

BIOTECHNOLOGY IN ANIMAL HUSBANDRY

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THE EFFECTS OF SOME MICROELEMENTS SUPPLEMENTATION – SELENIUM, ZINC AND COPPER INTO DAIRY COWS FEEDS ON THEIR HEALTH AND REPRODUCTIVE PERFORMANCES

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Review paper

Abstract: Microelements such as selenium, zinc and copper are indispensable nutrients for preserving major physiological functions, improving reproductive characteristics and overall health state. By their adequate use different ailments are prevented, while at the same time they have a positive effect on fertility and resistance. Insufficient quantities of these microelements, inadequate absorption and interaction with other microelements may lead to the impairment of the immune response due to metabolic and oxidative stress. In recent years, mainly organic forms of microelements have been administered to animals because they show better biological availability and can be retained longer in the organism. Besides, organic forms improve quality of products for human nutrition. Antioxidants should be added in optimal quantities in food for dairy cows with the aim of maximizing immune function and protection of tissues.

Key words: dairy cows, selenium, copper, zinc, health, reproductive performances

Introduction

Mineral matters play an important role in preserving animal immune function (*Shankar and Prasad, 1998*), fertility (*Rabiee et al., 2010*) and weight gain (*Enjalbert et al., 2006*). During transitional period dairy cows undergo a physiological stress when they prepare for parturition. In addition, significant metabolic changes can occur due to the nutrient needs of mammary gland necessary for the synthesis of milk. Simultaneously there occurs reduced uptake of food, negative energy balance (*Roche et al., 2009*) and oxidative stress (*Sordillo*

and Aitken, 2009). Transitional period is stressful for animals, therefore, decreased retention of mineral matters in the organism can be expected. Parturition and onset of lactation lead to decrease in the concentrations of calcium and zinc (Goff et al., 2002), what is observed also for other mineral matters (copper, selenium, cobalt, iodine) during lactation period. Mineral matters like selenium, zinc, copper, cobalt and iodine play an important role in synthesis of proteins, vitamin metabolism, forming of connective tissue and improvement of immune function. Proper supply of the organism by these minerals has an effect on health, fertility, lactation and immune functions (Griffiths et al., 2007). Moreover, some interactions between mineral matters in digestive tract of ruminants can lead to a significant fall in mineral level. The increase in the level of sulphur from 2 to 4 g/kg in dry matter can reduce the absorption of copper by 54.3 % when the level of molybdenum is 1 mg/kg dry matter. Increase in the concentration of molybdenum from 1 to 10 mg/kg dry matter can decrease absorption of copper by 48.3 % when the concentration of sulphur is 2.5 g/kg dry matter (NRC, 2001).

The role of selenium, zinc and copper in preserving the health status of dairy cows

Selenium is a key food ingredient which provides oxidative protection to the organism. Its biological role is performed through the enzyme glutathione peroxidase (GSH-Px). The activity of this enzyme depends on the level of selenium in food, what can be used as a reliable indicator of biological uptake of selenium. In animal nutrition the organic selenium in the form of selenomethionine and inorganic selenium in the form of Na-selenate or Na-selenite is used. Difference between these two forms is in metabolic pathway and efficiency of action. High levels of inorganic selenium are more toxic than the same levels of organic selenium (Todorović et al., 1999; Joksimović Todorović et al., 2006; Joksimović Todorović and Davidović, 2014). Antioxidative status is one of the factors which impacts the reproduction of dairy cows. Selenium reduces the occurrence of postpartum ailments in dairy cows such as retained placenta, mastitis, metritis, endometritis, ovarian cysts and increases the level of conception. High levels of superoxide ($O^{\cdot -}$) may decrease the function of neutrophils. Inadequate protection against free radicals leads to the reduction of neutrophil granulocytes what may have the incidence of a disease as a consequence. A primary function of selenium is to provide immunological defence and to increase the migration of neutrophil granulocytes into inflammatory region where they will ingest and destroy present bacteria. Selenium deficit leads to a functional disorder of all the cells of the immune system. The activity of T lymphocytes and NK cells (killer cells) is decreased, as well as the synthesis of some antibodies (Sordillo, 2005).

Zinc is an indispensable microelement for maintaining major physiological functions preserving in that way the health of individuals. By adequate zinc supplementation in feeds for dairy cows the incidence of either zinc surplus or deficit in the organism is being prevented (*Davidović et al., 2014, 2015*). Insufficient intake or disturbance in the zinc absorption can weaken the immune system due to metabolic and oxidative stress and development of mastitis (*Anton et al., 2013*). Zinc deficiency can cause the loss of appetite and metabolism disorder since this microelement is included in the synthesis of proteins, metabolism of carbohydrates and nucleic acids. Zinc plays a significant role in the synthesis of DNA and RNA by increasing the cellular replication and cellular proliferation (*Spears and Weiss, 2008*). It affects reproductive functions, secretion of gonadotropine, androgens, prostoglandin and prolactin, and also that of antioxidants (*Arthur, 2001*). *Kellogg et al. (2004)* report that zinc-methionine can significantly increase lactation performance and improve health of mammary gland since it reduces the number of somatic cells.

Copper is an important microelement for preserving health, reproductive status, immune functions and lactation performances in dairy cows. It is an integral part of a great number of metalloenzymes involved in numerous physiological processes such as: cellular respiration, lipids and carbohydrates metabolism, development of connective tissue, processes of myelinization, keratinization and pigmentation (*McDowell, 2003*). The adequate levels of copper in food are indispensable for optimization of immune system, since copper reduces the occurrence of development of metabolic and oxidative stress in dairy cows (*Cortinhas et al., 2010*).

Over a considerable time period these microelements were added into the animal feeds in the form of inorganic salts (sulphate and carbonates). In a last decade, organic sources of microelements (chelate, proteinates) which reduce the interaction of minerals with other matters in rumen preventing the accumulation of undissolving complexes and making in that way their intestinal absorption easier (*Spears, 2003*) were most used. Dairy cows fed rations that contain insufficient quantities of copper are often exposed to the risk of developing mastitis, metritis and disorder in locomotive system (*Enjalbert et al., 2006*). Copper overdose, in quantities higher than 40 mg/kg DM can cause toxicoses if this state is prolonged during lactation (*Engle et al., 2001*). The studies of *Kinal et al. (2007)* and *Joksimović Todorović et al. (2015)* indicate that there are statistically significant differences in the concentration of copper in blood plasma in the individuals fed copper inorganic sources in comparison with the individuals fed copper organic sources. However, *Cortinhas et al. (2012)* did not determine significant differences in the concentrations of copper in blood plasma in the individuals fed inorganic and individuals fed organic copper sources. *Anton et al. (2013)* discuss these results in following direction: the concentration of copper in blood plasma, besides its

adequate supplementation, can be affected by numerous various factors such as infection, inflammation and stress.

The use of microelements with the aim both of preventing postpartum ailments and improving reproductive characteristics

Placenta retention is a disease with a multifactorial etiology while a cause thereof is often unknown. During a transitory period in cows there occur metabolic, hormonal and biochemical changes. A prepartum function of neutrophiles decreases what can provoke the occurrence of postpartum diseases. The state in which placenta is retained for more than 12h is considered as having a disease. Numerous factors, including mechanical, nutritive and infective causative agents can impact the incidence of this disease. Incidence of placenta retention is often (about 50% animals) associated with zoonoses, such as brucellosis, salmonellosis, leptospirosis and listeriosis (*Gunay et al., 2011*). One of the studies indicated that placenta retention is one of the major causes of endometritis in cows (*Han and Kim, 2005*). This disease can either reduce fertility and therefore the percentage of conception, complicate conception or prolong a calving interval (*Bella and Roberts, 2007*). Selenium improves the function of neutrophiles, influences their migration and chemotactic activity. An increased concentration of cortisol represents a response to stress and inflammation of pregnant uterus. Cortisol can diminish the function of neutrophiles or completely prevent their activity and provoke this disease. The increased content of cytotoxic aldehyde (malondialdehyde) in erythrocytes and increased concentration of cortisol are considered as major causes of the onset of this disease. Many studies indicate that adequate levels of selenium, copper, zinc and certain vitamins, which play a role of antioxidants, can decrease the percent of individuals with retained placenta disease. (*Tillard et al., 2008; Sharma et al., 2011; Joksimović Todorović and Davidović, 2007a, 2013*).

The infections of mammary gland and uterus are frequent in dairy cows during peripartum period. Other health disorders which may happen in that period are milk fever and ketosis. Although the etiologies of the infections and metabolic disorders of these two diseases are different there is a significant connection between the incidence of these two ailments. In cows with milk fever clinical mastitis occurs five times more frequently compared to the cows which have no such health problems (*Curtis et al., 1985*). Dairy cows are exposed to numerous genetic, physiological and external factors which can endanger the immunity and increase the frequency of the incidence of mastitis (*Sordillo, 2011*). Selection directed to maximizing milk production increases metabolic stress caused by increased synthesis and secretion of milk and reduces resistance to mastitis.

The application of antioxidants in dairy cows reduces the duration and intensity of disease. Precise mechanisms into the improvement of health of mammary gland are not completely known but they are associated with their antioxidative functions (*Joksimović Todorović and Davidović, 2007b; Joksimović Todorović et al., 2012*). Antioxidants in dairy cows increase the resistance to mastitis, provide phagocytic capacity of neutrophils increasing the chemotaxis at the spot of the infection (*Spears and Weiss, 2008*). *Bicalho et al. (2007)* and *Formigoni et al. (2011)* established that supplementing the mineral matters to dairy cows can lead to decrease in the number of stillborn calves and the incidence of endometritis what is important for preserving proper reproductive performances. The study by *Machado et al. (2012)* showed that systemic addition of microelements to dairy cows` feeds can significantly reduce the number of cows with intrauterine contamination by *Fusobacterium spp.* and *Trueperella spp.* These bacteria are associated with the disease of uterus, particularly the onset of metritis and clinical endometritis. *Manspeaker et al. (1987)* suggest that dairy cows fed chelate minerals during late gestation and lactation have higher fertility due to increased ovarian activity, better involutions, regeneration of endometrial tissue, less periglandular fibrosis and lower mortality of embryos.

The results of the research into the effects of microelements supplementation on reproductive performances are controversial. Some authors reported that mineral matters supplementation can have either a negative (*Vanegas et al., 2004*), positive (*Sales et al., 2011*) or neutral effect (*Vanegas et al., 2004*) on reproductive performances. *Vanegas et al. (2004)* used copper, zinc and manganese in very low concentrations. These authors have determined that once administered mineral matters supplementation postpartum has no effect while two supplementations (the first prepartum and the second postpartum) can reduce reproductive performances. On the other hand, *Sales et al. (2011)* determined an increased levels of conception and embryonal survival in heifers that received subcutaneous injection of the same microelements 17 days before embryotransfer.

Conclusion

The application of organic form of mineral matters in the nutrition of dairy cows during pregnancy and lactation period has a positive effect on immune system and therefore on overall health status. Moreover, their application reduces the percentage of animals with postpartum diseases (placenta retention, mastitis, metritis, endometritis) improving, in that way, their reproductive performances.

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Efekti dodavanja pojedinih mikroelemenata – selen, cinka i bakra u hranu za mlečne krave na zdravlje i reproduktivne performanse

Mirjana Joksimović Todorović, Vesna Davidović, Mirjana Bojanić Rašović

Rezime

Mikroelementi selen, cink i bakar su neophodni nutrijenti za očuvanje osnovnih fizioloških funkcija, zdravlja i poboljšanje reproduktivnih karakteristika. Njihovom adekvatnom primenom preveniraju se različita oboljenja, a istovremeno pozitivno utiče na fertilitet i otpornost. Nedovoljna količina ovih mikroelemenata u obroku, neadekvatna apsorpcija i interakcija sa drugim mikroelementima, mogu dovesti do slabljenja imunskog odgovora usled metaboličkog i oksidativnog stresa. Poslednjih godina, uglavnom se daju organske forme mikroelemenata, jer imaju bolju biološku raspoloživost i duže se zadržavaju u organizmu. Pored toga, organske forme poboljšavaju kvalitet proizvoda namenjenih za ishranu ljudi. Antioksidanse treba dodavati u optimalnim količinama u hranu za mlečne krave u cilju maksimiziranja imunske funkcije i zaštite tkiva.

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THE QUALITY OF GOAT MEAT AND IT'S IMPACT ON HUMAN HEALTH

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Review paper

Abstract: Today, goats are spread throughout the world. They live in small or large herds and in different areas and environments. Because of its distinctive taste and desired chemical composition, goat meat is increasingly consumed in Serbia. As animal foods, it is rich in protein, vitamins and minerals, but contains very little fat, especially cholesterol. The aim of this review paper is to highlight some health benefits, nutritional values and potential use of goat meat. On the chemical composition of goat meat affect race, gender, productivity and adaptability to stress, environment, management, diet, weight at slaughter and health condition as well as slaughter and procedures with the carcasses after slaughter. Average chemical composition of lean goat meat contains about 75.42% water, 3.55% fat, 19.95 % protein and 1.06% mineral matter. The energy value is about 580 kJ per 100 g. The goat meat has about the same nutritional value as well as sheep meat. Due to low content of saturated fatty acids and cholesterol, goat meat in the human diet is healthier alternative compared to other types of red meat. Polyunsaturated fatty acids prevalent in goatmeat, and the diet rich in unsaturated fatty acids is correlated with a reduced risk of stroke and coronary disease. In addition, in goat meat are present the essential amino acids such as lysine, threonine and tryptophan. Regardless to the nutritional value, goat meat is still less appreciated due to their specific smell and taste, the more if the animal is older.

Key words: goat meat, quality, nutritional value, health

Introduction

Today, goats are spread throughout the world, with the exception of extreme cold areas. The goats are highly present in the countries with the extensive agricultural production, although their population rising in the richer countries, mainly due to the intolerance of certain groups of people to cow's milk.

They live in small or large herds and in different regions and environments: plain, desert, hilly and mountainous areas. *Devendra (2010)* states

that exist 1 156 different breeds of goats. The largest number of goat breeds are nourishing in Asia and Africa. According to *FAOSTAT (2012)* number of goats in the world is estimated to 957.40 million, while on the territory of Serbia in 2014 that number was 219 000 according to *Statistical Office of the Republic of Serbia (Republički zavod za statistiku, 2015)*.

Globally, consumption of goat meat is lower than consumption of beef (*Madruga and Bressan, 2011*), but goats undoubtedly serve as a major source of red meat for the people (*Webb et al., 2005*), particularly in developing countries.

Because of its distinctive taste and desirable chemical composition, goat meat is increasingly consumed in Serbia. As animal foods, it is rich in protein, vitamins and minerals, but contains very little fat, especially cholesterol. This type of meat is no opposing religious and cultural aspects of consumption. On the territory of the Republic of Serbia from 1954 to the nineties of past century, was a ban on the keeping of goats. Goats have not been eliminated as a kind of animal, but their production was not controlled (statistics on their numbers, selection, manner of keeping and slaughtering, meat quality) (*Ivanović and Pavlović, 2015*).

Differences in the quality of goat meat are result of age, gender and genetic groups (*Casey and Webb, 2010*).

In relation to the carcasses of sheep of the same age and sex, goat carcasses are smaller and have less fat on the surface (*Sheridan et al., 2003*). The carcasses of young goat containing a high percentage of muscle and a low percentage of bone, a lower percentage of intramuscular and subcutaneous fat, compared with the carcasses of sheep (*Santos et al., 2008; Stanis� et al., 2009*). On the other hand, a lower percentage of fat on the surface of carcasses can lead to cold shortening and significant loss during the post mortem cooling.

The aim of this review paper is to highlight some health benefits, nutritional values and potential use of goat meat.

The chemical composition of goat meat

On the chemical composition of goat meat affect race, gender, productivity and adaptability to stress, environment, management, diet, weight at slaughter and health condition as well as slaughter and procedures with the carcasses after slaughter. Because of these facts it is difficult to compare the results of the chemical composition of goat meat of different authors, who came to them under different conditions. According to research (*Ivanovic et al., 2012a, 2012b, 2014*), the average chemical composition of lean goat meat of the race Serbian white goat contains about 75.42% water, 3.55% fat, 19.95 % protein and 1.06 % mineral matter, while at the race Balkan goat, water was represented with 74.51 %, fat 3.92%, proteins 20.55% and mineral matter 1.04 %. The energy value was similar and is around 580 kJ per 100 g.

Water has influence on the quality of the meat, especially the juiciness, but also on the so-called technological quality. The muscles contain approximately 75% water, which is distributed within the myofibrils, between themselves, between the cell membrane (sarcolemma) and between the muscle bundles. Water is a bipolar molecule which is attracted by particulate species, such as proteins, and in the muscles may be related, reserved and free.

Genetics and treatment of live animals can greatly affect the future capacity of water in the product. The cooling or freezing mode after slaughter, especially during the rigor, is of great importance for the percentage of water that will remain in the meat. These factors have a significant impact on the pH. Retained water also affects the particular sensory properties (*Ivanović et al., 2012a*).

Adipose tissue and fat. Adipose tissue is not a passive depot of fat, it conducted a very extensive biochemical processes. In fat cells, glycerol is synthesizing, the fatty acids are linking into glycerides, the glycerides are converting to each other, the grease is synthesizing from glucose, fat mobilization is performing, whereby the fatty acids and glycerides are releasing which cause hyper lipemia. According to their chemical composition, the fats of adipose tissue are mainly triglycerides and are located in the cells, while the water, proteins and mineral matter are ingredients of connective tissue stroma. Adipose tissue of slaughtered animals contains 50-95% fat, 3-35% water, 2-15% protein and 0.1-0.6% mineral matter. The composition of fat in adipose tissue is highly variable and depends on nutrition, breeding, age, type of animal.

In water, fats are insoluble biomolecules. They are present in the muscles as a structural component of muscle membrane, between the muscle fibers as a droplets depot of triacylglycerol, and as adipose tissue, as to fat that gives the meat marbling. Fats represent about 5% of the cells organic matter. In the cell, there is 40-50 different types of fats. The cells of the brain and nervous tissue, are extremely rich in fats.

For cattle and sheep, diet has a great effect on the deposition of intramuscular fat (*De Smet et al., 2004; Grubić et al., 2005*), as well as on the concentration of saturated fatty acids (SFA) and polyunsaturated fatty acids (PUFA).

Lipids from meals are hydrolyzed in the rumen of ruminants. Unsaturated fatty acids from foods pass through the process of biohydrogenation by acting of microorganisms of the rumen. The result is that the ruminants predominantly absorb saturated fatty acids, so that the food that comes from ruminants contains a higher percentage of saturated fatty acids. When the biohydrogenation in the rumen is not complete, part of the conjugated linoleic acid (CLA) managed to avoid it and the animals absorb it in that form, so that the tissues and their products are supplied by isomers of CLA (mixture of isomers of conjugated linoleic acid).

Mainly, meat from ruminants fed with grass containing more PUFA, n-3 PUFA, CLA, vitamin E, beta-carotene and vitamin A compared to meat from ruminants fed with grains (*De Smet et al., 2004; Bressan et al., 2014*). Several studies are conducted with the aim to determine the fatty acid profile in goat meat (*Sheridan et al., 2003; Madruga et al., 2009*). However, biohydrogenation, transition of unsaturated into saturated fatty acids, elongation of fatty acid chain, as well as the metabolism and deposition rate, are not clear. After all, the composition of the fat in goats meat compared to monogastrics animals (e.g. swine.), is similar to other ruminants: large quantities of SFA, lower amounts of the PUFA, the presence of C18:1 and C18:2 trans and cis isomers of FA.

In animals, the main PUFA (C18:2n-6 and C18:3n-3) are obtained from the diet, but in ruminants, are biohydrogenized by ruminal bacteria into the intermediate FA and C18:0. The intermediate biohydrogenation includes cis and trans C18:1 isomers and C18:2 trans isomers, conjugated or unconjugated. Their concentration in the meat is related to reducing of C18 PUFA (*Bessa et al., 2000*). The grain feeds are considered as good source of C18:2n-6, while a green grass on pasture is rich in C18:3n-3 (*Goffman and Böhme, 2001; Boufaied et al., 2003*). More grazing in ruminant nutrition leads to higher percentage of omega-3 fatty acid, which is highly desirable.

C18:2 cis-9, trans-11 is quantitatively the largest isomer of CLA, and is more presents in the meat of ruminants fed on pasture compared to the meat of ruminants fed with grain feeds. Even it is thought, that this FA occurs in the rumen, about 70-80% of the acid is accumulated in the tissues as a result of endogenous transition into unsaturated C18:1 trans-11, catalyzed by the enzyme $\Delta 9$ desaturase (*Palmquist et al., 2004*). As a consequence of the difference in the concentration of CLA is a consequence of the difference in the quantities of C18:1 trans-11 absorbed in the rumen. The meat of ruminants is a natural source of conjugated linoleic acid (CLA), since CLA is a product of the rumen. Of all ruminant meat, the lamb meat is the richest source of CLA, when in question is the meat as a source.

Ivanovic et al. (2014) are examined the impact of race (Serbian white goat, Balkan goat and Bunte Deutsche Edelziege) on the fatty acid composition of goat meat and found that there was a statistically significant difference between the individual fatty acids in samples. According to the findings of these authors, the ratio of unsaturated to saturated fatty acids (UFA / SFA) in the meat was for Serbian white goat 0.96, for Balkan goat 0.92 and for German Spotted breed (Bunte Deutsche Edelziege) 0.46.

Genetic groups of ruminants, and therefore the goat, show a significant difference in the activity of $\Delta 9$ desaturase, which converts SFA to cis-9 monounsaturated fatty acids (MUFA), and elongase which converts C16:0 to C18:0. In these biotransformations participate enzymes desaturase and elongase and of their activity, as well as the amount of a substrate depends on the intensity and efficiency of these reactions. Desaturases are enzymes that are responsible for

the introduction of a new double bond in the fatty acid chain. Between fatty acids of n-3 and n-6 series there is a competition for these enzymes. Elongase are enzymes that are responsible for the chain extension of the fatty acid by two carbon atoms, i.e. for the introduction of two new methylene groups.

Proteins. After the water, the proteins are the most represented in the body. In the organisms of the animals and humans are entering through food. Each species of living organisms contains specific types of proteins. The proteins of the one species are chemically different from the proteins present in the other species. Therefore, it is right to say that specificity of some kinds of living beings comes from the specificity of the protein of which are built. The proteins are built from 20 correct building units - amino acids - that are called the building units of proteins.

There is not much data for the composition of amino acids in the goat meat that are bred in Serbia. *Ivanovic et al. (2014)* have examined the composition of amino acids in the meat of different races. They used as samples the meat (*m. longissimus dorsi*) which was from the two goat races (Balkan and Serbian white goat). The goats whose samples were tested are grown in the area of Stara Planina, and were four years old. In winter, the goats are fed combined by: hay and concentrate, and in summer were taken to pasture. The content of individual amino acids is shown in Table 1.

Table 1. The composition of amino acids (g/100 g) in protein of Serbian white and Balkan goats

Serbian white goat				Balkan goat			
Amino acid	Content	Amino acid	Content	Amino acid	Content	Amino acid	Content
Alanine	4.93	Lysine	8.36	Alanine	4.98	Lysine	8.11
Arginine	5.44	Methionine	3.20	Arginine	5.54	Methionine	3.51
Aspartic acid	8.66	Phenylalanine	4.22	Aspartic acid	8.74	Phenylalanine	4.55
Cysteine	1.02	Proline	3.45	Cysteine	1.04	Proline	3.37
Glutamic acid	14.41	Serine	3.92	Glutamic acid	13.95	Serine	3.89
Glycine	3.97	Threonine	4.84	Glycine	3.91	Threonine	4.97
Histidine	3.62	Tryptophan	1.19	Histidine	3.84	Tryptophan	1.27
Isoleucine	4.63	Tyrosine	4.17	Isoleucine	4.71	Tyrosine	4.44
Leucine	8.13 ^a	Valine	4.97	Leucine	8.38 ^b	Valine	5.05

Legend:^{a,b} - Mean values within a row with superscript differ significantly (P<0.05)

According to the results shown in Table 1, just between leucine amino acids were statistically significant difference between the samples. The absence of differences in the composition of amino acids in meat samples, confirms that proteins are typical for the species. The results obtained in research by *Brzostowski et al. (2008)*, *Webb et al. (2005)*, also indicate that the proteins, or amino acid

composition are typical for the species. Regarding to the essential amino acid composition, the goat meat closely looks like beef and lamb (*Correa, 2011*).

Vitamins and minerals. The meat and therefore goat meat is a good source of B vitamins (B1, B2, PP), minerals (P, Mg) and microelements.

Vitamins. In the nature, there is 13 vitamins and all are essential, without exception. The vitamins are not food, nor a substitute for food. The vitamins do not provide calories and energy directly, but are essential to body, especially vitamin B - to convert food into the energy. The only way to enter in the body is through food, both plant and animal origin. The meat can be one of the sources of vitamins, especially vitamins of B group. The confirmation of this are gave *Lawrie (1981)*, and *Holland et al. (1995)*.

Minerals. The minerals are inorganic chemical elements which in very small quantities are entered through food in the human and animals' body. They participate in metabolism, synthesis of many hormones and enzymes that ensure the normal growth of the organism and maintain it healthy. As well as vitamins and minerals must be constantly import with food and in a certain amount. In the food that is a source of minerals is includes the meat of different animal species (*Holland et al., 1995*). *Ivanovic et al. (2014)* have examined the content of microelements (mg/kg) in the meat of Serbian white goat and Balkan goat (4 years old). The obtained results are shown in Table 2.

Table 2. The content of microelements (mg/kg) in the kidneys of Serbian white and Balkan goats

Serbian white goat		Balkan goat	
Element	Content	Element	Content
Copper	0.73	Copper	0.58
Iron	7.25	Iron	6.51
Manganese	0.06	Manganese	0.07
Zinc	102.00	Zinc	114.00

According to (*Correa, 2011*), goat meat has higher levels of iron (3.2 mg) when compared to a similar serving size of beef (2.9 mg), pork (2.7 mg), lamb (1.4 mg), and chicken (1.5 mg). Comparatively, goat meat also contains higher potassium content with lower sodium levels.

Carbohydrates. The carbohydrates are derived from the muscle and connective tissues. In the muscle tissue, there is a polysaccharide - glycogen, wherein the D-glucose molecules are linked with 1,4- glucosidic bond, while at the branching sites of the backbone with 1,6- glucosidic bond. The inclusion of glycogen are suspended in sarcoplasm. The content of carbohydrates in the muscles of animals for slaughter is relatively low and ranges between 0.5% and 1.5%. The red muscles contain more glycogen than the white muscles. In the muscles, where is occurred post-mortem glycolysis a very small amount of glycogen, but the meat

containing intermediate products of metabolism:hexose-phosphates, triose-phosphates, pyruvic and citric acid and a significant amount of lactic acid (about 1%).The carbohydrates are also found in the glycoproteins and the mucopolysaccharides of the connective tissue (*Ivanović and Pavlović, 2015*).

The nutritional value and the impact on human health

The goat meat has about the same nutritional value as the sheep (more precisely, more protein and less fat compared to the sheep meat). *Anaeto et al. (2010)* state that, because of the molecular structure, the goat meat is more easily digested. Due to low content of saturated fatty acids and cholesterol, the goat meat in the human diet is healthier alternative compared to other types of red meat. According to the same author, polyunsaturated fatty acids prevalent in goat meat, and the diet rich in unsaturated fatty acids is correlated with a reduced risk of stroke and coronary disease. In addition, in goat meat are present the essential amino acids such as lysine, threonine and tryptophan.

The nutritional value of goat meat affects human health, because it is not only contains less total fat and cholesterol, but also content of saturated fatty acids are lower than in the traditional types of meat (Table 3).

Table 3. The nutrient composition of goat and other types of meat

Nutrient	Goat	Chicken	Beef	Pork	Lamb
Calories (kcal)	122	162	179	180	175
Fat (g)	2.6	6.3	7.9	8.2	8.1
Saturated Fat(g)	0.79	1.7	3.0	2.9	2.9
Protein (g)	23	25	25	25	24
Cholesterol (mg)	63.8	76.0	73.1	73.1	78.2

¹ Per 3 oz. of cooked meat

² USDA Nutrient Database for Standard Reference, Release 14 (2001)

Less saturated fats and a relatively high percentage of total unsaturated fats seems that the goat meat is very healthy.

The saturated fats are fats or fatty acids that do not include double bonds between carbon atoms in the fatty acid chain. They form a solid or semi-solid fat at room temperature, and are the cause of the increase in cholesterol levels. The amount of cholesterol in the food has only a moderate effect on the amount of cholesterol in the bloodstream.

The unsaturated fats are fats or fatty acids that contain one or more double bonds between the carbon atoms in the fatty acid chain. Where double bonds are formed, hydrogen atoms are eliminated. The fatty acids are monounsaturated when containing one double bond, and polyunsaturated if they contain more than one

double bond. Monounsaturated and polyunsaturated fats are in liquid form at room temperature. It is known that they reduce the risk of heart disease and stroke.

According to *the Harvard School of Public Health (2008)*, saturated fats (the bad fats) increase the risk of cardiovascular disease and other chronic diseases, while unsaturated fats (good fats) can improve cholesterol level in blood, alleviate inflammation, stabilize heart rhythm, and have numerous other useful effects.

When we talk about the effect of saturated and unsaturated fats on blood cholesterol levels and risk for heart disease, it is necessary to clearly understand the lipoproteins. The lipoproteins are complex particles consisting of a core of hydrophobic lipids that are surrounding by a layer of phospholipid and protein (lipid-binding proteins), that make up the particles are soluble in water. Due to the hydrophobic (water-repellent) nature of lipids, lipoproteins are the forms in which lipids as cholesterol, are transporting in the blood. The two main types of lipoprotein particles in the human blood are low density lipoproteins (LDL) and high density lipoproteins (HDL). Of these two lipoprotein holders of cholesterol, HDL contains relatively high concentration of protein and a small amount of cholesterol. Opposite, LDL contains a relatively low concentration of protein, and a large amount of cholesterol as a primary lipid. Usually, LDL transport cholesterol from the liver into cells throughout the body. The body is using cholesterol for the formation of cell membranes and the synthesis of vitamin D, estrogen, testosterone, and other steroid hormones. If is not used, LDL continues to bear the cholesterol through the blood. When too much LDL cholesterol circulates in the blood, these particles can attach themselves to the walls of arteries and form plaque that narrows arteries, restrict or block the flow of blood and thus cause heart attack or stroke. So, LDL cholesterol is often called "bad" cholesterol. Since HDL transports cholesterol from the cells, from the walls of the arteries and blood back to the liver for processing, HDL cholesterol is often called "good" cholesterol.

The clinical studies show that saturated fats originating from foods increase levels of LDL cholesterol, while monounsaturated and polyunsaturated fats can help in reduce LDL cholesterol and increase levels of HDL cholesterol in the blood. Based on these findings, it can be argue that the goat meat helps reduce blood cholesterol levels and thereby reduce the risk of atherosclerosis and coronary disease (*Correa, 2011*).

Regardless to the nutritional value, the goat meat is still less appreciated due to their specific smell and taste, the more if the animal is older (*Ivanović et al., 2011*).

Conclusion

The world's production of goat meat in the past ten years is increased by 30%. Compared to other types of meat (beef, lamb, chicken), goat meat has less acceptable taste and odor, but it is very similar nutritional value. The goat meat has

about the same nutritional and digestible value, as well as sheep. The nutritional value of goat meat affects human health, because it is not only contains less total fat and cholesterol, but also content of saturated fatty acids are lower than in the traditional types of meat. It is accepted that increasing levels of n-3 fatty acids (FA) in the human diet reduces the risk of heart problems and arteriosclerosis, and isomers of conjugated linoleic acids (CLA) have anticarcinogenic and antiarterogenic properties.

Due to the high nutritional value, the goat meat should be more promote in Serbia and thus improve his sale on the market.

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Kvalitet mesa koza i njegov uticaj na zdravlje ljudi

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Rezime

Danas su koze rasprostranjene u celom svetu. Žive u malim ili velikim stadima i to u različitim oblastima i okruženjima. Zbog svojeg karakterističnog ukusa i poželjnog hemijskog sastava, kozje meso se sve više konzumira u Srbiji. Kao namirnica životinjskog porekla bogata je proteinima, vitaminima i mineralima, sadrži vrlo malo masti, a posebno holesterola. Cilj ovog preglednog rada je da se istaknu neke zdravstvene prednosti, nutritivne vrednosti i potencijalna upotreba kozjeg mesa. Na hemijski sastav mesa koza utiču rasa, pol, proizvodnost i adaptiranost na stres, okolina, menadžment, ishrana, telesna masa prilikom klanja i zdravstveno stanje ali i klanje i postupci sa trupom posle klanja. Prosečan hemijski sastav krtog mesa koza sadrži oko 75.42 % vode, 3.55 % masti, 19.95 % proteina i 1.06 % mineralnih materija. Energetska vrednost je oko 580 kJ na 100 grama. Kozje meso ima otprilike istu hranljivu vrednost kao i ovčije. Zbog niske zastupljenosti zasićenih masnih kiselina i holesterola, kozje meso u ishrani ljudi, je zdravija alternativa u poređenju sa drugim vrstama crvenog mesa. Polinezasićene masne kiseline prevladaju u mesu koza, a ishrana bogata sa nezasićenim masnim kiselinama je u korelaciji sa smanjenim rizikom od moždanog udara i koronarnim bolestima. Pored toga, u mesu koza zastupljene su esencijalne amino kiseline kao

što su lizin, treoninitriptofan. Bez obzira na nutritivnu vrednost, meso koza je ipak manje cenjeno zbog specifičnog mirisa i ukusa, tim više ukoliko je životinja starija.

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GENETIC VARIABILITY AMONG CATTLE BREEDS OF NIGERIA USING THYROID HORMONE RESPONSIVE SPOT 14 ALPHA GENE (THRSP α) THROUGH POLYMERASE CHAIN REACTION (PCR)

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Original scientific paper

Abstract: The genetic variability among three Nigerian indigenous cattle breeds-White Fulani (WF), Red Bororo (RB) and Sokoto Gudali (SG) was carried out using Thyroid Hormone Responsive Spot 14 Alpha Gene (THRSP α) through Polymerase Chain Reaction (PCR). A total of sixty-seven (67) cattle blood samples comprising 30 WF, 25 RB and 12 SG were used. The DNA were extracted from blood samples using Zymobead Genomic DNA extraction kit after which PCR was carried out using 50 ng template DNA, 1.0 μ M primer (THRSP α), 16 μ l Nuclease-free water in a BIONEER Accupower premix. Gel electrophoresis was carried out and the gels scored. Statistical analyses were carried out using GENEPOP, PAST, SPSS version 16 and Tools for Population Genetic Analysis (TFPGA) version 1.3. The allelic frequency ranged from 0.3600 to 0.6400 in A and B alleles, average heterozygosity ranged from 0.4608 to 0.4861. Lowest genetic distance of 0.0010 between WF and RB and highest genetic distance of 0.0058 between SG and RB were identified. Lowest genetic identity of 0.9942 between RB and SG and highest genetic identity of 0.9990 between WF and RB was also identified. Two (2) genetic population clusters were identified in the dendrogram; WF and RB are in cluster 1 while SG is in cluster 2. Test of Hardy Weinberg equilibrium revealed variation in genotypic frequencies. These results therefore demonstrate variation among these three Nigerian indigenous cattle breeds which is attributed to the response of the breeds to various stimuli which also enhance their survivability and adaptation to their specific environments. Such variation can also be harnessed for conservation and improvement of our indigenous breeds through selection and breeding strategies.

Keywords: Genetic identity, Heterozygosity, Indigenous cattle, Allelic frequency

Introduction

Livestock play a vital role in the agricultural and rural economies of the developing world. Not only do they produce food directly, they also provide key inputs to crop agriculture through animal power (*ILRI, 1997*). In Nigeria, cattle production represents one of the major protein suppliers to the populace. It has been estimated that there are 13.9 million cattle in Nigeria (*Lawal-Adebowale, 2012*). Generally, cattle are raised for meat, milk, leather, dung, and draft purposes. Cattle as an economically important farm animal in Nigeria requires sound management and adequate conservation strategy through characterization at the molecular level for sustainable development. Genetic variability exists between and within individual livestock genotypes in a population. Such genetic variability can be measured through genetic markers, mitochondrial DNA, DNA sequence or specific genes that produce a detectable trait with a known location on a chromosome and that can be used to study family and population characteristics. The genetic variability found in domestic breeds allows farmers to develop new characteristics in response to changes in environment, diseases or market conditions (*Georg and Christina, 2007*). Variations at DNA level contribute to the genetic characterization of livestock populations and this may help to identify possible hybridization events as well as past evolutionary trends (*Choudhary et al., 2006*).

Thyroid Hormone Responsive Spot 14 Alpha gene (THRSP α) is primarily a nuclear protein which is important in the regulation of lipid metabolism. It is a gene that encodes a small acidic protein; it is a transcription factor that controls expression of blood and major lipogenic tissue such as liver and fat. The gene is highly correlated with intramuscular fat content (*Wang et al., 2009*). The expression of this was determined in bovine mammary gland and mapped the THRSP gene to bovine chromosome 29 nearest microsatellite marker RM179 on the USDA linkage map. THRSP α gene have been used successfully in estimating genetic relatedness and variability among various breeds and populations of goats and chicken (*Hirwa et al., 2009; Xiaopeng et al., 2012*) respectively. Most of the indigenous livestock populations in developing countries such as Nigeria have not yet been characterized and evaluated at phenotypic and genetic molecular levels (*Hannotte and Jianlin, 2005*), it is therefore necessary to determine genetic variability among cattle breeds indigenous to Nigeria. This research therefore is aimed at determining genetic variability among White Fulani, Red Bororo, and Sokoto Gudali breeds of cattle by Thyroid Hormone Responsive Spot 14 Alpha gene (THRSP α) through Polymerase Chain Reaction (PCR).

Materials and Methods

This research was carried out at the Animal Science laboratory, Faculty of Agriculture University of Port Harcourt. Blood samples were collected from sixty seven (67) cattle which comprised the following Nigerian indigenous breeds- 25 Red Bororo (RB), 30 White Fulani (WF), and 12 Sokoto Gudali (SG). Three (3) ml of blood was collected from each cattle in a 5ml EDTA bottle and kept at -4°C until the extraction of DNA commenced.

DNA Extraction and Polymerase Chain Reaction (PCR)

DNA from these blood samples of cattle was extracted using ZymoBead Genomic DNA kit protocol. The PCR followed as described by *Hirwa et al.* (2009). The DNA was amplified via PCR in a PCT- 100 Thermal Cycler (Biorad, Hercules, CA) using forward primer (Deletion F: 5'-GCC TCC GTC ACC GAT CAG- 3'). The 20 μl amplification reactions contained 50 ng templates DNA, 1.0 μl of each primer. 16 μl Nuclease-free water in a BIONEER AccuPower® TLA PCR Premix. PCR was performed for 33 cycles for 30 sec at 59.5°C , and 1 min at 72°C , after denaturation at 94°C for 2 minute, final extension was carried out for 10 minutes. The forward and reverse primers produce a 127 or 136bp is representative of THRSP α AA genotype and 127bp is representative of THRSP α BB genotype which is indicated by 96bp deletion.

Gel Electrophoresis and scorings of gels

10 μl of PCR product was loaded in a 1.5% agarose gel pre-stained with 0.5 $\mu\text{g}/\text{ml}$ ethidium bromide. Electrophoresis was carried out at room temperature for 1 hour at 100 volts using a Biorad Power Pac™ electrophoresis apparatus (Biorad, Hercules, CA, USA). The resulting amplified bands were visualized with UV light and photographed and were scored using GENE Mate Quanti- Marker 100bp DNA ladder (Bioexpress, UT, USA).

Statistical Analysis

The allelic frequencies and genotype frequencies were estimated by GENEPOP Software package (*Raymond and Rousset, 1995*). Other genetic analyses of data were performed using PAST, SPP version 16 and Tools for Population Genetic Analyses (TFPGA) version 1.3 (*Miller, 1997*).

Results and Discussion

The results of analysis of data are shown in Table 1-3 and Figure 1. Table 1 shows allelic frequency, heterozygosity and percent polymorphic loci in the three

cattle breeds used. Allele frequency is highest in Red Bororo at allele B with the value 0.6400 and lowest at allele A with the value 0.3600. These values are however lower than those reported by *Douglas (2008)* and *Bessa et al. (2009)* who observed allele frequency ranges of 10.16 to 89.84 and 2% to 10% respectively. Variations observed may be attributable to differences in environmental and /or genetic factors. Sokoto Gudali recorded the highest average heterozygosity of 0.5833 at B allele. The result is in consonance with earlier reports on mean observed heterozygosity of 0.574 and 0.5401 for Kherigarh and Kenkatha cattle respectively (*Pandey et al., 2006*). A 100% polymorphic loci was recorded in this study. This value is at variance with earlier reports of 75% and 98.3% respectively by *Nguyen et al. (2007)* and *Makkawi et al. (2007)*.

Table 1. Allele, heterozygosity and percentage polymorphic loci for the entire population

	Allele	Number of observed allele	Allele frequency	Number of heterozygosity	Heterozygosity frequency	Average heterozygosity	Average heterozygosity (unbiased)	Average heterozygosity (direct count)	% polymorphic loci
Entire population	1	51	0.3806	41.0000	0.6119	0.4715	0.4750	0.6119	100.000
	2	83	0.6194	41.0000	0.6119				
White Fulani	1	23	0.3833	19.0000	0.6333	0.4728	0.4808	0.6333	100.000
	2	37	0.6167	19.0000	0.6333				
Red Bororo	1	18	0.3600	14.0000	0.5600	0.4608	0.4702	0.5600	100.000
	2	32	0.6400	14.0000	0.5600				
Sokoto Gudali	1	10	0.4167	8.0000	0.6667	0.4861	0.5072	0.6667	100.000
	2	14	0.5833	8.0000	0.6667				

The genetic distance and Nei's identities for similarities and relatedness in the breeds are shown in Table 2. Genetic distance is lowest between White Fulani and Red Bororo with the value of 0.0010 and highest between Red Bororo and Sokoto Gudali with the value of 0.0058. A probable reason for the low genetic distance observed between White Fulani and Red Bororo could be attributed to homogeneity in the environmental and management factors prevalent in similar agro-ecological zones where these breeds are found in Nigeria. The two breeds – White Fulani and Red Bororo also recorded highest genetic similarities/ identities (0.9990). This result could be an indication of gene flow due to hybridization between populations (*Oliveira et al., 2005*).

Table 2. Nei's Identities/distances

Populations compared	Distances	Identities	Unbiased distances	Unbiased identities
White Fulani vs Red Bororo	0.0010	0.9990	-.0155	1.0156
White Fulani Vs Sokoto Gudali	0.0021	0.9979	-.0266	1.0270
Red Bororo Vs Sokoto Gudali	0.0058	0.9942	-.0240	1.0243

The population structures of breeds of cattle used in this study are presented in Figure 1 by the dendrogram. Two clusters were identified. White Fulani and Red Bororo are in Cluster 1 with pair wise distances of 0.010. This shows that the two breeds are closely related. Sokoto Gudali is in Cluster 2 with a pair wise distance of 0.020 from Cluster 1 which implies it is not closely related to the other two breeds. Test of Hardy Weinberg's Equilibrium is as indicated in Table 3. Variations in allelic and genotypic frequencies were observed for the AA, AB and BB genotypes across breeds. The gel electrophoresis band shown in Figure 2 also confirm further that polymorphism exist across the cattle breeds being studied. Such variations could be attributed to the response of the breeds to various stimuli which also enhance their survivability and adaptation to the environment. This information could be relevant in breeding planning and strategy for genetic improvement of the Nigerian indigenous cattle breeds.

Table 3: Test of Hardy-Weinberg's Equilibrium

	White Fulani			Red Bororo			Sokoto Gudali		
	Genotype	Observed number of genotype	Expected number of genotype	Genotype	Observed number of genotype	Expected Number of genotype	Genotype	Observed number of genotype	Expected number of genotype
	AA	9	11.4083	AA	9	10.2400	AA	3	4.0833
	AB	19	14.1833	AB	14	11.5200	AB	8	5.8333
	BB	2	4.4083	BB	2	3.2400	BB	1	2.0833
Exact probability			0.1219			0.4095			0.5477

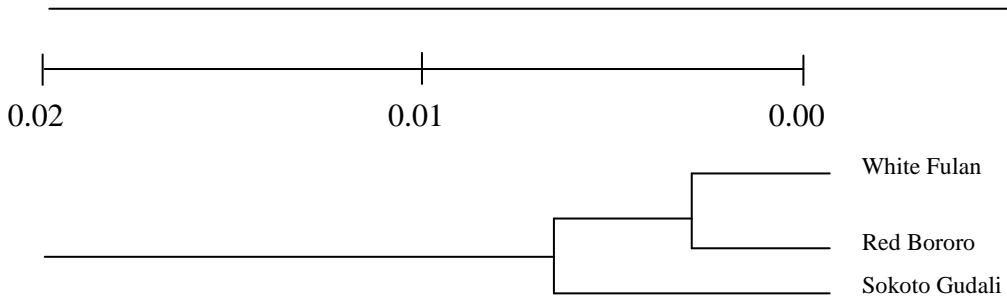


Figure 1. Dendrogram showing genetic variability among White Fulani, Red Bororo and Sokoto Gudali indigenous cattle breeds of Nigeria.

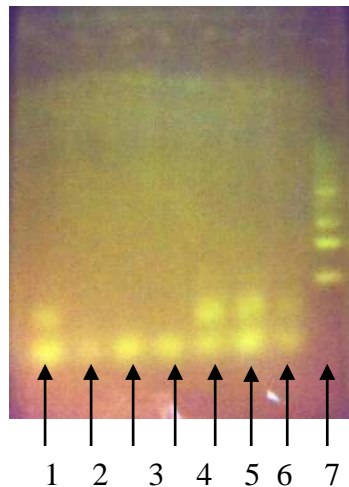


Figure 2. Gel electrophoresis of Thyroid Hormones Responsive spot 14 Alpha Gene (THRSP α) showing both for 7 individuals – 1 is AB, 2 is BB, 3 is BB, 4 is BB, 5 is AB, 6 is AB, 7 is AB and 8 is 100pb Molecular marker

Conclusion

This study revealed that genetic variation exists in White Fulani, Red Bororo and Sokoto Gudali Nigerian indigenous cattle breeds. This study contributes to the knowledge of the genetic variability among the three cattle breeds studied. It also shows that THRSP 14 alpha gene can be used to construct an appropriate measure of variability function through the genetic relationships between these breeds.

The existence of genetic variability allows for the organization of rational conservation and improvement programmes of these breeds based on greater knowledge of their genetic structuring and the relationships between these populations. It was recommended that the number of cattle used be increased in further studies in order to unravel the genetic potentials of Nigerian indigenous cattle breeds for conservation and improvement.

THRSP α u ispitivanju genetske varijabilnosti rasa goveda u Nigeriji metodom lančane reakcije polimeraze (PCR)

Folasade Olubukola Ajayi, Brilliant Ogagaoghene Agaviezor, Ebere Nnah

Rezime

Genetska varijabilnost tri nigerijske autohtone rase goveda - White Fulani (WF), Red Bororo (RB) i Sokoto Gudali (SG), ispitana je pomoću hormona štitne žlezde responsivne tačke 14 Alfa gena (THRSP α), korišćenjem lančane reakcije polimeraze (PCR). Ukupno šezdeset sedam (67) uzoraka krvi goveda su korišćeni u istraživanju, od kojih je 30 WF, 25 RB i 12 SG. DNA je ekstrahovana iz uzoraka krvi korišćenjem Zymobead Genomic DNA extraction kit-a nakon čega je izveden PCR korišćenjem 50 ng template DNA, 1.0 μ M primer (THRSP α), 16 μ l nukleaze – bez vode u BIONEER Accupower premix-u. Gel elektroforeza je izvedena i gelovi ocenjeni. Statističke analize su izvedene korišćenjem GENEPOP, PAST, SPSS verzija 16 i Tools for Population Genetic Analysis (TFPGA) verzija 1.3. Učestalost alela je bila u rasponu od 0.3600 do 0.6400 u A i B alela, prosečna heterozigotnost u rasponu od 0.4608 do 0.4861. Najmanja genetska distanca od 0.0010 između WF i RB i najveća genetska distanca od 0.0058 između SG i RB su identifikovane. Najniži genetski identitet 0.9942 između RB i SG i najviši genetski identitet 0.9990 između WF i RB je takođe identifikovan. Dva (2) klastera genetske populacije su identifikovana u dendrogramu; WFi i RB su u klasteru 1, dok je SG u klasteru 2. Test Hardy Weinberg equilibrium-a otkriva varijacije u frekvencijama genotipova. Ovi rezultati pokazuju varijaciju između ove tri nigerijske autohtone rase goveda koja se pripisuje odgovoru/reakciji rasa na razne stimuluse čime unapređuju svoje preživljavanje i prilagođavanje njihovim specifičnim sredinama. Takva promena može se iskoristiti za očuvanje i unapređenje naših autohtonih rasa kroz selekciju i strategiju odgoja.

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COMPOSITION OF RAW MILK FROM CONVENTIONAL AND ORGANIC DAIRY FARMING

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Abstract: Possible differences between composition of raw milk due to dairy farming system (organic vs conventional) as well as seasonal variations were investigated. The samples were analysed during one year. A total of 6.782 samples of raw milk were collected (4.496 from organic farming). Dairy farms were located in the northern part of Republic of Serbia (Province of Vojvodina). The principle of analysis of raw milk samples was in accordance with the methodology by mid-infrared spectrometry and flow cytometry. The fixed effect of system of farming and season (winter, spring, summer and fall) have shown a high statistical significance ($P < 0.01$) on all examined milk parameters except fat, total solids and somatic cell count, where the impact was slightly lower ($P < 0.05$). Significant difference wasn't found in number of bacterial colonies ($P > 0.05$). Composition of milk is also affected by a number of other factors, therefore it is recommended to involve factors such as nutrition of dairy cows, breed and farm management.

Key words: milk composition, organic milk, conventional milk, dairy farming, Holstein- Friesian dairy cows

Introduction

Milk production in accordance with the principles of organic production demands that the livestock production system adapt to specific technology-in first line origin of feed, reproduction, health, welfare and behavior of animals (*Rosati and Aumaitre, 2004*). Organic agriculture has achieved a positive contribution to the preservation of the environment, reduction of arable land and residual pesticides and thus has a direct effect on the improvement of human health (*Bloksma et al. 2008*). Based on the available data it can be concluded that organic farming globally is growing fastest in the Europe (*FiBL & IFOAM, 2015*). The number of cattle in organic farming in the EU for the last decade had a growth rate of over 12%. The share of this production in 2014 accounted about 3% compared

to conventional cattle, the share of dairy cows accounted for about 0.7 million certified animals (*Eurostat data, 2014*). Organic raw milk compared with milk from conventional production has a better fatty acid composition i.e. contains more polyunsaturated fatty acids - PUFA with a higher proportion of omega-3 fatty acids and conjugated linoleic acid (CLA) (*Ellis et al. 2006; Prandini et al. 2009*). The mentioned group of fatty acids (especially omega-3) has positive effects on human health. Their activity is associated with improvement of neurological function (*Contreras and Rapoport, 2002*), decreases the risk of diabetes, prevents the occurrence of cardiovascular diseases, improves function of the immune system (*Paris, 2003; Wahl et al. 2004*). Besides the increased content its also important ratio between fatty acids (*Prandini et al. 2009*). Milk from organic production contains more α - tocopherol and vitamin A (*Pentelescu, 2009*). For determination of fatty acids (FA) content the gas-liquid chromatography is most often used. This method requires time-consuming procedure, expensive reagents, and qualified staff. The mid-infrared (MIR) spectrometry method could be a good alternative for assessing the fatty acid profile of dairy products (*Soyeurt et al. 2006*). On the nutritional value of milk, the most direct impact has a diet and method of rearing (*Slots et al. 2009; Stergiadis et al. 2012*). Diet and nutrition in organic farming are based on the increased using of pasture, grass silage, hay and less on the use of concentrates and maize silage. Therefore, a positive influence of nutrition can be expected on content and ratio of fatty acids, omega-3 fatty acids and CLA, lipophilic vitamins, β -carotene, lutein as well as high level of antioxidants (*Bloksma et al. 2008; Sakowski et al. 2012; O'Donnell et al. 2010; Popović-Vranješ et al. 2011*).

The aim of this paper is to investigate possible differences between composition of raw milk due to dairy farming system (organic vs conventional) as well as seasonal variations.

Materials and Methods

Animals and System of farming

Three conventional and one organic dairy farm located in the northern part of Republic of Serbia (Province of Vojvodina) were included for the twelve -month research. Dairy farms were distributed in different regions and housed from 100 to 900 Holstein-Friesian (black and red) dairy cows. All cows were loose housed-free stall stable. Cows in conventional farming (CF) were fed a total mixed ration (TMR) consisting mostly of maize silage, lucerne silage, straw, lucerne hay, concentrates, and mineral supplements. The feeding of the cows in organic farming (OF) was based on home-grown fodder and mainly grass and grass-clover products (including home-grown grains). In both of farming systems the composition of the

diet corresponded to the daily milk yield of cows, so that feed rations were completely balanced in accordance with the cow's needs.

Sampling and Instrumental analysis

Sampling of each cow's milk started in December 2014 and lasted until December 2015. All samples were delivered for analysis to the central laboratory for milk quality at Faculty of Agriculture Novi Sad. The laboratory is accredited in accordance with the international standard (ISO 17025). The collection of samples is carried out in compliance with the regulations of the International Committee for Animal Recording (ICAR- AT4). For preservation of milk samples Asidiol was used in accordance with ISO 13366-2:2006 and IDF 148-2:2006. A total of 6.782 samples of raw milk were collected, 4.496 of which were from OF and the rest from CF. The analyses of raw milk samples were carried out on the FOSS instruments-CombiFoss™ FT+. This device is a combination instrument consisting of the MilkoScan™ FT+ and the Fossomatic™ FC. The principle of measurement of raw milk samples is based in accordance with the methodology by mid-infrared (MIR) spectrometry method and flow cytometry (FC). The following parameters were analyzed: fat, protein, lactose, total solids, somatic cell count (SCC), number of bacterial colonies (CFU-colony forming unit), milk urea (MU), and contents of fatty acid (FA): Saturated (SFA), unsaturated (UFA), polyunsaturated (PUFA) and monounsaturated Fatty Acids (MUFA).

Statistical analysis

The data was evaluated by the *STATISTICA* statistical software (Ver. 10 StatSoft Company, 2011). The average values and variability of examined parameters as well as the effect of factors (system of farming and season as a fixed effect) on investigated milk traits were studied by means of the procedures PROC UNIVARIATE and PROC GLM-General linear model. The model equation used for the evaluation was as follows:

$$Y_{ijk} = \mu + S_i + R_j + e_{ijk};$$

where:

Y_{ijk} – dependent variable (fat, protein, lactose, total solids, SCC, CFU, MU, SFA, UFA, PUFA, MUFA);

μ – mean value of dependent variable;

S_i – fixed effect of the System i ($i = 1,2$);

R_j – fixed effect of the Season j ($j = 1,2,3,4$);

e_{ijk} – other random effects.

In order to properly adjust SCC and CFU parameters to normal distribution, logarithmic transformation was used as follows:

- 1) $SCC = \text{Log}_2 (SCC / 100000) + 3$
- 2) $CFU = \text{Log}_{10} (CFU)$

Results and Discussion

The results from the GLM model are given in Table 1. The fixed effect of system of farming (OF vs CF) and season (winter, spring, summer and fall) have shown a high statistical significance ($P < 0.01$) on all examined milk parameters except fat, total solid and somatic cell count (as a LogSCC), where the impact was slightly lower ($P < 0.05$). Significant difference wasn't found in the number of bacterial colonies ($P > 0.05$).

Table 1. The effect of system of farming and season on examined milk parameters

Source of variation	System OF vs CF	Season	System x Season
Parameters	F – value		
Fat	11.77*	75.83*	34.44*
Proteins	140.89**	46.94**	13.2**
Lactose	88.52**	45.84**	17.73**
T.Solids	17.15**	49.04*	62.81*
LogSCC	226.83*	3.59*	3.07*
Urea	336.07*	293.97**	29.58**
SFA	125.49*	57.89	21.65**
UFA	91.57**	64.69**	44.30**
MUFA	153.81**	108.83	35.71**
PUFA	55.67**	213.83	76.07**
LogCFU	0.47 ^{n.s.}	50.66 ^{n.s.}	11.25 ^{n.s.}

The results are in accordance with other studies comparing OF vs CF milks parameters (Kouřimská et al. 2014; Popović-Vranješ et al. 2014; Toledo et al. 2002). The average value of fat content (4.23 – 4.26%) has shown very small absolute differences between investigated type of farming. The fat content was slightly lower in OF which is in line with some studies (Hanuš et al. 2008; Nauta et al. 2006). Diet and breed have a major influence on milk fat content. Fluctuation of milk fat content and fatty acid composition is under great influence of these two factors (Jenkins and McGuire, 2006; Kelsey et al. 2003; Kučević et al. 2014).

Values of the coefficient of variation (CV) and standard deviation (SD) indicate that the variability of this trait is more under the influence of biological and breed characteristics of dairy cows, than rearing conditions present on the farms (*Kučević et al. 2011*). The protein content was significantly higher in organic milk while lactose and total solids were lower (Table 2).

Table 2. Results of analyses for raw milk parameters

Milk Components	Organic dairy farming				Conventional dairy farming			
	n	mean	SD*	CV(%)**	n	mean	SD	CV(%)**
Fat (g/100 g)	4496	4.23 ^a	0.65	15.1	2286	4.26 ^b	0.67	16.2
Proteins (g/100 g)	4496	3.53 ^A	0.37	11.2	2286	3.43 ^B	0.30	9.2
Lactose (g/100 g)	4496	4.59 ^a	0.20	4.4	2286	4.65 ^b	0.19	4.3
T.solids (g/100 g)	4496	13.12 ^a	0.80	6.1	2286	13.17 ^b	0.85	6.3
LogSCC	4493	2.86 ^a	1.22	43.1	2286	3.31 ^b	1.16	35.2
LogCFU	4496	4.89 ^{aa}	0.38	7.3	2286	4.88 ^{aa}	0.38	8.6
Urea (mg/dL)	4496	25.49 ^a	8.15	32.2	2286	29.21 ^b	6.51	22.3

Occurrence of the higher value of the proteins in the organic milk isn't expected because diet doesn't contain concentrate feeds. High content of raw protein in fresh and dried fodder probably influenced the amount of protein in the raw milk. In relation to these parameters, there is a lot of contradictory results in the literature. There are studies conducted between two systems of farming relative to comparison of the protein content. Results of these researches showed low content of protein in OF (*Kouřimská et al. 2014; Hanuš et al. 2008; Kuczynska et al. 2012*). Some studies show similar values or no difference in protein contents between organic vs conventional milk as obtained in our research (*Zagorska and Ciprovica, 2008; Butler et al., 2008*). In addition to already mentioned factors, important are impacts by sire, year of birth, length of lactation, calving season, breeding region etc. These factors also significantly affect milk yield, milk fat, and protein content in standard lactation - 305 days (*Petrović et al., 2006; Sekerden, 1997; Kučević et al., 2014*). These references are pointing to the fact that comparing the quality of milk in two different types of farming should include numerous genetic and paragenetic factors in research. Significant difference between systems of farming was found in the somatic cell count but not in the number of bacterial colonies ($P > 0.05$). Average number of SCC ranged from 97.441/ml (log 2.86) to 161.874/ ml (log 3.31) while average number of bacterial colonies were 97.400 /ml (log 4.89) to 128.73/ ml (log 4,88). In relation to the number of SCC numerous of studies showed opposite results and conclusion.

Toledo et al. (2002), Cermanova et al. (2011) found a lower SCC in organic milk while higher or no statistically significant difference of SCC was found by (Kuczynska et al., 2012). Right away after leaving the udder, the milk of healthy cows, kept in adequate breeding conditions, is almost sterile and contains the minimum number of microorganisms (8.933 CFU / ml) (Kučević et al., 2013). Inflammation of the mammary gland is accompanied by changes in the number of SCC, mainly as an increase in SCC in diseased quarters of udder (Fregonese and Leaver, 2001). By monitoring the change in SCC can successfully manage the health of the udder. Occurrence of diseases of the mammary gland is usually associated with low level of hygiene during the breeding and milking (Schreiner and Ruegg, 2003). To contamination comes mainly during and after milking (after leaving the udder) due to activity of microorganisms from the environment.

Statistically significant difference was found in milk urea (MU). The mean value of MU was higher in CF (29.21 vs 25.49 mg/dL). Our results are similar to the values for MU obtained by Čobanović et al. (2015), Bastin et al. (2009) and Bandelja et al. (2011) for Holstein breed. Higher values of MU indicate an imbalance of protein and energy, but MU concentration is also influenced by a whole range of factors (feeding, breed, stage and number of lactations, body weight, daily production and chemical composition of milk, somatic cell count, season and milking) (Čobanović et al., 2015). Considering that results of this study showed a higher protein content in raw milk and lower value of MU in OF. It can be concluded that cows in OF had greater access to highly degradable carbohydrates in the diet than the cows in the CF. The average content of saturated, unsaturated, monounsaturated and polyunsaturated fatty acids (FA) in the raw milk (organically and conventionally produced) is presented in Table 3. According to the results, it is evident that there was a highly statistically significant difference between dairy farming in all tested parameters.

Table 3. Concentrations of fatty acid in raw milk

FA (g/dL of Milk)	Organic dairy farming				Conventional dairy farming			
	n	mean	SD	CV(%)	n	mean	SD	CV(%)
SFA	4496	2.52 ^A	0.45	18.1	2286	2.64 ^B	0.46	17.2
UFA	4496	1.38 ^A	0.28	20.3	2286	1.29 ^B	0.29	22.1
MUFA	4496	1.55 ^A	0.23	15.5	2286	1.46 ^B	0.24	16.8
PUFA	4496	0.38 ^A	0.04	11.2	2286	0.37 ^B	0.03	9.3

Milk produced in organic dairy farming had a significantly lower concentrations of SFA (2.52 – 2.64) but more UFA (1.38 -1.29). On the other hand, participation of PUFA was higher in the organic milk (0.38 – 0.37). This ratio of FA in organic milk is preferred in terms of positive impact on human health. These

results are partially in compliance with *Ellis et al. (2006)*, *Popović-Vranješ et al. (2011)*. In their research recorded that conventional and organic milk didn't differ with respect to milk SFA content, but organic milk had a higher percentage of PUFA compared with conventional milk.

Seasonal variability in the composition of FA is highly expressed during the year. Data are presented in Figure 1 and 2.

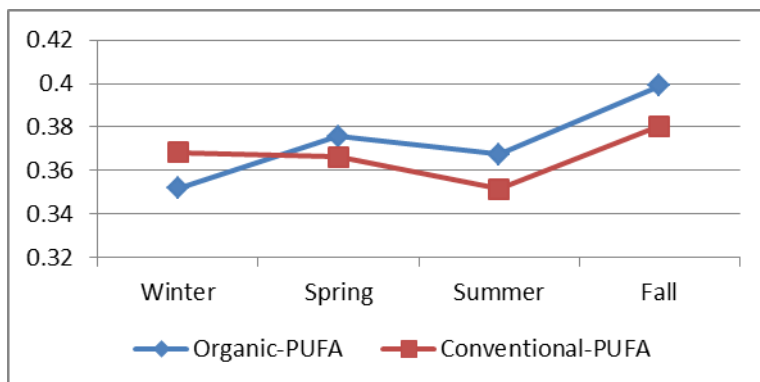


Figure 1. Seasonal variability of concentrations of PUFA in organic and conventional milk

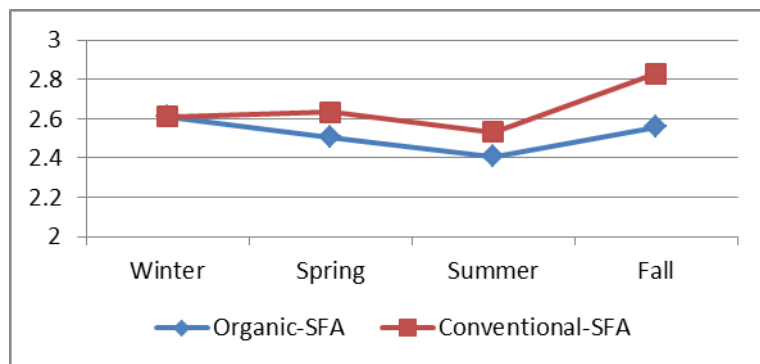


Figure 2. Seasonal variability of concentrations of SFA in organic and conventional milk

In organic dairy farming, the content of PUFA started increasing in late spring to early summer when the cows were put out on pastures or received fresh grass-clover products. Simultaneously with the increase of the content of PUFA, content of SFA in OF was gradually decreasing by the beginning of the fall. On the Figure 1 and 2 it can be clearly noted that the season identically affects conventional dairy farming too. A result of the effect of seasonality (variability by month of year) on milk FA content is in a line with other studies (*Ellis et al., 2006*; *Popović-Vranješ et al., 2011*; *Pentelescu, 2009*). All the authors pointed out that

FA content of milk is mostly influenced by nutrition. Using of TMR increased the proportion of milk SFA even during spring and summer. *Ellis et al. (2006)* emphasizes that knowledge of the effects of season, access to fresh grazing, or usage of specific silage types could be used by producers to enhance the content of beneficial FA in milk.

Conclusion

Composition of raw milk is affected by farming system (organic vs conventional), as well as seasonality during the year. Milk produced in organic dairy farming had a significantly lower concentration of saturated fatty acids but higher share of polyunsaturated fatty acids. This distribution of fatty acids in organic milk is preferred in terms of positive impact on human health. Composition of milk is also affected by a number of other factors. Therefore it is recommended to involve factors such as nutrition of dairy cows, breed and farm management during the comparison of conventional and organic dairy products. Regarding the diet of cows, special consideration should be given to the access to fresh grazing, silage type, cereal feeding etc., because nutritional factor takes a greatly impact on the composition of milk.

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Sastav sirovog mleka dobijen u sistemu konvencionalne i organske proizvodnje

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Rezime

Cilj rada je bio da se ispituju razlike između sirovog mleka dobijenog u različitim sezonama i sistemima proizvodnje (organska/ konvencionalna). Uzorci mleka su sakupljeni u toku čitave godine. Ukupno je sakupljeno 6782 (od čega 4496 iz organske proizvodnje). Farme su bile locirane na severnom delu Srbije (AP Vojvodina). Sirovi uzorci mleka su analizirani u skladu sa metodologijom infracrvene spektrometrije i protočne citometrije. Fiksni uticaji sistema proizvodnje i sezone (zima, proleće, leto i jesen) su pokazali visoku signifikantnu značajnost (P

< 0,01) na sve ispitivane parametre mleka osim na mlečnu mast, ukupnu suhu materiju i ukupan broj somatskih ćelija, gde je uticaj bio signifikantan na nivou ($P < 0,05$). Statistički značajna razlika nije pronađena u ukupnom broju bakterija (kolonija) ($P > 0,05$). Na sastav mleka takođe utiče i veliki broj drugih faktora, stoga je za preporuku da se u ispitivanje uključe i faktori poput ishrane mlečnih krava, rase i farmskog menadžmenta.

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FACTORS INFLUENCING PRODUCTIVE TRAITS OF AWASSI CROSSBREDS IN MACEDONIA

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Abstract: The purpose of the survey was to determine a bond between certain factors that affect most of the production traits in crossbreeds between Awassi and Ovchepolian indigenous sheep in Macedonia. We implemented 1145 individual lactation controls in two years of production (2012 and 2013). The impact of individual factors is studied using F-test and the analyses are made using SPSS set of programs. Many factors (lactation, lambing month and number of milk controls) had highly significant impact ($P < 0.001$) on daily milk production (morning, evening and total milk, fat percentage and fat kg) in this breed of sheep. Only fertility had no impact on any of the examined factors, with the exception of total daily milk, on which manifested with highly significant impact ($P < 0.01$). The average milk yield in examined crossbred sheep was 109 ± 0.479 l in two years of production, while the production of milked milk was 72 ± 0.421 l for the same period of time. The length of lactation period in these sheep was average 203 ± 0.61 days, for two analysed years. In relation to the age, in second lactation sheep was determined somewhat higher daily milk yield (0.478 ± 0.01 l.), compared to sheep in first lactation (0.475 ± 0.01 l.). This is quite logical, keeping in mind the lactation curve in sheep, especially those of dairy breeds to which Awassi breed belongs.

Key words: Awassi crossbreeds, production traits, impact of factors, daily milk yield

Introduction

The attempts for advancing the genetic potential of certain sheep production traits in Macedonia exist for 60 years now. Awassi breed of sheep has

special significance in this context. It is the first dairy sheep breed used for cross-breeding in order to increase the production of milk in indigenous and other breeds of sheep in Macedonia. This breed is imported in Macedonia for the first time in 1969. Since then it has been assumed that the percentage of crossbreeds between Awassi with Ovchepolian sheep is around 4% of the current number of sheep in Macedonia (689.938 sheep), while the percentage of purebred Awassi in the state in this moment is 0.3% (*www.fva.gov.mk*).

The Awassi is a fat-tailed breed found extensively in Syria, Lebanon, Iraq, Jordan, Southern Turkey, Israel and to a lesser extent in some other WANA countries (West Asia and North Africa), (*Gursoy et al., 2001*). The breed adapts to a wide range of environmental conditions from the steppe to the highly intensive system (*Galal et al., 2008*).

The improved type of Awassi sheep found on intensive dairy farms in Israel has been selected for increased milk production for over 50 years (*Epstein, 1985*). The use of improved type of Awassi sheep has become widespread in many countries, but very few of the environments where they are kept, are the same as found in Israel (*Gursoy et al., 2001*). The intensively managed dairy ewe flocks in Israel have somewhat different characteristics to the majority of other dairy flocks in the Mediterranean region (*Pollott and Gootwine, 2001*).

The Awassi breed has proliferated, for breeding purposes, into at least 30 countries in all continents. The great majority of these proliferations were from Israeli Improved Awassi, Turkish Awassi and the Syrian Awassi either directly or indirectly through a third country. Some of the countries introducing Awassi used it as an improver breed (*Galal et al., 2008*).

Awassi is present in Spain, imported in 1971 (*De la Fuente et al., 2006*), in Bulgaria in 1977, in Romania in 1973, in Hungary in 1989 and also present in Greece and Albania (*Kukovics et al., 2006*).

In Australia Awassi has been imported in 1987 (*Jassim et al., 2006*), and in Turkey the number of Awassi in 90s was around 1 million heads (*Pekel et al., 1994*).

In the intensive dairy flocks found in Israel, the ewe's milk is collected from birth until drying off, with no lamb suckling. This milking regime allows the study of the complete lactation of dairy sheep and coupled with the complete pedigree recording in such flocks, makes a study of the genetics of complete dairy ewe lactations possible (*Pollott and Gootwine, 2001*).

According the same authors (*Pollott and Gootwine, 2001*), there is widespread interest in using Israeli Awassi genes in different environments and in grading-up programmes. Given the high level of permanent effects found in many studies, we might expect to see recombination losses in milk yield in Awassi crosses. Some evidence for this has been presented by *Gootwine et al., (2001)* when introgressing the Booroola Merino into the Awassi.

However, besides diffuseness and results, in our scientific and specialist work, attitudes and opinions are divided in relation to this breed of sheep. According to some authors, this breed is suitable for cross-breeding with Macedonian indigenous breeds of sheep, because its origins are from Middle East and it adapts easily to the dry and hot climate in Macedonia, while significantly affects the increase of milk production in offspring. According to others, this breed is not the most appropriate for cross-breeding, because lambs from these half-breeds have increased percentage of fat tissues in tail, which is an undesirable trait among the largest consumers of Macedonian lamb (Italy and Greece).

However, our intention was to determine average annual and milked milk yield in these crossbreeds using proper monitoring and measurement and the degree of advancing the crossbreeds in milk production. At the same time, the goal was to determine the impact of certain factors (year, lactation, lambing month, month of milk controls, number of milk controls and fertility), affecting the traits of daily milk yield.

Materials and Methods

We took crossbreeds from F1 generation as analysis material, generation derived from Awassi rams and Ovchepolian sheep population, located on a farm near Gevgelija, southeastern Macedonia.

The analyses lasted 2 years (2012 and 2013). There were 89 heads included in 2012 and 88 in 2013 (Table 1).

Table 1. Number of tested sheep per year

Population	Year		Total
	2012	2013	
Crossbreeds F ₁ generation	89	88	177

Sheep controlled during 2012 were in first lactation, while in 2013 were in second. Table 2 shows the age structure of tested sheep per year.

Table 2. The age structure of the tested sheep per year

Year	Lactation (Parity)		Total
	I	II	
2012	89	/	89
2013	/	88	88
Total	89	88	177

During the experiment there were 575 lactation controls performed in these crossbreeds in first lactation and 570 controls in second lactation or a total number of 1145 individual lactation controls (Table 3).

The farming system was combined (stall and pasture) with usage of available vegetation on pastures the most of the year (7-8 months). Besides pasturing, sheep had alfa-alfa hay (500 gr. daily) during the period from November to February and concentrated product (300 gr./daily) during the period from November to April.

Milking period began after refusing the lambs (2 months after lambing) and lasted from August to October, depending on individual milk production.

Individual milk yield was controlled according A4 standard method (ICAR, 2009) which includes daily milk measurement that lasted 28 to 34 days. Milk recordings were initiated 10 days after lambing and lasted until the moment of drying, in the middle of October. The measurement of the milked milk was carried out with laboratory jigger with an accuracy of 10 milliliters.

During the suckling period, milk control regarding the specificity of the moment (lamb suckling), was carried out in such way that before morning milk control, lambs were separated from their mothers for 12 hours. Afterward, the lambs were returned to their mothers for 24 hours and the next morning were separated again, 12 hours before evening milk control.

The average number of performed controls was 7. Total individual sample of milk was taken in each milk control. Milk samples were collected in 50 ml plastic bottles, (25ml from the morning and evening milking) for the analyses of milk fat.

Based on these measurements of milk yield, the following was calculated:

- Total milk production in one lactation, in liters (l);
- Total milked milk of one lactation, in liters (l);
- Amount of milk consumed by lambs, in liters (l);
- Suckling period length in days;
- Length of lactation, in days.

Concerning the statistical processing, the traits of daily milk production (morning, evening and total amount of milk, % of fat and kg fat) were analysed using the following model:

$$Y = \mu + Y_j + L_k + MBl + TDm + TMn + Fo + ejklmno$$

where:

Y is the individual observation of each trait during a test (daily) control (morning, evening and total amount of milk, % of fat and kg fat);

μ is the general common average of the tested traits;

Y_j is the effect of the j-th year with (j=2012 and 2013);

L_k is the effect of the k-th lactation with (k=1,2);

MBl is the effect of the l-th lambing month (l= January, February);

TDm is the effect of the m-th recording month, with (m= February, March, April, May, June, July, August);

TMn is the effect of the n-th recording day, with (n= 1, 2, 3, 4, 5, 6, 7);

Fo is the effect of the o-th number of newborn lambs, with (o=1, 2);

eijklmno is the residual influence

The impact of certain effects was studied using the F-test, while statistical analyses were performed with the Statistical Package SPSS, version 13, (2004).

Results and Discussion

According to data in Table 3, the lactation yield for two analysed years in crossbreeds from F1 generation between Awassi rams and Ovchepolian indigenous ewes was average 109 ± 0.479 l, while the amount of milk consumed by lambs was average 37 ± 0.253 l. The production of milked milk in this population is average 72 ± 0.421 l for two analysed years, while the average length of suckling period is 54 ± 0.377 days. The length of lactation period in this population is average 203 ± 0.61 days for two analysed years. The table shows large variation between minimal (59.37 l) and maximal (169.33 l) amount of lactation yield in these crossbreeds, which points to disagreement. This is quite logical for crossbreeds from F1 generation.

Table 3. Descriptive statistics data of tested sheep population, LS-mean \pm SE

Parameter	N	Mean	Min	Max	Std. deviation	Cv
Lactation milk (litres)	89	109 \pm 0.479	59.37	169.33	20.17	18.50
Suckling milk (litres)	89	37 \pm 0.253	17.83	68.87	10.25	27.70
Milking milk (litres)	89	72 \pm 0.421	13.97	126.84	18.23	25.32
Length of suckling period (days)	89	54 \pm 0.377	33.5	86	13.54	25.07
Length of lactation (days)	89	203 \pm 0.61	198	235	29.46	14.51
Morning milk yield (litres)	1145	0,254 \pm 0.002	0.1	0.8	0.05	19.69
Evening milk yield (litres)	1145	0,282 \pm 0.002	0.12	0.65	0.082	29.08
Total daily milk yield (litres)	1145	0,536 \pm 0.04	0.22	1.45	0.028	5.22
Fat (%)	1145	7,36 \pm 0.02	5.41	9.22	0.678	9.21
Daily fat yield (kg)	1145	0,039 \pm 0.003	0.017	0.112	0.010	25.64

The determined annual milk production in our analyses was slightly lower (109 l), compared to the analyses of *Tokovski et al., (1977)* where it was determined 128.5 l milk yield in crossbreeds from F1 generation between Awassi rams and Ovchepolian ewes. In the same analyses, in other group of crossbreeds in first lactation, these authors determined lower milk yield (83.98 l) compared to the amount of milk yield from our analyses. Like us, these authors determined large variations between minimal and maximal lactation yield, ranging from 26.57 l to 114.75 l.

Todorovski et al. (1979) found significant variations between minimal (84 l) and maximal (195 l) annual milk yield in crossbreeds between Awassi and Kosovo sheep. These authors found lactation milk yield between 128 l and 261 l in

the same crossbreeds in second lactation. They also found highly significant lactation yield of 193 kg in crossbreeds between Awassi and Domestic sheep population. In some analyses the same authors found higher milk yield compared to our findings (*Todorovski et al., 1979; Todorovski et al., 1985*) where average lactation yield in these crossbreeds is 180 l.

Comparing the milk yield in our analyses (109 l) with the common one to Ovchepolian sheep as the largest indigenous breed of sheep (68.31 l), according to many authors (*Tashkovski et al., 1968; Shokarovski, 1957; Tashkovski, 1961; Tashkovski and Tokovski, 1969; Tokovski et al., 1977; Shokarovski et al., 1992*), we come to a conclusion of significant increase of milk yield, using Awassi as breed for advancement of this trait.

Seeing that Macedonia is not the only country where Awassi is used as breed improver, the milk yield in crossbreeds between Awassi and Kazakh fat-tailed sheep was 119.2 l, while in indigenous Kazakh sheep is only 42.6 l (*Malmakov et al., 2006*).

According to *Iniguez et al. (2006)*, in crossbreeds between Turkish and Syrian Awassi the milk yield is 119.18 kg, while in similar feeding conditions the milk production in Syrian sheep was 109.41 kg. *Gursoy et al. (2001)*, found an increase of 14% milked milk in first 120 days of lactation in F1 ewes, between Israeli Improved Awassi rams and pure Ceylanpinar Turkish ewes. To study the effect of crossbreeding Awassi with Romanov sires on litter size and milk production, *Kutluca et al. (2011)*, in crossbreeds was determined average milk yield of 104.3 kg in 157.2 days of lactation. Keeping in mind the milk production of Romanov sheep (44 kg milk), (*Boylan, 1998*), we found an increase of 137 % of lactation milk in these crossbreeds in relation to Romanov sheep.

Analysing these results, we come to a conclusion that they are close to ours, which indicates the degree of genetic improvement in low yield capacity sheep, when Awassi is used as breed improver. Herewith goes the assertion of *Galal et al. (2008)* that using Awassi as an improver breed outside the countries of origin had generally favorable effect on milk production with varied effects on fertility and lamb production.

Impact of factors

The results from these analyses are obtained on the basis of determined individual impact of mentioned factors (year, lactation, lambing month, month of milk recording, number of milk recordings and fertility) on daily milk production. By appointing the impact of the year, it is the impact of diet that is actually determined, while lactation represents the impact of age on milk yield. By determining the impact of lambing month, from the aspect of annual milk production in sheep it is actually the most convenient moment for lambing that is determined. The knowledge of oestrus control and fertilization in sheep is additionally enriched, using artificial methods to determine the moment of partus

(mating of sheep) and thus influence directly on the amount of obtained lactation milk.

The number of milk recording represents the stage of lactation and this factor is especially important for obtaining knowledge about lactation curve in each head, but on the other side represents important information in individual selection of sheep.

The month of milk recordings also determines the influence of diet, climate and other paragenetical conditions affecting sheep productivity.

While determining the impact of fertility on milk production, we tried to obtain additional knowledge about whether the number of lambs per sheep, affects daily milk yield in this genotype of sheep.

Analysing the results in Table 4, we can conclude that most of the factors (lactation, lambing month and number of milk recordings) had highly significant influence ($P < 0.001$) on daily milk production (morning, evening and total amount of milk, % of fat and kg fat) in tested crossbreeds. The year also manifested with highly significant impact on most of the traits ($P < 0.001$), with the exception of level influence on morning milking which was lower ($P < 0.05$). The month of milk recording also had highly significant influence ($P < 0.001$) on all traits, with the exception of milk fat percentage, on which this factor had no impact at all ($P > 0.05$).

Table 4. Factors influencing daily milk production in F_1 crossbreeds, F-test and its significance (F-statistics)

Factor	Df	Morning	Evening	Total	Fat (%)	Fat (kg)
Year	1	3.694*	18.240***	5.525***	79.164***	13.829***
Lactation	1	98.114***	156.477***	140.096***	29.597***	116.759***
Month of lambing	1	10,361***	9.367***	10.763***	9.503***	11.123**
Month of milk recording	6	54.256***	35.890***	49.180***	1.004 ^{ns}	30.941***
No of milk recordings	6	52.572***	50.380***	56.046***	9.304***	33.870***
Fertility	1	0.00 ^{ns}	0.273 ^{ns}	0.078**	1.117 ^{ns}	0.446 ^{ns}
R^2 – Coef. determination	/	0.646	0.685	0.689	0.326	0.627

ns – $P > 0.05$, * - $P < 0.05$, ** - $P < 0.01$, *** - $P < 0.001$

Only fertility had no impact on variation of parameters, with the exception of total daily milk on which manifested with high significant influence ($P < 0.01$).

The determination coefficient of examined factors in these sheep, ranged from 0.326 for milk fat percentage, to 0.689 for total daily milk. *Pacinovski et al. (2012)* found highly significant impact on almost all factors, with the exception of fertility, that had no impact on daily milk production in indigenous Ovchepolian

sheep in Macedonia. Same authors (*Pacinovski et al., 2014*), in other analyses found similar impact on almost all factors mentioned, with the exception of fertility on daily milk production in Awassi sheep.

Kastelic et al. (2013) found significant impact on milk production and milk fat in Istar sheep as indigenous breed in Slovenia and Croatia. *Pacinovski et al. (2013)* found similar year impact on lactation peak in East Friesian ewes in Macedonia.

The poor control of diet, typical of dairy sheep production systems based on grazing, can substantially limit the ability of sheep producers to control and modify the quantity and quality of milk (*Pulina et al., 2006*).

As housing, flock management, especially feeding (*Bencini, 2001; Pulina et al., 2006*), all strongly affect the quantity and composition of ewes' milk, it is possible to improve characteristics of produced milk by manipulating the above mentioned factors.

The impact of all these factors is particularly shown in the following tables (Tab. 5 – Tab.10).

Impact of year on daily milk production in tested sheep population

The year had highly significant impact ($P < 0.0001$) on most analysed traits of daily milk production in tested crossbreeds, except on morning milk where it had lower influence ($P < 0.05$), (Tab. 4).

According to data in Tab. 5, the average daily milk yield in these sheep is somewhat higher in 2013, (0.546 ± 0.011), unlike in 2012 with lower daily milk yield (0.526 ± 0.011). This is quite logical if we take in consideration that with sheep ageing (to certain level) also increases the annual milk production, and the crossbreeds in our experiment in 2012 were in first lactation, while in 2013 in second lactation.

Same refers to morning and evening milking as for the amount of milk fat obtained, which showed certain degree of higher values in 2013 compared to 2012. Only the percentage of milk fat was in opposite tendency as a result of logic consequence by increasing milk production, decreasing the percentage of milk fat, which is observed in this case.

Table 5. The impact of the year on daily milk production in F₁ crossbreeds, LS-mean \pm SE

Year	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
2012	89	0.247 \pm 0.004	0.279 \pm 0.01	0.526 \pm 0.01	7.27 \pm 0.03	0.038 \pm 0.001
2013	88	0.261 \pm 0.004	0.285 \pm 0.01	0.546 \pm 0.01	7.46 \pm 0.03	0.041 \pm 0.001

Effect of year on composition of milk as reported by *Ploumi et al. (1998)* for Chios sheep, while *Macciotta et al. (2005)*, for Sarda goats, proved the effect of year on fat and protein content.

Hassan (1995), did not find any significant effect of the year on composition of milk of Ossimi and Saidi sheep and their crosses with Chios. Also, any significant effect of the year on fat content of milk did not find *Matutinovic et al. (2011)*, at Dalmatian Pramenka (indigenous sheep breed in Croatia).

Analysing the daily milk yield in crossbreeds between Awassi and Ovchepolian sheep, *Tokovski et al. (1977)* found average daily milk yield of 714 ml, for 180 days of lactation. In the same analyses they found average milk yield of 515 ml for another group of crossbreeds.

Todorovski et al. (1979) found significantly higher daily milk yield (1.15 l) compared to the milk yield from our analyses in first lactation in crossbreeds between Awassi and Kosovo sheep, while still higher in second lactation 1.49 l.

Same authors (*Todorovski et al., 1985*) found 1 liter of daily milk yield in crossbreeds between Awassi and Ovchepolian sheep, which is also significant advantage compared to our analyses.

Impact of lactation (parity) on daily milk production in tested sheep population

Lactation (parity) had highly significant impact ($P < 0.001$) on all tested traits of milk production (Tab. 4) in these analyses. The average daily milk yield in these crossbreeds insignificantly increases or grows from first (0.475 ± 0.011) to second lactation (0.478 ± 0.011), (Table 6). Same happens with morning milk and the amount of daily milk fat obtained. Only a slight decline was observed in evening milk, but the difference was not significant. The percentage of milk fat is quite illogical, which instead of decreasing in some value according to the amount of total milked milk during the day, it has increased from first (7.45 ± 0.051) to second lactation (7.67 ± 0.061). For such inconsequence, the most likely cause is the paragenetical factor – diet, which can significantly influence on the content of milk fat and thus on the quality of milk.

Table 6. The impact of the lactation on daily milk production in F₁ crossbreeds, LS-mean \pm SE

Lactation	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
1	575	0.224 \pm 0.01	0.251 \pm 0.01	0.475 \pm 0.01	7.45 \pm 0.05	0.035 \pm 0.001
2	570	0.230 \pm 0.01	0.248 \pm 0.01	0.478 \pm 0.01	7.67 \pm 0.06	0.037 \pm 0.001

In relation to the increase of milk production and age, similar results are obtained in other breeds of sheep from Mediteranean region, such as Massese and Comisana sheep, as Italian indigenous breeds of sheep (*Sevi et al., 2000; Carnicella et al., 1989*) and Latxa sheep as Spanish indigenous breed (*Ruiz et al., 2000*). *Sevi et al. (2000)* found tendency to increase the amount of milk fat produced by increasing lactation, same authors (*Sevi et al., 2004*) observed similar

results during the study of effects of stage of lactation on ewe's milk composition in other analyses.

Gootwine and Goot (1996) and *Pollot and Gootwine (2001)* found significant influence of lactation (age) on average milk production in Awassi, concluding that age and lactation increase.

Impact of lambing month on daily milk production in tested sheep population

In relation to all tested traits of daily milk production, the month (season) of lambing had highly significant influence ($P < 0.001$), (Tab.4).

Analysing this factor, higher daily milk yield is recorded in tested sheep population in those who lambed in January (0.496 ± 0.01), (Tab. 7). Same happens with the morning and evening milk. Analogously, sheep lambed in February had higher ($7.59 \pm 0.07\%$), and sheep lambed in January had lower percentage of milk fat ($7.53 \pm 0.04\%$). The amount of milk fat produced is 0.037 kg daily in sheep lambed in January or 0.035 kg in sheep lambed in February.

Table 7. The impact of month of lambing on daily milk production in F₁ crossbreeds, LS-mean \pm SE

Month of lambing	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
1	924	0.236 ± 0.01	0.260 ± 0.01	0.496 ± 0.01	7.53 ± 0.04	0.037 ± 0.001
2	221	0.218 ± 0.01	0.238 ± 0.01	0.456 ± 0.02	7.59 ± 0.07	0.035 ± 0.001

In *Kastelic et al. (2013)* and *Pollot and Gootwine (2001)* analyses is determined impact of month or season of lambing on milk production. *Sevi et al. (2004)*, observed similar results during studying of effects of lambing season on ewe's milk composition in the region of Southern Italy. Also similar results were observed by *Carta et al. (1995)* who studied the effects of seasonal changes on Sarda dairy sheep whose diet is also based on pasture.

According to *Maria and Gabina (1993)* effect of lambing season is one of the most significant effects on quantity and composition of produced milk.

In *Pacinovski et al. (2014)* analyses is determined similar impact of lambing month factor on daily milk production.

Impact of month of milk recording on daily milk production in tested sheep population

Month of milk recording showed highly significant impact ($P < 0.001$), compared to almost all traits in crossbreeds tested. Only this factor had no impact on milk fat percentage ($P > 0.05$), (Tab. 4).

Analysing the period from second (February) to eighth (August) month of milk control, the amount of total daily milk was constantly growing from second to sixth month, when reached maximal daily lactation (0.518 ± 0.021), and decreased in

next two months (July and August), (Table 8). Analogous to total amount of milk, same happened with morning and evening milk.

Unlike this, the percentage of milk fat was constantly growing from February to April and had identical value in June and July ($7.51 \pm 0.06\%$), to reach the highest value in August ($7.59 \pm 0.09\%$). Concerning the produced amount of milk fat per head, it ranges from ($0.030 \pm 0.02\text{kg}$) in February to $0.039 \pm 0.001\text{kg}$ in June (Table 8).

Table 8. The impact of the month of milk recording on daily milk production in F1 crossbreeds
LS-mean \pm SE

Month of test	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
2	103	0.193 ± 0.02	0.202 ± 0.02	0.395 ± 0.03	7.67 ± 0.14	0.030 ± 0.002
3	152	0.217 ± 0.01	0.227 ± 0.01	0.444 ± 0.03	7.58 ± 0.11	0.034 ± 0.002
4	178	0.222 ± 0.01	0.252 ± 0.01	0.474 ± 0.02	7.56 ± 0.09	0.036 ± 0.001
5	178	0.238 ± 0.01	0.263 ± 0.01	0.501 ± 0.02	7.51 ± 0.07	0.038 ± 0.001
6	178	0.247 ± 0.01	0.271 ± 0.01	0.518 ± 0.02	7.51 ± 0.06	0.039 ± 0.001
7	178	0.243 ± 0.01	0.266 ± 0.01	0.509 ± 0.01	7.51 ± 0.07	0.038 ± 0.001
8	178	0.232 ± 0.01	0.264 ± 0.01	0.496 ± 0.02	7.59 ± 0.09	0.038 ± 0.001

According to *Matutinovic et al. (2011)* the content of milk fat was lowest in April and May during the early stages of vegetation growth, while pasture is rich in easily soluble nitrogen components, but poor in fibre and energy (*Kalit, 2008; Mikolayunas et al., 2008*). *Pollot and Gootwine (2001)* determined significant influence of month of milk control on milk production.

Impact of the number of milk recordings on daily milk production in tested sheep population

The number of milk recordings had highly significant impact ($P < 0.001$), on all analysed parameters in crossbreeds between domestic sheep population and Awassi (Tab. 4).

According this factor, the total milk production was constantly decreasing from the first to the last (seventh) milk control. Same happens with morning and evening milk.

Analogous on daily milk production, the percentage of milk fat is lowest in first milk control ($6.87 \pm 0.09\%$) and highest in last (seventh) milk control ($8.23 \pm 0.14\%$). The produced amount of milk fat ranges from $0.021 \pm 0.002\text{ kg}$ in seventh, to $0.054 \pm 0.001\text{ kg}$ in first milk control (Tab. 9).

In similar analyses, *Pacinovski et al. (2014)*, found highly significant impact on previous factors (month and number of milk control) in relation to all traits of daily milk production.

Table 9. The impact of the number of milk recordings on daily milk production in F1 crossbreeds, LS-mean \pm SE

Number of milk control	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
1	178	0.387 \pm 0.01	0.397 \pm 0.01	0.784 \pm 0.02	6.87 \pm 0.09	0.054 \pm 0.001
2	178	0.309 \pm 0.01	0.344 \pm 0.01	0.653 \pm 0.02	7.20 \pm 0.07	0.047 \pm 0.001
3	178	0.253 \pm 0.01	0.273 \pm 0.01	0.526 \pm 0.02	7.36 \pm 0.06	0.039 \pm 0.001
4	178	0.208 \pm 0.01	0.232 \pm 0.01	0.440 \pm 0.02	7.56 \pm 0.07	0.033 \pm 0.001
5	178	0.167 \pm 0.01	0.200 \pm 0.01	0.367 \pm 0.02	7.73 \pm 0.09	0.028 \pm 0.001
6	152	0.142 \pm 0.01	0.167 \pm 0.01	0.309 \pm 0.03	7.97 \pm 0.11	0.025 \pm 0.002
7	103	0.126 \pm 0.02	0.130 \pm 0.02	0.256 \pm 0.03	8.23 \pm 0.14	0.021 \pm 0.002

Impact of fertility on daily milk production in tested sheep population

Fertility or the number of lambs per sheep, had no impact on any of the tested traits of daily milk production, with the exception of total daily milk with impact value of $P < 0,01$ (Tab. 4).

Those who gave birth to one lamb from these sheep had higher daily lactation (0.503 \pm 0.011), unlike those who gave birth to two lambs (0.451 \pm 0.021), (Tab. 10).

Same happens with morning and evening milk. The percentage of milk fat is in opposite situation, which is quite logical that there is higher value in those who gave birth to two lambs (7.69 \pm 0.08), while lower value in those with one lamb (7.43 \pm 0.031). The produced amount of milk fat ranges from 0.035 \pm 0.001kg in crossbreeds with two lambs, to 0.037 \pm 0.001 in crossbreeds with one lamb, (Tab. 10).

Table 10. The impact of the fertility on daily milk production in F1 crossbreeds, LS-mean \pm SE

Number of lambs	N	Morning (litres)	Evening (litres)	Total (litres)	Fat (%)	Fat (kg)
1	1083	0.239 \pm 0.01	0.264 \pm 0.01	0.503 \pm 0.01	7.43 \pm 0.03	0.037 \pm 0.001
2	62	0.216 \pm 0.01	0.235 \pm 0.01	0.451 \pm 0.02	7.69 \pm 0.08	0.035 \pm 0.001

Pollot and Gootwine (2001) found significant impact of fertility or litter size on total milk production

Conclusion

Awassi breed represents a suitable breed for improving lactation with cross-breeding of indigenous population of sheep in Macedonia. Keeping in mind the percentage of blood in these crossbreeds (50% blood – Awassi and 50% blood – indigenous breed of sheep) the question whether to increase the percentage of blood in Awassi with cross-breeding of crossbreeds with purebred Awassi ewes or selection in F1 population is often challenging.

Our suggestion is that further keeping shall be in direction of selection in F1 population in order to avoid an increase in percentage of fat in lamb tails, a trait which is a big problem in selling traditional Macedonian lamb, exported more than 50 years in several European countries where the same trait is considered as disadvantage by consumers.

Out of analysed factors, lambing month is quite important for longer lactation and bigger amount of annually obtained milk, and the number of milk recordings which represents the course of lactation curve, and the basis on which further selection of sheep shall be based.

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Uticaj faktora na proizvodna svojstva kod meleza Awassi ovce i ovčepoljske pramenke u Makedoniji

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Rezime

Cilj ovih istraživanja, bio je da se utvrdi veza između nekih faktora koji imaju uticaj na proizvodna svojstva kod meleza između rase Awassi i autohtone ovčepoljske ovce u Makedoniji. Uključili smo ukupno 1145 individualnih laktacijskih kontrola, u toku dve proizvodne godine (2012 i 2013).

Uticaj posebnih faktora ispitivan je pomoću F-testa, dok su analize dobijenih podataka urađene pomoću programskog paketa SPSS. Veći broj faktora (laktacija, mesec jagnjenja i broj kontrole mlečnosti) imali su visoko značajan uticaj ($P < 0.001$) na dnevnu proizvodnju mleka (jutarnje, večernje i ukupno mleko, % mlečne masti i kg mlečne masti). Jedino plodnost nije imala nikakav uticaj na ispitivane faktore, osim na ukupnu dnevnu produkciju mleka, gde je manifestovan visoko značajan uticaj ($P < 0.01$).

Prosečna laktacijska mlečnost kod ispitivanih meleza, bila je 109 ± 0.479 l za dve proizvodne godine, dok je proizvodnja mznog mleka u istom periodu bila $72 \pm 0,421$ l. Dužina laktacijskog perioda u toku dve ispitivane godine u proseku je bila $203 \pm 0,61$ dana.

U odnosu na uzrast, kod meleza u drugoj laktaciji je utvrđena nešto veća dnevna mlečnost (0.478 ± 0.01 l.) u poređenju sa melezima u prvoj laktaciji (0.475 ± 0.01 l.). Ovo je sasvim logično imajući u vidu laktacijske krive kod ovce, posebno kod mlečne rase, gde pripada i Awassi rasa.

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COMPARATIVE STUDY ON CARCASS CHARACTERISTICS IN LAMBS FROM THE BULGARIAN DAIRY SYNTHETIC POPULATION AND ITS F₁ CROSSES WITH ILE DE FRANCE AND MUTTON CHAROLAIS

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Abstract: The objective of this study was to determine the effects of the father's breed onto the carcass characteristics in lambs from the Bulgarian Dairy Synthetic Population and its F₁ crosses with Ile de France and Mutton Charolais breeds. The scientific study took place at the Sheep Farm with the Agricultural Institute of Stara Zagora, Bulgaria. Object of the study were lambs from the Bulgarian Dairy Synthetic Population and its crosses with Ile de France and Mutton Charolais breeds. After reaching a live weight of 21-24 kg, 9 male lambs of different crosses were slaughtered. Slaughter took place at a licensed slaughter house. The lamb carcasses were chilled for 24 hours. Following this, linear measurements were made on the left carcass halves. The father's breed has a significant proven effect onto the large and small circumference of the leg ($P \leq 0.001$) and a proven effect onto the carcass length ($P \leq 0.01$). The Ile de France crosses feature 17.44% bigger leg circumference in comparison with the animals from the reference group. Similar is the situation with the Mutton Charolais crosses where we have 14.93% bigger leg circumference compared to that in the reference group. Determined is also an effect onto the weight of the chilled carcass and the carcass yield. The lambs from the trial groups have a proven bigger leg weight ($P \leq 0.001$), proven bigger chilled carcass weight ($P \leq 0.01$) and they show a trend for proving the effect on the carcass yield ($P \leq 0.05$).

Key words: crosses, carcass, linear measurements, cuts of meat

Introduction

In order to increase the production and improve meat quality, industrial crossing with various meat sheep breeds is most often used. In Bulgaria, Ile de France and

Mutton Charolais are the two breeds most commonly used for crossing. Their meat is more tender, more juicy, without strong, haunting smell, it has high protein content and a lower content of the fats (Nedelchev, 2005; Dimitrov et al., 2009; Jandasek et al., 2014; Slavov et al., 2015). This is of key importance for the consumers since according to doctors and nutritioners, healthy meat should be lean (Simopoulos, 2002; Janiszewski et al., 2016).

Both Ile de France and Mutton Charolais cross lambs reach their target weight for a shorter period of time (Cividini et al., 2005; Paim et al., 2013). Their faster growth together with their better muscle gain result in better carcass quality (Banks and Ross, 2003; Pannier et al., 2014), which means better efficiency on the farm and better lean meat yield (Pethick et al., 2006). Using the Mutton Charolais and Ile de France breeds for industrial crossing significantly increases meat productivity. This becomes possible thanks to the better muscle gain in those parts of the carcass producing the primal meat cuts (Slavov et al., 2005; Laleva et al., 2007; Pascal et al., 2009; Partida et al., 2012). With regard to the leg circumference and eye muscle area, the Mutton Charolais crosses are performing better than the Ile de France crosses, but worse in terms of percentage of the carcass deboned meat and carcass yield (Yankov, 2008).

The objective of this study was to determine the effect of the father's breed onto the carcass characteristics in lambs from the Bulgarian Dairy Synthetic Population and their F₁ crosses with Ile de France and Mutton Charolais.

Material and methods

The scientific study took place in 2015 at the Sheep Farm with the Agricultural Institute of Stara Zagora, Bulgaria. Object of the study were male lambs from the Bulgarian Dairy Synthetic Population and its crosses with Ile de France and Mutton Charolais breeds. The method of analogues was used to set up those three groups of animals. At the start of the study, the animal average live weight was respectively 14.86 kg for the first group, 14.11 kg for the second group and 13.80 kg for the third group. The lambs were raised in boxes until they reached a live weight of 21-24 kg. The boxes were equipped with feeders and drinkers. The animals from the three groups were fed with concentrate mixture (acc. Table 1) and alfalfa hay free. After the animals reached the target weight, 3 male lambs from each group were slaughtered. Slaughter took place in May 2015 in a Slaughterhouse near Stara Zagora. The lambs were transported to the Slaughterhouse in the early hours of the day using specialized transportation vehicle, company's property.

After slaughtering, carcasses were divided into 3 groups:

- first group (reference group)- 3 lamb carcasses from the Bulgarian Dairy Synthetic Population;

- second group- 3 lamb carcasses F₁ crosses of the Bulgarian Dairy Synthetic Population with Ile de France;
- third group- 3 lamb carcasses F₁ crosses of the Bulgarian Dairy Synthetic Population with the Mutton Charolais.

Table 1. Lamb fattening concentrate mixture-composition

Ingredients	Content, %
Corn	63,5
Barley	18,0
Sunflower groats	15,0
Calcium carbonate CaCO ₃	2,5
Salt	0,5
Premix (lambs)	0,5

The carcasses were chilled for 24 hours and then the linear dimensions of each carcass left half were measured. We used a linear measurement device and followed the methodics of *Zahariev and Pinkas (1979)*.

Carcass length (cm): determined was the distance from the front end of the pelvic symphysis to the middle of the first rib front side (Fig. 1, line A-B). *Chest width* (cm): measured at the level of the 5th thoracic vertebrae. Determined was the distance from the 5th thoracic vertebrae to the caudal end of the sternum (breastbone) from the ventral side (Fig. 1, line C-D).

Leg length (cm) measured was the distance from the carpal joint to the front end of the pelvic symphysis (Fig. 1, line A-E).

Leg circumference (cm): two measurements were made. The first measurement (large circumference) was made at the widest part of the leg (Fig. 1, line F-G). The second measurement (small circumference) (Fig. 1, line H-I) was made at the middle of the line determining the leg length (A-E). The carcasses were carved in accordance with *BDS 4348-78* into the following parts: lamb neck, lamb shoulder, cutlet, leg and loin (Fig. 2). Each cut of meat was then weighed using electronic scales.

Lamb neck (Fig. 2a): these are all cervical vertebrae (without the first cervical vertebra) and the adjoining muscle mass.

Lamb shoulder (Fig. 2b): this is the muscle mass in the front leg with the shin, the sides half of the thoracic vertebrae with the adjoining ribs and the sides half of the breastbone.

Cutlet (Fig. 2c): The front boundary of the cutlet coincides with the rear boundary of the shoulder. The cut outlining the rear boundary of the cutlet goes between the last and the last but one lumbar vertebrae. The cutlet contains the side halves of the lumbar vertebrae.

Leg (Fig. 2d) the front boundary of the leg coincides with the rear boundary of the cutlet. The rear boundary goes across the carpal joint.

Loin (Fig. 2e): containing the soft abdominal wall.

After weighing, the meat cuts were deboned in order to find out the weight of the meat and bones.

The results from our trial were processed by MY STAT and STATISTIKA AX.

Results and Discussion

The carcass linear measurement results are presented in Table 2. Data reveals that the lambs from the Bulgarian Dairy Synthetic Population stand out with the greatest carcass length (55.83 cm) in comparison to the animals from the other groups. Also, comparing the lambs from the second and the third group in terms of carcass length, there is a slight superiority of the Mutton Charolais crosses over Ile de France crosses (carcass length respectively 52.83 cm and 52.00 cm). Expressed as a percentage, the difference is 1.57%. Our results with regard to carcass length differ from the results obtained by *Yankov (2008)*. The author has determined that the Ile de France crosses feature longer carcass in comparison with Mutton Charolais crosses. In terms of the leg length, the lambs from the first group show the greatest leg length (35.00 cm), followed by the Ile de France crosses (34.17 cm) and the Mutton Charolais crosses (33.83 cm). Similar are the results presented by *Yankov (2009)*.

By measuring the leg circumference, it is possible to determine the muscle gain and the muscle development in lamb carcasses. The linear measurements based on this trait (leg circumference-large and small), showed that the lambs from both the second and the third group perform better than the animals from the Bulgarian Dairy Synthetic Population. The results from the leg small circumference measurement of the Ile de France crosses are 17.44% higher compared to the animals from the reference group. When measuring the leg large circumference, it can be seen that the Mutton Charolais crosses do better (38.50 cm) in comparison with the lambs from the Bulgarian Dairy Synthetic Population (34.50 cm). Expressed as a percentage, the difference is 10.39%. Similar results are presented by *Nedelchev (2005)*; *Slavov et al. (2005)*; *Laleva et al. (2007)*.

Table 2. Linear Carcass Measurements for the three groups

Carcass characteristics	Animal Groups											
	1 st group- Bulgarian Dairy Synthetic Population				2 nd group- Bulgarian Dairy Synthetic Population x Ile de France				3 rd group- Bulgarian Dairy Synthetic Population x Mutton Charolais			
	n	\bar{x}	$S\bar{x}$	V_c	n	\bar{x}	$S\bar{x}$	V_c	n	\bar{x}	$S\bar{x}$	V_c
Carcass length, cm	3	55.83	1.44	3.00	3	52.00	0.87	2.00	3	52.83	0.58	1.00
Leg length, cm	3	35.00	0.87	3.00	3	34.17	0.58	2.00	3	33.83	0.58	2.00
Leg circumference, cm	3	26.03	0.46	2.00	3	31.53	0.81	3.00	3	30.60	0.00	0.00
small circumference, cm												
large circumference, cm	3	34.50	0.00	0.00	3	38.00	0.87	2.00	3	38.50	0.00	0.00
Chest width, cm	3	15.87	1.44	9.00	3	15.90	0.69	4.00	3	15.90	0.69	4.00

The father's breed (Table 3) has a high proven effect onto the small and large leg circumference ($P \leq 0.001$) and a proven effect onto the carcass length ($P \leq 0.01$).

Table 3. Variance components of the characteristics studied according to the group

Characteristics	F
Carcass length, cm	11.553**
Leg length, cm	ns
Leg small circumference, cm	89.958***
Leg large circumference, cm	57.000***
Chest width, cm	ns

ns- not significant

**-. $P \leq 0.01$

***-. $P \leq 0.001$

Carcass characteristics for all three groups of lambs are presented in Table 4. It is seen that the Mutton Charolais crosses come first in terms of weight of the chilled carcass (11.03 kg), while the lambs from the first group come last (9.50 kg). There is only a slight difference between the two trial groups in terms of carcass weight. Expressed as a percentage, this difference is 2.36% and it is in favour of the Mutton Charolais crosses. *Laleva et al. (2007)* have found out that the F₁ crosses with Mutton Charolais stand out with bigger weight of the chilled carcass in comparison with the source breed (Trakian merino lambs). In his study *Yankov (2008;2009)* has found out that the Ile de France crosses have bigger weight of the chilled carcass in comparison with the Mutton Charolais crosses. The carcass yield

for both trial groups is significantly higher in comparison with that of the reference group. The Ile de France crosses have up to 8.42%, and Mutton Charolais - 5,86% better carcass yield in comparison with the lambs from the Bulgarian Dairy Synthetic Population. The better carcass yield in crosses with the meat sheep breeds in comparison with the source breeds is also confirmed by a number of other authors (*Slavov et al., 2005; Laleva et al., 2007; Anev, 2009*). Comparing the animals from the trial groups we have found out that the Ile de France crosses have higher carcass yield than the Mutton Charolais crosses. With regard to the weight of the various meat cuts, it is seen that the two trial groups feature better performance than the reference group. The weight of the neck cut in Mutton Charolais crosses is 26.14% higher compared to that of the Bulgarian Dairy Synthetic Population. In Ile de France crosses the weight of the neck cut is 16.67% higher than the weight of the neck cut in the Bulgarian Dairy Synthetic Population. In terms of meat cuts, the leg in the Mutton Charolais crosses weighs the heaviest (1.99 kg), Ile de France crosses coming second (1.97 kg), followed by the Bulgarian Dairy Synthetic Population (1.57 kg). In their study *Cividini et al. (2005)* and *Pajor et al. (2009)* have found out that the Mutton Charolais and Ile de France crosses feature heavier leg, shoulder and cutlet compared with the source breed. With regard to the weight of the different meat cuts, there are no significant differences between the two trial groups.

Table 4. Carcass characteristics of the lambs from the three groups

Carcass Characteristics	Animal Groups											
	1 st group- Bulgarian Dairy Synthetic Population				2 nd group- Bulgarian Dairy Synthetic population x Ile de France				3 rd group- Bulgarian Dairy Synthetic population xMutton Charolais			
	n	\bar{x}	$S\bar{x}$	Vc	n	\bar{x}	$S\bar{x}$	Vc	n	\bar{x}	$S\bar{x}$	Vc
Weight of the chilled carcass, kg	3	9.50	0.00	0.00	3	10.77	0.40	4.00	3	11.03	0.65	6.00
Carcass yield, %	3	43.85	0.58	1.00	3	47.88	1.79	4.00	3	46.58	1.91	4.00
Lamb neck, kg	3	0.65	0.08	12.00	3	0.78	0.09	12.00	3	0.88	0.12	14.00
Lamb shoulder, kg	3	2.11	0.05	2.00	3	2.28	0.28	12.00	3	2.33	0.18	8.00
Cutlet, kg	3	0.62	0.11	18.00	3	0.62	0.06	10.00	3	0.67	0.03	4.00
Leg, kg	3	1.57	0.07	5.00	3	1.97	0.09	5.00	3	1.99	0.10	5.00

The cross type has a high proven effect onto the weight of the leg cut (Table 5) ($P \leq 0.001$), proven effect onto the weight of the chilled carcass ($P \leq 0.01$) and there is a trend for proving the effect onto the carcass yield ($P \leq 0.05$).

Table 5. Variance components of the characteristics studied according to the group

Characteristics	F
Weight of the chilled carcass, kg	10.295**
Carcass yield, %	5.382*
Neck, kg	ns
Shoulder, kg	ns
Cutlet, kg	ns
Leg, kg	21,277***

ns- not significant

*- $P \leq 0.05$

**- $P \leq 0.01$

***- $P \leq 0.001$

After carving and deboning of the left half of the carcasses, we found out that with regard to the meat yield, the Mutton Charolais crosses perform better (3.98 kg) while the lambs from the Bulgarian Dairy Synthetic Population have the smallest meat yield (3.21 kg) (fig. 3). The difference in the meat yield between the Ile de France and the Mutton Charolais crosses is 3.44%. With regard to the weight of bones, after deboning the carcass halves, the animals from the first and the second group show nearly the same values. The weight of the bones in the animals from the reference group is 1.32 kg, while in the Ile de France crosses, it is 1.32 kg. More significant is the difference in the weight of the bones between the Ile de France and the Mutton Charolais crosses. Expresses as a percentage, it is 10.70% and it is in favour of the Ile de France crosses. The differences in the weight of the bones from the left half of the carcass determined by us is supported by *Yankov (2009)*. The author has found out that the weight of the bones in the Mutton Charolais crosses is higher.

Conclusion

The father's breed has a high proven effect onto the leg's weight ($P \leq 0.001$), proven effect onto the weight of the chilled carcass ($P \leq 0.01$) and a trend for proving the effect onto the carcass yield ($P \leq 0.05$). The Mutton Charolais crosses feature the biggest weight of the chilled carcass (11.03 kg), the Ile de France crosses coming second (10.77 kg), followed by the lambs from the reference group (9.50 kg).

Ile de France and Mutton Charolais breeds have a high proven effect onto the linear measurements of the leg circumference (both large and small circumference) ($P \leq$

0.001) and a proven effect onto the carcass length ($P \leq 0.01$). With regard to the small leg circumference, the Ile de France crosses show 17.44%, and the Mutton Charolais crosses 14.93% higher values than those of the Bulgarian Dairy Synthetic Population.

After deboning the carcass halves, we have found out that the Mutton Charolais crosses produced the higher amount of deboned meat (3.98 kg), followed by the Ile de France crosses (3.84 kg), and the Bulgarian Dairy Synthetic Population (3.21 kg).

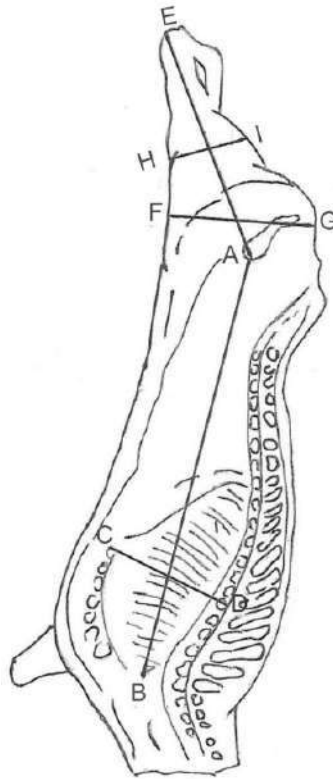


Fig. 1. Linear carcass measurements (*Zahariev and Pinkas, 1979*)

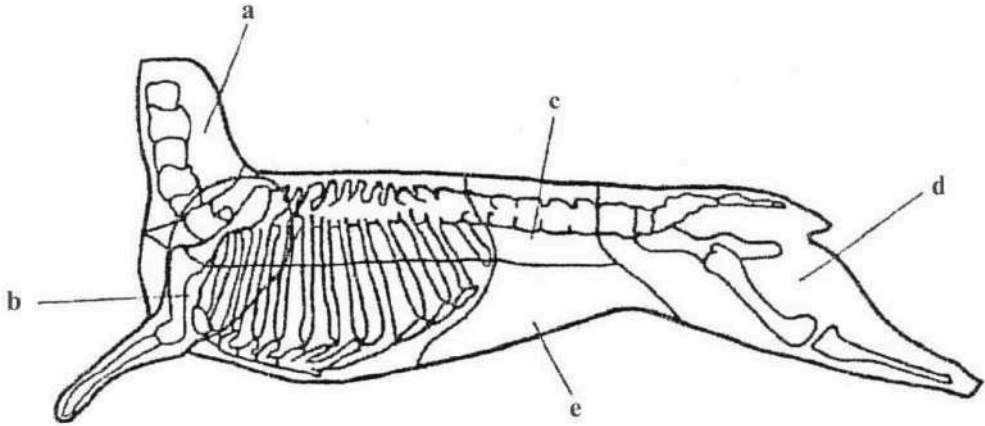


Fig. 2. Lamb cuts chart (Marinova and Popova, 2011)

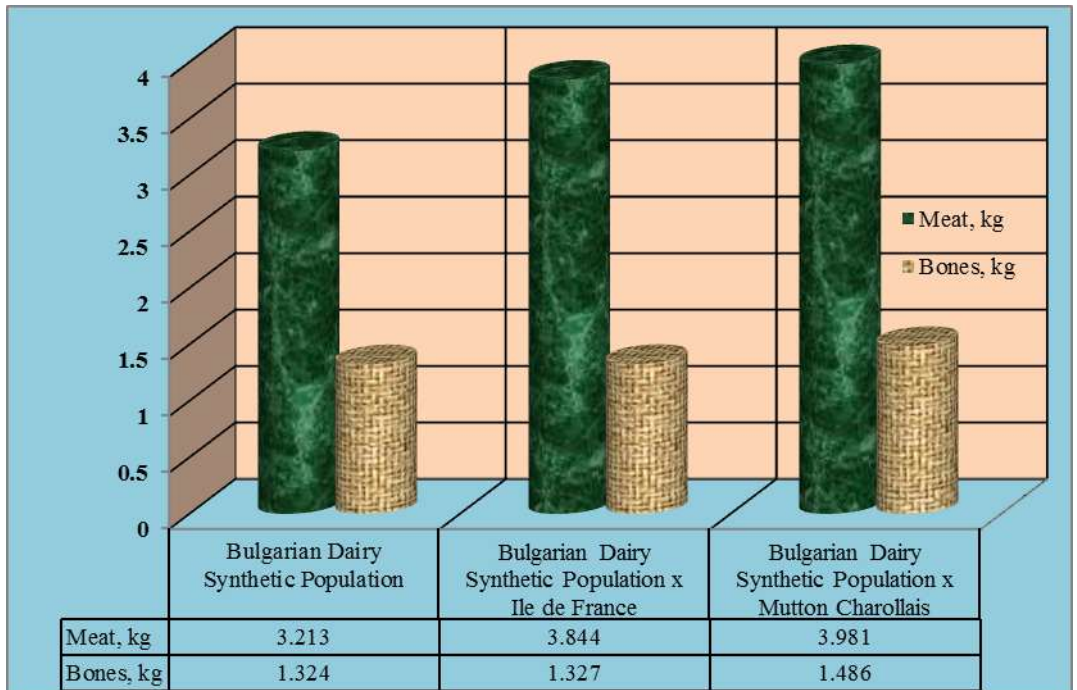


Fig. 3. Weight of the meat and bones after deboning carcass left halves for the lambs from all three groups

Uporedna analiza osobina trupa jagnjadi bugarske mlečne sintetičke populacije i F₁ meleza sa rasom Ile de France i Charollais

Nikolay Ivanov, Staika Laleva, Stefan Ribarski, Teodora Angelova

Rezime

Cilj ovog istraživanja je bio da se utvrdi uticaj rase oca na osobine trupa jagnjadi bugarske mlečne sintetičke populacije i njenih F₁ meleza sa Ile de France i Charollais rasama. Istraživanje je sprovedeno na farmi ovaca Poljoprivrednog instituta u Staroj Zagori u Bugarskoj. Predmet istraživanja su bila jagnjad bugarske mlečne sintetičke populacije i njeni melezi sa Ile de France i Charollais rasama. Nakon dostizanja težine grla od 21-24 kg, 9 muških jagnjadi različitih kombinacija ukrštanja su zaklani. Klanje je sprovedeno u licenciranoj klanici. Trupovi jagnjadi su hladeni 24 sata. Nakon toga, linearna merenja su izvršena na levim polutkama. Rasa oca ima dokazano značajan uticaj na obim noge ($P \leq 0,001$) i dokazani uticaj na dužinu trupa ($P \leq 0,01$). Ile de France melezi imaju 17,44% veći obim nogu u poređenju sa životinjama iz referentne grupe. Slična je situacija sa Charollais melezima gde imamo 14,93% veći obim nogu u poredjenju sa referentnom grupom.

Utvrđen je takođe efekat na težinu ohlađenog trupa i prinos trupa. Jagnjad iz oglednih grupa su imali dokazano veću težinu nogu ($P \leq 0,001$), dokazano veću težinu ohlađenih trupova ($P \leq 0,01$) i pokazuju trend kojim se dokazuje i efekat na prinos trupa ($P \leq 0,05$).

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PHENOTYPIC CORRELATION OF HENS BODY WEIGHT AND REPRODUCTIVE TRAITS OF BROILER PARENTS

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Abstract: Examinations were conducted in two flocks of broiler parents' hybrids Ross 308 and Cobb 500. At the beginning of the production cycle (24th weeks of age) was determined that the average hens body weight of hybrid Ross 308 was 2680.40 g, and to hybrid Cobb 500 was 2697.80 g. At 42nd weeks of age (middle of the production cycle) hens body weight was 3565.10 g (Ross 308) and 3599.05 g (Cobb 500), while at the end of the production cycle (61st weeks of age) hens body weight of hybrid Ross 308 was 3841.50 g, and to the Cobb 500 was 3850.00 g. Determined differences of hens body weight (17.40 g, 33.95 g and 8.50 g) in specific periods of the production cycle, and the difference in hens body weight for the entire cycle (23.26 g) weren't statistically significant ($P > 0.05$). Specific consideration of the impact of hens' body weight on reproductive performances of broiler parents was determined by calculating the coefficients of phenotypic correlation among the tested indicators. Thus, between hens body weight and most reproductive indicators of broiler parents were determined statistically very significant ($P < 0.001$) coefficients of phenotypic correlation, while between hens body weight and the percentages of chickens feasibility from fertilized eggs were determined significant ($P < 0.001$; $P < 0.01$; $P < 0.05$) correlation coefficients for a slightly shorter period than anticipated production cycle.

Key words: hens, body weight, reproductive traits, broiler parents, correlation.

Introduction

On reproductive performances of broiler parents, besides age and optimal gender ratio, significant impact has a body weight of hens during the production cycle (*Savić et al., 2004; Ciacciariallo and Gous, 2005; Djermanović et al., 2005; Vieira et al., 2005; Almeida et al., 2006; Djermanovic et al., 2008; Djermanovic et*

al., 2009; Djermanovic, 2010; Đermanović et al., 2010; Mitrovic et al., 2010; Mitrovic et al., 2011; Đermanović et al., 2012). Proper hormonal functioning of the endocrine system of hens except age and photostimulation (Lewis et al., 2005; Lewis and Gous, 2006; 2007; Usturoi et al., 2007) to a large extent depends on body's growth of breeding animal. In particular age with the optimal body weight ovary functioning is stimulated, and thus accelerates the maturation of oocytes, i.e. egg production. It is necessary to ensure to the fertilized ovum proper conditions for embryo development to bring offspring from fertilized egg.

Only proper feeding and the technology of exploitation parental flock can provide prerequisite for the maximum percentage of feasibility and necessary vitality and quality of eggs for incubation hatched offspring (Barnett et al., 2004; Maiorka et al., 2004; Enting et al., 2007; Wolanski et al., 2007). To be production period of fertilized eggs, i.e. day-old chickens, longer it is necessary to permanently keep hens in breeding shape, with special care to their development. Also, should have in mind that flock uniformity in terms of body weight is especially important factor in the second half of production cycle. Among other factors, body weight of breeding hens directly influences on reproductive performances. It is similar to the production of eggs for incubation, i.e. broiler chickens of different genotype. In most line hybrids eggs production for incubation begins in 24th weeks of age when the intensity of capacity is about 5% and higher. Since that period eggs production, fertility and chickens feasibility gradually increases to the maximum, and after that productivity of broiler parents reduces to a lesser or greater extent. Therefore it can be said that the period of exploitation of broiler parents depends to a very large extent by this time interval. As an indicator to give estimates to what period is reasonable to use the broiler parents in production of hatching eggs and day-old chickens, a significant contribution can provide calculated coefficients of phenotypic correlation between hens body weight and reproductive traits in the final period of the production cycle, that represents a turning phase in the in the utilization of parent flocks.

Material and Methods

Examinations were conducted in two parent flocks of heavy hen hybrids Ross 308 and Cobb 500. During the production cycle, was used the technology proposed by the breeders of the mentioned hybrids. Both broiler parent flocks were kept on the floor of the deep litter, and feeding, watering, ventilation and lighting were automatically regulated. Studied flocks were grown to the 61st week of age, i.e. both flocks have laid eggs in the beginning of the 22nd week, and for incubation were used eggs that have been laid from 24th week of age to the end of the production cycle, because it satisfied the minimum weight suitable for incubation (>50.00 g). From the results it follows that the period of egg production, i.e. production of day-old broiler chickens has lasted 38 weeks.

As the initial sample material were used 5200 birds of both genders of hybrids Ross 308 and 5430 broiler breeders of hybrids Cobb 500. Hybrids were kept in two separate objects. In the first object were stationed 4750 ♀ and 450 ♂ Ross 308 hybrids, and in the second 4960 ♀ and 470 ♂ Cobb 500 hybrids, so the gender ratio was 1 : 10.56 (Ross 308) and 1 : 10.55 (Cobb 500). In the preparation period from the 21st till the 24th week of the flock age mortality and cast aside were 13 birds (0.297%) to hybrid Ross 308, and to hybrid Cobb 500 12 birds (0.24%). That means that at the beginning of utilization of eggs for incubation to broiler parents of hybrid Ross 308 in the flock was 4737 hens, i.e. 4948 hens to hybrid Cobb 500. In order to control body weight each week individually was measured per 200 hens of hybrid Ross 308 and Cobb 500, selected randomly.

By this measuring was followed uniformity of hens tested flocks during the production cycle, furthermore was tested the influence of hens body weight on the reproductive performances of broiler parents: eggs weight, chickens feasibility from fertilized eggs, day-old chicken weight, relative share of chicken in egg weight, absolute weight loss of eggs and relative weight loss of eggs. Basic data processing was performed using variation statistical methods, and testing the differences between hybrids by using t-test. Nevertheless, the obtained results were used to calculate the correlation of tested traits by weeks of the production in the last third of the production cycle, i.e. from the 50th to the 61st weeks of age, by using correlation analysis. Statistical data processing was performed by using the program SAS/STAT (*SAS Institute, 2000*).

Results and Discussion

Average values, variability, importance of the difference in body weight of hens at certain times of the production cycle and for the whole period of egg production, are shown in the Table 1.

Table 1. Average values, variability, importance of the difference in body weight of hens at certain times of the production cycle

Production cycle period	Weeks of age (production)	Hybrid	$\bar{x} \pm \text{SEM}$	S	\bar{d}
Beginning	24 (1)	Ross 308	2680.40±14.63	206.93	17.40 ^{ns}
		Cobb 500	2697.80±17.09	241.66	
Middle	42 (19)	Ross 308	3565.10±19.86	280.92	33.95 ^{ns}
		Cobb 500	3599.05±20.12	275.28	
End	61 (38)	Ross 308	3841.50±21.39	302.56	8.50 ^{ns}
		Cobb 500	3850.00±21.68	306.59	
Entire production cycle	61 (38)	Ross 308	3411.15±61.58	394.33	23.26 ^{ns}
		Cobb 500	3434.41±61.03	390.76	

^{ns} P>0.05.

Table 1 data show that the average body weight of hens of both hybrids is increased gradually during the production cycle. Body weight of hens in the 24th week was 2680.40 g (Ross 308) and 2967.80 g (Cobb 500), and at the end of utilizing 3841.50 g to hybrid Ross 308 and 3850.00g to hybrid Cobb 500. During the production cycle Cobb 500 hybrid hens compared to hens Ross 308 had a higher average body weight, which was not statistically confirmed ($P>0.05$). Average body weight of Ross 308 hybrid hens for the whole exploitation period was 3411.15 g, and to hybrid Cobb 500 3434.41 g, the difference in body weight (23.26 g) between the tested hybrids hens was not statistically significant ($P>0.05$), indicating that genotype had no significant effect on body weight of hens.

The body weight was slightly higher than the genetic potential of the tested hybrids. To the similar results, in terms of average body weight of hens had come *Djermanovic et al. (2009)*, *Djermanovic (2010)* and *Mitrovic et al. (2010)*. *Usturoi et al. (2007)* during the breeding of the Ross 308 broiler parents found slightly lower average body weight of hens, depending on which groups of hens, in the 60th week of age was between 3988.95 g and 3990.44 g *Lewis et al. (2005)* and *Lewis and Gous (2006)* are in the 60th week of age Cobb 500 hybrid hens determined slightly higher average body weight of hens, between 4.21 and 4.25 kg, while *Lewis and Gous (2007)* in the 59th week of age determined slightly higher average body weight of hybrid Ross 308 (4.43 kg) and Cobb 500 (4.56 kg).

Beside established variation measures of hens body weight of the analyzed parent flocks, in order to gain better insight of the impact of body weight on reproductive performances of hens, coefficient of phenotypic correlation among tested traits in the last third of the production cycle was calculated (Table 2).

Table 2. The phenotypic correlation of hen's body weight and reproductive traits

Age (weeks)	Hybrid	BW, g	Coefficients of phenotypic correlation					
			r ₁	r ₂	r ₃	r ₄	r ₅	r ₆
50	Ross 308	3685.50	0.989***	0.677***	0.961***	0.763***	0.650***	-0.763***
	Cobb 500	3710.00	0.989***	0.657***	0.961***	0.765***	0.719***	-0.765***
51	Ross 308	3703.50	0.992***	0.620***	0.966***	0.781***	0.581***	-0.781***
	Cobb 500	3722.00	0.994***	0.585***	0.969***	0.792***	0.654***	-0.792***
52	Ross 308	3710.50	0.991***	0.556***	0.966***	0.797***	0.485***	-0.797***
	Cobb 500	3732.50	0.994***	0.525***	0.970***	0.809***	0.574***	-0.809***
53	Ross 308	3743.00	0.989***	0.495***	0.964***	0.812***	0.369***	-0.812***
	Cobb 500	3755.00	0.993***	0.466***	0.969***	0.815***	0.490***	-0.815***
54	Ross 308	3754.00	0.989***	0.440**	0.959***	0.809***	0.264 ^{ns}	-0.809***
	Cobb 500	3767.50	0.992***	0.413*	0.967***	0.817***	0.398*	-0.817***
55	Ross 308	3770.00	0.987***	0.375*	0.957***	0.812***	0.174 ^{ns}	-0.812***
	Cobb 500	3777.50	0.991***	0.362*	0.963***	0.821***	0.293 ^{ns}	-0.821***
56	Ross 308	3782.50	0.986***	0.315 ^{ns}	0.951***	0.805***	0.095 ^{ns}	-0.805***
	Cobb 500	3792.50	0.991***	0.316 ^{ns}	0.957***	0.817***	0.172 ^{ns}	-0.817***
57	Ross 308	3797.00	0.988***	0.254 ^{ns}	0.944***	0.794***	-0.003 ^{ns}	-0.794***
	Cobb 500	3805.00	0.991***	0.261 ^{ns}	0.950***	0.812***	0.032 ^{ns}	-0.812***
58	Ross 308	3805.50	0.986***	0.183 ^{ns}	0.938***	0.786***	-0.067 ^{ns}	-0.786***
	Cobb 500	3812.50	0.989***	0.203 ^{ns}	0.943***	0.803***	-0.062 ^{ns}	-0.803***
59	Ross 308	3812.50	0.985***	0.091 ^{ns}	0.929***	0.774***	-0.129 ^{ns}	-0.774***
	Cobb 500	3825.00	0.987***	0.121 ^{ns}	0.934***	0.791***	-0.157 ^{ns}	-0.791***
60	Ross 308	3827.50	0.983***	-0.016 ^{ns}	0.919***	0.752***	-0.134 ^{ns}	-0.752***
	Cobb 500	3835.00	0.984***	0.015 ^{ns}	0.923***	0.781***	-0.252 ^{ns}	-0.781***
61	Ross 308	3841.50	0.986***	-0.126 ^{ns}	0.909***	0.724***	-0.135 ^{ns}	-0.724***
	Cobb 500	3850.00	0.981***	-0.097 ^{ns}	0.910***	0.756***	-0.253 ^{ns}	-0.756***

BW – Body weight (g). * P<0.05; ** P<0.01; *** P<0.001; ^{ns} P>0.05.

r₁-hens body weight (g) x eggs weight (g); r₂-hens body weight (g) x chickens feasibility from fertilized eggs (%); r₃-hens body weight (g) x day-old chickens weight (g); r₄-hens body weight (g) x relative share of chicken in egg weight (%); r₅-hens body weight (g) x absolute egg weight loss (g); r₆-hens body weight (g) x relative egg weight loss (%).

Between body weight and weight of hens eggs, i.e. chickens weight determined absolute correlation connection (P<0.001). However, between hens body weight and chickens feasibility from fertilized eggs, to the both hybrids, calculated coefficients of phenotypic correlation were statically significant (P<0.001; P<0.01; P<0.05) to the 55th week of age (Table 2). This was reached because of slightly higher body weight of Cobb 500 hybrid hens, compared to Ross 308, during the production cycle (Table 1). Despite of this, it can be said that in term of this indicator achieved results were solid to the both tested hybrids.

Based on the data presented in the Table 2, to the both parent flocks was determined increasing trend of relative share of chicken in egg weight with increasing hens' body weight, similar to egg weight, i.e. chickens weight. Determined coefficients of phenotypic correlation between these indicators, to the both tested genotypes, were statistically very significant (P<0.001). Analogue to

the values for the relative share of chicken in egg weight was determined, but negative, statistically very significant values ($P < 0.001$), between hens body weight and relative egg weight loss to the both hybrids. Beside of this, between hens body weight and absolute egg weight loss were determined positive statistically significant ($P < 0.001$; $P < 0.01$; $P < 0.05$) coefficients correlation connection till the 53rd weeks of age (Ross 308), i.e. till the 54th weeks of age (Cobb 500).

Since this periods till the end of the production cycle determined coefficients of correlation connection between analyzed parameters weren't statistically significant ($P > 0.05$), but to Ross 308 hybrids from the 57th weeks of age, and to Cobb 500 from 58th week, to the end of the production cycle were negative.

The most of the authors, in their researches, mainly have been dealt with impact of age on productive and reproductive indicators of broiler parents, and in less extent with impact on hens' body weight. However, to the similar but also to the opposite results in terms of correlation connection of reproductive indicators in the first place age of broiler parents have come *Savić et al. (2004)*, *Đermanović et al. (2005)*, *Djermanović et al. (2008)*, *Djermanović et al. (2009)*, *Mitrović et al. (2009)*, *Sahin et al. (2009)*, *Djermanović (2010)*, *Đermanović et al. (2010)*, *Mitrović et al. (2011)* and *Djermanović et al. (2012)*.

Conclusion

Based on the obtained results can be ascertained that average hens body weight to the both tested hybrids, in relation to technological normative, were lower in the beginning and at the end of the production cycle. However, the differences between hens body weight to the both tested hybrids weren't statistically significant ($P > 0.05$), i.e. genotype had no significant effect on body weight of hens.

Based on calculated coefficients of phenotypic correlation and its relevance, we can say that the hens body weight significantly affected their breeding ability because to the both parent flocks, between hens body weight and eggs weight, day-old chickens weight, relative share of chicken in egg weight and relative egg weight loss were determined statistically very significant ($P < 0.001$) coefficients of correlation. But, between hens body weight and chickens feasibility from fertilized eggs to the both tested genotypes statistically significant ($P < 0.001$; $P < 0.01$; $P < 0.05$) coefficients of correlation were determined for a slightly shorter period (55th week of age) than expected production cycle, leading to the fact that increase in body weight decreases the ability of hens breeding. This also indicates that increase in hens' body weight causes reducing of the production cycle than anticipated, i.e. existence of the turning phase in the last third of the production cycle.

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Fenotipska povezanost telesne težine nosilja i reproduktivnih osobina brojlerskih roditelja

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Rezime

Ispitivanja su sprovedena na dva jata brojlerskih roditelja hibrida Ross 308 i Cobb 500. Na početku proizvodnog ciklusa (24. nedelja starosti) kod hibrida Ross 308 utvrđena je prosečna telesna težina nosilja 2680,40 g, a hibrida Cobb 500 2697,80 g. U 42. nedelji starosti (sredina proizvodnog ciklusa) telesna težina nosilja iznosila je 3565,10 g (Ross 308) i 3599,05 g (Cobb 500), dok je na kraju proizvodnog ciklusa (61. nedelja starosti) telesna težina nosilja hibrida Ross 308 iznosila 3841,50 g, a Cobb 500 3850,00 g. Utvrđene razlike telesne težine nosilja (17,40 g, 33,95 g i 8,50 g) u određenim periodima proizvodnog ciklusa, kao i razlika u telesnoj težini nosilja za ceo proizvodni ciklus (23,26 g) nisu bile statistički signifikantne ($P > 0,05$). Konkretnije sagledavanje uticaja telesne težine nosilja na reproduktivne performanse brojlerskih roditelja utvrđeno je izračunavanjem koeficijenata fenotipske korelacije između ispitivanih pokazatelja. Tako su između telesne težine nosilja i većine reproduktivnih pokazatelja brojlerskih roditelja utvrđeni statistički vrlo signifikantni ($P < 0,001$) koeficijenti fenotipske korelacione povezanosti, dok su između telesne težine nosilja i procenta izvodljivosti pilića od oplodjenih jaja utvrđeni statistički signifikantni ($P < 0,001$; $P < 0,01$; $P < 0,05$) koeficijenti korelacije za nešto kraći period (55. nedelja starosti) od predviđenog proizvodnog ciklusa.

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EFFECT OF DIETARY PROTEASE SUPPLEMENTATION AND SEX ON DRESSING PERCENTAGE AND BODY CONFORMATION IN BROILERS

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Original scientific paper

Abstract: This paper presents research results on the effect of protease on the dressing percentage of conventionally dressed carcass and body conformation in broiler chickens. Broiler diet was supplemented with 0.2% protease (group E-I) and 0.3% protease (group E-II), and protein content in the feed was reduced by 4% (E-I) and 6% (E-II) through a decrease in soybean meal content. Fast-growing Cobb 500 broilers were used for a 63-day fattening trial. Body conformation measurement included absolute carcass conformation measures (metatarsus length, keel length, breast depth, breast angle, thigh girth) and relative body conformation measures – conformation indices (body weight/metatarsus length, body weight/keel length, body weight/breast depth, body weight/thigh girth). Results showed a significant effect of sex on the dressing percentage of conventionally dressed carcass and all body conformation measures, whereas diet had a significant effect on the dressing percentage of conventionally dressed carcass and breast angle values.

Key words: broilers, protease, dressing percentage, body conformation

Introduction

The main goal of modern broiler production is to ensure maximum use of the genetic potential of broiler hybrids. An increased demand for breast, thigh and thigh meat has led to research on ways to increase the yield of primal cuts of broiler carcass through production i.e. fattening. Intensive selection over the last decades has focused on the traits: weight gain, final body weight, feed conversion ratio, vitality, feathering, body conformation, skin color and, to some extent, meat quality. The results of the selection and production work include increasing growth rate and increasing body weight, along with improvement in feed conversion ratio,

over a relatively short fattening period of 42 days. *Leeson (2007)* emphasized that modern broiler hybrids have a high growth rate and a good feed conversion ratio, but often suffer from metabolic diseases, leg problems and increased fat deposition. Changes in basic principles of chicken meat production in EU countries, primarily regarding the maintenance and improvement of farm animal welfare, environmental protection and food safety, have prompted intensive development of various natural (ecological, biological, organic, biodynamic etc.) food production programs (*Škrbić et al., 2011*), involving a prolonged fattening period, lower stocking densities per unit area, modifications in existing poultry feed formulations, use of biologically active ingredients in poultry diet (enzymes, antioxidants, organic acids).

For their normal growth and development, chickens require optimization of a large number of factors, both nutritional and ambient. The most important nutritional requirement for optimum animal performance is to provide appropriate dietary levels of proteins (*Bregendahl et al., 2002*) i.e. amino acids (*Wijten et al., 2004*). Young fattening poultry need relatively high amounts of protein in the diet for optimum growth. On a global scale, the majority of protein ingredients incorporated into animal feeds are obtained from vegetable proteins, with oilseed crops and legumes being the main sources. Vegetable proteins are inadequate for poultry, not only in terms of sufficiency - quantity, but also in terms of quality – amino acid balance. Moreover, the vegetable sources of protein vary both in crude protein concentration and in the amino acid composition of the protein (*Thorpe and Beal, 2001*).

More recently, a significant number of researchers have opted to focus on the effect of protease supplementation of broiler diets on production performance, nutrient digestibility and meat quality (*Maiorka et al. 2009; Fidelis et al. 2010; Angel et al. 2011; Frietas et al. 2011; Dosković et al. 2015*).

The objective of this study was to examine the effect of protease (Ronozyme ProAct) on the dressing percentage of conventionally dressed carcass and body conformation of broilers.

Materials and Methods

A total of 300 day-old Cobb 500 broilers were allocated to three groups i.e. boxes (4.1m x 2.6m), each containing 100 birds at a stocking density of 10 birds/m². The broilers were randomly grouped, giving a random ratio of male to female birds across groups. Chicks had free access to water and feed, and a 24-h photoschedule was applied. Ad libitum feeding was used. Optimal microclimate conditions were provided by ventilation system (through roof openings and using ventilation fans).

The experimental period was 63 days.

Dietary treatments

A three-stage feeding/fattening program was used, including starter (0-21 days), grower (22-42 days) and finisher (42-63 days).

One group of broilers was a control - C (fed a diet with a normal nutrient composition, in accordance with dietary requirements for individual fattening stages), and the other two groups were experimental groups i.e. E-I and E-II (receiving a diet with a 4% and 6% reduction in crude protein content, and with 0.2% and 0.3% protease supplementation, respectively).

Broiler feeds were in powdered form. Feed ingredients (used across fattening stages and test groups) and the chemical composition of feeds are presented in Table 1.

The enzyme protease commercially available as Ronozyme ProAct (DSM, The Netherlands) was used. Ronozyme ProAct is a preparation of serine protease produced by a genetically modified strain of *Bacillus licheniformis*. It is produced in solid and liquid forms and is intended for use as a feed additive in chickens for fattening at a recommended dose of 200 mg kg⁻¹, within the category of zootechnical additives, under the functional group of digestibility enhancers.

Table 1. Ingredients and nutrient composition of experimental diets for broilers¹

Ingredient, %	Starter stage (1 to 21 d)			Grower stage (22 to 42 d)			Finisher stage (43 to 49 d)		
	C	E-1	E-2	C	E-1	E-2	C	E-1	E-2
Treatments									
Maize	52.49	54.92	56.26	63.15	65.28	66.34	68.62	70.60	71.59
Soybean meal	22.24	19.79	18.44	13.00	10.85	9.78	9.10	7.10	6.10
Soybean groats	18.50	18.50	18.50	17.00	17.00	17.00	15.40	15.40	15.40
Feeding yeast	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
L-Lysine (78%)	0.10	0.10	0.10	0.20	0.20	0.20	0.23	0.23	0.23
DL-Methionine (99%)	0.22	0.22	0.22	0.30	0.30	0.30	0.30	0.30	0.30
Limestone	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10	1.10
Monocalcium phosphate	1.30	1.30	1.30	1.20	1.20	1.20	1.20	1.20	1.20
Salt	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Calcium formiate (30.5%)	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Captex T	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Premix	1	1	1	1	1	1	1	1	1
Protease	0.00	0.20	0.30	0.00	0.20	0.30	0.00	0.20	0.30
Calculated composition									
ME, kcal/kg	3.081	3.100	3.112	3.157	3.174	3.183	3.181	3.198	3.207
Crude proteins, %	22.59	21.72	21.24	18.99	18.22	17.84	17.16	16.45	16.09
Crude fats, %	5.59	5.55	5.70	5.67	5.73	5.76	5.55	5.61	5.64
Ca, %	0.96	0.95	0.95	0.91	0.91	0.90	0.90	0.89	0.89
Total P, %	0.73	0.72	0.72	0.68	0.67	0.67	0.66	0.65	0.65
Available P, %	0.44	0.44	0.43	0.40	0.40	0.40	0.39	0.39	0.39
Total lysine, %	1.33	1.27	1.24	1.15	1.10	1.08	1.05	1.00	0.98
Methionine+cystine, %	0.92	0.90	0.89	0.91	0.89	0.88	0.86	0.84	0.83

¹ Treatments: C-control group, standard broiler diet, without protease; E-I- broilers fed a diet with a 4% reduction in crude protein level compared to control diet, and with 0.2% protease supplementation; E-II broilers fed a diet with a 6% reduction in crude protein level compared to control diet, and with 0.3% protease supplementation.

Data collection

At 63 days, 10 male and 10 female broilers were randomly selected from each group, tagged, weighed and slaughtered after a fasting period of 10 hours.

At slaughter, ready-to-grill carcass weight and abdominal fat weight were measured. Then, each carcass was dissected into primal cuts i.e. breast, drumsticks, thighs, wings, back, pelvis (according to the *Commission Regulation (EC) No. 543/2008*), and the following carcass conformation measures were taken (using the method of *Pavlovski and Mašić, 1983*): breast angle BA (degrees), metatarsus length ML (mm), keel length KL (mm), breast depth BD (mm) and thigh girth TG (mm). These measures indicated the body structure and development of certain i.e. major carcass parts of broilers. In order to eliminate the influence of body weight on these measures, their index values were determined (body weight/metatarsus length (g/mm) BW/ML, body weight/keel length (g/mm) BW/KL, body weight/breast depth (g/mm) BW/BD and body weight/thigh girth (g/mm) BW/TG) with the exception of breast angle, which is a genetically determined trait and, as such, only slightly affected by body weight (*Škrbić et al. 2011*).

Statistical analysis

The data obtained in this study were subjected to conventional statistical methods.

The significance of differences for meat quality parameters (dressing percentages, body conformation measures) was tested by the mathematical model of a two-factor analysis of variance (2x3 design – 2 sexes-S and 3 feeding treatments-FT).

The significant differences determined by the analysis of variance and the results of F-exp values were evaluated using Tukey's test. Significance was accepted at $P < 0.05$.

The test parameters were examined by the analysis of variance (ANOVA) using the statistical software *Statistica for Windows Release 6.0 (1995)*.

Results and Discussion

The dressing percentage of conventionally dressed carcass and relative body conformation measures are given in Table 2.

The analysis of the data in Table 2. shows that all carcass quality parameters were significantly affected by sex, with females having a significantly higher dressing percentage for conventionally dressed carcass, but lower values of all body conformation measures ($P < 0.05$), except keel length in E-II broilers ($P > 0.05$), which was due primarily to higher body weight at slaughter. Similarly, some other authors (*Pavlovski et al. 2007; Hopić et al. 1996; Hopić et al. 2000; Blagojević et al. 2009*) have reported higher values for all absolute body conformation measures and better body conformation scores in male broilers

compared to females, regardless of age. The different diet formulations used for the experimental groups of broilers had a considerably lower effect on the carcass traits tested. Namely, significance was observed only when comparing the dressing percentage of conventionally dressed carcass (control males had a higher dressing percentage compared to E-I and E-II male broilers) and breast angle values (the highest breast angle was obtained in control males, with significance determined compared to E-II male broilers and also between control and E-I females). *Dosković et al. (2012)* found that different protein levels used in broiler diet with or without protease supplementation had no effect on the dressing percentage of conventionally dressed carcass, linear conformation measures and conformation indices in Cobb 500 broilers on day 49 of the fattening trial.

Table 2. Dressing percentage of conventionally dressed carcass and body conformation (absolute values) of broilers across experimental groups

Treatment		Yield "CP", %	ML mm	KL mm	BD mm	BA degrees	TG mm	
Protease	Sex							
No	♂	\bar{x}	84.97 ^c	91.30 ^{ab}	130.20 ^{ab}	118.80 ^a	161.70 ^a	173.10 ^a
		Sd	0.81	2.75	1.55	4.08	5.64	6.84
	♀	\bar{x}	87.19 ^a	80.80 ^c	126.80 ^c	108.40 ^b	154.10 ^{bc}	158.40 ^b
		Sd	0.49	3.76	1.81	3.78	6.81	7.44
0.2%	♂	\bar{x}	84.94 ^c	93.60 ^a	130.60 ^a	116.40 ^a	159.20 ^{ab}	171.40 ^a
		Sd	0.51	2.37	1.07	6.67	7.15	5.29
	♀	\bar{x}	86.57 ^b	80.40 ^c	126.30 ^d	110.50 ^b	147.90 ^d	154.80 ^b
		Sd	0.70	2.76	3.59	4.50	7.61	7.27
0.3%	♂	\bar{x}	84.61 ^c	90.30 ^b	129.70 ^{ab}	116.30 ^a	154.50 ^b	167.10 ^a
		Sd	0.33	4.19	1.89	3.16	6.79	6.71
	♀	\bar{x}	86.47 ^b	81.00 ^c	128.30 ^{bc}	109.40 ^b	148.50 ^{cd}	159.40 ^b
		Sd	0.78	2.36	2.06	1.78	6.62	7.46
p-value								
Source of variation								
Protease		0.030	0.377	0.670	0.841	0.014	0.398	
Sex		0.000	0.000	0.000	0.000	0.000	0.000	
Protease x sex		0.333	0.137	0.100	0.224	0.454	0.108	

CP- "conventional dressing", ML-metatarsus length, KL-keel length, BD-breast depth, BA-breast angle, TG-thigh girth

^{a-d} Means within columns with different superscripts differ significantly ($P < 0.05$)

Index values of body conformation measures are presented in Table 3.

All body conformation indices (body weight/metatarsus length - BW/ML, body weight/keel length - BW/KL, body weight/breast depth - BW/BD and body weight/thigh girth - BW/TG) showed significant differences ($P < 0.05$) when the effect of sex was considered, whereas no effect of diet ($P > 0.05$) was observed.

Male broilers had higher relative body conformation scores compared to female birds, which was in agreement with the results of *Dosković et al. (2012)* in Cobb 500 broilers at 49 days of age, whereas *Blagojević (2011)* found that sex had a significant effect on BW/ML index in Master Gris broilers.

Table 3. Index values of body conformation measures in broilers across experimental groups

Treatment			BW/ML	BW/KL	BW/BD	BW/TG
Protease	Sex		g/mm	g/mm	g/mm	g/mm
No	♂	\bar{x}	48.50 ^a	33.99 ^a	37.28 ^a	25.59 ^a
		Sd	2.61	1.74	1.96	1.45
	♀	\bar{x}	44.33 ^b	28.19 ^b	32.94 ^b	22.54 ^b
		Sd	4.20	2.30	1.94	1.06
0.2%	♂	\bar{x}	46.94 ^a	33.62 ^a	37.73 ^a	25.64 ^a
		Sd	3.54	2.37	1.69	1.93
	♀	\bar{x}	44.57 ^b	28.35 ^b	32.40 ^b	23.15 ^b
		Sd	3.21	1.70	1.57	1.43
0.3%	♂	\bar{x}	47.21 ^a	32.78 ^a	36.56 ^a	25.51 ^a
		Sd	3.66	1.47	1.62	2.06
	♀	\bar{x}	43.63 ^b	27.53 ^b	32.28 ^b	22.17 ^b
		Sd	2.52	1.51	1.56	0.99
p-value						
Source of variation						
Protease			0.635	0.236	0.381	0.527
Sex			0.000	0.000	0.000	0.000
Protease x sex			0.687	0.870	0.559	0.680

BW – body weight at slaughter, ML – metatarsus length, KL – keel length, BD – breast depth, TG – thigh girth

^{a-b} Means within columns with different superscripts differ significantly ($P < 0.05$)

Conclusion

The results suggest that Cobb 500 male broilers at slaughter on day 63 of the fattening trial had a significantly lower dressing percentage of conventionally dressed carcass compared to female broilers, and higher values of all linear body conformation measures ($P < 0.05$), except keel length (group E-II). Moreover, all relative body conformation measures – conformation indices were significantly affected by broiler sex ($P < 0.05$). The effect of reduced crude protein levels in broiler diets (a 4% and 6% reduction compared to protein content in control diet) supplemented with protease (0.2% and 0.3% protease, respectively) exhibited significant differences when comparing the dressing percentages of conventionally dressed carcasses (control males had a higher dressing percentage than E-I and E-II male broilers) and breast angle values (between control and E-II males, and

between control and E-I females). Diet had no effect on the other absolute body conformation measures and conformation indices.

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Uticaj proteaze dodate u hranu i pola na randman i konformaciju trupova tovnihi pilića

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Rezime

U radu su prikazani rezultati ispitivanja enzima proteaze na randman klasično obrađenog trupa i konformaciju trupova pilića u tovu. Enzim proteaza dodat je u hranu za piliće u tovu u koncentraciji 0,2% (O-I grupa), odnosno 0,3% (O-II grupa), uz istovremeno smanjivanje sadržaja ukupnih proteina u smešama za 4% (O-I grupa), odnosno 6% (O-II grupa), preko smanjenog učešća sojine sačme. U ogledu je korišćen brzorastući hibrid Cobb 500, a tov pilića trajao je 63 dana. Za ocenu konformacije trupova utvrđene su apsolutne mere konformacije trupa (dužina piska, dužina kobilice, dubina grudi, grudni ugao, obim bataka) i relativne mere konformacije trupova – indeksi mera konformacije (telesna masa/dužina piska, telesna masa/dužina kobilice, telesna masa/dubina grudi, telesna masa/obim bataka). Rezultati istraživanja su pokazali da je pol pilića imao značajan uticaj na randman klasično obrađenog trupa i na sve mere konformacije trupova, a da je ishrana pilića različitim formulacijama obroka imala signifikantan efekat na randman klasično obrađenog trupa i vrednost grudnog ugla pilića.

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EFFECT OF STRAIN AND AGE ON BONE INTEGRITY OF COMMERCIAL BROILER CHICKENS

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Abstract: Skeletal disorders and leg problems cause varying degree of economic losses in broiler birds. This study was aimed at investigating the effect of strain and age on bone integrity of some broiler strains available to poultry farmers in Nigeria using morphometric and mechanical indices. Four hundred (400) one-day old chicks comprising of 100 each of Arbor Acre (AA), Hubbard (HB), Marshal R (MR) and Marshal Y (MY) strains were raised for a period of 42 days. The birds were fed similar diet at the starter and finisher phases. At the end of each week, 4 birds were randomly selected from each of the group and sacrificed, femur and tibiotarsal (left and right) bones were obtained from each of the bird for analyses. Body Weight (BW), Femur Weight (FW), Femur Length (FL), Tibia Weight (TW), Tibia Length (TL), Weight/Length Index (WLI), Diaphysis Diameter (DD), Relative Bone Density (RBD), Robusticity Index (RBT) and Tibiotarsal Index (TI) were recorded each week. Mechanical parameters were evaluated using the universal testing machine: Force, Moment of Inertia (MI), Stress and Modulus of Elasticity (ME). Mean values of Mechanical indices of the femur bone was significant at Day 42 with AA Strain having the least mean value of force and MY Strain with highest value of ME. The results of tibiotarsal bone revealed that MY Strain had highest mean value of force, ME and Stress across the weeks. Conclusively, Marshal Y strain was found to have better bone integrity than the other groups

Key words: Broiler Chicken, Bone integrity, Femur bone, Tibiotarsal bone, Morphometric mechanical

Introduction

In broilers, fast growth rates are generally correlated with musculoskeletal weakness since the development of bone in these animals is not well organized, less dense and more porous than slow growing ones (*Bennett, 2008; Williams et al., 2004*). Thus, tibia bones are strongly loaded by muscles and more prone to

various mineralization disorders and even fractures (*Charuta et al., 2011*). Market age poultry often suffer from lameness and bone deformities, which can cause bone breakage during catching and transportation and which create problem during processing. Around one third of broilers that died during transportation to processing plants were found to have died from hemorrhaging associated with femur dislocation (*Gregory and Wilkins, 1992*). In addition, skeletal deformities slow down automatic processing lines and increase the requirement of manual trimming during deboning (*Oviedo-Rondón, 2007*). Another concern that arises from leg disorders at the processing plant is food quality. Bone fragility and porosity are correlated with the incidence of bone fragments in deboned meat products and discolouration of meat adjacent to bone due to the leaching of blood (*Gregory and Wilkins, 1992*). Moreover, lame broilers that spend more time lying in the litter (*Oviedo-Rondón et al., 2009*) have more breast blisters, scratches, inflammatory processes, and muscle atrophy (*Julian, 1998; Vaillancourt and Martinez, 2002*). A secondary consequence of poor leg health is the increment of contaminants carried by lame broilers into the processing plant which threatens food safety (*Oviedo-Rondón, 2007*). All these carcass quality defects plus emaciation originates when broilers cannot walk to feeders and drinkers, which results in increased meat losses due to condemnations of carcasses (*Pattison, 1992; Yogaratnam, 1995*). There are different strains of broiler birds available to commercial poultry farmers in Nigeria, these constitute different broiler genotypes, with implications on differences in their performance characteristics such as growth rate, weight gain, feed utilization and efficiency, liveability, meat quality, skeletal abnormalities and bone integrity even when raised under the same environmental condition. This can be attributed to the differences in animal expression as a result of differences in both their genetic make-up and environmental factors. Therefore this study was aimed at investigating the bone integrity of some strains of broiler chicken available to commercial poultry farmers in Nigeria.

Materials and Methods

Four hundred (400) unsexed One-day-old broiler chickens comprising of 100 each of Arbor Acre, Hubbard, Marshal R and Marshal Y strains were used for this study. The strains were allotted into four different groups to constitute the treatment. Each group consisted of two (2) replicate pens with fifty (50) birds per pen. The experiment lasted for 42 days, during which bone samples (tibia and femur) were collected on weekly basis starting from the Day 21. All the necessary management practices were strictly adhered to. At the end of each week, four (4) birds were randomly selected from each treatment, weighed and sacrificed.

Tibiotarsal and femur (right and left) were removed from individual broilers and de-fleshed by hand, the bone caps were removed by hand and tibial and femur bones without muscles, ligament and tendons were obtained. The right and left tibiae as well as femur were then weighed and total length and bone shaft widths measured by means of a digital caliper with an accuracy of 0.001 mm according to the method described by (Zhang and Coon, 1997). The left tibiotarsal and femur bones were individually sealed in plastic bags to minimize moisture loss, and stored in a freezer at -18 °C until the end of the seventh week for mechanical testing. Morphometric measurements were then carried out on the right tibiotarsal and femur bones with the aid of a digital caliper. Parameters measured on the two bones include: Tibia weight (g), femur weight (g), Tibia length (mm), femur length (mm), Tibiotarsi Weight/Length index (g/mm), femur weight/length index (g/mm), Robusticity index, Diaphysis diameter(mm), Tibiotarsal index, relative bone density.

- Robusticity index, tibiotarsal index, relative bone density were calculated using the following formula respectively.
- Robusticity index = bone length/ cube root of bone weight (Tohid et al., 2014)
- Tibiotarsal index = diaphysis diameter – medullary canal diameter/diaphysis diameter × 100 (Tohid et al., 2014)
- Relative bone density = bone weight/ live body weight x 100 (Charuta et al., 2013).

Measurements of the bones mechanical properties were taken by means of the three-point bending test, using a universal testing machine. After breaking, diameter measurements were made inside and outside the mid-shaft of the bone both perpendicular and parallel to the direction of the applied force to calculate the area moment of inertia, stress as well as modulus of elasticity as described by (Kocabagli, 2001).

Statistical Analysis: The data on tibia and femur bone parameters were analyzed as a completely randomized design using the analysis of variance procedure of the SAS 9.2. Differences between means were compared using the Duncan Multiple Range Test (DMRT).

Results

The effect of strain on morphometric and mechanical parameters of femur and tibiotarsal bones at Day 42 are presented in Tables 1 and 2 respectively.

Table 1. Morphometric and Mechanical Parameters of Femur Bone at Day 42

Parameters	Arbor Acre	Hubbard	Marshal R	Marshal Y
Body weight(g)	1798.00±167.33	1720.75±113.96	1687.75±24.04	1643.75±108.63
Femur weight(g)	7.73±0.67	7.94±0.76	8.33±0.48	7.78±0.50
Femur length(mm)	65.95±1.10	67.34±1.91	69.69±1.09	67.45±0.69
Weight/length index	0.11±0.009	0.117±0.009	0.119±0.006	0.115±0.007
Robusticity index	33.47±0.69	33.90±0.33	34.44±0.59	34.13±0.83
Diaphysis diameter(mm)	8.12±0.25	8.76±0.14	8.93±0.49	8.63±0.29
Relative bone density	0.43±0.02	0.46±0.02	0.49±0.03	0.48±0.03
Force(Kg)	12.01 ^b ±0.34	12.75 ^a ±0.06	12.90 ^a ±0.16	12.91 ^a ±0.07
Moment of inertial	0.02±0.003	0.03±0.005	0.002±0.003	0.02±0.003
Stress (kg/cm ²)	156.17±20.47	196.74±19.75	204.05±25.40	216.54±32.91
Modulus of Elasticity (kg/cm ²)	6213.23 ^b ±538.64	7061.11 ^{ab} ±1119.69	8287.79 ^{ab} ±915.02	9459.10 ^a ±448.94

a, b Means with the same superscript across the rows were not significantly

Table 2. Morphometric Parameter of Tibia Bone at Day 42

Parameters	Arbor Acre	Hubbard	Marshal R	Marshal Y
Body weight(g)	1798.00±167.33	1720.75±113.96	1687.75±24.04	1643.75±108.63
Femur weight(g)	10.34±0.82	10.25±0.60	10.38±0.53	9.89±0.74
Femur length(mm)	89.30±1.30	91.29±1.67	92.48±2.04	91.36±0.62
Weight/length index	0.12±0.008	0.11±0.007	0.11±0.005	0.11±0.008
Robusticity index	41.12±0.68	42.12±0.95	42.45±0.88	42.73±1.26
Diaphysis diameter(mm)	7.87±0.25	8.43±0.17	8.28±0.28	7.81±0.48
Tibiotarsal index	34.68±1.61	30.24±2.10	26.67±3.92	25.19±2.26
Relative bone density	0.58±0.01	0.60±0.03	0.62±0.04	0.60±0.04
Force(Kg)	11.67d±0.10	12.06c±0.08	12.54b±0.10	13.10a±0.11
Moment of inertial	0.02±0.00	0.02±0.00	0.02±0.00	0.02±0.00
Stress (kg/cm ²)	239.08b±12.10	270.43b±2.47	249.94b±1.89	325.73a±27.05
Modulus of Elasticity (kg/cm ²)	10652.00b±1227.32	12534.99b±415.27	12661.84b±901.45	16655.30a±1289.97

a, b, c, d Means with the same superscript across the rows were not significantly different

Discussion

The bone weight/ bone length index is an index of bone density (*Seedor et al., 1991*), the higher the bone weight/ bone length index, the denser is the bone (*Monteagudo et al., 1997*), on the contrary, low Robusticity index indicates a strong bone structure (*Reisenfeld, 1972*). The results obtained at Day 21, 28 and 35 for both bone types in the parameters of Body Weight, Femur Weight, Femur Length, Weight/Length Index and Relative Bone Density did not reveal any significant differences across all the stains. This implied that the rate of weight gain in all the broiler strains at this stage were similar which resulted in their corresponding tibia and femur weight and length. However, Arbor Acre had the least robusticity index and a slightly higher value of relative bone density in the femur and tibiotarsal bones, this may implied that this strain had a better bone mineralization and density than other strains at this age. This may likely be attributed to the shorter length of the bone of Arbor Acre strain as compared to the other groups. The result of relative bone density revealed that Marshal Y had a slightly denser bone structure as adjudged by its slightly higher value for this

parameter. The strain difference was in tandem with the report of (*Vitorović et al., 2008*) who reported strain differences in ninety-one day old Master Gris, Red Bro, Farm Q and Hubbard Classic strains of broiler chickens in the parameters of Weight, Length, Breaking Force, Cross-Sectional Diaphysis Area, Cross-Sectional Medullary Area, Cross-Sectional Cortical Area.

Arbor Acre strain had the least mean value of Robusticity index and relative bone density with Marshal Y having a higher mean value of these parameters in the two bone types. Femur and tibiotarsal bone parameters increased arithmetically as the birds' age suggesting possible positive correlation between the parameters and the age of the bird. This is in tandem with the report of (*Krupski and Tataru, 2007*) that the values of densitometric, morphometric and mechanical bone parameters in mid-tibia increased with the age of the birds.

The values of stress parameters obtained from femur bones revealed that there were no significant differences but Marshal Y Strain had the highest mean value with Arbor Acre Strain having the least across the weeks. The results of tibiotarsal bones however indicated that there were significant differences in the mean value which is consistent with the femur bone, suggesting a better bone integrity of Marshal Y Strain compared to others. According to (*Lott et al., 1980*), bone strength is measured as required force (in Kg) to break a bone. The results of Force parameters revealed that there were no significant differences across the strain in the femur bone at Day 21 and 28 but the Day 42 result showed a significant higher mean value in the Marshal Y Strain compared to others suggesting that more The force required to break the bones of Marshal Y Strain was higher than that required for others, hence strong bone structure of this strain, similar conclusion was achieved for the tibiotarsal bone. *Patterson et al. (1986)* pointed out that stress and modular elasticity are more appropriate terms to describe bone strength with respect to different weight and length measures of bones at different bird species. *Rath et al. (2000)* also reported that modulus of elasticity describes bone hardness and its constitutional materials while yield stress is associated with only bone hardness and more hardness of a bone is related to a greater value of modular elasticity. In line with this postulations, it was observed that significant higher values of Modulus of Elasticity were recorded in the Marshal Y strain with the least value in the Arbor Acre Strain suggesting that the bone of Marshal Y Strain are stronger and less prone to breakage than the other strains.

Conclusion

This study indicated that there were strain differences in the morphometric and mechanical parameters of long bone of pelvic limbs of broiler chickens with

Marshal Y strain displaying a better bone integrity index as evident in all the indices adopted in this study.

Uticaj linije i starosti na integritet kostiju komercijalnih brojerskih pilića

Zubair Kayode Salaam, Mabel Omolara Akinyemi, Osaiyuwu Henry Osamede

Skeletni poremećaji i problemi sa nogama uzrokuju različite stepene ekonomskih gubitaka u odgoju brojerskih pilića. Ova studija je imala za cilj da ispita uticaj linije i starosti na integritet kosti nekih linija brojlera koje su na raspolaganju odgajivačima živine u Nigeriji, koristeći morfometrijske i mehaničke indekse. Četiri stotine (400) jednodnevnih pilića od kojih je po 100 bilo Arbor Acre (AA), Hubbard (HB), Marshal R (MR) i Marshal I (MI) linija, su odgajani u periodu od 42 dana. Pilići su hranjeni sličnim načinom ishrane u starter i finišer fazi. Na kraju svake nedelje, po 4 pileta je nasumično odabrano iz svake grupe i žrtvovano, kako bi se butne kosti i tibiotarzalne kosti (leva i desna) svakog brojlera koristile za analize. Telesne težine (BW), težina femura (FW), dužina femura (FL), težina tibije (TW), dužina tibije (TL), Indeks težina/dužina (WLI), prečnik dijafize (DD), relativna gustina kostiju (RBD), indeks robustnosti (RBT) i tibiotarzalni indeks (TI) su evidentirani svake nedelje. Mehanički parametri su procenjeni korišćenjem univerzalne mašine za testiranje: sila loma, moment inercije (MI), stresa i modula elastičnosti (ME). Srednje vrednosti mehaničkih indeksa butne kosti su bile značajne u uzrastu od 42 dana sa AA linijom sa najmanjim srednjim vrednostima sile loma i MY linija sa najvećom vrednošću ME. Rezultati za tibiotarzalne kosti otkrili su da je MY linija imala najviše srednje vrednosti sile loma, ME i stresa tokom celog perioda. Konačno, utvrđeno je da je Marshal I linija imala bolji integritet kostiju od ostalih grupa.

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ATTITUDES OF CONSUMERS IN SERBIA TOWARDS THE IMPORTANCE OF A BALANCED DIET AND TABLE EGGS AS FOODSTUFF

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Abstract: To investigate consumers' attitudes towards a balanced diet and eggs as foodstuff, 509 consumers have been surveyed. Through this survey, the consumers answered questions about the importance of a balanced diet, importance of having eggs in diet, the most common way of egg consumption, benefits and drawbacks of eating eggs. The responses were analysed by gender, age, occupation, education and place of living of the respondents (city of Belgrade, other cities and other populated places). Based on these results, it can be concluded that most of the respondents (79% males and 87% females) think a balanced diet is important, whereas 41% males and 52% females think it is very important. Moreover, most of the respondents find it important to have eggs in diet, and males and females do not differ in their responses (69%). The most common way of cooking and consuming eggs is frying, according to 68% male and 50% female respondents from all the places (60% from Belgrade, 55% from other cities and 62% from other populated places). As a benefit of eating eggs, the most frequent response is "nutritive value" (43% respondents), whereas, the most frequent drawback is "a risk of cholesterol" (19% respondents). Hence, one should pay particular attention to education on the importance of diet, at the level of the whole community.

Key words: diet, eggs, habits, consumer, survey

Introduction

Throughout all life stages, diet is very important for keeping good health. It is considered to be a significant factor in preventing chronic non-communicable diseases (NCDs), which were responsible for about 60% of the 56.5 million of

deaths reported around the world in 2001 and accounted for about 46% of the disease burden (Šipetić-Grujičić, 2012). Novaković et al. (2012) suggest that according to relevant international health organisations, over 40% malignant diseases and about 80% of all cardio-vascular diseases can be prevented with a proper diet, regular physical activity and smoking cessation. From the aspect of dietitians, it is necessary to provide food that complies with principles of modern medicine. However, economic and other circumstances make people conform to the existing ways of producing and providing food, and according to Mijatović and Mirčevski (2013), it is necessary to find a balance between those opposite ends. The same authors imply that dietetics should be an interesting topic not only for experts from this area but also for each individual, and in regard to that the authors suggest we need a systemic approach when analysing the existing eating habits and perceptions of food. Over the last decades, a growing importance in different areas of research has been given to habits and attitudes of consumers. Consumers are no longer seen as passive spectators, but as someone who plays an important role, actively participating in creating the environment they live in. Jovović and Femić (2006) imply that the basis of good marketing is to be familiar with consumers' needs and expectations. Furthermore, Savović et al. (2012) point out how important it is to include consumers in the system of quality insurance, in terms of food quality and safety, and to respect their perception as users. According to Zarić and Mijajlović (2009), eating habits significantly affect the choice of food, while Pavlovski et al. (2003) point out that consumers are growingly worrying about the way food is produced, as well as food safety and harmlessness. When studied the attitude of table egg consumers, surveys have investigated their preferences, as well as factors that affect consumers' decisions regarding table eggs (Kralik et al., 2014; Mizrak et al., 2012; Pavlovski et al., 2007; Huang, 2013). Based on those studies, it can be concluded that consumers are growingly demanding, having some specific requests in terms of producing eggs with special characteristics (functional food), animal welfare, food safety, etc. Consumers' attitudes towards eggs in diet have changed several times over the last decades and in one period consumers were focused on the risk of cholesterol, which resulted in the fear of coronary diseases and a decline in egg consumption (Ruxton et al., 2010). Bertechini and Mazzuco (2013) state that egg consumption varies from country to country, and according to IEC (2012), the highest annual consumption per capita is in Mexico (365), Japan (355), and in China and Hungary (350). Egg consumption in Serbia in 2007 was about 8kg (135 eggs per capita), whereas the same year egg consumption in the EU and the USA was 12 kg (200 eggs per capita) and 14 kg (240 eggs per capita), respectively (Milošević and Perić, 2011). In the next four years eggs consumption in the EU stagnated and 2011 it was 12.9 kg, while in the same period egg consumption in the Russian Federation increased from 12.7 kg to 15.2 kg (*Trend in poultry production in Europe and around the world, 2015*). Zlatanović (2015) implies that in 2013 egg consumption in the Republic of Serbia was 222 eggs per

capita, being highest when compared to the neighbouring countries - in Macedonia it was 168, Croatia 153 and in Slovenia 76 eggs per capita. A certain number of consumers are still suspicious towards eggs, although the latest studies imply the fear of cholesterol is not grounded in science, and *Gray and Griffin (2009)* point out that cholesterol in food, such as eggs, has little and clinically insignificant effect on cholesterol in blood. The same authors suggest this is the reason why official food organisations recommend that limitations in egg consumptions should be abolished.

The goal of this paper is to identify attitudes of consumers in the Republic of Serbia towards the importance of a balanced diet and eggs as foodstuff, the most common way of cooking and consuming eggs and benefits/drawbacks of eating eggs.

Material and Methods

The survey was conducted in the first half of 2016, on the territory of the Republic of Serbia. The survey comprised a random sample of 509 table egg consumers, bearing in mind to include different categories that are the subject-matter of the research (consumers' gender, age, education and place of living). The survey questionnaire included the city of Belgrade, other cities (52) and other populated places (38), being in line with the structure of populated areas in Serbia given by the Register of spatial units and GIS (*Statistical Office of the Republic of Serbia*). This research is a pilot research on attitudes and eating habits of the population in Serbia, as well as their preferences in terms of table eggs, conducted on the Belgrade area and in 14 districts (Braničevo, Južna Bačka, Kolubara, Mačva, Moravica, Pčinja, Podunavlje, Pomoravlje, Rasina, Raška, Srem, Šumadija, Toplica, Zlatibor).

To obtain the objectivity of data, the respondents were asked to fill in the survey questions without the presence of the surveyors. The survey questionnaire comprised the following:

1. Data on the respondents. The respondents had to circle the answers to the questions on their gender (male, female), education (primary school, secondary agricultural school, other secondary school, faculty of agriculture/veterinary science, other faculty, master's/doctoral degree in agriculture/veterinary science, master's/doctoral degree in other field), occupation/status, age (under 18, 18-25, 26-35, 36-45, 46-55, 56-65, 66-75, over 76) and place of living (the respondents had to fill in the blank space with their place of living);

2. Close-ended questions, with a Likert-type scale (from 1 – not important at all to 5 – very important (applied for the questions on the importance of a balanced diet and the importance of having eggs in diet);

3. Multiple-choice questions, from 1 to 7, where 1 means most frequent and 7 less frequent (applied on the question on the most frequent way of cooking/consuming);

4. Open-ended questions - where the respondents were asked to write their answers, i.e. to state their own opinions/attitudes (applied on the question on benefits/drawbacks of eating eggs).

The responses were analysed by respondents' gender, education, occupation/status, age and place of living. The choice of variables was based on a number of studies (*Jovičić et al. 2015; Kralik et al. 2014; Rodić-Trmčić et al., 2015*) and a starting point was that consumers' attitudes towards diet and their eating habits was determined by gender and age, social structure, demographic and social factors.

Only completed questionnaires (where the respondents marked categories and gave the answers on the questions) were analysed. The researchers used standard methods of analysis in Microsoft Excel.

Results and Discussion

Table 1 gives an overview of the results regarding the attitude of consumers towards the importance of a balanced diet. It shows the responses on the survey question: "How important is a balanced diet to you?"

Based on the results and the responses of the consumers who circled "4" and "5" on the marking scale, it can be ascertained that most of the respondents (79% males and 87% females) think a balanced diet is important and 41% males and 52% females think it is very important (they circled "5" on the scale). The results are in line with the research of *Arganini (2013)*, who points out women generally pay more attention to diet than men and thinks that nutrition and health are related. Furthermore, the results are also in line with the Research on health of the population of the Republic of Serbia in 2013 (*Ipsos Strategic Marketing, 2013*), which also shows that 19.7% of adults in Serbia do not consider health aspects when it comes to nutrition, and it is even more noticeable in the male population (26.3%).

Table 1. Responses on the survey question: “How important is a balanced diet to you?”

Category of the respondents	How important is a balanced diet to you?					
	Marking scale*					
	%	1	2	3	4	5
Gender	100					
Male	60	3	3	15	38	41
Female	40	1	0	12	35	52
Education	100					
Primary	2	0	0	13	37	50
Secondary agricultural school	3	7	0	27	53	13
Other secondary school	26	2	2	18	32	46
Faculty of agriculture/veterinary science	33	1	0	13	37	49
Other faculty	29	2	2	9	37	50
Master's/doctoral degree in agriculture/veterinary	5	0	4	4	33	59
Master's/doctoral degree in other fields	2	0	0	20	30	50
Occupation/Status	100					
Students	13	0	0	16	47	37
Employed	72	1	2	13	35	49
Unemployed	6	7	3	10	40	40
Retired	9	2	0	12	31	55
Age	100					
18-25	14	0	0	21	41	38
26-35	16	1	1	14	39	45
36-45	25	1	2	15	38	44
46-55	29	2	3	10	32	53
56-65	12	1	0	10	38	51
66-75	4	5	0	10	14	71

*1 – not important at all; 5 – very important

When observed by education level of the respondents, diet is most important to the respondents with master's and doctoral degree in agriculture and veterinary science (59%), and observed by occupation/status, it is most important to the retired (55%). Having analysed data on age, it can be seen that attitudes towards the importance of a balanced diet changes with age – the percentage of the respondents who think diet is very important increases, ranging from 38% (age 18-25) to 71% (age 66-75). The data show that particular attention should be paid to the education of younger generations, but also education in general, including all age categories, and bearing in mind that diet is a very important factor in preventing chronic non-communicable diseases (NCDs) (*Šipetić-Grujičić, 2012*). Moreover, a need for continuous education of wider structures on the importance of diet is also in line with *Nikolić (2011)*, who implies that the responsibility for a proper diet should be shared between individuals, society, family and health service.

Table 2 gives an overview of the results regarding the attitude of both male and female consumers of different age towards the importance of a balanced diet. It shows the responses on the survey question: “How important is a balanced diet to you?”

Table 2. Responses on the survey question: “How important is a balanced diet to you?” given by gender

How important is a balanced diet to you? (%)						
Age						
Male						
Scale	18-25	26-35	36-45	46-55	56-65	66-75
1	0	3	2	3	5	0
2	0	3	2	7	0	0
3	20	24	19	10	9	0
4	42	35	41	37	48	13
5	38	35	36	43	38	87
Female						
Scale	18-25	26-35	36-45	46-55	56-65	66-75
1	0	0	0	1	0	8
2	0	0	1	0	0	0
3	22	8	13	10	10	15
4	40	41	37	30	32	15
5	38	51	49	59	58	62

*1 – not important at all; 5 – very important

The respondents aged 18-25, both males and females (38% of males and 38% of females) regardless of their education and occupation/status have the same attitude towards a balanced diet, and the importance of a diet grows with age, for both male and female respondents. The obtained data are in line with research that shows the awareness on the importance of a healthy diet rises with age (*Fraza and Allshouse, 2003*). In the category of age 25-65, more female respondents assessed a balanced diet as “very important”, which implies that diet becomes more important to women after adolescence (*Jovičić, 2015*).

Table 3 gives an overview of the results regarding the attitude of consumers towards the importance of eating eggs. It shows the responses on the survey question: “Do you think you should have eggs in your diet?”

Based on the results, it can be ascertained that females think it is important to have eggs in diet - 38% marked “5”, which is slightly higher than in case of males (33%). If we take into account the responses marked with “4” and “5”,

however, there is no difference between males and females (69%). In available literature no data on gender preferences for egg consumption in Serbia have been found. When analysed by respondents' education, we can see that eggs in diet are very important (marked with "5") to people with the lowest level of education (primary education) and the highest level of education (master's/doctoral degree in agriculture or veterinary). The obtained results can be explained by the fact that experts in agriculture and veterinary have the most extensive knowledge on eggs as highly valuable foodstuff, whereas when it comes to the respondents with the lowest level of education, some additional research is needed to explain their attitude towards the importance of having eggs in diet.

Table 3. Responses on the survey question: "Do you think you should have eggs in your diet?"

Category of the respondents	Do you think you should have eggs in your diet?					
	Marking scale*					
	%	1	2	3	4	5
Gender	100					
Male	40	1	7	22	37	33
Female	60	2	3	26	31	38
Education	98					
Primary	2	0	0	13	37	50
Secondary agricultural school	3	0	13	34	20	33
Other secondary school	26	2	3	27	27	41
Faculty of agriculture and veterinary science	33	1	4	25	39	31
Other faculty	29	2	5	22	34	37
Master's/doctoral degree in agriculture/veterinary	5	0	4	17	29	50
Master's/doctoral degree in other science fields	2	0	30	30	30	10
Occupation/Status	100					
Students	13	0	3	38	42	17
Employed	72	1	5	23	31	40
Unemployed	6	3	7	23	37	30
Retired	9	5	5	18	29	43
Age	100					
18-25	14	0	4	37	41	18
26-35	16	2	8	30	36	24
36-45	25	0	2	23	36	39
46-55	29	3	5	22	25	45
56-65	12	2	6	16	33	43
66-75	4	5	0	10	33	52

*1 – not important at all; 5 – very important

Moreover, when observed by respondents' occupation/status, having eggs in diet is most important to the retired (43%), and least important to students (17%). The obtained results can be related to the lifestyle of these categories, since the retired have more time to prepare food themselves and the younger population unfortunately, more often based their diet on fast food (*Mijatović and Mirčevski,*

2013). Furthermore, it can be seen that the attitude towards the importance of having eggs in diet changes with age. The percentage of those who think it is very important increases, from 18% (age 18-25) to 52% (age 66-75). In this survey, the nutritive value of eggs was the most commonly listed benefit (219 respondents, i.e. 43%).

The importance of eggs in diet has also been pointed out in some research. *Mijatović and Mirčevski (2013)* mention that eggs are a healthier morning meal than pastries made of white flour; and in a healthy-eating pyramid eggs are placed on the top (*Healthy Eating Plate & Healthy Eating Pyramid, 2005*). According to *Rodić-Trmčić et al. (2015)* egg consumption in Serbia has shown a trend of slight growing, amounting to 4.3, 4.4 and 4.5 eggs per capita per week in 2011, 2012 and 2013, respectively. What would help increase egg consumption is better knowledge of all structures of the population, especially of young population, since there are still some concerns in our country regarding a negative impact of eggs on the level of cholesterol in blood. In this survey, 19% of the respondents (95 people) mentioned fear of increased cholesterol levels as a drawback of eating eggs. When analysed by age structure, the results show that fear of increased cholesterol is highest (28%) in the age group 35-55, which can be related to earlier misconceptions about cholesterol. The younger population had half as less percentage of those who expressed the fear of cholesterol, i.e. 14% respondents in the age group 18-25 stated a high level of cholesterol as a drawback of eating eggs. Numerous studies have shown, however, that cholesterol level in food is neither associated with cholesterol level in blood (*Song and Kerver, 2000*), nor with an increased risk of heart diseases (*Barraj et al., 2009*), and that is the reason why focus should be put on educating people in Serbia on this topic. Furthermore, *López-Sobaler and González-Rodríguez (2015)* point out that leaving eggs out from diets is unnecessary and undesirable.

Table 4 gives an overview of the results regarding the attitude of both male and female consumers of different age towards the importance of eating eggs. It shows the responses on the survey question: "Do you think you should have eggs in your diet?"

Table 4. Responses on the survey question: “Do you think you should have eggs in your diet?” given by gender

Do you think you should have eggs in your diet? (%)						
Age						
Male						
Scale	18-25	26-35	36-45	46-55	56-65	66-75
1	0	3	0	3	0	0
2	8	12	6	9	5	0
3	27	35	20	19	14	0
4	38	35	37	28	48	50
5	27	15	37	41	33	50
Female						
Scale	18-25	26-35	36-45	46-55	56-65	66-75
1	0	2	0	2	2	8
2	3	6	0	3	8	0
3	42	24	25	25	17	15
4	42	37	35	22	25	23
5	13	31	40	48	48	54

*1 – not important at all; 5 – very important

More female respondents aged 25-75, regardless of their education and occupation/status, find it important to have eggs in diet (Table 4). In available literature, no data were found on differences in attitudes of male and female consumers of different age towards having eggs in diet. However, what can be important is the research of *López-Sobaler and González-Rodríguez (2015)* on nutritive requirements of men and women, indicating that women in certain life stages and at a certain age have specific requirements. According to these authors, egg consumption can prevent certain chronic diseases and improve general health of women at an old age.

Table 5 gives an overview of the attitudes towards a balanced diet and eating eggs, given by the place of living. Differences in diet in different regions in Serbia were determined in the research of *Rodić-Trmčić et al. (2015)*. On the other hand, the aim of this paper was to identify differences in consumers' attitudes towards the importance of a balanced diet and eggs in diet, depending on their place of living – a city with over a million people, other cities with a much smaller number of people or other populated places with the smallest number of people. As a starting point, the authors made an assumption that different place of living differed in attitudes and eating habits. Data found in the research of *Mijatović and Mirčevski (2013)* support this assumption, stating that egg consumption in agricultural households is higher (181 egg per person per year) than the consumption in non-agricultural households (171.6 eggs per person per year).

Taking into account the answers marked “4” and “5”, 84% of respondents from the Belgrade area assessed a balanced diet as “very important”, whereas in

other cities (83%) and other populated places (81%) that percentage was slightly smaller. The obtained result shows that the population in Serbia, whether they live in the capital, smaller cities, or other populated places, has a similar opinion on the importance of a balanced diet. The importance of having eggs of diet was poorest assessed by the respondents from “other populated places” (58%) and highest assessed by the respondents from “other cities” (73%), when observed the answers marked with “4” and “5”.

Table 5. Responses of the consumers from cities and other populated places, regarding a balanced diet and eggs in diet

Place of living	Part of the sample %	How important is a balanced diet to you?					Do you think you should have eggs in your diet?				
		Marking scale*					Marking scale*				
		1	2	3	4	5	1	2	3	4	5
Total	100	1	1	14	36	48	2	5	25	32	36
Područje grada Beograda	41	2	0	14	36	48	1	6	26	30	37
Other cities	47	1	3	13	36	47	2	4	21	35	38
Other populated places	12	3	2	14	39	42	2	8	32	34	24

*1 – not important at all; 5 – very important;

Table 6 gives an overview of the results regarding the ways of consuming eggs. Frying (fried eggs and scrambled eggs) are most common way of consumption for all the respondents, regarding of their gender, occupation/status and place of living.

Table 6: Ways of cooking / consuming eggs

Category of the respondents	Ways of cooking / consuming eggs							
	Part of the sample %	Boiling		Frying			Other	
		Soft-boiled	Hard-boiled	Fried	Scrambled	bled with ham, mushrooms, ooms,	Eggs as an ingredient (in cakes, salads, etc.)	Other
Gender	100							
Male	60	11	11	30	22	16	8	2
Female	40	10	18	22	18	10	19	3
Occupation/Status	100							
Students	13	1	9	25	24	24	12	5
Employed	73	11	15	26	19	11	16	2
Unemployed	6	14	25	11	32	18	0	0
Retired	8	13	22	20	20	2	19	4
Place of living	100							
Belgrade area	41	8	15	22	22	16	15	2
Other cities	47	10	17	28	16	11	17	1
Other populated areas	12	11	12	24	24	14	10	5

Previous research has shown that eggs consumption varies significantly between different counties (*Magdelaine, 2011*). Moreover, available literature suggests that there are differences among countries in the way of cooking/consumption (*Mizrak, 2012; Zelić, 2015*), which can be explained by differences in traditional cuisine. In that sense, the results of this research indicate similarity with the results from the neighbouring countries (*Zelić, 2015*) and differences when compared to Turkey (*Mizrak i sar., 2012*).

Conclusion

According to the results of the survey about the importance of a balanced diet and eggs as foodstuff, it can be concluded that most consumers in Serbia consider a balanced diet as important. More female respondents (87%) give importance to balanced diet than male respondents (79%). Particular attention should be paid to education on the importance of diet as an important factor in preventing diseases, on the level of the whole community. A special focus should be put on younger generations, putting a focus on the importance of eggs as highly-valuable foodstuff. Since there are still some concerns and confusion among the population of Serbia regarding egg consumption and cholesterol increase, special attention should be given to eliminate this unreasonable fear. Egg consumption in Serbia is lower than in some developing and developed countries. In this sense, educational and other measures should be taken in order to increase the percentage of eggs as a highly nutritious foodstuff in the diet of all population categories.

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Stavovi potrošača u Srbiji o značaju uravnotežene ishrane i konzumnim jajima kao namirnici

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Rezime

U cilju ispitivanja stavova potrošača o značaju uravnotežene ishrane i jajima kao namirnici anketirano je 509 potrošača, na području Republike Srbije.

Kroz anketno ispitivanje potrošači su se izjasnili o značaju uravnotežene ishrane, značaju jaja u ishrani i najčešim načinima konzumiranja, prednostima i nedostacima jaja. Odgovori anketiranih potrošača obrađeni su po polu, starosti, zanimanju, obrazovanju i sredini u kojoj žive (grad Beograd, ostali gradovi, ostala naseljena mesta). Na osnovu rezultata ispitivanja može se zaključiti da je većini anketiranih potrošača važna uravnotežena ishrana (79% za muški i 87% za ženski pol), pri čemu se o ishrani kao veoma važnoj izjasnilo 41% muškaraca i 52% žena. Takođe, za većinu anketiranih potrošača zastupljenost jaja u ishrani je važna, a po ovom pitanju među pripadnicima različitih polova nema razlika (69%). Najčešći način pripreme, odnosno konzumiranja, su pržena jaja, kod oba pola (za muškarace je to 68%, a kod žena 50%) i u svim životnim sredinama u Beogradu 60%, ostalim gradovima 55% i ostalim naseljenim mestima 62%). Kao prednost jaja u ishrani potrošači su najčešće naveli hranljivost (43% ispitanika), a kao najčešći nedostatak strah od povećanja holesterola (19% ispitanika). Edukaciji o značaju ishrane, sa akcentom na značaj jaja kao visokovredne namirnice, trebalo bi posvetiti posebnu pažnju, na nivou celokupne društvene zajednice.

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MORPHOMETRIC CHARACTERIZATION OF THE LIPIZZANER HORSE BREED IN THE STUD „VUČIJAK“

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Abstract: The research was conducted on 10 stallions and 31 mares of the Lipizzaner breed in the stud “Vučijak” Prnjavor. In general, 28 morphometric measures of stallions and mares were taken. Morphometric characterization shows that the “Vučijak” stud farm owns Lipizzaner with smaller body form comparing to other Lipizzaner around Europe. The body of the Lipizzaner from “Vučijak” has a rectangle shape which is in accordance with the appearance of the majority of Lipizzaner in Europe. Based on these measures, it is confirmed that there is a statistically significant difference between stallions and mares in: withers height, width of lower jaw, length of neck and shoulders, length of radius, width of chest, length and width of pelvis, volume of front leg tibia, and length of hind leg long pastern (proximal phalanx). Out of all named, in comparison to stallions, mares have bigger morphometric measures of length of shoulders, width of pelvis and length of hind leg long pastern (proximal phalanx). In the remaining 18 measures stallions and mares of the “Vučijak” stud show certain homogeneity.

Keywords: stud Vučijak, Lipizzaner breed, morphometric characterization, stallion, mare

Introduction

The Lipizzaner is one of the oldest European horse breeds. The Lipizzan breed dates back to 1580, when it has been established at Lipizza (now Lipica). They are trained for riding schools, classical dressage and other equestrian sports as well as recreation (*Pallottino et al., 2015*). Lipizzaner has had a specific development: due to desires of the Habsburgs, high number of different breeds participates in its perfectly composed creation. From Andalusian Baroque stallions it gets tranquility and obedience, from Arabian stallions it gets loyalty, from Italian stallions it gets graciousness and from Karst stallions it gets stamina and firmness. The strict systematic selection during 450 years of its existence, as well as breeding the same breed on a higher number of stud farms with different purpose, results in higher number of lines and genera and higher phenotype diversity.

Lipizzaner horse have been previously studied with respect to morphometric variability (Zehner et al., 2001a, 2001b; Zehner 2001c; Rastija et al., 1991, 2004; Zohmann et al., 2001). The most famous Lipizzaner stud are: Baclean, Đakovo, Fagaras, Monterotondo, Piber, Szilvasvarad and Topol'cianky. Zehner et al. (2001b) have done a morphometric characterization of Lipizzaner breeds from all seven stud. Characterization and comparison of horses from different stud farms is done on the basis of 37 morphometric measures and formed angles. Its conclusion is that there are 34 differences out of 37 measures for mares and 29 differences out of 37 measures for stallions. The extremes of morphometric results are horses from Piber and Szilvasvarad; their breeding is different which brings to morphometric differences between them.

The stud "Vučijak" is the only Lipizzan stud in Bosnia and Herzegovina, founded in 1946. Reproductive material has been taken from breeding stables in Croatia. The purpose of founding stud has been to enrich the existent horse population of horses in northern parts of Bosnia and Herzegovina. Soon enough, breeding of Lipizzaners spread to almost entire Bosnia thanks to selective work at the "Vučijak" stud. Praček (1999) notes that the breeding of Lipizzaners in Yugoslavia in 1970s has been planned in three directions: classical dressage (Lipica), driving and weight carrying (Bosnia) and field works (Slavonia, Posavina, Vojvodina and other parts of Serbia).

The aim of this work is a morphometric characterization of Lipizzan horses from the stud "Vučijak" and finding differences in morphometric measures between stallions and mares.

Material and methods

Morphometric measures are taken at the stud "Vučijak". Ribbon and height measuring stick have been used for measuring. On each individual animals 28 physical measures has been found, following the standard procedure which was developed by Oulehla (1996). Every measured animal has finished its development. Horses have been brought on flat and sturdy base and held by the workers of the haras. They have all been measured by one person. In general, 41 animals (10 stallions and 31 mares) have been measured, i.e. around 54% of all horses predicted for further reproduction by the Commission for the annual overview of the haras. Statistical data is done in ANOVA program, and the difference in morphometric measures between stallions and mares is tested by t-test. The difference between environments is tested for probability of 0.05 and 0.01.

Results and discussion

Results of morphometric characterization of stallions and mares in the stud "Vučijak" can be seen in tables 1 and 2. In comparison to mares, stallions have bigger measures for: height at withers (tape), width of head (lower jaw), length of

neck and length of upper arm 2. Beside these, stallions have bigger measures for: length of rearquarters, circumference of cannon bone (metacarpal), width of chest, and the found difference is statistically relevant. In comparison to stallions, mares have more emphasized length of shoulders, length of pastern (hind leg) and width of hips. The difference between the first two measures is statistically of high importance, while for the third it is statistically relevant. The differences for other found measures between stallions and mares are not statistically relevant. *Rastija et al. (1991)* claim that the height at withers (tape) for stallions at the stud Vučijak has been between 148.00 and 161.67 cm, unevenly and significantly lower than for stallions at the Đakovo in Croatia. The same author claims that the circumference of chest for stallions at the stud Vučijak has been between 158.00 and 188.33 cm, while the volume of tibia has been between 19.69 and 20.00 cm. The average value for the first two morphometric measures in this research is in the range of these values. The only difference is the circumference of cannon bone (20.03 cm) which is above the range given by the same author.

Table 1. Morphometric characteristics of stallions on the stud "Vučijak", cm (n=10)

Morphometric measurements	X	S	V	Relative measurement, %
Height at withers (measuring stick)	148.80±1.02	2.23	1.58	100
Height of back	140.40±1.32	4.19	2.98	94.35
Height of rump	148.20±0.95	3.10	2.03	99.60
Body length	153.50±1.85	5.85	3.18	103.16
Length of forequarters	36.40±0.81	2.59	7.11	24.46
Length of barrel	69.70±1.01	3.19	4.57	46.84
Length of rear quarters	51.50*±0.74	2.36	4.64	34.61
Depth of chest	66.70±0.84	2.66	3.98	44.82
Width of chest	39.30* ±0.57	1.82	4.63	26.41
Width of hips	48.20*±0.95	3.01	6.24	32.39
Width of thurls	50.00±0.47	1.49	2.92	33.60
Length of neck	76.50**±1.05	3.34	4.63	51.41
Length of shoulder	56.90**±1.01	3.21	5.64	38.24
Height at withers (tape)	159.80**±1.39	4.41	2.75	107.39
Circumference of chest	176.90±1.57	4.97	2.80	118.89
Circumference of cannon bone (metacarpal)	20.03*±0.52	1.65	8.12	13.46
Circumference of cannon bone (metatarsal)	21.60±0.33	1.07	4.95	14.52
Length of head	52.89±0.57	3.28	6.21	35.54
Width of head (lower jaw)	17.30**±0.51	1.63	9.42	11.63
Length of upper arm 1	34.00±0.64	2.05	6.02	22.85
Length of upper arm 2	40.00**±0.64	2.05	5.20	26.88
Length of forearm	38.40±0.76	2.41	6.27	25.81
Length of cannons (front leg)	21.10±0.67	2.13	10.09	14.18
Length of pastern (front leg)	15.40±0.40	1.26	8.18	10.35
Length of thigh	40.80±0.55	1.75	4.28	27.42
Length of second thigh	42.80±0.80	2.52	5.88	28.76
Length of cannons (hind leg)	26.40±0.54	1.71	6.47	17.74
Length of pastern (hind leg)	15.01**±0.17	0.56	3.70	10.15

If we compare morphometric measurements of stallions in the Vučijak with the stallions of other Lipizzan stud by various authors it can be found that the stallions of the Vučijak have smaller shape. *Rastija et al. (2004)* claim that the height at withers for the stallions from Đakovo, measured with stick, has been 159.20 cm, and with ribbon, 168.10 cm. The body length has been 155.45 cm, depth chest 72.30 cm, circumference of chest 184.25 cm and circumference of cannon bone 21.22 cm. According to *Zechner (2001c)* research, the greatest height at withers is noted for the stallions of the Szilvasvarad haras (158.20 cm), followed by the stallions from Topol'cianky (156.80) and Fagaras (156.80), while the smallest measure is in the Piber (153.60). The same author finds that the stallions from Topol'cianky have the longest body (165.20 cm), followed by those from Szilvasvarad (163.10), Đakovo (161.10), while those from Monterotondo have the shortest body (155.20). Found differences between the horses from Vučijak and other Lipizzan stud farms is in accordance with *Zehner et al. (2001a, 2001b) and Zehner (2001c)* where the differences in morphometric measurements between horses of different stud farms are credited to different goals of breeding. Horses from the Vučijak have rectangle shape of the body, which is in accordance with the mentioned research where authors have found that horses from different Lipizzan stud have rectangle body shape and that the difference in the body length and height at withers is between 3 cm and 8 cm. Only the horses from the Monterotondo haras are not in that frame. *Sölkner et al. (2001a)* claim that Lipizzan stables in Slovenia, Croatia and Slovakia breed modern horses for riding while those in Romania breed studs for improvements in field works.

Table 2. Morphometric characteristics of mares on the stud "Vučijak", cm (n=31)

Morphometric measurements	X	S	V	Relative measurement %	Ratio studs-mares
Height at withers (measuring stick)	147.12±0.61	3.44	2.32	100	98.87
Height of back	142.14±0.57	3.20	2.24	96.61	101.24
Height of rump	149.41±0.56	3.14	2.10	101.56	100.82
Body length	154.61±0.85	4.73	3.05	105.09	100.72
Length of forequarters	36.58±0.47	2.66	7.28	24.86	100.49
Length of barrel	72.74±0.91	5.09	6.99	49.44	104.36
Length of rear quarters	49.19±0.74	2.36	4.64	33.43	95.51
Depth of chest	67.03±0.52	2.92	4.35	51.68	100.49
Width of chest	36.90±0.60	3.37	9.13	25.08	93.89
Width of hips	50.83±0.49	2.78	5.46	34.55	105.46
Width of thurls	48.48±0.42	2.36	4.86	32.95	96.96
Length of neck	66.45±0.77	4.32	4.96	45.17	86.86
Length of shoulder	61.63±0.83	4.63	7.58	41.89	108.31
Height at withers (tape)	154.67±0.78	4.34	2.80	105.13	96.79
Circumference of chest	175.32±1.37	7.63	5.25	119.17	99.11
Circumference of cannon bone (metacarpal)	19.51±0.14	0.82	4.20	13.26	97.40
Circumference of cannon bone (metatarsal)	21.80±0.16	0.94	4.31	14.82	35.39
Length of head	50.12±0.59	3.30	6.58	34.07	94.76
Width of the head (lower jaw)	15.45±0.24	1.33	8.60	10.50	89.31
Length of upper arm 1	32.64±0.24	1.37	4.19	22.18	96.00
Length of upper arm 2	37.06±0.28	1.61	3.44	25.19	92.65
Length of forearm	37.80±0.42	2.35	6.21	25.69	98.44
Length of cannons (front leg)	21.87±0.39	2.18	9.96	14.86	103.64
Length of pastern (front leg)	15.25±0.25	1.41	9.24	10.37	99.02
Length of thigh	39.45±0.52	2.94	7.45	26.81	96.69
Length of second thigh	44.41±0.51	2.86	6.43	30.18	103.76
Length of cannons (hind leg)	24.96±0.49	2.76	11.05	16.96	94.54
Length of pastern (hind leg)	17.00±0.29	1.64	9.28	11.55	113.26

Mares, just like stallions, in comparison to other Lipizzan mares, have smaller body frame. The average height at withers measured with stick is 147.12 cm, while the average height at withers measured with ribbon is 154.67 cm. Mares from the Vučijak have rectangle body shape which proves that the body length is 154.61 cm, which is more than height at withers by 7.49 cm, and that is in accordance with *Zehner et al. (2001a, 2001b); Zehner (2001c) and Zohmann et al. (2001)*. Depth of chest for mares from Vučijak is 67.03 cm in average, while the circumference is 175.32 cm. According to *Zehner et al. (2001a)* the greatest height at withers is noted for mares from the Piber (156.80 cm), followed by those from Đakovo (155.40) and Fagaras (154.70), while the smallest is noted for mares from Szilvasvarad (153.20) and Lipica (153.20). The same author concludes that

the longest body is found for mare from Piber (164.80 cm), and the shortest for mares from Lipica (158.50 cm). The results of the measurements of mares at the Đakovo, according to *Rastija et al. (2004)*, exceed the morphometric measures of mares at the Vučijak found in this research, which is also confirmed by the research from *Baban et al. (2006)*. According to these authors, the values measured with ribbon: height at withers, circumference of chest and circumference of cannon bone are: 164.73 cm, 185.68 cm and 20.83 cm – significantly higher than the results in this work.

Sölkner et al. (2001a) give three basic measures (height at withers, circumference of chest and circumference of cannon bone) of mares from seven Lipizzan horse stables: Beclean (153.70 - 178.90 -19.50), Fagaras (154.70 - 181.60 - 19.60), Đakovo (155.40 - 193.30 - 20.40), Lipica (153.20 - 188.00 -19.20), Piber (153.30 - 190.10), Szilvasvarad (156.80 - 189.30 - 20.50), Topol'canky (153.20 - 191.40 - 19.50). Mares from these Lipizzan horse stables are higher at withers and have bigger circumference of chest than mares from Vučijak. On the other hand, mares from Vučijak have bigger circumference of cannon bone (front leg) than mares from Beclean, Lipica and Topol'canky. According to *Čačić (2003)*, the fundamental morphometric measures of Lipizzan horses from Croatia are: height at withers (with measurement stick) – 150.38 cm, height at withers (tape) – 160.59 cm, circumference of chest – 184.50 cm and circumference of cannon bone – 19.39 cm. Following the morphometric measurements of the horses from the Vučijak in this work, it can be claimed that the Lipizzaner from Prnjavor is the most similar to Lipizzaners from Croatia.

Sölkner et al. (2001a) and *Sölkner (2001b)* find that the basic population of Lipizzaners is in numerous, mostly big and state-owned, stud farms (Lipica in Slovenia, Đakovo in Croatia, Piber in Austria, Beclean, Fagaras and Simbata de Jos in Romania, Monterotondo in Italy, Szilvasvarad in Hungary and Topol'canky in Slovakia) and private breeding farms around the world. Besides the mentioned Lipizzan studs, we can add Karađorđevo (Serbia), Lipik (Croatia) and Vučijak (Republic of Srpska, Bosnia and Herzegovina) as significant horse farms for the breeding of this breed. Lipizzan horses from Lipik have been saved at Karađorđevo during the tragic wars in former Yugoslavia, which is commendable, knowing how small population of Lipizzaners is. This is confirmed by *Habe et al. (2002)*, emphasizing the history of breeding of Lipizzaners and small number of such horses.

Morphometric differences between the horses from different stud depend on the goals of breeding which differs from one country to another or even one stud to another. In Hungary and Romania, they breed bigger and rougher horses with emphasized length of format, capable of field working and equestrian sports. Italians have no specific goal in Lipizzan breeding and they aim at preserving the lines and genera which they have. Austrians and Slovenians breed Lipizzaners for general dressage for the needs of the Spanish Riding School of Vienna, Austria.

Slovakians aim to breed a heavier Lipizzaner in order to use it for work and riding and other needs. Croats aim to breed a Lipizzaner which would be bigger and used for riding and cart driving.

These researches show the morphometric differences between the Lipizzaner from Vučijak and Lipizzan horses from other notable stud in Europe. It is similar with smaller stud, as it was aimed at the Vučijak in Prnjavor.

Conclusion

In conclusion, according 28 taken measurements there is a statistical difference between stallions and mares in 10 measurements: height at withers (tape), width of head (lower jaw), length of neck, length of upper arm 2, length of rearquarters, width of chest, length of pastern (hind leg), circumference of cannon bone (metacarpal), width oh hips and lenght of shoulders. Out of 10 named measurements, in comparison to stallions, mares have bigger morphometric measures of length of shoulders, width of hips and length of pastern (hind leg). In the remaining 18 measures there is no specific differences noted between stallions and mares. Lipizzaner of the Vučijak has a rectangle body shape, which is in accordance with the appearance of Lipizzaners in the most of European stud farms. Horses from Vučijak are smaller than other Lipizzan horses in Europe, which is in accordance with the breeding aim of this stud farm, which is to create a smaller horse for field works and carrying weight.

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Morfometrijska karakterizacija lipicanera ergele "Vučijak"

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Rezime

Istraživanje je sprovedeno na 10 pastuva i 31 kobili rase lipicanera na ergeli "Vučijak" Prnjavor. Ukupno je izmereno 28 morfometrijskih mera na konjima navedene ergele. Morfometrijska karakterizacija je pokazala da ergela Vučijak poseduje lipicanera manjeg okvira tela u odnosu na druge konje poznatih ergela lipicanera u Evropi. Pored toga telo lipicanera iz Prnjavora ima izgled pravougaonika, što je u saglasnosti sa izgledom tela lipicanera većine drugih ergela lipicanera iz Evrope. Na osnovu izmerenih morfometrijskih mera utvrđeno je da

postoji statistički značajna razlika između pastuva i kobila na mere: visina grebena (merena pantljikom), širina donje vilice, dužina vrata, dužina ramena, dužina nadlaktice 2, širina grudi, dužina karlice, širina karlice, obim cevanice prednje noge, dužina kičične kosti zadnje noge. Od svih navedenih morfometrijskih mera, kobile su imale veće mere: dužina ramena, širina karlice i dužina kičice zadnje noge u odnosu na pastuve. Na ostalih 18 mera pastuvi i kobile ergele Vučijak su pokazali određenu homogenost.

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CONDITION FACTOR AND ORGANOSOMATIC INDICES OF RAINBOW TROUT (*ONCHORHYNCHUS MYKISS*, WAL.) FROM DIFFERENT BROOD STOCK

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Abstract: Condition factor and organosomatic indices of fish represent a way of monitoring environmental factor influence on fish. The rainbow trout (*Oncorhynchus mykiss*, Wal.) individuals that were used in the work originated from different brood stock and different fish farms. Spawning was carried out on five selected fish farms. After that, the breeding eggs were delivered in Klasnik hatchery where the same environmental conditions were provided during embryonic development and cultivation of all five groups of individuals. As a conditioning factor in this work Fulton's condition factor and coefficients of the heart, liver and spleen were analyzed. In all individuals we determined the total length, standard length and body mass. In total, fifty individuals, which were divided into five groups from five brood stock, were analyzed. The results show significantly different values for the condition factor and organosomatic indices between some of the analyzed groups.

Key words: rainbow trout, brood stock, organosomatic indices

Introduction

Condition factor and organosomatic indices are usually used in assessing the general health of the fish, on individual and population level (Hoque *et al.*, 1998). Since they include many levels of processes in the organism at various levels of organization, indices like Fulton's condition factor can indicate nutritional status of individuals and the general health status of fish (Adams *et al.*, 1992).

The condition factor presents a degree of individual's response to the influences coming from the environment such as the quantity and quality of nutrients, the presence of pathogens, pollutants and toxic substances, which can result in a change in the mass of individuals and organs compared to individuals from the unchanged environment. The value of condition factor is an expression of the physical condition of the fish and it is suitable for comparison of individuals of the same species and demonstrates differences in relation to sex, season and place of sampling (Dekić, 2010).

Organosomatic indices can be described as the ratios of organs to body weight when the measured organ in relation to body mass can be directly linked to some environmental changes (Ronald and Bruce, 1990). It is manifested through changes in size that are reflected through a reduction or increase, influenced by environmental factors. Size and weight of the liver, spleen, heart, gonads and other organs are related to the overall length and weight of fish and indicate the general status of health of the fish. The aim of this research was to determine the values of condition factor and organosomatic indices at five groups of rainbow trout from different brood stock and to estimate their shape and health status.

Materials and Methods

The rainbow trout (*Oncorhynchus mykiss*, Wal.) individuals that were used in the work originated from different brood stock and different fish farms. Spawning was carried out on five selected fish farms. After that, the breeding eggs were delivered in Klasnik hatchery where the same environmental conditions during embryonic development and cultivation of all five groups of individuals were provided. Spawning of rainbow trout from five investigated groups was conducted from November 21, 2011 to December 13, 2011. As a conditioning factor in this work Fulton's condition factor and coefficients of the heart, liver and spleen were analyzed. In all individuals total length, standard length and body mass were determined. In total, fifty individuals, which were divided into five groups from five brood stocks, were analyzed.

Fulton's condition factor (K) were calculated by using the formula:

$$K = W * 100 / L^3, \text{ where:}$$

W - weight of the fish (g),

L - standard length of the fish (cm) (Akombo et al., 2013).

After that, the liver, spleen and heart were carefully removed and weighed. Organosomatic indices for liver (HSI), heart (CSI) and spleen (SSI) were calculated as follows:

$$\text{OSI} = [\text{weight of the organ (g)} / \text{weight of the fish (g)}] \times 100$$

Statistical analyses were obtained by using SPSS 15, and ANOVA and LSD test for data comparison ($p < 0.05$).

Results and Discussion

Comparing the average values for standard and total body length of rainbow trout from different fish farms, no significant differences were observed. Values for body mass showed significant difference between first and second group ($p=0.027$) and there were significant differences for Fulton's condition factor by comparing fifth with the first and the third group ($p = 0.030$; $p = 0.042$).

In regards with the coefficients of organs, we observed higher differences between groups. There were significant differences for CSI by comparing the fifth group with the second and the third groups ($p = 0.038$; $p = 0.019$), and for HSI, when we compared the first with the second ($p = 0.023$), third ($p = 0.026$), fourth ($p = 0.016$) and fifth groups ($p = 0.008$).

Spleen somatic index showed the highest differences compared in groups; significant differences were noticed between the first and fourth groups ($p = 0.034$), second and third ($p = 0.006$), third and fourth ($p = 0.003$) and between third and fifth group ($p = 0.010$).

Analysing the results of examined parameters, it can be said that the individuals in the first group had the highest values of total length, standard length, body mass, and Fulton's coefficient, whereby it is necessary to emphasize that the individuals of this experimental group were spawned a few days earlier than other groups. In contrast with the morphometric characteristics and condition factor, individuals from the third experimental group had the highest index values of organosomatic indices.

Values of examined parameters of rainbow trout are given in Table 1.

Table 1. Morphometric characteristics, condition factor and organosomatic indices of *Oncorhynchus mykiss* form different brood stocks

STATISTICAL PARAMETERS*		Total length (cm)	Standard length (cm)	Mass (g)	Fulton coefficient	Cardiosomatic index (%)	Hepatosomatic index (%)	Spelecosomatic index (%)
I	Mean	15.47	13.79	50.69	1.91	0.154	1.907	0.181
	SD	1.24	1.16	12.10	0.25	0.031	0.258	0.053
	MIN	13.80	12.20	28.63	1.58	0.119	1.609	0.119
	MAX	17.20	15.10	67.03	2.29	0.213	2.283	0.240
	CV	8.02	8.42	23.88	13.19	20.263	13.541	29.137
II	Mean	14.09	12.40	34.88	1.80	0.158	1.575	0.180
	SD	1.12	1.09	8.03	0.12	0.032	0.352	0.064
	MIN	12.00	10.50	21.23	1.61	0.099	1.052	0.107
	MAX	15.70	13.90	46.55	2.04	0.205	2.416	0.258
	CV	7.93	8.81	23.02	6.52	20.063	22.376	35.339
III	Mean	14.33	12.57	44.14	1.90	0.168	2.498	0.225
	SD	3.08	2.92	28.22	0.25	0.055	1.383	0.098
	MIN	9.90	8.40	10.40	1.44	0.120	1.344	0.133
	MAX	18.60	16.90	89.79	2.32	0.278	5.769	0.397
	CV	21.51	23.22	63.94	13.37	32.882	55.344	43.589
IV	Mean	14.83	13.25	42.36	1.82	0.138	1.555	0.124
	SD	0.95	0.90	7.18	0.18	0.020	0.283	0.025
	MIN	13.20	11.70	31.93	1.60	0.105	1.132	0.092
	MAX	16.30	14.50	52.12	2.10	0.174	2.151	0.174
	CV	6.44	6.77	16.94	9.76	14.236	18.168	20.247
V	Mean	14.49	12.99	38.68	1.72	0.124	1.515	0.138
	SD	1.30	1.20	11.41	0.10	0.021	0.198	0.042
	MIN	11.80	10.40	18.54	1.61	0.084	1.218	0.084
	MAX	16.80	15.20	63.23	1.87	0.151	1.940	0.236
	CV	8.98	9.23	29.50	6.02	17.248	13.066	30.557

Cardio somatic index (CSI) in the examined groups of rainbow trout originate from different brood stock and indicate the existence of significant differences when compared in groups, with the highest values among individuals from the third group. The values of the CSI can indicate various changes in the organism under the influence of abiotic and biotic factors.

It was found that the CSI values get lower in the individuals whose embryos were exposed to higher temperatures as compared to specimen whose embryos were exposed to lower temperatures. Temperature affects the myogenesis process, organelle structure, gene expression, size of distribution of muscle fibers, diameter of muscle fibers and heart development (Johnston, 2006). Stickland et al. (1988) are during experiments on *Salmo salar* and *Salmo trutta* embryos incubated on high temperatures notice significant reduction of muscle fibers than those, which were incubated at low temperatures. Heart abnormalities were observed in cultivated individuals compared to individuals that are sampled from natural habitats (Poppe and Seierstad, 2003).

In addition, values of CSI for *Oncorhynchus mykiss* infected with parasite *Ichthyophonus* were significantly higher. Because of the experiment, it was determined that heart mass and CSI of the infected fish were significantly higher than in individuals from the control group (for approximately 40 %). Increased mass of the heart can be explained with the parasite biomass, infiltration of immune cells and fibrous tissue surrounding the parasite and any of these events may compromise the heart's function, for example, reduction in cardiac capacity (Guyton, 1961).

Similar to the CSI, HSI showed significant differences in our research (between investigated groups), with the largest value in the third group. The liver coefficient (HSI) is a good indicator of the energy status and fish in poor habitat conditions have lower values of this index than fish species that inhabit the food rich habitats. Coefficient of the liver is considerably lower in fish exposed to toxic substances, such as cadmium, and zinc.

HSI of bony fish varied in the range from 1 to 2 %, and it was characteristic for the particular species, but within the same species it depended on the physiological and health status of fish, as well as their condition (Oguri, 1978). The values obtained by this research are in the range from 1 to 2%, with the exception of individuals from the third group, where average value was 2,498%. The liver is an energy reservoir and plays a significant role in the metabolism. The changes in its size and weight are directly related to the stressors that come from the environment. Considering all organosomatic indices, HSI is most often associated with exposure to contamination (Adams and McLean, 1985). HSI values vary with season (Sabrowski and Buchholz, 1996; Beamish et al., 1996), and on its relative size directly affects the quality and quantity of nutrients (Foster et al., 1993). HSI also varies depending on the gender and stage of gonads development (Fabacher and Baumann, 1985; Forlin and Haux, 1990).

Increased values of HSI and liver in general are noticed for *Oncorhynchus mykiss* (Oikari and Nakari, 1982) which were exposed contaminants from oil spills. Anderson et al. (1988) have suggested that the increased HSI is a result of increased production of the endoplasmic reticulum and increasing of protein synthesis under the influence of contaminants. This condition is associated with

hypertrophy of the hepatocytes and hyperplasia due to the presence of contaminants (*Elskus and Stegeman, 1989*). The HSI in our research showed significant differences of the respective groups, and the highest value of this ratio was in the third group.

Spleen has an important hematopoietic function in a vertebrate and because of that SSI is a good indicator of the state of activity of the immune system and the occurrence and intensity of infection and disease. Increased values of SSI indicate a bacterial or parasitic infection, while lower values indicate a complete absence or a mild infection and suggest a good general state of health of fish. The spleen somatic index is used as a reliable diagnostic tool because of its hematopoietic function (*Anderson, 1990*), and its dysfunction can lead to the changes in the level of the entire organism.

Different endogenous and exogenous factors have impact on SSI; it can vary depending on the taxonomic status of the species (*Anderson et al., 1982; Ruklov, 1979*) and within the same population (*Ruklov, 1979*). The relative size of the spleen may vary depending on fish sex, age, size, stage of development of the gonads and the growth (*Ruklov, 1979*). Nonspecific stressors, e.g. hypoxia may affect changes in the morphology of the spleen. Hypoxia is very common in the aquatic environment, and impact of moderate and acute hypoxia was studied with the rainbow trout, *Oncorhynchus mykiss*, which results with the contracted spleen (*Yamamoto, 1987; Randall and Perry, 1992*). In addition, if hypoxia takes a long time, it leads to the increase in number of erythrocytes, which is explained, with increased concentration of erythropoietin. Changes in the size of the spleen may be a sign of dysfunction that affects the general health of the individual. Reduction of the size of the spleen can be in connection with acute nonspecific stressors, as well as with a number of chronic exposures to chemical contaminants, which are responsible for the necrosis and changes in cellular processes (*Yamamoto, 1987; Randall and Perry, 1992*).

Conclusion

All experimental groups of rainbow trout have the same environmental conditions during the embryonic development and growing. Values of total and standard body length did not show any significant differences for all experimental groups but other investigated parameters showed statistically significant differences. Considering that all individuals were under the same conditions and the same treatment, the observed differences can be explained with conditions on the fish farms and different brood stock.

Koeficijent kondicije i organosomatički indeksi dužičaste pastrmke (*Oncorhynchus mykiss*, Wal.) iz različitih matičnih jata

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Rezime

Koeficijent kondicije i organosomatski indeksi riba predstavljaju jedan od načina praćenja uticaja faktora spoljašnje sredine na ribe. Jedinke dužičaste pastrmke (*Oncorhynchus mykiss*, Wal.) koje su korišćene u radu poreklom su od različitih matičnih jata, sa različitih ribogojilišta. Mrest je obavljen na pet izabраниh ribogojilišta, nakon čega je oplodena ikra dopremljena u mrestilište Klašnik gde su obezbeđeni isti uslovi sredine tokom embrionalnog razvoja i gajenja za svih pet grupa jedinki. U radu je kao kondicioni faktor analiziran Fultonov koeficijent uhranjenosti, te koeficijenti srca, jetre i slezine. Kod svih jedinki određene su vrednosti totalne dužine, standardne dužine i mase tela. Ukupno je analizirano 50 jedinki iz pet grupa, odnosno matičnih jata. Rezultati pokazuju postojanje razlika u vrednostima praćenih koeficijenata između pojedinih analiziranih grupa.

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POTENTIALS OF SERBIAN LIVESTOCK PRODUCTION - OUTLOOK AND FUTURE

Milan M. Petrović¹, Stevica Aleksić¹, Milan P. Petrović¹, Milica Petrović², Vlada Pantelić¹, Željko Novaković¹, Dragana Ružić-Muslić¹

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Review paper

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EFFECTS OF REARING SYSTEM AND BODY WEIGHT OF REDBRO BROILERS ON THE FREQUENCY AND SEVERITY OF FOOTPAD DERMATITIS

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ŠKRBIĆ Z., PAVLOVSKI Z., LUKIĆ M. (2007): Uticaj dužine tova u različitim sistemima gajenja na klanične osobine brojlerskih pilića genotipa Redbro. *Biotechnology in Animal Husbandry* 23, 3-4, 67-74.

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PhD Thesis:

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In Scientific Books:

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At Scientific Meetings:

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Institute for Animal Husbandry, Belgrade-Zemun
11th International Symposium
“Modern Trends in Livestock Production”
11th – 13th October 2017, Belgrade, Republic of Serbia



FIRST ANNOUNCEMENT

Institute for Animal Husbandry, Belgrade-Zemun is organizing traditional International Symposium, “Modern Trends in Livestock Production” with following topics:

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 - Genetics
 - Reproduction
 - Breeding
 - Selection
 - Nutrition
2. Production technology and quality of products
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Organizing Committee**



Dr. Milan M. Petrović,
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