion method. The activity of the teat dips was assessed in Sabouraud agar (BD DifcoTM, Franklin Lakes, NJ, USA), under aseptic conditions. Wells were formed in the inoculated medium in a Petri dish, each of which were filled with 100 μL of tested dips. The Petri dishes were incubated at 30°C. After 48 hours of incubation, antimicrobial activity was assessed by measuring the diameter of clear zones around wells.

Results. The study determined that both teat dips under investigation exhibited anti–*Prototheca* activity. *Profidip Chlorhexidine* inhibited the growth of *P. zopfii* strains, with an average clear zone at 19.1 ± 0.6 mm. *Profidip Iodine* was reliably more effective ($P < 0.05$), with an average clear zone at 31.4 ± 0.9 mm, and scanty algal growth on media.

Conclusion. Post milking teat dips *Profidip Iodine* and *Profidip Chlorhexidine* inhibit in vitro growth of *P. zopfii* strains. The teat dips exhibit different anti–*Prototheca* activity, with *Profidip Iodine* proving more effective ($P < 0.05$) against the algal cultures than *Profidip Chlorhexidine.*

Keywords: teat dip, P. zopfii, antimicrobial efficacy.

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A COMPARATIVE ANALYSIS OF THE PHYSICAL, CHEMICAL, AND SENSORY ATTRIBUTES OF CONVENTIONAL AND FREE-RANGE BROILER CHICKEN MEAT

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Investigating the physical-chemical traits and sensory qualities of conventional and free-range broiler meat was the aim of the current study. Broiler chicken carcasses from conventional and free-range broiler chickens were obtained from local suppliers. The analyses were performed at the Institute of Animal Rearing Technologies, Lithuanian University of Health Sciences (Kaunas, Lithuania). Evaluation of broiler chicken meat quality indices was performed after 48 hours of cooling at 4°C. The raw breast (*Pectoralis major*) meat was used for physicochemical characterization and sensory evaluation. Protein, fat, ash, dry meter content, pH, drip loss, water holding capacity, cooking loss, and color coordinates (CR-400, Minolta Camera, Osaka, Japan), measuring L* values of lightness, a* values for redness and b* values for yellowness were determined in broiler chicken breast meat. For sensory analysis, descriptive tests, and emotional expressions with Face Reader 6 software (Noldus Information Technology, Wageningen, Netherlands), 200 g of chicken breast meat was boiled at 100°C for 20 minutes. Ten participants were selected for the sensory analysis team. A one-way analysis of variance (ANOVA) was used to analyze the data. The Tukey test for comparison was used, with a 95% confidence interval of significance. Samples were prepared in triplicate for all the analyses done.

After examining the physical-chemical traits and properties of breast meat from conventional and free-range broilers, meat cooking losses were found to be statistically significant in both groups (difference between groups: 3.7% ($P < 0.05$); water holding capacity: 1.36% ($P < 0.05$)). Color indicators showed that the external surface of free-range broiler chicken breast meat was darker than conventionally reared meat. The study of sensory attributes revealed differences between samples in smell, color, tenderness, and juiciness. Consumers showed higher acceptability for free-range broiler meat in comparison with conventional. Patients for free-range broiler meat expressed higher "happy", "surprised", and "angry" emotions ($P \leqslant 0.05$)

Keywords: broiler chicken, free-range, conventional, meat quality, sensory attributes.

CONNECTIONS BETWEEN YOUNG HORSE PERFORMANCE AND TARSAL JOINT RADIOLOGICAL IMAGE MEASUREMENTS

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The study aimed to assess connections between tarsal dimensions and performance to understand changes connected with orthopedic status. Warmblood horses (1249 ± 114.6 days) on performance tests were x-rayed (19 stallions and 50 mares) and 14 measurements of the hock joint area were made (VetRayVision). The measurements were correlated with performance results: 3 basic body measurements, 3 conformation evaluations and performance traits (gaits and jumping: 2 in hand and 9 under saddle). The Pearson correlations ($P < 0.05$) of the tarsal measurements with basic body measurements were medium (0.3–0.6). The highest value was calculated between overall tarsal length and height at withers. Two significant correlations between conformation traits and the length of central/third tarsal bones were positive and low, while the others were low and negative (12 cases). The connections between measurements and movement in hand were significant for 17 cases from 28 possible and obtained low values (up to 0.35). The same level of correlation (0.10–0.38) was calculated for movement under rider (48 cases for 112 possible). Free jumping correlated only with one tarsal measurement (−0.25). Only one trait – rideability – was not connected with any tarsal measurement. The obtained results suggest that the meaning of the size of the tarsal joint is underevaluated by the judges' committees as the greater size of tarsal measurements is evaluated lower in the conformation evaluation.

Keywords: horse, tarsal joint, size measurement, performance.

ASSOCIATION BETWEEN DAIRY BULLS' FERTILITY INDEX AND COWS' CONCEIVING SUCCESS

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Ensuring optimal conditions can increase the chances of successful breeding and maintain a healthy and productive herd. The successful conceiving of cows involves various factors [1]. One of the ways to improve cows' fertility in the herd is to use the bulls' fertility index. The main trait of the fertility index is a non-return rate [2]. The aim of the study was to determinate how the fertility index is associated with a possibility to conceive from the first and the second insemination after spontaneous ovulation (AI) and timed artificial insemination (TAI). For this purpose, 243 healthy dairy cows with similar yielding (9000–10 000 kg per lactation), lactation (2–4) and BCS (3.75–4) were inseminated by different bulls with the fertility index −0.4, −0.1, 0.4 and 1.3. AI was performed after spontaneous oestrus when the standing reflex was determined (cows' oestrus was monitored 3 times per day for 15 min). TAI cows were inseminated by G7G protocol. Bulls with a 1.3 fertility index had a statistically significantly higher possibility to conceive cows after the first AI comparied with bulls with a 0.4 fertility index. AI success between cows inseminated by bulls with different fertility indexes was not statistically significant in the second AI. Insemination success was highest when cows were inseminated after TAI by a bull with a fertility index 1.3. Cows inseminated by the mentioned bull had a 13.9% and 12.5% higher statistically significant possibility to conceive after TAI compared with cows inseminated by bulls with the fertility index −0.4 and 0.4, respectively. Simillar differences in the possibility to conceive were determined between bulls after the second TAI. In conclusion, a higher fertility index does not always mean better conceiving success. Bulls with a higher fertility index are more appropriate for insemination for cows after AI and TAI.

Keywords: fertility index, spontaneous, TAI, AI, conceiving.

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DISTRIBUTION AND POTENTIAL ROLE OF AQUAPORINS IN SPERM: LESSONS FROM A BOVINE STUDY. PRELIMINARY RESULTS

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In view of the need to search for new parameters assessing semen quality, allowing for a precise evaluation of male reproductive potential, studies are conducted with the main goal of identifying and analyzing the expression of aquaporins (AQPs) in bovine sperm. AQPs are small transmembrane proteins that, according to many researchers, may be involved in maintaining proper viability and motility of sperm [1, 2]. The latest data indicate that these proteins may also play an important role in the process of sperm cryopreservation [3]. The research presented here is conducted on cryopreserved semen samples from 20 healthy and sexually mature Polish Holstein–Friesian black and white bulls. The ejaculates were classified into two groups: high–quality semen (n = 10) and low–quality semen (n = 10), based on the evaluation of sperm concentration, motility, vitality, morphology, hypoosmotic swelling test and mitochondria activity. Using immunofluorescence (IF), a detailed localization of all AQPs potentially present in bovine sperm is being carried out. Subsequently, their expression will be analyzed using Western blot (WB), and the results will be verified using high–performance liquid chromatography coupled with mass spectrometry (HPLC–MS). These analyses will be accompanied by measurements of selected metabolites (free amino acid and fatty acid profiles, biogenic amines, cholesterol, and malondialdehyde levels) both in sperm and seminal plasma. To date, the locations of AQP3, AQP7 and AQP8 have been identified and tentatively determined using IF and WB. Meanwhile, the presence of AQP1, AQP4, AQP5, AQP9 in bovine sperm was excluded. Currently, research on the search for a relationship between their expression and semen quality is ongoing. The presence of other aquaporins in bovine sperm, including AQP0, AQP11 and AQP12 is also verified.

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ANALYSIS OF MILKING INDICATORS OF DIFFERENT COW BREEDS

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Cow milking is considered as a laborious and time-consuming job at livestock farms [1]. The milking process represents one of the most important tasks on a dairy farm. On farms with conventional milking systems, it accounts for roughly a third of farm's total labor demand [2]. Increasing the milk flow rate at which milking is terminated can shorten milking time and increase milking efficiency [3]. It is important that milking speed-related traits present high heritability estimates for higher selection responses in breeding programs aiming to breed for more efficient and adapted animals for milking systems [4].

The aim of the study was to analyze the milking indicators of different cow breeds.

Materials and methods. In the study farm, cows are kept loose all year round, in a modern cold-type barn. During summer, the animals are not grazed, and they are fed a complete diet that meets their physiological needs. Cows are milked 2x20 side-by-side in a milking parlour. The milking data of 420 cows were collected, 140 of which were of the Lithuanian Red breed, 62 of the Swedish Red and White and 218 of the White and Red Holstein breeds. For the analysis of the data, statistical indicators were calculated for each evaluated trait (milk yield at milking, milking duration, average milk flow, maximum milk flow, milk yield in the first 2 minutes, milk flow in time intervals: 0–15 s, 15–30 s, 30–60 s, 60–120 s): arithmetic mean, mean error and statistical reliability of the data (P). The obtained results were considered statistically significant when $P < 0.05$.

Results and conclusion. We found that Lithuanian Red cows produced 0.51 kg more milk than White and Red Holsteins and 0.29 kg more milk than Swedish Red and White. The milk yield in the first two minutes of milking was 0.31 kg higher in the Lithuanian Red cows than in the Swedish Red and White cows and 0.38 kg higher than in the White and Red Holstein cows. The average milking time was 6.19 minutes. The milking time of the White and Red Holstein cows was 0.11 min longer than that of the Lithuanian Red cows ($P < 0.01$) and 0.17 min longer than that of the Swedish Red and White Holstein cows ($P < 0.01$). The average milk flow of Lithuanian Red cows was only 0.03 kg/min higher than that of Swedish Red and White and 0.11 kg/min higher than that of White and Red Holstein cows. The highest milk flow rate of the Lithuanian Red and Swedish Red and White breeds was 0.22 kg/min higher than that of the White and Red Holstein breed. The milk flow rates of the Lithuanian Red cows at the time intervals 0–15 s, 15–30 s, 30–60 s and 60–120 s were found to be 0.05, 0.23, 0.11 and 0.17 kg/min higher than that of the Swedish Red and White cows, and 0.04, 0.21, 0.21 and 0.2 kg/min higher than that of the White and Red Holstein cows. In conclusion, the Lithuanian Red cows on the farm produced more milk, milked faster and had higher milk flows.

Keywords: cows breed, milking duration, milk flow rate.

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ANALYSIS OF POPULATION STRUCTURE AND INBREEDING IN THE LATVIAN HEAVY WARMBLOOD HORSE POPULATION

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Latvian heavy warmblood horses (LSB) breeding program was developed in 2004. It is aimed at preserving this type, because the number of these horses is small and the status of the local endangered breed has been accepted. Conservation of the native population is important when thinking about genetic diversity today and in the future [1]. This study is relevant for those horse breeders who are working on the preservation program of the LSB horses breed in order to more efficiently plan their breeding work. The purpose of the study was to analyse the population structure and inbreeding of the LSB horse population.

The Latvian Horse Breeders Association provided the pedigree data of LSB horses. For LSB, the reference population included 257 females and 117 males that were alive at the time of data selection. For reference animals in the data set as many ancestor generations as possible were included. The software POPREP was used for the analysis of the population structure and the inbreeding coefficient.

In the LSB population, the average age of the breeding mares and stallions at the time of foal birth was between 8 and 15 years. The number of offspring raised for breeding varied from year to year, and during the last 10 years, the highest number of foals (24 foals) that were raised for breeding was in 2016.

In the LSB population, the average generation interval within the last 10 years was similar for stallions and mares (8.8 years on average). There were only 5 stallions in the LSB population, who produced 12–18 offspring during their lifetime; the contribution of the other stallions to the production of breeding animals was significantly lower. The largest number of offspring raised for breeding from one mare was 5 animals.

According to the pedigree analysis, the five generations' pedigree completeness was above 90% for animals born from 2000 and the first generation 100% completeness starting from 1985. Between 2000 and 2019, 374 animals were born, and the proportion of non–inbred animals in LSB population was only 9.09% (34 animals). The highest proportion of inbred animals (85.56 %) had an inbreeding coefficient up to 5%. There were some animals with inbreeding higher than 11%.

In conclusion, breeding organizations should control the LSB offspring inbreeding level and not mating related stallions and mares. The inbreeding coefficient can be decreased for the LSB population. Well–completed pedigree information will make it possible to control the level of inbreeding of the offspring.

Keywords: inbreeding, horse population, heavy warmblood, population structure.

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DETERMINATION OF *TG5* GENE POLYMORPHISM AND ITS INFLUENCE ON PRODUCTIVITY TRAITS OF BEEF CATTLE REARED IN LITHUANIA

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Thyroglobulin (TG) is the precursor of T3 and T4 thyroid hormones, which have an important role in metabolic regulation and affect lipid metabolism [1]. The results of various scientific studies have shown that the *TG5* gene should be considered a functional and positional candidate gene that affects the accumulation of fat in the cattle body [2]. Thus, a single-nucleotide polymorphism located in the 5 untranslated region of this gene is used for marker-based selection aimed at increasing marbling. At present, scientists are actively studying the genetic determination of the meat production characters of cattle using DNA markers. This allows supplementing traditional breeding methods with the method of selecting animals based on desirable genetic markers. Thus, it becomes possible to significantly accelerate the selection process by improving the genetic potential of cattle breeds [3, 4]. The aim of this study was to investigate the prevalence of *TG5* gene polymorphism and to determine its influence on productivity traits of beef cattle reared in Lithuania. Cattle hair follicle samples were collected from 30 bulls of Angus, Limousin, Aubrac and Charolais breed. Hair samples and the data on productivity trait records were obtained from Šilutė control bulls feeding station. Bovine genomic DNA was extracted from hair follicles using Chelex DNA extraction method. Polymorphism of *TG5* locus was identified using a PCR-RFLP method. PCR product of *TG5* gene was digested with *BStx2I* restriction nuclease. Investigation of polymorphism of *TG5* gene showed that allele C (frequency − 0.617) and genotype CC (frequency − 0.533) were the most common in the analyzed population of beef cattle. Meanwhile, the heterozygous CT genotype was rarest, with a frequency of 0.167. Evaluating the observed and expected heterozygosity in the investigated group of animals, the observed heterozygosity was found to be lower than expected, indicating an insufficient amount of genetic diversity in the loci studied. The difference was statistically significant ($P < 0.01$). When calculating the influence of *TG5* gene polymorphism on cattle productivity traits, it was observed that this polymorphism had a statistically significant ($P < 0.05$) effect for many productivity traits. The data on the influence of genotypes showed that cattle of CC genotype had the higher live weight, hot carcass weight and carcass weight and the highest calculated averages of the three indicators (average daily weight gain/live weight/age in days; average daily weight gain/warm carcass weight/age in days; and average daily weight gain/ carcass weight/age in days) than animals of CT or TT genotype. The data was statistically significant ($P < 0.05$). In conclusion, the results showed that polymorphism of the *TG5* gene influences many productivity traits of beef cattle.

Keywords: cattle, *TG5* gene, polymorphism, PCR-RFLP.

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EVOLUTIONARY TRANSITION FROM CONVENTIONAL TO EAZA-RECOMMENDED METHOD OF *TENEBRIO MOLITOR* CULTIVATION

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This study focuses on enhancing the efficiency of cultivating and breeding mealworms (*Tenebrio molitor*) to serve as a nutritious animal feed, particularly beneficial for captive exotic animals. The research aimed at transitioning from a traditional cultivation approach, which relied heavily on visual assessments to determine mealworm developmental stages and sieving timing, to a protocol recommended by the European Association of Zoos and Aquaria (EAZA). The EAZA-endorsed method prescribes a fixed egg-laying period of 7–10 days and enforces strict controls on the age and size of mealworms at harvest. Despite maintaining consistent cultivation parameters such as relative humidity, ambient temperature, and nutritional regime across both methods, the transition required significant adjustments. Key changes included moving to a complete substrate replacement at the end of each ten-week cycle and initiating synchronized breeding cycles to avoid overlapping developmental stages, with only the beetles being transferred to new containers to facilitate egg-laying. However, achieving the EAZA standard of reducing the egg-laying time to a maximum of 10 days for optimal productivity is an ongoing objective. While collecting data for ten months and evaluating the amount of mature mealworms during the sieving process, a notable increase ($P < 0.05$) in larval yield under the EAZA guidelines compared with the traditional mixed-stage breeding approach was determined. Although full adherence to EAZA standards is yet to be achieved, our findings underscore the potential of these advanced mealworm cultivation and breeding techniques in improving animal feed production.

Keywords: *Tenebrio molitor,* mealworm beetle, output augmentation, breeding method.

DETERMINING THE OPTIMAL LACTATION PHASE OF A COW FOR CHANGING THE MILKING SYSTEM FROM CONVENTIONAL TO AUTOMATIC

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Several studies have proven that changing the milking system from a conventional to an automatic one has a positive effect on the milk yield of Holstein–Friesian cows [1]. One of the reasons for this increase in milk yield is the increased number of milkings performed by cows [2]. However, there are no reports which indicate the phase of lactation favorable for changing the milking system. The aim of the study was to determine the time of lactation for changing the milking system from conventional to automatic. The study included 191 cows from the Polish Holstein–Friesian breed in 7 different herds, where farms were equipped with an automatic Lely Astronaut A4 system. The milking data from these cows were collected for 4 years from 2010 to 2014. The statistical analysis of the data was performed by using the analysis of variance. The significant differences between the selected groups were established by using the Scheffe test (SAS software). Based on statistical analysis, it was seen that the optimal lactation phase in which the milking system was changed had a highly significant effect on the yield of primiparous milk. The study showed that in the primiparous group, the highest milk yield (10 125.6 kg) was found in those who experienced a change from conventional to automatic milking between the 101–200th day of lactation. On the other hand, the lowest milk yield was found in primiparous cows who started automatic milking after the 200th day of lactation (8540.6 kg). The study did not show the lactation phase in which the milking system was changed to the milk yield of cows in the second lactation. It should be emphasized, however, that the best effects were observed among cows that were milked from the middle (101–200 days) of lactation (10 278.0 kg).

Our findings have shown that the change of the milking system is more effective if the milking system is changed from a conventional milking system to an automatic milking system during the 101–200 days.

Keywords: automatic milking system, Holstein–Friesian, milk yield, lactation phase.

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HERITABILITY OF POLISH HOLSTEIN-FRIESIAN COWS' MILK ELECTRICAL CONDUCTIVITY RECORDED BY MILKING ROBOTS FOR INDIVIDUAL UDDER QUARTERS

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Some research results suggest that the electrical conductivity of cow's milk (EC) could be successfully used in breeding programs as an additional indicator of mastitis [1]. The electrical conductivity increases in milk produced by cows with mastitis due to damage to cells in the mammary tissue [2]. Roy et al. [3] have reported an increase in EC of milk samples from mastitis. The aim of the study was to estimate the daily heritability EC of milk from quarter milkings of Polish-Holstein-Friesian cows milked in barns equipped with milking robots.

A total of 608 984 daily records of EC for the first and the second lactation were related to 2432 cows from 23 herds. The EC heritability was estimated individually for each udder quarter, i.e. left front – LF, right front – RF, left rear – LR, right rear – RR. Univariate random regression model (RRM) and fourth-order Legendre polynomials for the regression on the number of milking days (from test day 5 to test day 305) were applied. The heritability was estimated using the Wombat package [4].

It was shown that the average EC of milk 6.92 mS. The value of the trait varied depending on the quarter of the udder and the day of lactation, ranging from 6.92 mS (RF, RR) to 6.93 mS (LF, LR). Daily heritability indicators of EC showed high variation during lactation and udder quarters. The heritability values ranged from 0.106 to 0.353 for LF, from 0.062 to 0.253 for LR, from 0.103 to 0.376 for RF, and from 0.113 to 0.381 for RR. It was recorded that the average EC heritability for four udder quarters and the entire first and second lactation was at a level of 0.191. The corresponding indices determined for individual quarters of the udder were: LF – 0.253, LR – 0.161, RF – 0.168, RR – 0.181.

Summing up the results of the research, it should be stated that the heritability of milk electrical conductivity changed depending on the analysed quarter of the udder. At the same time, these results suggest that the best response to selection in this respect may be expected in the case of the left front quarter.

Keywords: automatic milking system, Holstein-Friesian, electrical conductivity, heritability.

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THE VAGINAL MICROFLORA IN ANESTRUS PERIOD OF HEALTHY FEMALE DOGS

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Presence of bacteria, free or inside the epithelial cells, is frequently reported in vaginal smears of healthy dogs [1]. Depending on the cycle stage from 50% to 100% of clinically healthy dogs are characterized by a vaginal bacterial population, usually mixed, consisting of both aerobic and anaerobic microorganisms often opportunistic pathogens [2]. The treatment of pets with antimicrobials may affect the population of commensal bacteria and increase the risk for colonization of the urogenital tract by potentially pathogenic bacteria [3].

The aim of this study was to isolate microorganisms from the vagina of female dogs in the anestrus period and to evaluate the antimicrobial susceptibility.

Materials and methods. Samples from vagina were taken from 45 healthy female dogs in anaestrus period. The samples were cultured on differential–diagnostic and selective nutrient media. Microorganisms were identified according to biochemical and antigenic characteristics. The antimicrobial susceptibility was determined by the disk diffusion method using the Kirby-Bauer technique (Bauer et al., 1966). Nine antimicrobial agents were used for susceptibility testing. The P value of < 0.05 was considered to be significant.

Results. The predominant microorganisms isolated from female dogs in the anestrus period (n = 45) were Streptococcus spp. (24.3%), Staphylococcus pseudintermedius (21.95%) and Escherichia coli (21.95%). Bacteria were not isolated from four female dogs. In a comparison of the effects of antimicrobial agents of the female dogs in the anestrus period, cefovecin and enrofloxacin were found to be more effective than amoxicillin. Enrofloxacin was also more effective than erythromycin and vancomycin. The obtained results were statistically reliable (P < 0.05)

Conclusions. A wide variety of microorganisms may be isolated from a female dog's vagina in the anestrus period. Bacteria from Streptococcus genus, Staphylococcus pseudintermedius and Escherichia coli were the most common microflora in our investigation. The highest sensitivity was detected to cefovecin and enrofloxacin.

Keywords: female dogs, microorganisms, vagina, antimicrobial susceptibility.

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ISOLATION OF MICROORGANISMS FROM CANINE SKIN AND EVALUATION OF ANTIMICROBIAL RESISTANCE

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Various bacterial agents are found on canine skin and fur, which creates natural individual skin microbiota, but when skin barrier and immunity are conflicted, natural microbiota can become potentially pathogenic and make new pathways for new pathogenic microorganisms [1]. Microbiota, which is present in individual canine skin, "trains" an innate and adaptive immune system. Antimicrobial resistance is gradually increasing, so it is important to investigate antimicrobial resistance in cases of bacterial skin diseases to ensure targeted treatment [2]. The aim of this study was to isolate microbiota from canine skin and determine antimicrobial resistance.

Materials and methods. 36 canines were tested. According to clinical symptoms, dogs were divided into two groups: clinically healthy canines (n = 18) and canines which had clinical symptoms of pyoderma (n = 18). The swabs were placed in "Amies" transport medium and taken to the laboratory. The obtained samples were cultured in aerobic conditions on selective and differential nutrient media. Bacteria were identified according to biochemical and antigenic characteristics, and their antimicrobial susceptibility was determined by the disk diffusion method using the Kirby-Bauer technique [3]. The *P* value of < 0.05 was considered to be significant.

Results. The following strains of microorganisms were isolated from the healthy skin of dogs: *Staphylococcus warneri* (33.3%), *Bacillus* spp. (27.7%), and *Staphylococcus pseudinermedius* (5.5%). The most prevalent species from pyoderma cases were *Staphylococcus pseudintermedius* (38,8%) and *Staphylococcus aureus* (27.7 %). *Staphylococcus warneri* isolates from healthy canine skin were resistant to oxacillin (89.5%), trimethoprim-sulphamethoxazole (78%) and cefovecin (61.5%). In dogs with pyoderma symptoms, 89.5% of *S. pseudintermedius* strains were resistant to trimethoprim–sulphamethoxazole and oxacillin; 100% of *S. aureus* strains were resistant to oxacillin and fusidic acid.

Conclusions. Coagulase negative *Staphylococcus warneri* was the most prevalent in dogs with healthy skin. Coagulase positive *Staphylococcus aureus* and *Staphylococcus pseudintermedius* were predominant species in dogs with pyoderma. Most isolates from clinically healthy canine skin were resistant to oxacillin, trimethoprim-sulphamethoxazole and cefovecin, while those from canines with pyoderma were resistant to oxacillin, trimethoprim–sulphamethoxazole and fusidic acid.

Keywords: canine, skin, microorganisms, antimicrobial resistance.

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ULTRA-FREEZING EFFECT OF CRYOPRESERVED BOVINE SEMEN ON THE MOTILITY AND VIABILITY OF POST-THAW SPERMATOZOA

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Advancements like artificial insemination (AI) have significantly boosted genetic improvement and cattle production, with semen quality being essential for a successful AI [1]. Cryopreservation and cooled semen technology have been explored for improved semen transportation and insemination. However, the cost, maintenance and availability of liquid nitrogen, crucial for cryopreservation, underscore the need for alternative semen storage solutions [2].

In light of this, our investigation was carried out on the effects of varied storage temperatures and durations on motility and viability of the bull's semen. Cryopreserved semen samples were obtained from 6 bulls. For each bull, 6 doses were retained in liquid nitrogen (at −196°C) as a control group, while 6 doses were transferred to −75°C (ultra-freezer "Sanyo"). The study assessed 72 cryopreserved straws at distinct intervals: 1 day, 10 days, 30 days and 90 days. To obtain a comprehensive evaluation of motility, we calculated the average motility by combining the results from the subjective and objective motility (computer-aided semen analysis using Sperm Class Analyzer) assessments. This approach allowed us to consider both qualitative and quantitative aspects of sperm motility. Viability was assessed with the staining method (Eosin/Nigrosine) and the hypo-osmotic swelling test (HOST) to account for potential variations in dye penetration and staining efficiency, and to provide a more accurate representation of the spermatozoa viability after thawing.

For longer storage durations of 90 days, the results indicated that liquid nitrogen storage at −196°C showed significantly higher sperm motility and viability ($P < 0.05$), with values of 92.12% and 45.58%, respectively, compared with those at −75°C (73.74% motility and 25.08% viability). This suggests that for long-term preservation −196°C storage remains optimal.

However, interestingly, short-term storage for up to one month, the ultra-freezer storage at −75°C could serve as a viable and cost-effective alternative to −196°C storage. The motility rates were not significantly different ($P > 0.05$) with 78.68% at −75°C and 83.89% at −196°C. Moreover, the viability rates were comparable, with 41.5% at −75°C and 40.58% at −196°C, indicating that the quality of semen does not significantly diminish under these conditions.

Keywords: bull, semen quality, cryopreservation, ultra-freezing, artificial insemination.

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ANALYSIS AND COMPARISON OF QUALITY PARAMETERS OF CRYOPRESERVED SEXED AND UNSEXED BULL SEMEN

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Sexing of sperm can be achieved with more than an 85% accuracy using the flow cytometry method [1]. But a challenge of this is that the stress placed on the sperm has detrimental effects on the quality and viability of the sperm [2]. The objective of this study was to investigate the differences in quality and viability of sexed and unsexed sperm and the effect of the length of removal from cryopreservation.

For this study, three bulls were selected, and a sexed and unsexed sample of their semen was taken and cryopreserved in liquid nitrogen. The samples were removed and defrosted in a water bath maintained with a hot plate at 37°C. The first six samples, three sexed and three non-sexed, where removed after 10 seconds. They were then all tested for motility, viability, concentration, morphology, and the hypo-osmotic swelling (HOS) test. The next six samples were removed from the water bath after five hours and motility, viability, and HOS test were repeated.

In the sample defrosted for 10 seconds, the motility was found to be on average 40% higher in the sexed sample and 47.37% higher in the sexed sample for the five hour defrosted samples compared with the respective unsexed samples ($P < 0.05$). Viability was tested using eosin and nigrosin staining with the 10-second defrost being 25.36% more dead in the sexed sample and with the 5-hour defrost being 19.9% more dead in the sexed semen samples ($P < 0.05$). Concentration was calculated and found to be 75.73% more concentrated in the sexed sample ($P < 0.05$). Morphology was calculated by looking for the number of defects per 100 spermatozoa, and it was found that 60.61% less defects were in the unsexed sperm ($P < 0.05$). Lastly, the HOS test was performed with the 10-second defrost having 21.51% more spermatozoa dead in the sexed sample; in the 5-hour defrost, 1.55% more dead spermatozoa were found to be dead in the sexed sample ($P > 0.05$).

From all these results we can conclude that unsexed sperm has a higher viability than sexed sperm. A previous study found successful pregnancy rates to be 84.42% in unsexed sperm and 75.33% in sexed sperm [3]. From the parameters examined in this study, we can understand the reasons of the differences in viability.

Keywords: viability, cattle, sex sorted semen.

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TLR4, *MEF2A*, *MEF2C* AND *MAPK1* GENE EXPRESSIONS IN DAIRY CATTLE UDDER PARENCHYMA INFECTED WITH COAGULASE-POSITIVE STAPHYLOCCI

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Mastitis is a complex disease affected by many factors related to several pathways connected with each other or not connected. Coagulase-positive staphylococci (CoPS) are one of the main mastitic pathogens. Based on the previous studies, we selected four (*TLR4, MEF2A, MEF2C, MAPK1*) interconnected genes where protein products play essential roles during inflammation. TLR4 is involved in a migration of neutrophils to and from the mammary gland during mastitis. It also detects bacterial ligands, such as lipopolysaccharide (LPS) [1]. All genes from the MEF2 family are transcription factors which regulate the expression of immune-related genes. Moreover, MEF2A and MEF2C regulate apoptosis, while MEF2C is also involved in the formation of B cells. MEF2A and MEF2C perform some functions such as that cannot be performed by other proteins from this family [2]. MAPK1, however, plays a main role in the inflammatory response and in autoimmune diseases [3]. The aim of the study was to compare the expressions of the studied genes in dairy cow udder parenchyma from quarters infected with CoPS (n = 10) and those adjacent to them (AHCoPS, n = 10) to those derived from healthy cows (H, n = 10), wherein healthy udder quarters taken as a control were derived from the whole healthy udders.

A total of 30 samples of dairy cow udder parenchyma were collected just after slaughter. Animals were culled at the end of lactation (286 days, SD = 27) and showed either reproductive difficulties but had healthy udders or chronic or/and subclinical mammary gland inflammation that was recurrent and incurable. RNA was extracted from frozen tissue samples. Only RNA with the integrity number (RIN) > 7.0 were selected for further analysis. The RT-qPCR method was used to establish gene expressions. The variance analysis was conducted using the MIXED procedure in SAS (SAS/STAT, 2002-2012, v. 9.14).

The expression of *MEF2A* and *MAPK1* genes did not differ between the groups while the *TLR4* and *MEF2C* expressions were higher in CoPS and H than in AHCoPS. TLR family plays an essential role in pathogen recognition and activation of innate immunity mediating cytokine production; thus, such processes do not occur in AHCoPS but do in CoPS and H. The lowest expression of *MEF2C* in AHCoPS may mean that apoptotic processes are much lower than in CoPS and even H, and the protection of the AHCoPS quarter against infectious agents by producing defensive antibodies is lowered.

Keywords: dairy cow, mastitis, adjacent quarters.

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CORRELATION BETWEEN SELECTED IMMUNE GENE EXPRESSIONS IN LIVER OF YOUNG CASTRATED BUCKS OF POLISH WHITE IMPROVED SUPPLEMENTED WITH *CURCUMIN-ROSEMARY* DRIED EXTRACT MIXTURE

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Expression profiles of cathelicidins (CATHs), acute phase proteins (APPs), and defensins (DEFs) can provide helpful information about the health state of goats [1, 2], e.g. after supplementation of the diet with pro-health substances. However, the synergic or antagonist activity of these proteins/peptides has not been studied yet. Therefore, the aim of the study was to analyze the effect of supplementation with the curcumin-rosemary dried extract mixture (a ratio of 896:19) on the correlations between APPs (serum amyloid (SAA)), haptoglobin (Hp), ceruloplasmin (Cp), C-reactive protein (CRP), alpha-1 acid glycoprotein (AGP), fibrinogen (Fb), α-lactalbumin (LALBA)), CATHs (capra hircus bacetnecin 3.4 (ChBAC3.4), cathelicidin 2 (BAC5), cathelicidin 3 (BAC7.5), cathelicidin 6 (MAP28), cathelicidin 7 (MAP34)), and DEFs (β-defensin 1 (GBD1), β-defensin 2 (GBD2)) gene expressions in the livers of young castrated bucks of Polish White Improved (PWI) breed. Two groups were distinguished: the control (CG), on the basal diet (n = 10), and the experimental (EG) (n = 10) with additive of 1.6 g/day/head of the mixture. The experiment lasted 124 days. At the start, bucks were 8 months of age with a live weight of 28.8 kg (± 4.9 kg) on average. A total RNA were isolated from their livers. The cyclophilin A (*PPIA*) and battenin (*CLN3*) were used as reference genes in the RT-qPCR method. The Pearson correlation was calculated (PROC CORR, SAS package). In the CG, relationships were found between *BAC7.5-GBD1* (0.74, $P = 0.04$), *BAC5-Hp* (0.74, $P = 0.04$), *BAC5-MAP28* (0.73, $P = 0.04$), *Hp-MAP28* (0.72, $P = 0.04$), *Fbα-Fbγ* (0.87, $P = 0.01$), *Fbα-Fbβ* (0.85, $P = 0.02$), and *Fbβ-Fbγ* (0.92, $P = 0.001$). The associations between gene expressions were much stronger in the EG than in the CG (all above 0.9). Moreover, several new associations appeared, especially with *MAP28* and *Fbs*. In opposite, the expression of *BAC5* in the EG was not correlated in any other gene. Much more and higher correlations between APPs, DEFs and CATHs gene expressions in the EG than in the CG may suggest that a supplementation with the studied mixture triggers the immune response in the livers of youngcastrated bucks of PWI breed, possibly causing mainly synergistic activity of their protein products.

Keywords: bucks, acute phase proteins, cathelicidins, defensins, correlations.

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COMPARISON OF THE NUTRITIONAL PROFILES OF EGGS PRODUCED BY LAYING HENS REARED UNDER INTENSIVE AND EXTENSIVE CONDITIONS

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Consumer demand for animal-based production has grown, emphasising product safety, quality, and animal welfare. Research has extensively explored the nutrition of laying hens in conventional, free-range, and organic systems [1]. However, the impact of outdoor access on egg-laying performance and quality traits remains inconclusive. So, the aim of this study was to perform a comparative analysis of the nutritional value of eggs from laying hens reared under extensive and intensive conditions. Eggs from domestically reared laying hens (extensive rearing with a known egg-laying period) and eggs purchased from a supermarket (intensive rearing of laying hens) were utilized for the research. The following indicators were measured in fresh and stored eggs (28 days at a refrigerator temperature of 4°C): egg height, yolk height, albumen height, Haugh unit, yolk colour intensity, eggshell thickness, yolk pH, albumen pH, dry matter (DM), fat, ash, and protein content in the yolk, as well as the sensory profile of fresh eggs. Comparing fresh and 28-day-old eggs from extensively and intensively reared laying hens, the extensive conditions resulted in higher essential indicators: fresh eggs were heavier by 24%, while stored eggs were heavier by 19% ($P < 0.05$). Extensively farmed fresh eggs showed a 7-point higher yolk colour intensity compared with intensive eggs, increasing by 8 points after storage ($P < 0.05$). The analysis of egg yolk colour characteristics using colour coordinates revealed that the yolk yellowness (b*) value was 27% higher in eggs from hens raised under extensive conditions than in intensively reared ones ($P < 0.05$). Extensively reared fresh eggs exhibited a 25% higher yolk weight, a 28% higher albumin weight, and a 57% higher albumin height compared with eggs from intensively reared hens ($P < 0.05$). Furthermore, the albumin weight of eggs stored for 28 days was 31% higher in eggs from extensively reared hens compared with eggs from intensively reared hens ($P < 0.05$). No significant differences were found in the chemical composition of eggs from laying hens raised under different conditions ($P > 0.05$). After the sensory profile assay, it was found that the intensity of the yolk smell was 10% more noticeable in the eggs of laying hens reared in extensive conditions compared with intensively reared eggs ($P < 0.05$). Eggs from extensively raised hens exhibited a 56% lower yolk colour uniformity compared with eggs from intensively reared hens, while the yolk fineness of eggs from intensively kept hens was 17% higher than that of eggs from extensively raised hens ($P < 0.05$). In conclusion, to provide a safe and high-quality alternative for egg consumption that meets acceptable standards, it is recommended to prioritize eggs from laying hens reared under extensive conditions.

Keywords: laying hens, rearing condition, egg quality, nutritional value.

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