UDC636

Print ISSN 1450-9156
Online ISSN 2217-7140

BIOTECHNOLOGY IN ANIMAL HUSBANDRY

CONTENTS

Review paper

Dušica Ostojić Andrić, Slavča Hristov, Radica Đedović, Vlada Pantelić, Dragan Nikšić, Blagoje Dimitrijević, Nataša Tolimir
FARM ANIMAL WELFARE CONCEPT: FROM BEGINNINGS TO INTEGRATION IN MODERN PRODUCTION SYSTEMS…… 269

Vladimir Kurćubić, Zoran Ilić, Radojica Đoković, Milan P. Petrović, Violeta Caro-Petrović
VACCINATION AND MEDICATION AGAINST BOVINE RESPIRATORY DISEASE COMPLEX (BRDC) …….. 279

Original scientific paper

Meysam Latifi, Ali Mohammadi
ANALYSIS OF GENETIC PARAMETERS AND GENETIC TRENDS FOR EARLY GROWTH TRAITS IN IRANIAN AFSHARI SHEEP ……..……. 289

Violeta Caro Petrović, Milan P. Petrović, Dragana Ružić Muslić, Nevena Maksimović, Nikola Delić, Bogdan Cekić, Zorica Bjelić
THE EFFECT OF SERVICE RAM ON REPRODUCTIVE PERFORMANCES AND BIRTH WEIGHT OF LAMBS……..……..… 303

Dragan Nikšić, Vlada Pantelić, Dušica Ostojić-Andrić, Predrag Perišić, Marina Lazarević, Ivan Ćosić, Maja Petričević
VARIABILITY OF FUNDAMENT TRAITS IN PRIMIPAROUS SIMMENTAL HEIFERS ……... 313

Nadya Mincheva, Mitko Lalev, Magdalena Oblakova, Pavlina Hristakieva
REGRESSION MODELS FOR ESTIMATING CHICK HATCHLING WEIGHT FROM SOME EGG GEOMETRY TRAITS…… 323

Simeon Rakonjac, Snežana Bogosavljević-Bošković, Zdenka Škrbić, Miloš Lukić, Vladimir Dosković, Veselin Petričević, Milun D.Petrović
QUALITY AND CHEMICAL COMPOSITION OF EGGS AFFECTED BY REARING SYSTEM AND HEN'S AGE ……...……. 335

Violeta Mandić, Zorica Bijelić, Vesna Krnjaja, Aleksandar Simić, Maja Petričević, Nenad Mićić, Violeta Caro Petrović
EFFECT OF HARVESTING TIME ON FORAGE YIELD AND QUALITY OF MAIZE…… 345

Tatyana Bozhanska, Boryana Churkova, Teodora Mihova
BASIC CHEMICAL COMPOSITION AND ENERGY NUTRITIONAL VALUE OF FODDER BIOMASS FROM ARTIFICIAL ECOSYSTEMS ……. 355

Retraction note…… 369

VOL 34, 3
Founder and publisher
INSTITUTE FOR ANIMAL HUSBANDRY
11080 Belgrade-Zemun
Belgrade 2018

Journal for the Improvement of Animal Husbandry

UDC636

Print ISSN 1450-9156
Online ISSN 2217-7140

BIOTECHNOLOGY IN ANIMAL HUSBANDRY

Belgrade - Zemun 2018

Biotechnology in Animal Husbandry 34 (3), p 269-369, 2018 ISSN 1450-9156
Publisher: Institute for Animal Husbandry, Belgrade-Zemun UDC 636

Address of the Editor's office
Institute for Animal Husbandry, Autoput 16, P. Box 23, 11080 Belgrade-Zemun, Republic of Serbia Tel. 381 11 2691 611, 2670 121; Fax 381 11 2670 164;
e-mail: biotechnology.izs@gmail.com; www.istocar.bg.ac.rs

Biotechnology in Animal Husbandry is covered by Agricultural Information Services (AGRIS) -Bibliographic coverage of abstracts; Electronic Journal Access Project by Colorado Altiance Research Libraries -Colorado, Denver; USA; Matica Srpska Library -Referal Center; National Library of Serbia; University Library "Svetozar Markovic", Belgrade, Serbia; EBSCO, USA; DOAJ and European Libraries
According to CEON bibliometrical analysis citation in SCI index 212, in ISI 9, impact factor (2 and 5) of journal in 2012: 0,667 and 0,467, - M51 category

Annual subscription: for individuals -500 RSD, for organizations 1200 RSD, -foreign subscriptions 20 EUR. Bank account Institut za stočarstvo, Beograd-Zemun 105-1073-11 Aik banka Niš Filijala Beograd.

Journal is published in four issues annually, circulation 100 copies.

The publication of this journal is sponsored by the Ministry of Education and Science of the Republic of Serbia.
Printed: "Mladost birošped", Novi Beograd, St. Bulevar AVNOJ-a 12, tel. 381 11 2601-506

Biotechnology in Animal Husbandry 34 (3), p 269-277 , 2018 ISSN 1450-9156
Publisher: Institute for Animal Husbandry, Belgrade-Zemun UDC 636.083
https://doi.org/10.2298/BAH1803269O

FARM ANIMAL WELFARE CONCEPT: FROM BEGINNINGS TO INTEGRATION IN MODERN PRODUCTION SYSTEMS

Dušica Ostojić Andrić[1], Slavča Hristov[2], Radica Đedović[2], Vlada Pantelić[1], Dragan Nikšić[1], Blagoje Dimitrijević[3], Nataša Tolimir[4]

[1]Institute for Animal Husbandry, Zemun-Belgrade, Republic of Serbia
[2]Faculty of Agriculture, University of Belgrade, Zemun-Belgrade, Republic of Serbia
[3]Faculty of Veterinary Medicine, University of Belgrade, Belgrade, Republic of Serbia
[4]Institute for the Application of Science in Agriculture, Belgrade, Republic of Serbia
*Corresponding author: andricdusica.iah@gmail.com
Review paper

Abstract: The farm animal welfare science has undergone a thorny path of development, often disputed because of its lack of measurability and the purpose of existence. At the very beginning, primarily based on moral and ethical attitudes, over time it pointed to the importance of meeting the needs of animals and the consequences of their neglect and exhaustion in the conditions of intensive livestock production. An important segment of its development was the definition and development of methodologies for the assessment of welfare indicators, which made it measurable and accepted as a scientific discipline with the knowledge applicable and useful in modern production systems. This paper is a concise review of the evolution of the animal welfare science, but also an indication of its future in the context of the development of "symbiotic" connections with the concepts of sustainable agriculture and food safety as integral parts of the modern ecological movement arose from a unified concern for the welfare of people and animals, a care for planetary welfare in general.

Key words: farm animal welfare, development, importance, legislation, production systems, achievements

Introduction

Moral origin and subject of animal welfare

Providing animal welfare has always been important, but people's knowledge of it has changed over time, especially since recently. The attitude of people about what is and what is not moral probable has changed to a lesser degree

during the millennium (*Broom, 2011*). According to utilitarianism, it is morally acceptable for humans to use animals for the production of food, work, entertainment and company, for the production of natural fibres and clothing and footwear materials and for scientific purposes in a way that does not cause animal suffering. This view is contrary to the attitudes of the advocates for animal rights, who consider it ethically unacceptable for humans to exploit animals as their property (*Vučinić, 2006).* Many animal welfare discussions are based on what people do or should do about it. Also, philosophers and the public are often concerned with the issue of killing animals for human consumption and clothing as well as for research purposes *(Fraser, 2008*). These are important ethical issues, but according to *Broom (2011),* not the essential issues of animal welfare. The subject of welfare is what happens to animals before they die, including their treatment just before death, often before the slaughter itself or the way they are killed. However, as *Haynes (2008)* points out, with this view there is a danger of neglecting or inadequately considering the ethical question: is it or is it not acceptable to kill animals? Therefore, scientific research on the welfare of animals should not be based solely on ethics, however scientific knowledge cannot be gained without understanding ethical attitudes.

The significance of welfare

Caring for the welfare of farm animals contributes not only to animals, but also to people while at the same time generating benefits for the environmental protection. Improving the welfare of farm animals increases their productivity (*McInerney, 1998*). This is to a lesser extent applicable in developed countries where land prices and labour costs are high, so the food production in conditions of welfare consideration is also expensive. However, many developing countries have extensive agriculture and low labour costs that give them an edge in the agricultural market, especially if they are oriented towards highly valued organic foods and the market for products deriving from production based on respect of basic welfare principles, the so called *animal-welfare market*. This is of particular importance given that in the developed countries, the demand for these types of products is growing, along with the development of consumer awareness of the welfare of farm animals, as well as the concern about the quality of food consumed on a daily basis (*Wandel and Bugge, 1997; Blokhuis et al., 2003; Ostojić Andrić et al., 2012*). A modern consumer defines the notion of food quality not only through its edible quality and hygienic correctness, but also through the state of welfare of the animal from which it was derived. In this way, animal welfare has become an important part of the general concept of food quality (*Blokhuis, 2007*). Numerous studies (*Gregory, 1993; Scanga et al., 1998; Lindenlauf et al., 2010*) have shown that the animal farming in inadequate conditions undoubtedly negatively affects the quality of the obtained foods and on human health. Animal products can have reduced nutritional and edible quality due to neuro-hormonal reactions resulting

from exposure of animals to acute or chronic stress (*Moberg and Mench, 2000*). The health status of animals as an important indicator of their well-being is also influencing the food quality *(Ostojić Andrić et al., 2015, 2016)*. Studies of the relationship of well-being and quality of milk (*Bergamo et al., 2003; Butler et al., 2011*) show that milk of cows reared on pasture contains a higher share of omega-3 fatty acids, as well as a more favourable omega-6 and omega-3 fatty acids ratio, higher vitamin E and beta-carotene content.

The history and definitions of welfare

From the 19th century to the sixties of the 20th century, knowledge of the biological functions of animals increased significantly. At the end of this period, scientific discipline such as ethology and neurobiology became accepted in the scientific community. *Ruth Harrison's* book *„Animal Machines“,* published in 1964, indicated that the animal production industry very often treats animals as machines rather than living beings. That is why the British government in 1965 set up a *Brambell committee* to report on this phenomenon. The famous ethologist *Thorp (1965),* a member of the committee, emphasized then that understanding animal biology is important as well as that they have their own biological needs, including the need to display appropriate behaviours, and that in case of their denial, frustrations and problems occur in animals. This view of Brambell's report became known as the concept of "*five freedoms*" - the basis on which all the following welfare concepts were developed.

In the 1960s of the 20th century, the focus of the discussion on this topic was more based on what people should do to protect animals, and much less on the welfare itself. In the seventies and early eighties, the term animal welfare was used, although not defined, and most researchers did not consider it scientific. Significant progress in the development of the concept of animal welfare was done by ethologist and psychologist research on motivational systems (*Miller, 1959; Hinde, 1970; McFarland and Sibly, 1975)*. In the book "Biology of Behavior" by *Broom (1981),* the animals described in it represent as sophisticated decision makers in almost all aspects of their action. This was completely contradictory to the previously very widespread, and later rejected, notion that the animals in their actions were driven exclusively by instincts.

Progress in understanding animal motivations, cognitive abilities and the complexity of social behaviour over the past 30 years has led to an accelerated development of animal welfare studies. A key point of consensus among scientists in this field that animal welfare is measurable and therefore has a scientific character, was achieved in the nineties. However, opinions on what was most important for the welfare were still divided. Veterinarians considered that welfare was adequate if the animal was healthy and, contrary to the ethologists, did not deal with animal feelings. Then, the ethologist Marion Dawkins pointed out that

'welfare' does not mean 'just health' and that an animal with poor welfare may be physically healthy and productive but still suffer from a wide range of negative psychological states like fear, frustration, which was a crucial starting point for this emergent discipline *(Dawkins, 1998)*.

Early attempts to define the welfare of animals represented it as a state of harmony of animals with nature or the environment (*Hughes, 1982*). This is a biologically correct fact and the forerunner of later definitions, but it is not measurable and therefore scientifically useless. As the term welfare was used more and more in science, legislation and discussions, there was a clear need for a more precise definition. Finally, *Broom (1986)* presents the definition that is today most widely accepted: *welfare represents the state of the animal created in response to its attempts to struggle with environmental impacts*. In doing so, to „struggle" means to establish control over mental and physical stability. The welfare can be scientifically measured and varies from very bad to very good. The welfare will be poor if the „struggle" is difficult, or even impossible. There are different strategies starting from behavioural, physiological, immunological, and others managed by the brain. Feelings, such as pain, fear and various forms of satisfaction, can be part of a combat strategy where feelings are a key factor of welfare. The system can function in a satisfactory way when the struggle is successfully completed or unsuccessful - when the animal suffers. Since one or more strategies can be used as a response to a particular environmental challenge, there is also a wide range of welfare indicators that can be assessed.

The concept and methods of welfare assessment

In the formation of a general concept of welfare, adaptation, stress, needs (freedom) of animals and their rights are key issues. The most important issue in terms of providing welfare is certainly a question of animal needs. According to *Broom and Johnson (1993),* this need is that part of the animal's biological basis to provide the appropriate resource or response to certain stimuli from the surrounding environment or its body. The idea of providing five basic needs to animals proposed for the first time in the Brambell report *(Brambell, 1965*) has been developed into the generally accepted concept of animal welfare, the so called "Five Great Freedoms" (freedom from discomfort, hunger and thirst, fear and distress, pain, injury and disease, and freedom to express natural behaviour) *(FAWC, 1979).* This concept applies to all animals whose survival depends on man, and the degree of provision of each of these freedoms can be determined by numerous physical, anatomical, physiological, pathological and ethological indicators *(Ostojić Andrić et al., 2013).* Based on this concept, important methods for the assessment of welfare, including the Animal needs index *(Bartussek, H., 1985),* Welfare Quality Assessment Protocol *(WQAP, 2009),* EFSA Risk

Assessment Method *(EFSA, 2012)* and the 'Life worth living' approach *(Mellor, D.J., 2016)* have been constructed.

The legislation, integration and the future of animal welfare

The legislations in the field of the protection of farm animal welfare are largely present and applied in a large number of countries, enabling a broad ethical and moral framework for public and private action. Within the European Union, a significant number of directives, regulations and strategies for the protection of the welfare of farm animals resulted from the recognition that they have feelings (*European Parliament, 2017*). Through collaboration with science, these national and supranational regulations and strategies aim to propagate and encourage modern production systems to function according to the principles of protection of welfare. Unfortunately, in some of the largest producing countries, such as China, India and Brazil, legislation in this area is very poor or even absent. In our country, a major shift in this area was made by the adoption of the first Law on Animal Welfare in 2009, which drew public attention, but also enabled better supervision and control of the protection of animal welfare in production.

On a global scale, it is essential that the United Nations (UN) with the document from 2016 *(FAO, 2016)* have recognized the welfare of farm animals as one of the key factors necessary for securing the concept of sustainable agriculture that is essential for the survival of humanity. In addition, there is an increasing tendency to scientifically analyse, research and present the interconnected animal and human health interactions (*Gibbs, 2014; Pinillos et al., 2016*). Also, the modern concept of food safety is an integral part of the global ecological modernization of society *(CIWF, 2012)* and is unambiguously related to the protection of the welfare of farm animals.

Priority, but not an easy task in the coming period, will be the design of production systems that will have to ensure an adequate level of animal welfare, the production of quality and safe food according to environmental principles, while at the same time increasing yields and profitability. As part of this "symbiosis," the science of welfare will have to deal with its biggest enemies: intensifying of the production, inadequate farm conditions as well as farm management, intensive selection, climate change and health and animal behaviour disorders that result from it (*Oltenacu and Broom, 2010; Ostojić Andrić et al., 2011, 2016, 2017; Rojas-Downing et al., 2017*).

Conclusion

The animal welfare science over a long period of its constitution succeeded in drawing public attention to the problems of modern animal breeding. Incorporation of the basic principles of welfare, as a result of public and scientific support, is now more and more present in farm animal production. It can be said that the welfare finally found its very important place and justification of existence not only in the ethical and moral sense, but also in the context of an important ally of other sciences in dealing with problems in the sphere of climate change, food safety and human health.

KONCEPT DOBROBITI FARMSKIH ŽIVOTINJA: OD ZAČETAKA DO INTEGRACIJE U SAVREMENE SISTEME PROIZVODNJE

Dušica Ostojić Andrić, Slavča Hristov, Radica Đedović, Vlada Pantelić, Dragan Nikšić, Blagoje Dimitrijević, Nataša Tolimir

Rezime

Nauka o dobrobiti farmskih životinja prošla je trnovit put razvoja, često osporavana zbog svoje nemerljivosti i svrsishodnosti postojanja. U samom začetku prvenstveno zasnovana na moralnim i etičkim stavovima, tokom vremena ukazala je na značaj zadovoljenja potreba životinja i posledice njihovog zanemarivanja i iscrpljivanja u uslovima intenzivne stočarske proizvodnje. Važan segment njenog razvoja bili su definisanje i razvoj metodologija za ocenu indikatora dobrobiti, čime je ona postala merljiva i prihvaćena kao naučna disciplina sa saznanjima primenljivim i korisnim u savremenim sistemima proizvodnje. Ovaj rad predstavlja sažet prikaz evolucije nauke o dobrobiti, ali i nagoveštaj njene budućnosti u smislu razvoja „simbiotičkih“ veza sa konceptima održive poljoprivrede i bezbednosti hrane kao integralnih delova savremenog ekološkog pokreta proisteklog iz objedinjene brige o dobrobiti ljudi i životinja, brige o dobrobiti planete generalno.

Ključne reči: dobrobit farmskih životinja, razvoj, značaj, zakonska regulativa, sistemi proizvodnje, dostignuća

Acknowledgement

This research was funded by the Ministry of Education, Science and Technological Development, Republic of Serbia within project TR-31053.

References

BARTUSSEK H. (1985): Vorschlag für eine Verordnung der Steiermärkischen Landesregierung für den Bereich der Intensivtierhaltung. Der österreichische Freiberufstierarzt, 9, 4-15.
BERGAMO P., FEDELE E., IANNIBELLI L., MARZILLO G. (2003): Fat-soluble vitamin contents and fatty acid composition in organic and conventional dairy products. Food Chemistry, 82, 625-631.
BLOKHUIS H.J, JONES, R.B., GEERS R., MIELE M., VEISSIER, I. (2003): Measuring and monitoring animal welfare: Transparency in the food product quality chain. Animal Welfare, 12, 4, 445-455.
BRAMBELL F.W.R. (1965): Report of the technical committee to enquire the welfare of animals kept under intensive livestock husbandry systems. (Cmnd. 2836) H.M.Stationary Office. London.
BROOM D.M. (1981): Biology of Behaviour: Mechanisms, Functions and Applications. Cambridge University Press, New York, 311.
BROOM D.M. (1986): Indicators of poor welfare. British Veterinary Journal, 142, 524–526.
BROOM D.M. (2011): A History of Animal Welfare Science. Acta Biotheoretica, 59, 121, 121-137.
BROOM D.M. AND JOHNSON K.G. (1993): Approaching questions of stress and welfare. In: Stress and Animal Welfare. Kluwer Academic Publishers, 1-7.
BUTLER G., STERGIADIS S., SEAL C., EYRE M., LEIFERT C. (2011): Fat composition of organic and conventional retail milk in northwest England. Journal of Dairy Science, 94, 24-36.
CIWF(2012): Compassion in World Farming. Food Security and Farm Animal Welfare; CIWF: Godalming, UK, 2012.
DAWKINS M.S. (1998): Evolution and animal welfare. The Quarterly Review of Biology, 73, 305–328.
EFSA PANEL ON ANIMAL HEALTH AND WELFARE - AHAW (2012): Guidance on risk assessment for animal welfare. EFSA Journal. 10, (1), 2513. 1-30
EUROPEAN PARLIAMENT (2017): Animal Welfare in the European Union; Directorate-General for Internal Policies: Brussels, Belgium.
FAO (2016): United Nations High Level Panel on Food Security and Nutrition. Sustainable Agricultural Development forFood Security and Nutrition, including the Role of Livestock; FAO: Rome, Italy.
FAWC (1979): Written reference in the enclosed press notice released by Farm Animal Welfare Council, London, United Kingdom in December 1979. Retrieved August, 2012 from http://www.fawc.org.uk/pdf/fivefreedoms1979.pdf
FRASER D. (2008): Understanding animal welfare: the science in its cultural context. Wiley Blackwell, Chichester.

GIBBS E.P.J. (2014): The evolution of One Health. Veterinary Record, 174, 85–91.
GREGORY N.G. (1993): Welfare and product quality: the need to be humane. In: Safety and Quality of Food from Animals. British Society of Animal Production Occasional Publication 17, 51–56.
HAYNES R.P. (2008) Animal and human health and welfare. A comparative philosophical analysis. Journal of Agriculture Environmental Ethics, 21, 91–97.
HINDE R.A. (1970): Animal behaviour: a synthesis of ethology and comparative psychology, 2nd edn. McGraw Hill, New York.
HUGHES B.O. (1982): The historical and ethical background of animal welfare. In: Uglow J (ed) How well do our animals fare? Proc. 15th annual conference of the reading University Agricultural Club, 1981, 1–9.
LINDENLAUF M.M., DEITTERT C., KÖPKE U. (2010): Assessment of environmental effects, animal welfare and milk quality among organic dairy farms. Livestock Science, 128,1,140-148.
ROJAS-DOWNING M.M., NEJADHASHEMI P.A., HARRIGAN T., WOZNICKI S.A. (2017): Climate change and livestock: Impacts, adaptation, and mitigation, Climate Risk Management, 16, 145-163.
MCFARLAND D.J., SIBLY R.M. (1975): The behavioural final common path. Philosophical Transactions of the Royal Society B., 270, 265–293.
MCINERNEY (1998): The economics of welfare. In Michell & Ewbank eds, Ethics, Welfare, Law and Market Forces: The Veterinary Interface. Universities Federation for Animal Welfare. Wheathampstead.
MELLOR D.J. (2016): Updating animal welfare thinking: Moving beyond the "Five Freedoms" towards a Life Worth Living. Animals, 6, 21.
MILLER N.E. (1959): Liberalization of basic S-R concepts: extensions to conflict behaviour, motivation and social learning. In: Koch S (ed) Psychology: a study of a science, vol II. McGraw Hill, New York.
MOBERG G. AND MENCH J. A. (2000): Biology of Animal Stress: Basic Principles and Implications for Animal Welfare. CABI Publishing. Wallingford, Oxon, GBR., 391.
OLTENACU P.A. AND BROOM D.M. (2010): The impact of genetic selection for increased milk yield on the welfare of dairy cows. Animal Welfare, 19(S), 39-49.
OSTOJIĆ-ANDRIĆ D., HRISTOV S., NOVAKOVIĆ Ž., PANTELIĆ V., PETROVIĆ M.M., ZLATANOVIĆ Z., NIKŠIĆ D. (2011): Dairy Cows Welfare Quality In Loose Vs. Tie Housing System. 3rd International Congress "New perspectives and Challenges of Sustainable Livestock production"Belgrade, Republic of Serbia, 5-7th October 2011. Biotechnology in Animal Husbandry 27, 3, Book 2, 975-984.
OSTOJIĆ ANDRIĆ D., NOVAKOVIĆ Ž., PETROVIĆ M.M., PANTELIĆ V., NIKŠIĆ D., LAZAREVIĆ M., ZLATKOVIĆ Z. (2012): Farm Animals Welfare:

Consumers View And Concern In Serbia. Proceedings of The 1st International Symposium on Animal Science, Belgrade, Republic of Serbia, 8-10th November, Book 1, 435-444.
OSTOJIĆ ANDRIĆ D., HRISTOV S., PETROVIĆ M.M., PANTELIĆ V., ALEKSIĆ S., NIKŠIĆ D., DOKMANOVIĆ M. (2013): Welfare Indicators Of Dairy Cows - Selection And Implementation In Assessment. Proceedings of the 10th International Symposium „Modern Trends in Livestock Production“, Belgrade, Serbia, October 2nd-4th, 66-79.
OSTOJIĆ ANDRIĆ D., ALEKSIĆ S., PETROVIĆ M.M., PANTELIĆ V., STANIŠIĆ N., CARO PETROVIĆ V., NIKŠIĆ D., PETRIČEVIĆ M. (2015): Beef Cattle Welfare - Risks And Assurance. Biotechnology in Animal Husbandry 31 (3), 313-326.
OSTOJIĆ ANDRIĆ D., PETROVIĆ M.M., HRISTOV S., STANOJKOVIĆ A., PANTELIĆ V., NIKŠIĆ D., PETRIČEVIĆ M. (2016): Key Health Issues Affecting Dairy Cows Welfare. Proceedings of The Second International Symposium of Veterinary Medicine - ISVM 2016, June 22-24, Belgrade, Serbia.
OSTOJIĆ ANDRIĆ D., HRISTOV S., PETROVIĆ M.M., PANTELIĆ V., BOJKOVSKI J., NIKŠIĆ D., MIĆIĆ N. (2017): Dairy Cows' Welfare Quality In Different Seasons. International Symposium On Animal Science (ISAS) 2017, 5th - 10th June 2017, Herceg Novi, Montenegro, 302-308.
PINILLOS R.C., APPLEBY M.C., MANTECA, X., SCOTT-PARK F., SMITH C., VELARDE A. (2016): One Welfare—A platform for improving human and animal welfare. Veterinary Record, 179, 412–413.
SCANGA J.A., BELK K.E., TATUM J.D., GRANDIN T., SMITH G.C. (1998): Factors Contributing to the Incidence of Dark Cutting Beef. Journal of Animal science, 76, 2040-2047.
THORPE W.H. (1965): The assessment of pain and distress in animals. Appendix III in report of the technical committee to enquire into the welfare of animals kept under intensive husbandry conditions, F.W.R.Brambell (chairman). H.M.S.O., London.
VUČINIĆ M. (2006): Ponašanje, dobrobit i zaštita životinja. Fakultet veterinarske medicine Beograd.
WANDEL M., BUGGE A. (1997): Environmental concern in consumer evaluation of food quality, Food Quality and Preference, Volume 8, Issue 1, 19-26.
WQAP-WELFARE QUALITY® ASSESMENT PROTOCOL FOR CATTLE (2009): Welfare Quality Consortium, Lelystad, Netherlands, 180.

Received 27 August 2018; accepted for publication 26 September 2018

Biotechnology in Animal Husbandry 34 (3), p 279-287 , 2018 ISSN 1450-9156
Publisher: Institute for Animal Husbandry, Belgrade-Zemun UDC 619 .636
https://doi.org/10.2298/BAH1803279K

VACCINATION AND MEDICATION AGAINST BOVINE RESPIRATORY DISEASE COMPLEX (BRDC)

Vladimir Kurćubić[1], Zoran Ilić[2], Radojica Đoković[1], Milan P. Petrović[3], Violeta Caro-Petrović[3]

[1]University of Kragujevac, Faculty of Agronomy Čačak, Cara Dušana 34, Čačak, Republic of Serbia
[2]University of Prishtina-Zubin Potok, Faculty of Agriculture, Jelene Anzujske bb, Republic of Serbia
[3]Institute for Animal Husbandry, 11080 Belgrade - Zemun, Republic of Serbia
Corresponding author: Vladimir Kurćubić, vkurcubic@kg.ac.rs
Review paper

Abstract: BRDC causes pneumonia of cattle, due to which high morbidity and mortality, decreasing food intake and daily growth, and quality of carcasses is significantly reduced. Preventive application of cattle vaccination and treatment of diseased animals with therapeutic agents, with the aforementioned losses, leads to the fact that BRDC is the most expensive cattle disease worldwide. Given the complex etiology (the influence of more viral and bacterial agents), it is characteristic that if viral agents cause disease, the morbidity level is high and the mortality rate is lower. Cattle affected by bacterial infections have a sporadic morbidity and a higher mortality rate. There are a number of predisposing factors originating from the environment or hosts that have a negative impact on the health status of cattle. So, there are a number of challenges that must be solved in the future, with the aim of developing new technologies that would contribute to increasing animal resistance, eliminating risk factors and reducing exposure to pathogens.

Key words: Bovine respiratory disease complex (BRDC), viruses, bacteria, vaccination, medication

Introduction

Bovine respiratory disease complex (BRDC) continues to be one of the most expensive syndromes in fattening and dairy cattle throughout the world. BRDC leads to huge economic losses due to high morbidity and mortality, weight loss, reduced food utilization, reduced quality of carcasses and extensive measures of prophylaxis and therapy *(Edwards, 2010)*. In the European Union, production losses (excluding livestock deaths) are about 576 million euros annually *(Barrett, 2000)*. The losses on the cattle industry in the US are calculated on US $1 billion, while prevention and treatment costs amounted to over 3 billion US dollars

annually *(Griffin, 2006; Snowder et al., 2007).* The losses in beef production in the US in 2010 (1,055,000 cattle) were estimated at $643 million *(NASS, 2011)*. An estimated 1.9 million animals *(Nicholas, 2011)* are affected by BRDC each year in the UK cattle industry with costs estimated at around £60 million annually *(NADIS, 2007).*

The major viral and bacterial pathogens that they presume are the causes of BRDC have been identified with largely clarified etio-pathogenesis. Numerous predisposing factors bear a high risk of development of BRDC, including environmental (weather and ambient temperature, humidity and dust), host factors (age, sex, race, genetics, immune status) and stressful management practices (transportation, changes in diet, high density of animals, handling and surgeries), as described by *Taylor et al. (2010).* The most common viruses that causes BRDC are bovine respiratory syncytial virus (BRSV), parainfluenza virus type 3 (PI3V), bovine herpes virus type 1 (BHV1) and bovine viral diarrhea virus (BVDV) *(Bednarek et al., 2012).* Bacterial infections causes by *Mannheimia haemolytica*, *Pasteurella multocida*, *Histophilus somni*, *Arcanobacterium pyogenes*, *Streptococcus pneumonie*, *Staphylococcus aureus, Chlamydiales spp.*, *Fusobacterium necrophorum*, *Corynebacterium bovis*, *Streptococcus spp*. and *Micrococcus spp*. *(Taylor et al., 2010).* A larger number of preventative tools (vaccines) and effective medicines (anti-microbial and anti-inflammatory products) have been developed with the potential to enable BRDC control. BRDC is complex, multi-factorial and despite the contemporary tools continues to endanger cattle health, welfare and farm profitability *(Caldow, 2011).*

Vaccination

One of the main ways to combat the emergence of BRDC is the vaccination of cattle against the most common viral and bacterial agents *(Urban-Chmiel and Grooms, 2012).* In the US, are currently available different combinations of vaccines against the viral agents BHV1, BVD, PI3, BRSV and the bacterial pathogens *Mannheimia haemolytica*, *Pasteurella multocida* and *Histophilus somnus*. BVDV is unique because intrauterine infection can result in persistently infection (PI) of cattle. Cattle that were PI are with the symptoms of chronic illness or dying in feedlots *(Loneragan et al., 2005).* In Serbia, determination of the presence of PI in cattle of various ages from a herd with a different purpose (fattening/milk) explored by *Kurćubić et al. (2010).* BVDV Ag was not confirmed in either of the cases, probably that the prevalence of PI cattle was extremely low (0.75-2%), and not examined sera of all animals from the farms on presence of BVDV Ag, whose age allowed this examine. Due to the specificity of viral agents, the attenuated live and killed vaccines can be found on the market. Depending on the type of vaccine, the development of immunity requires a period of one to three weeks, and it may be necessary to apply multiple doses of vaccines

("booster" immunization) to achieve the protective level of the immune response. The time that the initial vaccination will be carried out is variable. If the time of the transport of the cattle to the feedlot is longer than 12 hours, it is recommended that the animals rested a certain time before they are vaccinated. The safest preventive approach is if the calves are vaccinated before they are transported to the feedlots, especially if the vaccination is part of a pre-conditioning program (management strategies which allow creating an immune response at a time when the stress and the presence of infectious agents are minimal). *Richeson et al. (2008)* compared the results of the use of a polyvalent vaccine prepared from modified live viruses (MLV), which was administered upon arrival at the site or postponed after 14 days. They revealed improvement in daily body weight gain at day 0 to 14 and day 0 to 42 in the postponed procedure.

A very illustrative and interesting review of the study of bovine respiratory illness for 26 years (1983-2009) was announced by *Fulton (2009).* The list of vaccines and therapeutic agents in the *Veterinary Pharmaceuticals and Biologicals (1982/1983)* showed several MLV vaccines containing BHV1, BVDV with no genotype noted, and PI3V for parenteral use. Data for intranasal use were very small for MLV BHV1 vaccines. Only two vaccines from killed viruses (KV) were available: one KV vaccine with BHV1, PI3V and BVDV, and a second KV vaccine with BHV1 and PI3V. In that time, no licensed vaccines against BRSV. During 2009 (26 years later) in the *Compendium of Veterinary Products, 11th edition (2008),* lists a much larger number of vaccines. The contemporary vaccines listed in above mentioned *Compendium* reflect the innovation in the identification and the addition of selected strains of viruses. Such representatives are MLV vaccines for injection or intranasal use for BHV1, BVDV1a and BVDV2a, PI3V, and BRSV as well as KV vaccines for these same viruses for injection. Certain vaccine strains from 1983 are now described much more precisely, such as the BVDV1a and BVDV2a identified in many current vaccines, which was simply listed as BVDV in 1983. Although many changes have occurred in the period of 26 years and progress has been made in the research and formulation of vaccines, most vaccine strains of BHV1, BVDV, PI3V are isolates from 35 to over 50 years ago.

In Serbia, the immunogenicity of two (mono and polyvalent) experimentally prepared inactivated vaccines containing BVDV reference and field strains evaluated by *Kurćubić et al. (2011a).* They formed 3 experimental groups: Group I - 10 calves vaccinated twice (days 1 and 28) s/c with 2 mL of inactivated polyvalent vaccine per animal; Group II - 10 calves vaccinated in same manner with inactivated monovalent vaccine; Control group (C) consisted of 9 unvaccinated calves. Blood serums are taken from experimental animals on days 0, 14, 28, 42 and 56 post vaccinations, and examined by VNT. The immune response developed more rapidly and values of geometric mean titer (GMT) for BVDV neutralizing Ab were strongly higher in blood sera of calves from Group II. Second

part of the study *(Kurćubić et al., 2011b)* included evaluation of the immune response to same BVDV vaccine by the iELISA method. The occurrence of a specific Ab against BVDV was first detected on day 42 post immunization in both experimental groups of beef calves and remained steady until day 56. The statistically very significant differences observed between the experimental animals of groups I and II on days 42 and 56 after vaccination suggest considerably higher immunogenicity of the monovalent vaccine.

Bednarek et al. (2012) described the market situation when it comes to different types of vaccines against the infective agents which cause of BRDC. Inactivated vaccines for immunization of cattle against *P. multocida* and septicemic and pneumonic strains of *M. haemolytica* and *H. somni* are available under various commercial names: Hiprabovis pneumos, Pastobov, Bovilis Bovipast. Inactivated or modified live polyvalent vaccines that are used in a number of countries contain antigens such as BRSV, PI3 (Rispoval, Bovilis Bovipast), BVDV (Mucosiffa, Bovilis BVD) and BHV1. Recent research is an antigen of the IBR virus used in the formulation of marker vaccines, whether live vaccines (Rispoval-IBR marker vivum, Hiprabovis-IBR marker live) or inactivated (eg, Ibraxion, Rispoval-IBR-marker inactivatum, Bovilis IBR - marker inactivatum) applied in IBR eradication. The most common is the parenteral vaccine application, but it usually has to be twice as stimulating a quality protective response. Recent vaccine produced (Rispoval 3) have combined live and dead viral components of the four main antigens and is administered intramuscularly. They are also available MLV are administered intranasally (Rispoval RS+PI3 IntraNasal). More research has revealed that the intranasal immunization of calves gives active immunity in very young animals despite the presence of maternal antibodies and generates a significant systemic response and interferon induction *(Stokes, 2006).*

In Serbia, according to the *National Veterinary Medicines Register (2017)*, there are no registered and available vaccinations of domestic producers for the immunization of healthy cattle and calves against respiratory diseases, although they were available in the previous period *(Kurćubić et al., 2014).* The six available registered import vaccines are: Bovilis® IBR marker inac (Intervet International B.V., ATCvet QI02AA03), which contains inactivated BHV1, modified (gE-) strain GK/D; BOVILIS BVD (Intervet International B.V., ATCvet QI01AA01), vaccine which contains inactivated BVDV, strain C 86; BIOBOS RESPI 4 (BIOVETA, A.S., Czech Republic, ATCvet QI02AL**), which contains inactivated BRSV (strain BIO-24), inactivated PI3V (strain BIO-23), inactivated BVDV (strain BIO-25) and inactivated *Mannheimia* (*Pasteurella*) *haemolytica* (strain DSM 5283) serovar 1A; RISPOVAL 3-BRSV-PI3-BVD (ZOETIS BELGIUM S.A., Belgium, ATCvet QI02AH**), vaccine which contains modified live PI3V, strain RLB103, live BRSV, strain 375 and inactivated BVDV type 1, strains 5960 (cp) i 6309 (ncp); RISPOVAL IBR-MARKER VIVUM (ZOETIS

BELGIUM S.A., Belgium, ATCvet QI02AD01), vaccine which contains live BHV1, modified (gE-) strain Difivac; RISPOVAL RS+PI3 INTRANASAL (ZOETIS BELGIUM S.A., Belgium, ATCvet QI02AD07), vaccine which contains live modified PI3V, strain RLB103 and live modified BRSV, strain 375.

Therapeutic agents

The rapid destruction of the pathogenic bacteria of the BRDC, especially the *Pasteurellaceae* family, is the first step in the treatment of this complex syndrome, and prevents the formation of more serious lesions on the lungs. The use of antibiotics must be at an earlier stage after an infection (usually accompanied by fever, loss of appetite and nasal discharge), and certainly before irreversible changes (oral breathing, orthopnea, cyanosis). The most commonly used are antibiotics that have prolonged action and the spread of the antibacterial spectrum: tetracyclines (oxytetracycline), macrolides (florfenicol, tulathromycin, gamythromycin) and fluoroquinolones (enrofloxacin, marbofloxacin, danofloxacin). At the first European Buiatrics Forum in Marseille (2009), three new concepts of antibiotic application in the treatment of cattle affected by BRDC were presented. Single Injection Shot Acting AntiBiotic (SISAAB) primarily represents new formulations of fluoroquinolones (Marbocyl S - marbofloxacin), Baytril One or Enroxil Max (enrofloxacin). In this concept, tetracyclines (Tetradur) with long acting can be used here like single intramuscular injections. Single Injection Long Acting AntiBiotic (SILAAB) implies application of new generations of macrolides: tulathromycin (Draxxin) and gamythromycin (Zactran). Multiple Injection Long Acting AntiBiotic (MILAAB) is a risky concept due to increased potential for resistance to antibiotics.

Mannheimia haemolytica (formerly *Pasteurella haemolytica*) and *Pasteurella multocida* bacterins were available, but they were probably replaced by more modern products in the meantime. The most commonly used antibiotics for BRDC therapy in the period 1982-1983 *(Fulton, 2009)* were erythromycin, penicillin-dihydrostreptomycin, tylosin injectable and oral oxytetracycline and injectable sulfamethazine, while today they are almost forgotten. In the *Compendium of Veterinary Products, 11th edition (2008)* is displayed almost completely new list of drugs available in 2009. Modern antibiotics that are used today include the principal marketed injectable: ceftiofur, oxytetracycline, enrofloxicin, florfenicol, danofloxacin, tilmicosin and tulathromycin. Also listed in the 2008 *Compendium* are injectable tylosin, erythromycin and penicillin. A new drug available in 2009 is the non-steroidal anti-inflammatory product flunixin meglumine. They are also used oral medications such as sulfas, tetracyclines and tylosin.

Kurćubić et al. (2013) revealed that the most common bacteria in fattening cattle sick of BRDC were *P. multocida*, *Aeromonas viridans* and *Corynebacterium*

bovis, pointing to their greater importance in BRDC in Serbian beef cattle in regard to *Mannheimia haemolytica*, predominantly represented worldwide. Sensitivity testing showed that the most efficient antibiotics against *P. multocida* were enrofloxacin and florfenicol (100% of tested *P. multocida* isolates sensitive on both antibiotics). *Jezdimirović et al. (2011)* tested the clinical efficacy of tulathromycin (TU) and florfenicol (FL) in the treatment of bronchopneumonia (BP) caused by *Pasteurella multocida* isolated from diseased six months old Holstein calves. TU proved to be a drug of first choice in the treatment of BP.

In the study researcher found that the percent of morbidity was decreased about two-fold in the case of tulathromycin injection and the percent of occurrence of mortality was reduced to 3.6% and 13.5% for tulathromycin and tilmicosin respectively *(Nickell et al., 2008).* Administration of florfenicol (40mg/kg) in calves after transportation, especially in "high-risk" groups, significantly reduced (more than 35%) BRDC incidences during the first 3 week of feeding *(Booker et al., 2007).*

Anti-inflammatory agents of steroidal and non-steroidal origin are used to control the inflammatory reaction in the lungs of cattle *(Lekeux, 2006).* In BRDC supportive therapies were applied different kinds of corticosteroids: betamethasone, dexamethasone, prednisolone, cortisone, hydrocortisone, flumethasone and trimcinolone. Because of their strong immunosuppressive character were usually use only in a single administration. Nonsteroidal anti-inflammatory drugs are, with some exceptions, analgesic and antipyretic. The drugs most commonly used in BRD therapy in Europe are flunixin meglumine, carprofen, ketoprofen, meloxicam, tolfenamic acid and metamisole.

Conclusions

BRDC is still the most serious health problem in cattle breeding, especially in fattening animals. In order to reduce the economic losses to an acceptable level, it is necessary to reduce stress (caused by various predisposing factors), improve the quality of vaccines and design the most optimal programs for vaccination and application of therapeutic agents (metaphylaxis strategy), as well as to better understand the impact of genetics on resistance against BRDC.

Vakcinacija i terapija protiv kompleksa respiratornog oboljenja goveda (BRDC)

Vladimir Kurćubić, Zoran Ilić, Radojica Đoković, Milan P. Petrović, Violeta Caro-Petrović

Rezime

BRDC uzrokuje upalu pluća goveda, usled čega je visok morbiditet i mortalitet, i dolazi do smanjenog unosa hrane i dnevnog prirasta, kao i kvaliteta govođih trupova. Preventivna primena vakcinacije goveda i lečenje terapeutskim agensima obolelih goveda, uz prethodno navedene gubitke, dovode do činjenice da je BRDC najskuplje oboljenje goveda širom sveta. Obzirom na kompleksnu etiologiju (uticaj većeg broja virusnih i bakterijskih uzročnika), karakteristično je da ukoliko virusni uzročnici izazivaju oboljenje, nivo morbiditeta je visok a stopa mortaliteta niža. Goveda zahvaćena bakterijskim infekcijama imaju sporadičan morbiditet i višu stopu mortaliteta. Postoji niz predisponirajućih faktora poreklom iz okoline ili domaćina koji imaju negativan uticaj na zdravstveni status goveda. Dakle, postoji niz izazova koji u budućnosti moraju biti rešeni, sa ciljem da se razviju nove tehnologije koje bi doprinele povećanju otpornosti životinja, otklanjanju faktora rizika i smanjenju izloženosti patogenima.

Ključne reči: kompleks respiratornog oboljenja goveda (BRDC), virusi, bakterije, vakcinacija, lečenje

Acknowledgment

This review research was financed by the Ministry of Education, Science and Technological Development of Republic of Serbia, project TR 31001.

References

BARRETT D.C. (2000): The calf pneumonia complex - treatment decisions. Cattle Practise, 8, 135-138.

BEDNAREK D., SZYMAŃSKA-CZERWIŃSKA M., DUDEK K. (2012): Bovine Respiratory Syndrome (BRD) Etiopathogenesis, Diagnosis and Control. In: A Bird's-Eye View of Veterinary Medicine. Ed. Carlos C. Perez-Marin, InTech, 363-373.

BOOKER C.W., ABUTARBUSH S.M., SCHUNICHT O.C., JIM G.K., PERRETT T., WILDMAN B.K., GUICHON P.T., PITTMAN T.J., JONES C., POLLOCK C.M. (2007): Evaluation of the efficacy of tulathromycin as a metaphylactic antimicrobial in feedlot calves. Veterinary Therapeutics: Research in Applied Veterinary Medicine, 8, 3, 183-200.

CALDOW G. (2011): Control of the Bovine Respiratory Disease Complex. Cattle Practice, 16, 1, 1-10.

Compendium of Veterinary Products (2008): 11th edition. Port Huron, MI: North American Compendium Ltd, 1-2428.

EDWARDS T.A. (2010): Control methods for bovine respiratory disease for feedlot cattle. Veterinary Clinics of North America-Food Animal Practice, 26, 2, 273-284.
FULTON R.W. (2009): Bovine respiratory disease research (1983-2009). Animal Health Research Reviews 10, 2, 131-139.
GRIFFIN D. (2006): Antibiotic metaphylaxis to control respiratory disease. Cattle Production Library CL-606, 1-6.
KURĆUBIĆ V., ILIĆ Z., DJOKOVIĆ R., JEVTIĆ S., PETROVIĆ T. (2010). Determination of the presence and degree of infection with bovine viral diarrhea virus in cattle herds in central Serbia - possibilities for control. Veterinarski Glasnik, 64, 3-19.
KURĆUBIĆ V., PETROVIĆ T., ĐOKOVIĆ R., ILIĆ Z., PETROVIĆ M.D. (2011b): Antibody response of beef calves to experimental monovalent and multivalent inactivated bovine viral diarrhoea virus vaccines as measured by indirect ELISA method. Biotechnology in Animal Husbandry, 27, 901-991.
KURĆUBIĆ V.S., ĐOKOVIĆ R.D., ILIĆ Z.Ž., STOJKOVIĆ J.S., PETROVIĆ M.P., CARO-PETROVIĆ V. (2014): Modern approach to the enigma of bovine respiratory disease complex: a review. Pakistan Veterinary Journal, 34, 1, 11-17.
KURĆUBIĆ V.S., ĐOKOVIĆ R.D., VIDANOVIĆ D., ŠEKLER M., MATOVIĆ K., ILIĆ Z., STOJKOVIĆ J. (2013): Bovine respiratory disease complex (BRDC): viral and bacterial pathogens in Serbia. Biotechnology in Animal Husbandry, 29, 1, 29-36.
KURĆUBIĆ V.S., MILIĆ N., DJOKOVIĆ R.D., ILIĆ Z.Z. (2011a): Evaluation of immunogenic properties of monovalent and polyvalent inactivated bovine virus diarrhea virus (BVDV) vaccines. African Journal of Microbiology Research, 5, 2422-2427.
LEUKEUX P. (2006): A therapeutic strategy for treatment of the bovine respiratory disease complex: the rationale for the combination of a nonsteroidal antiinflammatry drug with an antibiotic. In: BRDC - setting a new therapeutic standard: a multimodal approach. Proceedings of the XXIV World Buiatrics Congress, 15-19 October, Nice, France, 8-11.
LONERAGAN G.H., THOMSON D.U., MONTGOMERY D.L., MASON G.L., LARSON R.L. (2005): Prevalence, outcome, and health consequences associated with persistent infection with bovine viral diarrhea virus in feedlot cattle. Journal of the American Veterinary Medical Association, 226, 595-601.
NASS (2011): Cattle and calf death losses. http://usda.mannlib.cornell.edu/usda/current/CattDeath/CattDeath-05-12-2011.pdf (accessed June 10, 2015)
National Animal Disease Information Service (NADIS) (2007): Health Bulletin: Respiratory Disease in Cattle. UK, NADIS, 1-3.

National Register of Veterinary Medicinal Products (2017): Medicines and Medical Devices Agency of Serbia. Available at: https://www.alims.gov.rs/ciril/files/2018/01/nrl-vet2017.pdf
NICHOLAS R.A.J. (2011): Bovine Mycoplasmosis: silent and deadly. Veterinary Record, 168, 459-432.
NICKELL J.S., WHITE B.J., LARSON R.L., BLASI D.A., RENTER D.G. (2008): Comparison of short-term health and performance effect related to prophylactic administration of tulathromycin versus tilmicosin in long-hauled highly stressed beef stocker calves. Veterinary Therapeutics: Research in Applied Veterinary Medicine, 9, 147-156.
RICHESON J.T., BECK P.A., GADBERRY M.S., GUNTER S.A., HESS T.W., HUBBELL D.S., JONES C. (2008): Effect of an-arrival versus delayed modified live virus vaccination on health, performance, and serum infectious bovine Rhinotrachitis titers of newly receive beef calves. Journal of Animal Science, 86, 999-1005.
SNOWDER G.D., VAN VLECK L.D., CUNDIFF L.V., BENNETT G.L., KOOHMARAIE M., DIKEMAN M.E. (2007): Bovine respiratory disease in feedlot cattle: Phenotypic, environmental, and genetic correlations with growth, carcass, and longissimus muscle palatability traits. Journal of Animal Science, 85, 1885-1892.
STOKES CH. (2006): Intranasal vaccination against respiratory disease in animals: a review. In: Intranasal BRSV and PI3 vaccination: innovation for the prevention of respiratory disease in cattle. Proceedings of the XXIV World Buiatrics Congress, 15-19 October, Nice, France, 7-9.
TAYLOR J.D., FULTON R.W., LEHENBAUER T.W., STEP D.L., CONFER A.W. (2010): The epidemiology of bovine respiratory disease: What is the evidence for predisposing factors? Canadian Veterinary Journal, 51, 1095-1102.
URBAN-CHMIEL R., GROOMS D.L. (2012): Prevention and Control of Bovine Respiratory Disease. Journal of Livestock Science, 3, 27-36.
Veterinary Pharmaceuticals and Biologicals 1982/1983 (1983): Aronson CE (ed.). 1/1-A/64. Veterinary Medicine Publishing Co.

Received 6 June 2018; accepted for publication 14 August 2018

Biotechnology in Animal Husbandry 34 (3), p 289-301, 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 575.17'636.38(5)
https://doi.org/10.2298/BAH1803289L

ANALYSIS OF GENETIC PARAMETERS AND GENETIC TRENDS FOR EARLY GROWTH TRAITS IN IRANIAN AFSHARI SHEEP

Meysam Latifi[1], Ali Mohammadi*[2]

Faculty of Agriculture, University Of Kurdistan, Kurdistan, Iran.
Faculty of Agriculture, University of Tabriz, Tabriz, Iran.
*Corresponding author: alimohamadi36@gmail.com
Original scientific paper

Abstract: The purpose of the present study was estimation of genetic parameters and genetic trends of early growth traits using Bayesian approach by Gibbs3f90 software in Iranian Afshari sheep. The data set [birth weight (BW), weaning weight (WW) and pre-weaning daily weight gain (PWDG)] were collected during the period 1999 to 2010 from Agriculture Jahad of Zanjan province, Iran. The fitted fixed effects were herd–year–season as interactions, sex (male, female), birth type (single, multiple) and age of dam. Based on Derivative Information Criteria (DIC), for studied traits the most appropriate model was determined. Therefore, based on the most appropriate fitted model, the direct additive heritabilities estimate for BW, WW and PWDG were 0.32±0.02, 0.05±0.01 and 0.24±0.02, respectively. The estimates of maternal heritabilities were 0.17±0.04, 0.07±0.02 and 0.12±0.05 and total heritabilities 0.11±0.05, 0.08±0.02 and 0.08±0.03 for BW, WW and PWDG, respectively. Direct genetic trends were positive for all traits but only significant for BW 0.75±0.31 g/year (P <0.05). Also, maternal genetic trends were for all traits negative and was significant for BW -0.63±0.27 g/year (P <0.05). The moderate estimates of heritabilities for early growth traits indicate that in Afshari sheep faster genetic improvement through selection is possible for these traits. Furthermore, the results genetic trends in this current study indicated that genetic improvement through selection is suitable only for BW in Afshari sheep.

Keywords: Afshari sheep, early growth, heritability, genetic trend

Introduction

An advantage of a breeding program can be assessed by change in breeding value expressed as a proportion of expected theoretical change of the mean breeding value for the trait under selection (*Mokhtari and Rashidi, 2010*).

Sheep meat production is one of the most important parts of Iranian livestock industry. However, improving growth performance by selection programs is an important method to increase meat production in lamb breeding herds (*Gholizadeh and Ghafouri-Kesbi, 2015*). In this regard, one of the heavy, fat-tail, meat and high lamb growth rate breeds in Iran is Afshari sheep. This sheep has high potentials of growth and reproduction and adapt to cold climate and must be considered in breeding program (*Ghavi Hossein-Zadeh, 2012; Mohammadi et al., 2009*). Afshari breed is distributed in the origins of harsh environmental condition Zanjan province and in some parts of Eastern and Western Azarbayjan and Kordistan of Iran. The purpose of Afshari sheep breeding were selection for increasing body weight and phenotypic characteristics. However, in Afshari sheep faster genetic improvement through selection is possible for body weight (*Eskandarinasab et al., 2010*).

Body size varies between 45 and 57 kg in adult ewes and frequency of ewes lambing is 65 to 75 percent *(Mohammadi et al., 2009*). The birth weight (BW) of Afshari sheep breed were reported 4.3 and 4 kg, weaning weight (WW) 29.6 and 26.5 kg for male and female lambs, respectively *(Monem et al., 2005*). Estimates of genetic parameters of various traits of Iranian Afshari sheep have been reported in different studies, such as: Litter size *(Mohammadi et al., 2011*), direct heritability of BW, and post average daily gain 0.231 and 0.07, respectively (*Khorsand et al., 2014*). Also, direct heritability of kleiber ratio at weaning and kleiber ratio at 6 months of age 0.13 and 0.06, respectively reported by *Eskandarinasab et al. (2010).* Moreover, in literature several were used for growth traits in others Iranian breeds of sheep. Such as; Mehraban sheep (*Zahamin and Mohammadi, 2008*); Makuei sheep (*Ghafouri-Kesbi and Baneh, 2012*); Baluchi sheep (*Gholizadeh and Ghafouri-Kesbi, 2015*); Kermani sheep (*Bahreini Behzadi et al., 2005*).

The main objective of the current study was to estimate the genetic parameters and genetic trends for early growth traits in Afshari sheep, by fitting different six models, including direct and maternal additive genetics and maternal permanent environmental effects.

Materials and methods

Data recording and management

The Afshari sheep is mostly local breed and kept on extensive production. The Jahad of Agriculture in Iran put in place a breeding program to enhance the efficiency of output by improving growth traits, wool production and milk yield that are economically important (*Mokhtari and Rashidi, 2010*). The data set include birth weight (BW), weaning weight (WW) and pre-weaning daily weight gain (PWDG) records available of Afshari sheep. The records were retrieved of 6912 lambs that descended from 3116 dams and 201 sires were collected between 1999 and 2010 from Agriculture Jahad of Zanjan province, Iran. The descriptive statistics of the data structure are summarized in Table 1.

Table 1. The characteristics of the data structure for early growth traits of Afshari sheep

Item	Traits		
	BW (g)	WW (g)	PWDG (g)
No. of records	6912	4511	4511
Average weight	3.81	25.15	213.80
No. of sires with record	201	159	159
No. of dams with record	3116	2555	2555
Standard deviation (SD)	0.90	4.63	59.39
Coefficient of variation, % (CV)	23.75	18.42	27.78

BW: birth weight; WW: weaning weight; PWDG: pre-weaning daily weight gain.

Statistical analysis

The GLM procedure of SAS software was applied to identify the fixed effects have significant influences on traits (*SAS, 2004*). The fitted fixed effects were herd–year–season as interactions, sex (male, female), birth type (single, multiple) and age of dam (2-7 years old). The variance components were estimated with six different univariate animal models as follows: (1)

$$\boldsymbol{Model\ 1}: \boldsymbol{y = Xb + Z_1a + e}$$
$$\boldsymbol{Model\ 2}: \boldsymbol{y = Xb + Z_1a + Wc + e}$$
$$\boldsymbol{Model\ 3}: \boldsymbol{y = Xb + Z_1a + Z_2m + e} \qquad \boldsymbol{Cov\,(a,m) = 0}$$
$$\boldsymbol{Model\ 4}: \boldsymbol{y = Xb + Z_1a + Z_2m + e} \qquad \boldsymbol{Cov\,(a,m) = A\sigma_{am}}$$
$$\boldsymbol{Model\ 5}: \boldsymbol{y = Xb + Z_1a + Z_2m + Wc + e} \qquad \boldsymbol{Cov\,(a,m) = 0}$$
$$\boldsymbol{Model\ 6}: \boldsymbol{y = Xb + Z_1a + Z_2m + Wc + e} \qquad \boldsymbol{Cov\,(a,m) = A\sigma_{am}}$$

Where $\boldsymbol{y}$; is the vector of observations; $\boldsymbol{b}$, is vector of fixed effects; $\boldsymbol{a}$ and $\boldsymbol{m}$ are vectors of random direct and maternal additive genetic effects; $\boldsymbol{c}$, is the vector of random maternal permanent environmental effects; $\boldsymbol{e}$ is vector of residual effects and $\boldsymbol{X}$, $\boldsymbol{Z_1}$, $\boldsymbol{Z_2}$ and $\boldsymbol{W}$ are incidence matrices for $\boldsymbol{b}$, $\boldsymbol{a}$, $\boldsymbol{m}$ and $\boldsymbol{c}$, respectively. $\boldsymbol{Cov\,(a,m)}$; indicates whether covariance between direct and maternal additive genetic effects was considered. Generally, the (co)variance structure for studied traits will be as follows:

(2)

$$var\begin{bmatrix} a \\ m \\ pe \\ e \end{bmatrix} = \begin{bmatrix} A\sigma_a^2 & A\sigma_{am} & 0 & 0 \\ A\sigma_{am} & A\sigma_m^2 & 0 & 0 \\ 0 & 0 & I\sigma_{pe}^2 & 0 \\ 0 & 0 & 0 & I\sigma_e^2 \end{bmatrix}$$

Where, **A** is the additive numerator relationship matrix coefficients among animals; σ_a^2, σ_m^2, σ_{pe}^2 and σ_e^2 are direct additive genetic, maternal additive genetic, maternal permanent environmental and residual variances, respectively, $\boldsymbol{\sigma_{am}}$ is the covariance between direct and maternal effects and **I** is the identity matrix. The (co)variance components were estimated using Gibbs sampling methodology using Bayesian approach by Gibbs3f90 software (*Mistzal 2002*). The number of samples, length of burn-in and sampling interval were 300000, 30000 and 100, respectively.

Estimates of total heritability (h_t^2) are of value to predict phenotypic response to selection. Total heritability estimates were calculated according to *Willham, (1972)* as: (3)

$$h_t^2 = \frac{\sigma_a^2 + 0.5\sigma_m^2 + 1.5\sigma_{a,m}}{\sigma_p^2}$$

1. Model comparison criteria

Goodness of fit for the models was examined using Deviance Information Criterion (DIC), as follows: (4)

$$DIC = 2 \times \overline{D(\theta)} - D(\bar{\theta})$$

Where $\overline{D(\theta)}$ is the posterior expectation of Log-likelihood and $D(\bar{\theta})$ is Log-likelihood evaluated at the posterior mean of the parameters.

Results and Discussion

Among traits studied, PWDG (27.78) had the maximum coefficient of variation (CV %). Standard deviation (SD) for BW, WW and PWDG in this current study, were observed 0.90, 4.63 and 59.39, respectively (Table 1). Maximum growth rate observed in the PWDG, Similarly, *Prince et al. (2010)* in Avikalin sheep and *Ghafouri-Kesbi et al. (2011)* in Zandi sheep. The reason of less CV % for WW and BW may be due to effect of outside environmental in these traits. Also, due to that during the final months of the maintenance and in the winter, the ewes are fed inside. These results are in agreement with those reported by *Miraei Ashtiani et al. (2007)*; *Boujenane; Diallo (2017).*

The least square means and standard errors for fixed effect of studied traits are presented in Table 2. Herd-year-season as interactions significantly affected all studied traits (P <0.001). Gender and birth type had a significant effect on BW,

WW and PWDG traits (P <0.001). All traits were significantly influenced by age of dam (P <0.001). These fixed effects also were shown to be significant in previous studies, such as: Abbasi et al. (2012), in Baluchi sheep; *Boujenane; Diallo (2017)*, Sardi sheep; *Eskandarinasab et al. (2010)*, Afshari sheep; *Jafari and Razzagzadeh (2016)*, Makuie sheep. In this research, six model different were compared for better fitting performance of WW, BW and PWDG traits in Afshari sheep. These results comparison of the models are in agreement with those reported by *Khorsand et al. (2014); Ghafouri-Kesbi and Baneh (2012).*

Table 2. Number of observations and least square means ±SE of early growth traits

Fixed effects	Traits		
	BW	**WW**	**PWDG**
Herd*Year*season	**	**	**
Gender	**	**	**
Male	3.61 ± 0.02^{a}	25.31 ± 0.11^{a}	211.57 ± 1.48^{a}
Female	3.43 ± 0.02^{b}	24.73 ± 0.09^{b}	207.25 ± 1.42^{b}
Birth type	**	**	**
single	3.87 ± 0.01^{a}	25.62 ± 0.10^{a}	214.26 ± 1.21^{a}
twin	3.17 ± 0.02^{b}	24.42 ± 0.16^{b}	204.54 ± 1.86^{b}
Age of dam (Year)	**	**	**
2	3.44 ± 0.02^{a}	24.62 ± 0.16^{c}	204.22 ± 1.88^{b}
3	3.51 ± 0.02^{a}	25.14 ± 0.16^{a}	209.21 ± 1.85^{ab}
4	3.54 ± 0.01^{a}	25.40 ± 0.15^{a}	211.28 ± 1.83^{a}
5	3.57 ± 0.03^{a}	25.33 ± 0.17^{ab}	212.03 ± 1.90^{a}
6	3.56 ± 0.02^{a}	25.01 ± 0.18^{abc}	210.64 ± 2.15^{ab}
7	3.52 ± 0.02^{a}	24.64 ± 0.22^{bc}	209.04 ± 2.66^{ab}

*****: Significant at P < 0.05.
******: Significant at P < 0.001.
A same letter in each column means not-significant differences of least square means in letters are not significant at
P < 0.05.

Selection of a best model depends partly on the DIC that fitted on the data. For analyze BW, WW and PWDG, 4th, 3rd and 6th models had the lowest DIC values and were selected the most appropriate model (Table 3).

Table 3. Deviance information criteria (DIC) values for early growth traits under six models (The most appropriate model in bold)

Model	Traits		
	BW	**WW**	**PWDG**
1	11225.26	24640.54	46927.05
2	11222.84	24491.91	46635.49
3	10401.10	**24366.13**	46924.60
4	**9416.53**	24676.14	46266.31
5	11378.09	24401.93	46534.08
6	10217.70	24492.76	**41249.34**

Estimates of (co)variance components, based on the most appropriate model for each trait are shown in Table 4. The direct additive genetic variance was highest at PWDG (534.10) and lowest in BW (0.11). The direct additive heritabilities (h_a^2) estimate for BW, WW and PWDG were 0.32±0.02, 0.05±0.01 and 0.24±0.02, respectively. An estimate of direct additive heritability for BW was in agreement with reported by *Boujenane and Diallo (2017)* in Sardi sheep; *Abbasi et al. (2012)* in Baluchi sheep and *Eskandarinasab et al. (2010)*, in Afshari sheep; *Jafaroghli et al. (2010)* in Moghani sheep. Whereas, finding by *Gizaw et al. (2007)*, in Menz sheep (0.46) for direct additive heritability were higher than this results present study in the case BW trait. Various estimates of the WW trait have been reported at other Iranian sheep's, such as: Mehraban sheep 0.22 (*Zamani and Mohammadi, 2008*); Makooei sheep 0.28 (*Ghafouri-Kesbi and Baneh. 2012*); Kermani sheep 0.19 (*Bahreini Behzadi et al., 2005*); Moghani sheep 0.07 (*Jafaroghli et al., 2010*). Different values reported by this study can be due to different breeds, fitted statistical models, data structure and different environmental conditions. The estimates of direct additive heritability for PWDG were also similar to range of published estimates, as *Zamani and Mohammadi (2008)* and *Khorsand et al. (2014).*

Furthermore, the estimates of maternal heritabilities of BW, WW and PWDG were 0.17±0.04, 0.07±0.02 and 0.12±0.05, respectively. Based on obtained results, magnitude of maternal heritabilities declined from birth to weaning. The regarding to results available in this study, maternal heritabilities were in accordance with other reports (*Jafaroghli et al., 2010; Mohammadi et al., 2015*; for BW and WW 0.18 and 0.06; 0.11 and 0.03, respectively). Estimated maternal permanent environmental effect (c^2) were 0.08±0.02 for PWDG by best model for this trait. Maternal permanent environmental effect was estimated to be lower than direct and maternal heritabilities for PWDG trait by best model. The maternal permanent environmental effect estimates for PWDG agreeing with the finding presented by *Zamani and Mohammadi (2008)* and *Zamani and Almasi (2017)* in Mehraban sheep, *Gholizadeh and Ghafouri-Kesbi (2015)* in Baluchi sheep.

Table 4. Estimates of (co)variance components and genetic parameter estimates for early growth traits

Trait	σ_a^2	σ_m^2	σ_{pe}^2	σ_e^2	σ_p^2	σ_{am}	h_a^2 ± SE	h_m^2 ± SE	c^2	r_{am}	h_T^2 ± SE
BW	0.11	0.06	-	0.18	0.36	-0.07	0.32±0.02	0.17±0.04	-	-0.76	0.11±0.05
WW	0.59	0.94	-	11.65	13.18	-	0.05±0.01	0.07±0.02	-	-	0.08±0.02
PWDG	534.10	252.23	165.34	1235.20	2186.87	319.40	0.24±0.02	0.12±0.05	0.08±0.02	-0.77	0.08±0.03

σ_a^2: additive genetic variance, σ_m^2: maternal additive genetic variance, σ_{pe}^2: maternal permanent environmental variance, σ_e^2: residual variance, σ_p^2: phenotypic variance, σ_{am}: direct-maternal genetic covariance, respectively; h_a^2: direct additive heritability, h_m^2: maternal heritability, c^2: maternal permanent environmental effect, r_{am}: correlation between direct and maternal genetic effects and h_T^2: total heritability

The fitting the direct and maternal genetic covariance resulted to estimates of correlation between direct and maternal genetic effects (r_{am}) of -0.76 and -0.77 for WW and PWDG, respectively. *Boujenane and Diallo (2017)* in Sardi sheep reported that correlation between direct and maternal genetic effects -0.67 and -0.1 for BW and WW, respectively. That is close to our results in this regard, which estimates may be attributed to the data structure and due to natural selection for an intermediate optimum (*Tosh and Kemp, 1994*).

The estimates of total heritabilities for BW, WW and PWDG were obtained 0.11±0.05, 0.08±0.02 and 0.08±0.03, respectively. The estimates of direct and maternal annual genetic trends for early growth traits (g/year) in this current study were given in Table 5. Estimates of total heritability for all traits are low, suggesting that mass selection would not be very effective in improving these traits. With regard to published estimates, the total heritability obtained in this current study was comparable with the results of several studies (e.g. *Boujenane and Diallo (2017)* in Sardi sheep*; Zamani and Almasi (2017)* in Mehraban sheep; *Rashidi et al. (2008)* in Kermani sheep). However, these results were inconsistent with the results of some studies (*Ekiz et al., 2004; Mandal et al., 2006*) in different breeds of sheep.

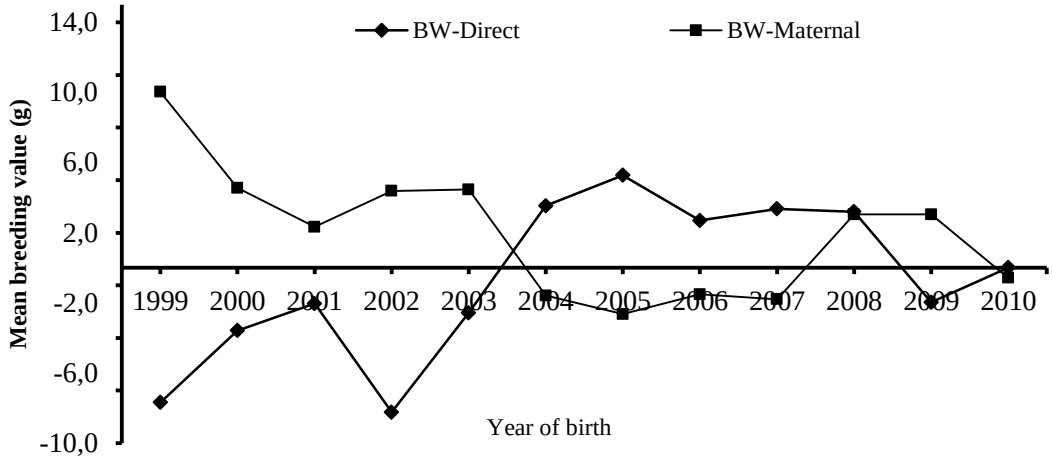
Table 5. Estimates of genetic trends (g/year) for early growth traits of Afshari lambs from 1999 - 2010

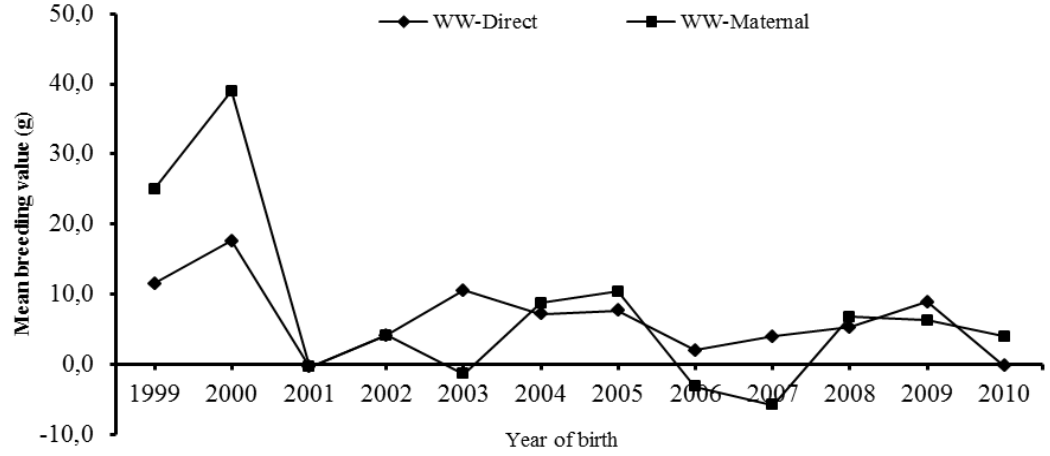
Trait	DGT±SE	R^2	MGT±SE	R^2
BW	0.75^{*}± 0.31	36.36	-0.63^{*}± 0.27	34.87
WW	0.71^{ns}± 0.41	23.42	-1.87^{ns}± 0.94	28.43
PWDG	1.82^{ns}± 17.96	0.01	-5.90^{ns}± 13.35	0.02

DGT, MGT and R^2: Direct genetic trend, Maternal genetic trend and regression fit for genetic trend, respectively.
*: Significant at P < 0.05.
Ns: Not significant.

Positive genetic trend was observed for BW 0.75±0.31 and significant (*P* <0.05), and for WW and PWDG not significant. Generally, plots of the mean predicted breeding values on year of birth (1999-2010) of studied traits for direct and maternal are given in Figure 1. As indicated in Figure 1. for BW-Maternal mean breeding value since the beginning of study period positive were observed (from 1999 year of birth) then sudden drop in 2003 year of birth and again trend increase showed the end (close to zero). BW-Direct mean breeding value at early of period was negative genetic trend and then trend increased. For WW-Direct and maternal mean breeding values at early of period were positive and high and at the end of the period it was low and close to zero. In addition to, for PWDG-Direct and maternal mean breeding value it has been various slopes over the years. Estimate of direct genetic trend for BW obtained in the present research (0.75 g/year) was low and generally agrees with that reported by *Mokhtari and Rashidi (2010)* in

Kermani sheep (2 g/year); *Hanford et al. (2005)* in Rambouillet sheep (0.4 g/year). However, higher estimates for genetic trends of BW in other sheep breeds were reported by several authors (0.23, 20, 25 and 128 g/year were reported by *Shrestha et al. (1996)* in Suffolk, and Finn sheep; *Shaat et al.* (2004) in Ossimi breed, *Rashidi and Akhshi, (2007)* in kurdi sheep, respectively). The direct genetic trends for WW and PWDG were not significant similar to the *Boujenane and Diallo (2017)* in Sardi sheep. Further, maternal genetic trend was negative for all traits this percent study, and only significant for BW (P <0.05). Our results of maternal genetic trend for BW were close to *Boujenane and Diallo (2017)*; in Sardi sheep. A higher value for maternal genetic trend (3 g/year) was reported in Iranian Kermani sheep (*Mokhtari and Rashidi, 2010*). Also, the estimated maternal genetic trend for WW was consistent with estimates of *Boujenane and Diallo (2017)* in Sardi sheep (-0.26 g/year). The low annual genetic trends for studied traits in this study could be attributed mainly to the low heritability estimates that it depends mostly on phenotypic characteristics instead of additive genetic values. Also, due to the low nutritional level and harsh climate condition of Afshari sheep that is not very favorable for the expression of sheep's genetic potential. These results were similar to those reported by *Boujenane and Diallo (2017), Mokhtari and Rashidi (2010)*. The obtained results of the mean breeding values during the year of birth (Maternal and direct) of BW, WW and PWDG were consistent with the results by *Boujenane and Diallo (2017), Mokhtari and Rashidi (2010).*





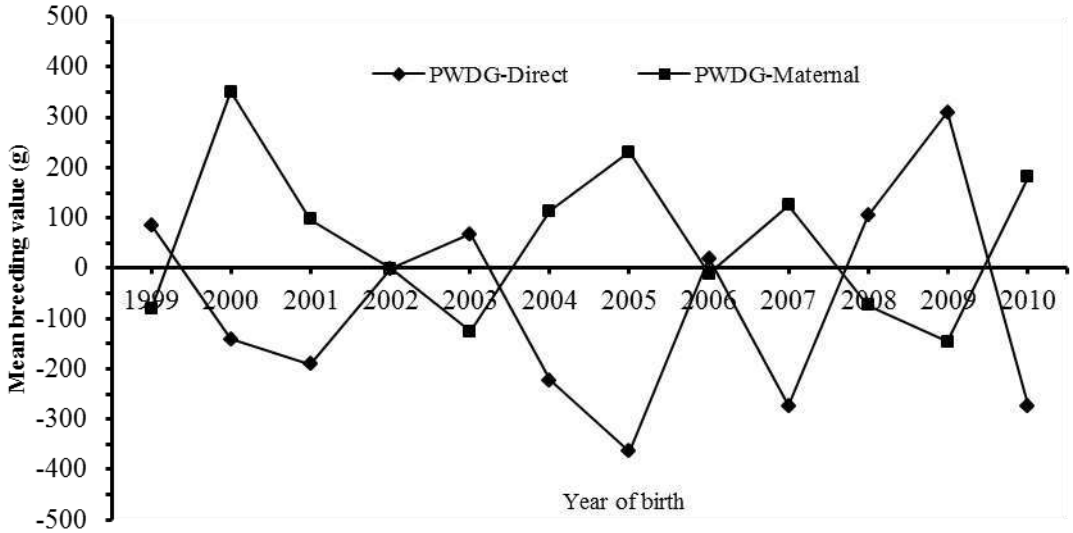


Figure 1. Means of predicts of breeding values BW, WW and PWDG at year of birth.

Conclusion

The low heritability estimates obtained for investigated traits in this research suggest that mass selection based on these traits may result in slow genetic progress for growth traits. Also, only BW was affected by genetic and maternal trends. Generally the low annual genetic trends for studied traits were observed. Eventually, recommended to improve the management of flock and also to select males based on their genetic merit.

Analiza genetskih parametara i trendova za osobine ranog porasta iranske afshari ovce

Meysam Latifi, Ali Mohammadi

Rezime

Svrha ove studije bila je procena genetičkih parametara i genetskih trendova osobina ranog porasta korišćenjem Baiesov-og pristupa sa softverom Gibbs3f90 kod iranske afshari ovce. Skup podataka (težina na rođenju (BW), težina na odbijanja (WW) i dnevni prirast telesne težine pre odbijanja (PWDG)) su sakupljeni u periodu od 1999. do 2010. godine od strane poljoprivrednog jahada pokrajine Zanjan, Iran. Prilagođeni fiksni efekti bili su zapat-godina-sezona kao interakcije, pol (muški, ženski), vrsta rođenja (jedinac, blizanci) i starost majke. Na osnovu Kriterija izvedenih informacija (Derivative Information Criteria - DIC), za ispitivane osobine određen je najprikladniji model. Prema tome, na osnovu najprikladnijeg prilagođenog modela, procena neposrednih aditivnih heritabilita za BW, WW i PWDG bila je 0,32±0,02; 0,05±0,01 i 0,24±0,02; respektivno. Procene heritabiliteta sa majčinske strane su 0,17±0,04; 0,07±0,02 i 0,12±0,05 i ukupnog heritabiliteta 0,11±0,05; 0,08±0,02 i 0,08±0,03 za BW, WW i PWDG, respektivno. Direktni genetski trendovi bili su pozitivni za sve osobine, ali samo za BW 0,75±0,31 g/godišnje (P<0,05). Takođe, genetski trendovi sa majčinske strane bili su negativni za sve osobine i značajan za BW -0,63 ± 0,27 g/godišnje (P<0,05). Umerene procene heritabiliteta za osobine ranog porasta pokazuju da je kod afshari ovaca brže genetsko poboljšanje kroz selekciju moguće za ove osobine. Osim toga, genetski trendovi u ovoj studiji ukazali su na to da je genetsko poboljšanje kroz selekciju pogodno samo za BW kod afshari ovaca.

Ključne reči: afshari ovce, rani porast, heritabilitet, genetski trend

References

ABBASI M.A., ABDOLLAHI-ARPANAHI R., MAGHSOUDI A., VAEZ TORSHIZI R., NEJATI-JAVAREMI A. (2012): Evaluation of models for estimation of genetic parameters and maternal effects for early growth traits of Iranian Baluchi sheep. Small Ruminant Research, 104, 62–69.

BAHREINI BEHZADI M.R., EFTEKHAR-SHAHROUDI F., VAN VLECK L.D. (2005): The study of maternal effects on estimation of heritabilities and determination of environmental factors on early growth traits in Kermani sheep. Journal of Science and Technology of Agriculture and Natural Resources, 9, 195–202.

BOUJENANE, I., DIALLO, I.T. (2017): Estimates of genetic parameters and genetic trends for pre weaning growth traits in Sardi sheep. Small Ruminant Research, 146, 61–68.

EKIZ B., OZCAN M., YILMAZ A. (2004): Estimates of Genetic Parameters for Direct and Maternal Effects with Six Different Models on Birth and Weaning Weights of Turkish Merino Lambs. Turkish journal of Veterinary and Animal Science, 28, 383-389.

ESKANDARINASAB M., GHAFOURI-KESBI F., ABBASI M.A. (2010): Different models for evaluation of growth traits and Kleiber ratio in an experimental flock of Iranian fat-tailed Afshari sheep. Journal of Animal Breeding and Genetics, 127, 26–33.

GHAFOURI-KESBI F., ABBASI M.A., AFRAZ F., BABAEI M., BANEH H., ABDOLLAHI ARPANAHI R. (2011): Genetic analysis of growth rate and Kleiber ratio in Zandi sheep. Tropical Animal Health and Production, 43, 1153-1159.

GHAFOURI-KESBI F., BANEH H. (2012): Genetic parameters for direct and maternal effects on growth traits of sheep. Archiv Tierzucht, 55, 603–611.

GHAVI HOSSEIN-ZADEH N. (2012): Bayesian estimates of genetic changes for body weight traits of Moghani sheep using Gibbs sampling. Tropical Animal Health and Production, 44, 531–536.

GHOLIZADEHA M., GHAFOURI-KESBI F. (2015): Estimation of genetic parameters for growth-related traits and evaluating the results of a 27-year selection program in Baluchi sheep. Small Ruminant Research, 130, 8–14.

GIZAW S., LEMMA S., KOMEN H., VAN ARENDONK J.A.M. (2007): Estimates of genetic parameters and genetic trends for live weight and fleece traits in Menz sheep. Small Ruminant Research, 70, 145–153.

HANDFORD K.J., VAN VLECK L.D., SNOWDER G.D. (2002): Estimates of genetic parameters and genetic change for reproduction, weight, and wool characteristics of Columbia sheep. American Society of Animal Science, 80, 3086–3098.

JAFARI S., RAZZAGZADEH R. (2016): Genetic analysis and the estimates of genetic and phenotypic correlation of growth rates, Kleiber ratios, and fat-tail dimensions with birth to yearling live body weight traits in Makuie sheep. Tropical Animal Health and Production, 4, 667–672.
JAFAROGHLI M., RASHIDI A., MOKHTARI M.S., SHADPARVAR A.A. (2010): (Co)Variance components and genetic parameter estimates for growth traits in Moghani sheep. Small Ruminant Research, 91, 170–177.
KHORSAND A., HAFEZIAN S.H., TEIMOURI-YANSARI A., FARHADI A. (2014): Genetic Parameters of Direct and Maternal Effects for Growth Traits of Afshari Sheep. Iranian Journal of Applied Animal Science, 4, 69–74.
MANDAL A., NESER F.W.C., ROUT P.K., ROY R., NOTTER D.R. (2006); Estimation of direct and maternal (co)variance components for pre-weaning growth traits in Muzaffarnagari sheep. Livestock Science, 99, 79–89.
MIRAEI-ASHTIANI S.R., SEYEDALIAN S.A.R., MORADI SHAHRBABAK M. (2007): Variance components and heritabilities for body weight traits in Sangsari sheep, using univariate and multivariate animal models. Small Ruminant Research, 73, 109–114.
MISTZAL I., TSURUTA S., STRABEL T., AUVRAY B., DRUET T., LEE D.H. (2002): BLUPF90 and related programs. Communication no. 28-07, In: Proceedings of the 7th World Congress for the Genetic Applied Livestock Production, Montpellier, France.
MOHAMMADI A.R., ABBASI M.A., MOGHADDAM A.A. (2009): ZARE SHAHNEH, A. Estimation of growth traits in Iranian Afshari sheep breed under rural production system. Journal of Animal and Veterinary Advances, 8, 1449–1454.
MOHAMMADI K., ABDOLLAHI-ARPANAHI R., AMRAEIC F., MIRZA MOHAMADI E., RASHIDI A. (2015): Genetic parameter estimates for growth and reproductive traits in Lori sheep. Small Ruminant Research, 131, 35–42.
MOKHTARI M.S., RASHIDI A. (2010): Genetic trends estimation for body weights of Kermani sheep at different ages using multivariate animal models. Small Ruminant Research, 88, 23–26.
MONEM M., KIYANZAD M.R., GHARAHDAGHI A.A. (2005): Afshari sheep breed. Domestic Animal Genetic Resources Information System.
PRINCE L.L.L., GOWANE G.R., CHOPRA A., ARORA A.L. (2010): Estimates of (co)variance components and genetic parameters for growth traits of Avikalin sheep. Tropical Animal Health and Production, 42, 1093–1101.
RASHIDI A., AKHESHI H. (2007): Genetic and environment trends estimates of some growth traits in Kurdi sheep. Journal of Agricultural Science, 38, 329–335.
RASHIDI A., MOKHTARI M.S., SAFI JAHANSHAHI A., MOHAMMAD ABADI M.R. (2008): Genetic parameter estimates of pre-weaning growth traits in Kermani sheep. Small Ruminant Research, 74, 165–171.
SAS. User's Guide, version 9. SAS Institute, Cary, NC, 2004.

SHAAT I., GALAL S. (2004): Mansour, H. Genetic trends for lamb weights in flocks of Egyptian Rahmani and Ossimi sheep. Small Ruminant Research, 51, 23–28.
TOSH J.J., KEMP R.A. (1994): Estimated of variance components for lamb weights in three sheep population. Journal of Animal Science, 72, 1184–1190.
ZAMANI P., ALMASI M. (2017): Estimation of autosomal and sex-linked heritabilities for growth related traits in Markhoz breed of goats. Iranian Journal of Applied Animal Science, 48, 109–117. [Persian]
ZAMANI P., MOHAMMADI H. (2008): Comparison of different models for estimation of genetic parameters of early growth traits in the Mehraban Sheep. Journal of Animal Breeding and Genetics, 125, 29–34.

Received 13 July 2018; accepted for publication 20 September 2018

Biotechnology in Animal Husbandry 34 (3), p 303-311 , 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 636.082'636.3
https://doi.org/10.2298/BAH1803303C

THE EFFECT OF SERVICE RAM ON REPRODUCTIVE PERFORMANCES AND BIRTH WEIGHT OF LAMBS

Violeta Caro Petrović*, Milan P. Petrović, Dragana Ružić Muslić, Nevena Maksimović, Nikola Delić, Bogdan Cekić, Zorica Bjelić

Institute for Animal Husbandry 11080 Belgrade, Serbia
Corresponding author: violycaro@yahoo.com
Original scientific paper

Abstract: The investigation was conducted in population of Mis breed of sheep, divided into three groups. For reproduction had use rams of Ile de France breed. Induction and synchronization of oestrus was done off-season in October. The percentage of lambing sheep, sheep fertility, and number of lambs acquired per father, sex and birth type of lambs were observed. Ram 1 had a highest number of ewes lambed with an excellent percentage of mating success and highest fertility rate of ewes with differences on fertility rate of 17% (between group of ewes in ram 1 and ram 2), 36% (group of ewes in ram 1 and ram 3) and 19% (group of ewes in ram 2 and ram 3).The highest birth weight were on lambs born single from ram 3, born twins from ram1, triplets of ram 3. For the lambs born quadruplets (unfortunately only in rams 1 and 2 with one lamb of each was alive) the lamb of ram 2 was higher in birth weight. The male and female lambs of ram 3 got the highest birth weight while the female lambs of ram 1 acquired the lowest birth weight. The highest average birth weight of lambs was born single, male; born triplets' male from lambs of ram 3 having an average birth weight of 6.36kg and 4.0 kg but got the lowest for lambs born twins on female lambs 3.66 kg. The male lambs of ram 1 got the highest average birth weight on lambs born twins with an average birth weight of 4.38 kg but the lowest on triplets born 3.3kg. Unfortunately only one each of the two ram with lambs born quadruplets alive both male and the higher was that lamb of ram 2 with a birth weight of 3.3 kg. Result showed the influence of ram on number of lambs born and lambs' birth weight. Between subject effects Father* birth type showed a significant effect on birth weight of lambs.

Key words: ram, reproductive performances, lambs, birth weight

Introduction

The most important factor determining the success of sheep production is reproductive efficiency which includes successful lambing (*Kutluca Korkmaz and Emsen, 2016).* The type and continuation of production and the majority of the sheep breeds differ in reproductive behavior depending on the changes in season latitude/longitude, the length of the photoperiod and other factors (*Dogan and Nur, 2006; Abu Gazal, 2010, Laleva et al., 2014*). Most of the characteristics of domestic animals are quantitative-measurable. The animal's production properties are controlled by hundreds, perhaps a thousand genes (*Falconer and MacKay, 1996*). The main criteria used to evaluate, and thus the selection of the individuals to be used for breeding, is their breeding value for properties of interest *(Petrovic et al., 2011).*

In order for genetic progress to be more effective for features that are difficult to measure or have low heritability, a more complex and expensive process is needed which is not desirable from the point of production economics at the farm. The success of genetic improvement is based on expectations that the descendants by their phenotypic values will be above the average values of parents. Expected genetic progress - selection success, it is valued depending on the value of heritability for the given property and the selection differential. Since the degree of inheritance that we call heritability is the ratio of genetic and total phenotypic variance, which means that it is not a biological constant, its value can be different in populations, (*Hill, 2014*; *Petrović and Pantelić, 2015)*.

Bearing in mind that in sheep production, the number and quality of offspring in a much larger extent depends on the genetic potential of ram, it is of particular scientific and practical importance to test the fathers. Mis breed of sheep is a new Serbian population and widely experimental research are very important for its future use in farms.

The aim of this paper was to determine the impact of ram on some reproductive performances, body weight of lambs at birth and test the interaction between subject effects.

Material and Methods

The research was carried out at the experimental farm of the Institute for Animal Husbandry, Belgrade. The material for testing was 142 Mis breed of sheep, divided into three groups. Each group is assigned a ram of Ile de France for insemination. Induction and synchronization of oestrus was done off-season in October. Vaginal sponges with flugestone acetate (Syncro-Part 30mg) are used to induce estrus and stays in place for 14 days. To achieve optimal synchronization of

ovulation, after the removal of vaginal sponge, injection of PMSG with a dosage of 600 IU is administered intramuscularly for each sheep. The resulting lambs of the F1 generation are recorded and measured at birth. It has observed the percentage of lambing sheep, sheep fertility, and number of lambs acquired per father, sex and litter size (birth type) of lambs. The statistical processing of data was performed using the SPSS software version 20.

Results and Discussion

The ewes mated with the assigned ram and the percentages of mating success are shown in table 1. It indicated that ram 1 had a highest number of ewes lambed with an excellent percentage of mating success while the ram 3 got the lowest number of ewes lambed and the poorest percentage of mating success.

Table 1. Rams with their corresponding number of ewes

Ram	Number of ewes mated	Number of ewes lambed	Percentage of mating success, %
1	48	45	93.75
2	47	30	63.83
3	47	22	46.81

As seen in table 2, the highest fertility rate of ewes was on the group of ram 1 with differences on fertility rate of 17% (between group of ewes in ram 1 and ram 2), 36% (group of ewes in ram 1 and ram 3) and 19% (group of ewes in ram 2 and ram 3).

Table 2. Number of lambs acquired by ram with the designated ewes lambed

Ram	Number of lambs born	Fertility, %
1	80	177
2	48	160
3	31	141

Table 3. Average Body weight of lambs at birth (kg.), according to ram

Ram	Mean	N	Std. Deviation	Minimum	Maximum	Std. Error of Mean
1	4.12	80	.84	2.60	6.70	.09
2	4.33	48	.88	3.00	6.70	.13
3	4.92	31	1.27	2.10	6.70	.23
Total	4.34	159	.99	2.10	6.70	.08

The lambs of ram 3 got the highest average birth weight while the lowest average birth weight was in lambs of ram 1 (Table 3). The analysis of variance showed significant effect of father on lambs' birth weight ($P<0.05$).

The lambs' average body weight at birth according to birth type per ram in table 4, it represented that the highest weight were on lambs born single from ram 3, born twins from ram1, triplets of ram 3. For the lambs born quadruplets (acquired both in rams 1 and 2 but only 1 lamb of each was alive) higher lamb birth weight from lamb of ram 2.

The test of between subject effects Father* birth type showed significant effect on birth weight of lambs ($P<0.05$).

Table 4. Average birth weight of lambs (kg.), according to ram and litter size

Ram	BT	Number of lambs	Mean	Std. Deviation
1	single	8	5.31	.78
	twins	48	4.30	.66
	triplets	23	3.36	.34
	quadruplets	1	2.60	-
	TOTAL	80	4.11	.84
2	single	8	5.71	.81
	twins	22	4.23	.52
	triplets	17	3.88	.63
	quadruplets	1	3.30	-
	TOTAL	48	4.33	.88
3	single	16	5.85	.92
	twins	13	3.91	.74
	triplets	2	4.00	.56
	quadruplets	-	-	-
	TOTAL	31	4.92	1.27

The sex of lambs born and their average birth weight per father (table 5). The male and female lambs of ram 3 got the highest birth weight while the male lambs of rams 1 and 2 had the same average birth weight. The female lambs of ram 1 acquired the lowest birth weight. The test of between – subjects effects Father* Sex of lambs showed no significant effect on lambs birth weight. ($P>005$)

Table 5. Average birth weight of lambs (kg.), according to ram and sex of lambs

Ram	Sex	Number of lambs	Mean	Std. Deviation
1.0	Male	47	4.18	.86
	Female	33	4.02	.82
2.0	Male	25	4.18	.74
	Female	23	4.50	1.01
3.0	Male	15	5.00	1.25
	Female	16	4.84	1.33

Table 6. Average weight of lambs (kg) according to ram, litter size and sex of lambs

Ram	BT	Sex	Number of lambs	Mean	Std. Deviation
1.0	Single	Male	6	5.16	.86
		Female	2	5.75	.35
	Twins	Male	28	4.38	.66
		Female	20	4.20	.67
	Triplets	Male	13	3.30	.36
		Female	10	3.44	.32
	Quadruplets	Male	1	2.60	-
2.0	Single	Male	2	5.65	.64
		Female	6	5.73	.91
	Twins	Male	12	4.23	.46
		Female	10	4.22	.62
	Triplets	Male	10	3.92	.71
		Female	7	3.84	.55
	Quadruplets	Male	1	3.30	-
3.0	Single	Male	6	6.36	.81
		Female	10	5.55	1.04
	Twins	Male	7	4.11	.66
		Female	6	3.66	.81
	Triplets	Male	2	4.00	.56

The average birth weight of lambs depending on their birth type and sex of lambs per father (Table 6), it can noticed that the highest average birth weight of lambs born singles was the male lambs of ram 3 with an average weight of 6.36 kg and the lowest was 5.16 kg the male lambs of ram 1. The male lambs of ram 1

born twins was the highest 4.38 kg on the average birth weight while the lowest was the female lambs of ram 3 with an average birth weight of 3.66 kg. For the lambs born triplets the highest was 4.0 kg the male lambs of ram 3 and the lowest 3.3 kg the male lambs of ram 1.Unfortunately only one each of the two ram had quadruplets born alive both male and the higher was that lamb of ram 2 with a birth weight of 3.3 kg. The interaction between subject effects Father * BT * Sex showed no significant effect on lambs birth weight ($P>0.05$).

The result acquired in the experiment indicated the influence of ram on number of lambs born and lambs' birth weight. Complying in our result was that by *Sánchez-Davila et al. (2015)* in Saint Croix hair sheep informed the effect of ram was significant ($P < 0.05$) on litter size and birth weight of lambs. Likewise agreeable with our result by *Schmidova et al. (2016)* informed that service rams in Suffolk sheep also have a clearly detectable influence on number of born. *Hagger (2002)* found out the effect of service ram on litter size was also in accordance with our result. *Freking et al.* (2000) stated the main effect of ram breed was significant for number born, litter birth weight, birth weight, of which it was absolutely compatible with ours. *Assan and Makusa (2005*) found the e effect of sire was significant on birth weight in indigenous Sabi sheep and Mutton Merino, however a non-significant effect on birth weight in Dorper sheep.

Assan and Makusa, (2005) suggest that selection of rams should be given a priority in any sheep production system making it useful in the improvement of overall economic efficiency. *Yaqoob et al. (2004)* was concluded that breed of sire can affect the birth weight of lambs and this factor should require careful consideration in practical lamb husbandry. Our research result can confirmed their ideas that ram have very important role in sheep production.

Dakhlan et al. (2015) noted the contribution of sire by birth type interaction to the expression of birth weight was significant, also supported the result of our study.

Conclusion

Our research showed that effect of service ram on reproductive performances and birth weight of lambs is important for considering of sheep production and flock improvement programs. Each ram showed differently as in ram 3 in spite showed the poorest percentage of mating success the males' offspring single and triplets born are the heaviest on average birth weight. On the other hand, ram 1 showed the best in mating success with highest birth weight on male lambs born twins but the lowest on female lambs born triplets. Two rams had quadruplets born with only one lamb alive each both male and the higher was that

lamb of ram 2. Generally the ram have influence on number of lambs born and lambs' birth weight.

In this connection the ram therefore does not only contribute half of his genetics but its success as a breeder.

Uticaj priplodnih ovnova na reproduktivne performanse i težinu jagnjadi na rođenju

Violeta Caro Petrović, Milan P. Petrović, Dragana Ružić Muslić, Nevena Maksimović, Nikola Delić, Bogdan Cekić, Zorica Bjelić

Rezime

Ispitivanje je sprovedeno u populaciji ovaca rase Mis, podeljenoj u tri grupe. Za reprodukciju korišćeni su ovnovi Il de frans rase. Indukcija i sinhronizacija estrusa obavljena je u oktobru van sezone. Posmatrani su sledeći parametri: procenat jagnjenja, plodnost ovaca i broj potomaka po ocu, polu i tipu jagnjenja. Ovan 1 je imao najveći broj ovaca koje su se jagnjile sa visokim procentom uspeha parenja i najvišom stopom plodnosti ovaca sa razlikama u stopi plodnosti od 17% (između grupe ovaca ovna 1 i ovna 2), 36% (grupa ovaca ovna 1 i ovna 3) i 19%. Najveće težine na rođenju su utvrđene kod jagnjadi jedinaca od ovna 3, blizanaca od ovna 1, trojki ovna 3. Za jagnjad rođenu kao četvorke (nažalost samo kod ovnova 1 i 2 sa jednim jagnjetom od svakog koje je bilo živorođeno) jagnjad ovna 2 su bila najveća na rođenju. Muška i ženska jagnjad ovna 3 su imala najvišu težinu pri rođenju, dok su ženska jagnjad ovna 1 imala najnižu težinu na rođenju. Najviša prosečna težina pri rođenju jagnjadi rođene kao jedinci, muškog pola; rođeni kao trojke muškog pola ovna 3 imali su prosečnu težinu na rođenju od 6,36 kg i 4,0 kg, ali i najnižu za jagnjad blizance ženskog pola - 3,66 kg. Muška jagnjad ovna 1 su imali najveću prosečnu težinu na rođenju jagnjadi rođenih kao blizanci sa prosečnom telesnom težinom od 4,38 kg, a najmanju jagnjad trojke - 3,3 kg. Rezultat je pokazao uticaj ovna na broj rođene jagnjadi i težinu jagnjadi na rođenju. Između efekata otac * tip jagnjenja utvrđen je značajan uticaj na težinu jagnjadi na rođenju.

Ključne reči: ovan, reproduktivne performanse, jagnjad, težina na rođenju

Acknowledgment

The study is part of the projects TR 31053 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.

References

ABU GAZAL B.M.O. (2010): Different Estrous Induction Protocols During the Non-Breeding Season in Assaf Ewes. Thesis- Degree of Master in Animal Production, Faculty of Graduate Studies at An-Najah National University, Nablus, Palestine.

ASSAN N., MAKUZA S.M. (2005): The Effect of Non-genetic Factors on Birth Weight and Weaning Weight in Three Sheep Breeds of Zimbabwe Asian-Australian Journal of Animal Science,18, 2: 151-157.

DAKHLAN A., MOGHADDAR N., VAN DER WERF J.H.J. (2015): Genotype by Birth or Rearing Type Interaction in Merino Sheep. Proceedings of Association for the Advancement of Animal Breeding and Genetics, 21: 405-408.

DOGAN I., NUR Z. (2006): Different estrous induction methods during the non-breeding season in Kivircik ewes. Veterinarni Medicina, 51, (4): 133–138.

FALCONER D.S., MACKAY T.F.C. (1996):Introduction to Quantitative Genetics, 4th ed. Longman, UK.

FREKING B.A., LEYMASTER K.A., YOUNG L.D. (2000): Evaluation of Dorset, Finnsheep, Romanov, Texel, and Montadale breeds of sheep: I. Effects of ram breed on productivity of ewes of two crossbred populations. Journal of Animal Science, 78:1422–1429.

HAGGER C. (2002): Multitrait and repeatability estimates of random effects on litter size in sheep. Animal Science, 74, 209–216.

HILL W.G. (2014): Review paper - Applications of Population Genetics to Animal Breeding, from Wright, Fisher and Lush to Genomic Prediction. Genetics, 196, 1–16 .

KUTLUCA KORKMAZ M., EMSEN E. (2016): Growth and reproductive traits of purebred and crossbred Romanov lambs in Eastern Anatolia. Animal Reproduction, 13, 1, 3-6.

LALEVA S., SLAVOVA P., PACINOVSKI N, BONEV G., CILEV G., POPOVA Y. (2014): Comparison of the ovulation rate, fertility and birth weight in sheep of Trakian Merino Breed and their crosses with Booroola. Macedonian Journal of Animal Science, 4, 2, 49-53.

PETROVIĆ P.M. (2000): Genetika i oplemenjivanje ovaca (Monografija). Naučna knjiga, Beograd, 365 pp.

PETROVIĆ P.M., PANTELIĆ V. (2015): Modern Selection of Farm Animals. Publisher: Institute for Animal Husbandry, Belgrade – Zemun 320 pp.

PETROVIC P. M., SRETENOVIC LJ., MUSLIC RUZIC D., PACINOVSKI N., MAKSIMOVIC N. (2011): The effect of crossbreeding systems on lamb meat production. Macedonian Journal of Animal Science,1,1, 57-60.

SÁNCHEZ-DÁVILA F., BERNAL-BARRAGÁN H., PADILLA-RIVAS G., DEL BOSQUE-GONZÁLEZ A.S., VÁZQUEZ-ARMIJO J.F., LEDEZMA-TORRES R.A. (2015):Environmental factors and ram influence litter size, birth, and weaning weight in Saint Croix hair sheep under semi-arid conditions in Mexico**.** Tropical Animal Health Production, 47(5):825-31.
SCHMIDOVÁ J., MILERSKI M., SVITÁKOVÁ A., NOVOTNÁ A., VOSTRÁ-VYDROVÁ H., VOSTRÝ L. (2016): THE INFLUENCE OF RAM ON LITTER SIZE IN SUFFOLK SHEEP. Acta argiculturae Slovenica, Supplement 5, 109–112.
SPSS software version 20.
YAQOOB M., MERRELL1 B.G., SULTAN J.I. (2004): Comparison of Three Terminal Sire Breeds for Birth Weight of Lambs Kept Under Upland Grassland Conditions in the Northeast of England. Pakistan Veterinary Journal, 24(4):196-198.

Received 23 August 2018; accepted for publication 21 September 2018

Biotechnology in Animal Husbandry 34 (3), p 313-322 , 2018 ISSN 1450-9156
Publisher: Institute for Animal Husbandry, Belgrade-Zemun UDC 636.06'636.2
https://doi.org/10.2298/BAH1803313N

VARIABILITY OF FUNDAMENT TRAITS IN PRIMIPAROUS SIMMENTAL HEIFERS

Dragan Nikšić[1], Vlada Pantelić[1], Dušica Ostojić-Andrić[1], Predrag Perišić[2], Marina Lazarević[1], Ivan Ćosić[1], Maja Petričević[1]

[1] Institute for Animal Husbandry, Autoput 16, P. Box 23, 11080, Belgrade-Zemun, Republic of Serbia
[2] University of Belgrade, Faculty of Agriculture, Nemanjina 6, 11080, Belgrade-Zemun, Republic of Serbia
Corresponding autor:draganniksic84@gmail.com
Original scientific paper

Abstract: Visual evaluation and recognition of dairy traits of cows are preliminary indicators of milk yield, longevity, and reproductive ability of the individual animal, which is very important from the aspect of the economics of milk production. The deficiencies in the fundament traits lead to poor production, poor health and premature culling of cows from the herd. The paper examines the frequency of preferred scores of a certain trait in the first calving heifers according to the housing method (animals reared by individual agricultural producers and farm animals) and their origin (domestic and imported animals), as well as the impact of these two factors on the observed properties. Four fundament traits were analysed: the position of the hind legs, the development of the hocks/joints, the pastern joints and the height of the feet on a total of 954 first-calving Simmental heifers. Observed by the housing method, the higher frequency of the preferred scores for all of the fundament traits, was achieved by the farm cows, while according to the origin of the cows, the higher frequency of the preferred scores for all of the fundament traits was realized by imported animals in relation to domestic cows. The influence of the factors of the housing method and origin of animals examined by $\chi 2$ test on all the tested linear scores (frequency of scores) of the fundament traits was statistically very significant ($p \leq 0.001$), while the analysis of the variance (F test) determined high significance ($p \leq 0.001$) of the interaction between the origin and housing method on the height of the feet, as well as significant effect ($p \leq 0.05$) on the position of the hind legs, while on other linear scores of the fundament traits it did not exhibit statistical significance ($p > 0.05$).

Key words: Simmental breed, fundament, rearing method, origin, linear scores

Introduction

The evaluation of the exterior of animals is based on knowledge of the structure and function of individual organs as well as the most important relationships between the individual parts of the body.

Assessment of body development of cattle is a critical evaluation of whether the affected animal, by its overall appearance and individual parts of the body, can guarantee good health and high production, or longevity. Inclusion of a linear type score, contributes to the estimation of the cow's breeding value, which positively reflects on the overall results of the selection and the success of production (*Pantelić et al., 2006*).

Linear assessment includes the evaluation of each trait in its biological extremes, according to the scale ranging from 1 to 9 in the scorecard, and it should be emphasized that the highest score (9) is not the most favourable for each trait, because for some traits the average grades (5) is the best (*Petrović and Pantelić, 2015*).

A minimum set of feet and leg traits required in an effective selection program include the results or measurements of one or more feet traits and the assessment of the leg traits. The combination of angles, length and diagonals of the hind feet seems to be the best combination that allows the ease and precision of scoring or measurement and predictive values for the lameness traits of the feet. In fact, some data suggest that only the diagonal of the feet is sufficient measurement (*Boelling, 1994; McDaniel and Vilk, 1996*). Which traits of the hind legs should be included is not so well established. Most programs use the position of the rear legs - side view, which primarily reflects the angle of the hock joint. *Boelling (1994)* finds that it is more related to locomotion results than to feet measurements. Other data show that the position of the hind legs is more informative for the prediction of survival (*McDaniel et al., 1993*), although less hereditary than the side view. When available, estimates of locomotive properties look like useful predictors. The actual incidence of lameness should be included when present, but the studies do not compare its value with respect to the indirect selection based on scores for feet and leg traits and locomotion results.

By studying the incidence of deformity of the extremities of the Simmental cows depending on the type of bedding, *Stojanović et al. (2018)* reached the following results: of the total number of studied cows 3.45% had "X" front leg position, 14.8% had "X" position of the hind legs. The convergent position of the front legs was recorded in 35.86%, and divergent in 8.28% of animals. The convergent position of the hind legs was established in 16.55% of cows, and divergent in 2.76%. The forward protruding position of the front legs was recorded in 4.14% of cows, inducted position in 11.03%, and a broad position in 4.14% of animals. The sickle position of hind legs was recorded in 17.24% of the cows, and 7.59% of animals were post-legged. The pronounced soft front leg pasterns were

determined in 7.59% of the cows, and the soft pasterns of the hind legs in 33.79% of the total number of estimated cows. The observed changes in the joints in the shape of swellings were recorded in 1.38% of cattle on the carpal joint and 2.76% on the tarsal joint. The affected blade and body joint (scurry shoulder) was recorded in 43.45% of the total number of cows. The body weight of Busha cows in the area of the Pirot district amounted to 226.07 kg, the height to withers 104.33 cm, the height to rump 104.12 cm, pelvis width 32.52 cm, breast depth 53.97 cm, breast girth 130.48 cm, and body length 119.67 cm.

By studying correlations between the milk performance and the fundament traits, results obtained show mainly negative correlations (*Boelling and Pollott, 1997*). Their results suggest that lameness will increase as a result of the selection on increased milk yields, unless taking into account the traits of the fundament. *Uribe et al. (1995)* find that genetic correlations of production with "troubleshooting of leg problems" ranged from 0.20 to 0.27 and indicate that long-term selection on yield has probably reduced the problems that arise in the fundament traits. Foot and leg disorders were correlated with milk performance in the United States, r = 0.48 (*Lions et al., 1991*) and the Netherlands r = 0.26 (*Groen et al., 1994*).

Material and methods

The basic data on the traits of the fundaments, as well as the data on the origin of all examined cows, were collected in cooperation with the farm "Lazar" Blace housing a number of animals included in this research. For animals housed on individual farms, data on these traits were collected in cooperation with breeding organizations, which implement the breeding program in the area of Toplica district.

The total number of animals (n = 954) used in the analysis of morphometric traits were divided into two groups based on the housing method, and two groups based on origin, in the following way:

Based on the housing method:

Group 1: animals reared by individual producers (n = 504);

Group 2: animals reared on the farm (n = 450);

Based on the origin:

Group 1: animals of domestic origin (n = 718);

Group 2: imported animals (n = 236).

All of the properties of the fundament were evaluated linearly after the first calving: the position of the hind legs, the development of the hocks, the pastern joints and the height of the hoof.

The processing of collected data consisted in determining the frequency for each assessment individually, the frequency of the preferred scores for all the traits of the fundament and the comparison of the received frequencies by groups.

Subsequently, the influence of the housing method (applied $\chi 2$ test) and the influence of the origin ($\chi 2$ test applied) on the frequency of linear scores for each property of the fundament was examined, while the analysis of variance examined the influence of the housing method and origin, as well as their interaction with all linear scores, in the following model with a fixed influence on the housing method and origin and their interaction:

$$Y_{ij}=\mu+N_i+P_j+NP_{ij}+e_{ijk}$$

- Y_{ij}: examined trait,
- μ: population average for given trait,
- N_i: fixed effect of i housing method (i=1,2),
- P_j: fixed effect of j origin (j=1,2),
- NP_{ij}: effect of interaction between factors (housing method and animal origin), eij: random error

For statistical data processing and application of the specified model, the software SPSS Statistics for windows, Version 23.0 was used.

Results and Discussion

Based on the results shown in Table 1, it can be concluded that the majority of animals had preferable scores for studied trait, with animals reared on the farm (79.56%) and animals originating from import (83.05%) had a higher frequency of desirable scores compared to animals reared by individual producers (60.12%), and animals of domestic origin (64.76%). The effect of animal origin and housing method was statistically very significant ($p\leq0.001$) on the frequency of the rear leg assessment, examined by the $\chi 2$ test. By variance analysis (F test), significant influence ($p\leq0.05$) of origin and interaction of origin and housing method was determined.The frequency of preferable scores for the development of the hock joints was twice as high in farm animals compared to the first-calving heifers reared by individual producers and amounted to 49.33%. A higher percentage of preferred scores was also recorded in imported animals (40.25%) compared to animals of domestic origin (33.85%).

On the basis of the $\chi 2$ test of the independence of the trait, a statistically very significant correlation ($p\leq0.001$) of the origin and score for the development of the hock joints was established in the first place, as well as the housing method and scores for the same trait. The variance analysis (F test) showed a high significance of the housing method ($p\leq0.001$), while the origin of the animal and the interaction of origin and housing method did not have a statistically significant effect ($p> 0.05$) on the development of the joints.

Table 1. Linear estimates and their frequencies for the position of hind legs and the development of hock joints in the first calving heifers of Simmental breed

Scores	Housing						Origin					
	Animals reared by individual producers			Farm animals			Domestic			Imported		
	N	%	% of the group	N	%	% of the group	N	%	% of the group	N	%	% of the group
REAR LEG POSITION												
1	0	0.00		0	0.00		0	0.00		0	0.00	
2	21	4.17	20.63	0	0.00	5.33	21	2.92	15.18	0	0.00	8.05
3	83	16.47		24	5.33		88	12.26		19	8.05	
4	**2**	**0.40**		**26**	**5.78**		**7**	**0.97**		**21**	**8.90**	
5	**80**	**15.87**	**60.12**	**262**	**58.22**	**79.56**	**225**	**31.34**	**64.76**	**117**	**49.58**	**83.05**
6	**221**	**43.85**		**70**	**15.56**		**233**	**32.45**		**58**	**24.58**	
7	97	19.25		63	14.00		142	19.78		18	7.63	
8	0	0.00	19.25	5	1.11	15.11	2	0.28	20.06	3	1.27	8.90
9	0	0.00		0	0.00		0	0.00		0	0.00	
χ2 Test												
Housing χ2=259.311*** p=0.000							Origin χ2=86.763*** p=0.000					
F Test												
Housing F=1.097^{nz} p=0.295							Origin F=6.281* p=0.012					
Housing x Origin F=4.498* p=0.034												
HOCK JOINT DEVELOPMENT												
1	0	0.00		0	0.00		0	0.00		0	0.00	
2	0	0.00	0.59	0	0.00	1.11	0	0.00	0.97	0	0.00	0.43
3	3	0.59		5	1.11		7	0.97		1	0.43	
4	1	0.20		11	2.44		4	0.56		8	3.39	
5	153	30.36	76.39	153	34.00	49.56	226	31.48	65.18	80	33.90	59.32
6	231	45.83		59	13.11		238	33.15		52	22.03	
7	**110**	**21.83**		**153**	**34.00**		**198**	**27.58**		**65**	**27.54**	
8	**6**	**1.19**	**23.02**	**68**	**15.11**	**49.33**	**44**	**6.13**	**33.85**	**30**	**12.71**	**40.25**
9	**0**	**0.00**		**1**	**0.22**		**1**	**0.14**		**0**	**0.00**	
χ2 Test												
Housing χ2=168.306*** p=0.000							Origin χ2=29.770*** p=0.000					
F Test												
Housing F=18.871*** p=0.000							Origin F=0.499 ^{nz} p=0.480					
Housing x Origin F=0.660^{nz} p=0.417												

***- p≤0.001; ** - p≤0.01; * - p≤0.05; ns - p>0.05

By analysing the pastern joints and the height of hoofs, according to the groups (Table 2), the lowest percentage of cows with favourable scores for pastern joints were cows grown by individual producers (52.78%), then cows of domestic origin (63.37%), (74.15%) and the highest percentage with favourable scores for the pastern joints were cows reared on the farm (80.89%).

By testing the frequencies using the $\chi 2$ test, it was found that there was statistically very significant variation ($p \leq 0.001$) for pastern joints influenced by the method of rearing of cows and their origin. By analysing the variance (F test), a highly significant influence of the rearing method was determined ($p \leq 0.01$), significant influence of the animal origin ($p \leq 0.05$), while the interaction of origin and housing did not have a statistically significant effect ($p > 0.05$) on the pastern joints.

The frequency of the preferred scores for the observed trait was considerably higher in farm animals (67.11%), than in the animals reared by individual producers, where only 18.25% of first calvers obtained preferred scores. If the observed population is viewed on the basis of animal origin, it can be noted that the frequency of preferred scores was slightly higher for the imported animals (41.95%), than for the animals of domestic origin, where 41.09% of first calving heifers received preferred scores for the height of the hoofs.

The influence of the animal origin and the method of housing was statistically very significant ($p \leq 0.001$) on the frequency of scores for the height of the hoofs, examined by the $\chi 2$ test. By variance analysis (F test), a very significant influence ($p \leq 0.001$) of the way of housing, the origin of the animals and their interaction was also determined.

Table 2. Linear estimates and their frequencies for the properties of pastern joints and height of the hoofs in first calving heifers of Simmental breed

Scores	Housing						Origin					
	Animals reared by individual producers			Farm animals			Domestic			Imported		
	N	%	% of the group	N	%	% of the group	N	%	% of the group	N	%	% of the group
PASTERN JOINTS												
1	0	0.00		0	0.00		0	0.00		0	0.00	
2	21	4.17	22.42	0	0.00	0.44	21	2.92	12.67	0	0.00	10.17
3	92	18.25		2	0.44		70	9.75		24	10.17	
4	**5**	**0.99**		**30**	**6.67**		**12**	**1.67**		**23**	**9.75**	
5	**40**	**7.94**	**52.78**	**125**	**27.78**	**80.89**	**121**	**16.85**	**63.37**	**44**	**18.64**	**74.15**
6	**221**	**43.85**		**209**	**46.44**		**322**	**44.85**		**108**	**45.76**	
7	59	11.71		61	13.56		106	14.76		14	5.93	
8	66	13.10	24.80	23	5.11	18.67	66	9.19	23.96	23	9.75	15.68
9	0	0.00		0	0.00		0	0.00		0	0.00	
$\chi 2$ Test												
Housing $\chi 2=188.453^{***}$ p=0.000							Origin $\chi 2=50.272^{***}$ p=0.000					
F Test												
Housing $F=6.595^{**}$ p=0.010							Origin $F=4.542^{*}$ p=0.033					
Housing x Origin $F=0.073^{nz}$											p=0.788	
HOOF HEIGHT												
1	0	0.00		0	0.00		0	0.00		0	0.00	
2	0	0.00	3.17	0	0.00	5.56	0	0.00	4.60	0	0.00	3.39
3	16	3.17		25	5.56		33	4.60		8	3.39	
4	3	0.60		33	7.33		9	1.25		27	11.44	
5	82	16.27	78.58	49	10.89	27.33	86	11.97	54.31	45	19.07	54.66
6	311	61.71		41	9.11		295	41.09		57	24.15	
7	**86**	**17.06**		**185**	**41.11**		**186**	**25.91**		**85**	**36.02**	
8	**6**	**1.19**	**18.25**	**117**	**26.00**	**67.11**	**109**	**15.18**	**41.09**	**14**	**5.93**	**41.95**
9	**0**	**0.00**		**0**	**0.00**		**0**	**0.00**		**0**	**0.00**	
$\chi 2$ Test												
Housing $\chi 2=376.879^{***}$ p=0.000							Origin $\chi 2=87.933^{***}$ p=0.000					
F Test												
Housing $F=28.317^{***}$ p=0.000							Origin $F=22.492^{***}$ p=0.000					
Housing x Origin $F=19.328^{***}$											p=0.000	

***- p≤0.001; ** - p≤0.01; * - p≤0.05; nz - p>0.05

Conclusion

By examining the fundament traits in the first calving heifers of Simmental breed observed according to the method of housing, the higher frequency of the preferred scores for all the fundament traits was recorded for farm cows, while in regard to the origin, a higher frequency of the preferred scores for all fundament properties was recorded in imported cows compared to cows of domestic origin. The influence of the factors of housing method and origin on all tested linear estimates (frequency of estimation) of the fundament traits, examined by $\chi 2$ test, was statistically very significant ($p \leq 0.001$), while the analysis of the variance (F test) determined high significance ($p \leq 0.001$) of the effect of origin and housing method on the height of hoofs, as well as significance ($p \leq 0.05$) of the effect on the position of the hind legs, while on other linear scores of the fundament traits it did not exhibit statistical significance ($p > 0.05$).

Genetic improvements in fundament traits are unlikely to prevent all foot problems, but the choice of animals with preferred scores for the height of hoofs, pastern joints, the development of the hock joints, and the position of the hind legs can reduce their incidence in the herd. By examining the scores of the fundament traits, it can be concluded that their significance lies in the longevity of the cows and, consequently, on the livestock production.

Problems with extremities are more pronounced than stated in the literature, since these problems are often the primary reason for the incidence of sterility that is not recorded when cows are culled from production, and as the reason their consequence is stated, i.e. sterility.

Varijabilnost osobina fundamenta kod prvotelki simentalske rase

Dragan Nikšić, Vlada Pantelić, Dušica Ostojić-Andrić, Predrag Perišić, Marina Lazarević, Ivan Ćosić, Maja Petričević

Rezime

Vizuelna procena i prepoznavanje mlečnih karakteristika krava su preliminarni pokazatelji mlečnosti, dugovečnosti, kao i reproduktivnih sposobnosti grla, što je veoma važno sa aspekta ekonomičnosti proizvodnje mleka. Nedostaci u osobinama fundamenta dovode do slabije proizvodnje, lošeg zdravstvenog stanja i preranog isključenja krava iz zapata. U radu je ispitivana učestalost poželjnih ocena određene osobine kod prvotelki podeljenih po načinu držanja (grla kod individualnih proizvođača i grla sa farme) i podeljenih po poreklu (grla domaćeg odgoja i grla iz uvoza), kao i uticaj ova dva faktora na posmatrane osobine.

Analizirane su četiri osobine fundamenta: položaj zadnjih nogu, razvijenost skočnog zgloba, kičični zglobovi i visina papaka na ukupno 954 prvotelke simentalske rase. Posmatrano prema načinu držanja, veću frekvenciju poželjnih ocena za sve osobine fundamenta iskazane u ocenama ostvarile su krave sa farme, dok su prema poreklu krava, veću frekvenciju poželjnih ocena za sve osobine fundamenta iskazane u ocenama ostvarile krave poreklom iz uvoza u odnosu na krave domaćeg porekla. Uticaj faktora načina držanja i porekla grla ispitivani $\chi 2$ testom na sve isptitivane linearne ocene (frekvenciju ocena) osobina fundamenta bio je statistički vrlo visoko značajan ($p \leq 0,001$), dok je analizom varijanse (F test) utvrđena visoka značajnost ($p \leq 0,001$) interakcije porekla i načina držanja na visinu papaka, kao i značajnost ($p \leq 0,05$) na poziciju zadnjih nogu, dok na ostale linearne ocene osobina fundamenta nije ispoljila statističku značajnost ($p > 0,05$).

Ključne reči: simentalska rasa, fundament, način držanja, poreklo, linearne ocene

Acknowledgment

The study is part of the projects TR 31053 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia

References

BOELLING D. (1994): Correlations of locomotion and body and claw traits with regard to production in dairy cattle. VIII th International Symposium on Diseases of the Ruminant Digit, Banft, Canada. June 27-30.

BOELLING D., POLLOTT G.E. (1997): The genetics of feet, legs, and locomotion in cattle. Animal Breeding Abstracts, 65, 1-11.

GROEN A.F., HELLINGA L., OLDENBROEK J.K. (1994): Genetic correlations of clinical mastitis and feet and leg problems with milk yield and type traits in Dutch black and white dairy cattle. Netherlands Journal of Agricultural Sciences 42, 371-378.

LYONS D.T., FREEMAN A.E., KUCK A.L. (1991): Genetics of health traits in Holstein cattle. Journal of Dairy Science 74, 1092-1100.

McDANIEL B.T., TABBAA M.J., WILK J.C., CLAY J. (1993): Relationships among sire evaluations for linear conformation traits and survival Holsteins from the South Eastern United States. Report Select Sires (unpub. mimeo).

McDANIEL B.T., WILK J.C. (1996): Genetics of foot diagonal and its relation to survival. Journal of Dairy Science 79 (Suppl. 1), 217.

PANTELIĆ V., SKALICKI Z., PETROVIĆ M.M., LATINOVIĆ D., ALEKSIĆ S., MIŠČEVIĆ B., OSTOJIĆ D. (2006): Linearno ocenjivanje bikovskih majki simentalske rase. Simpozijum "Stočarstvo, veterinarstvo i agroekonomija u

tranzicionim procesima", Herceg Novi 18-25.06.2006., Zbornik kratkih sadržaja,61.
PETROVIĆ P.M., PANTELIĆ V. (2015): Savremena selekcija domaćih životinja. Monografija. Institut za stočarstvo.
STOJANOVIĆ M., PERIŠIĆ P., NIKŠIĆ D., PANTELIĆ V., OSTOJIĆ-ANDRIĆ D., LAZAREVIĆ M., PETRIČEVIĆ M. (2018): Incidence of deformations of the extremities of Simmental cows in different types of stalls. Biotechnology in Animal Husbandry 34 (2), 189-198.
URIBE, H.A., KENNEDY, B.W., MARTIN, B.W. AND KELTON, D.F. (1995): Genetic parameters for common health disorders of Holstein cows. Journal of Dairy Science 78, 421-430.

Received 14 August 2018; accepted for publication 13 September 2018

Biotechnology in Animal Husbandry 34 (3), p 323-334 , 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 637.07'636.5
https://doi.org/10.2298/BAH1803323M

REGRESSION MODELS FOR ESTIMATING CHICK HATCHLING WEIGHT FROM SOME EGG GEOMETRY TRAITS

Nadya Mincheva, Mitko Lalev, Magdalena Oblakova, Pavlina Hristakieva

Agricultural Institute - Stara Zagora, 6000, Bulgaria
Corresponding author: minchevan@yahoo.bg
Original scientific paper

Abstract: The prediction of chicks' weight before hatching is an important element of selection, aimed at improving the uniformity rate and productivity of birds. With this regards, our goal was to develop and evaluate optimum models for similar prediction in two White Plymouth Rock chickens lines – line L and line K on the basis of the incubation egg weight and egg geometry characteristics – egg maximum breadth (*B*), egg length (*L*), geometric mean diameter (*Dg*), egg volume (*V*), egg surface area (*S*). A total of 280 eggs (140 from each line) laid by 40-week-old hens were randomly selected. Mean arithmetic values, standard deviations and coefficients of variation of studied parameters were determined for each line. Correlation coefficients between the weight of hatchlings and predictors were the highest for egg weight, geometric mean diameter, volume and surface area of eggs (r=0.731-0.779 for line L; r=0.802-0.819 for line K).

Nine linear regression models were developed and their accuracy evaluated. The regression equations of hatchlings' weight vs egg length had the lowest coefficient of determination (0.175 for line K and 0.291 for line L), but when egg length and breadth entered the model together, its value increased significantly up to 0.541 and 0.665 for lines L and K, respectively. The weight of day-old chicks from line L could be predicted with higher accuracy with a model involving egg surface area apart egg weight (ChW=0.513EW+0.282S - 10.345; R^2=0.620). In line K a more accurate prognosis was attained by adding egg breadth as an additional predictor to the weight in the model (ChW=0.587EW+0.566B - 19.853; R^2=0.692). The study demonstrated that multiple linear regression models were more precise that single linear models.

Key words: chick weight, egg weight, egg geometry parameters, regression models

Introduction

The avian egg is a biological system whose purpose is to guarantee the proper development of the embryo and successful hatching as a fully developed chick (*Narushin and Romanov, 2002*). *Tahir et al.* (*2011*) outlined that incubation egg weight and hatchlings' weight were important for modelling or predicting slaughter weight and economic efficiency.

Normally, the shape of avian eggs is oval. The mathematical description of egg profile allows calculation of its volume and surface area on the basis of its breath and length. The results from various experiments showed that this geometry traits could be used for prediction of the weight of hatchlings (*Narushin et al. 2002*), the weight of eggs (*Rashidi and Gholami, 2011*), internal properties and composition of eggs (*Shafey et al., 2014*), hatchability (*Narushin and Romanov, 2002*), eggshell quality (*Altuntaş and Şekeroğlu, 2008*). In comparison with these geometry traits, egg weight was more important for hatchling's weight (*Narushin et al., 2002; Sahin et al., 2009*), with specific effect of the line and breed. Numerous studies have shown that egg weight had a substantial influence on the weight of day-old chickens (*Mitrović et al., 2011***;** *Traldi et al., 2011***;** *Mukhtar et al., 2013***;** *Ng'ambi et al., 2013***;** *Mbajiorgu and Ramaphala, 2014***;** *Iqbal et al., 2016; 2017*).

Egg length and breadth are traits that are easy to determine and therefore, often used in experiments with poultry eggs. They could influence the weight of day-old chicks. *Khurshid et al.* (*2003*) demonstrated that these parameters were reliable for predicting the weight of hatchling quails. *Farooq et al.* (*2001*) reported significant correlation coefficients between aforementioned dimensions and chick weight - r=0.58 with egg length and r=0.78 with egg breadth. Experiments with eggs of different fowl species (goose, quail and chicken) indicated that egg shape index (*Saatci et al., 2005*; *Yilmaz and Caglayan, 2008*; *Sahin et al., 2009; Lotfi et al., 2011*), and egg density (*Narushin et al., 2002*) did not have an effect on hatchling's weight.

To predict chickens' weight before the hatching, various models have been developed on the basis of linear and non-linear equations from which the weight is associated with incubation eggs' weight (*Tahir et al., 2011*; *Ng'ambi et al., 2013*; *Ramaphala and Mbajiorgu, 2013*; *Rashid et al., 2013*). *Rashid et al.* (*2013*) calculated that the weight of day-old chickens from three studied breeds increased by 0.595-1.361 g for every 1 g increase in egg weight.

Obviously, the weight of eggs is a more accurate parameter for day-old chickens' weight that egg geometry characteristics, but better results could be obtained when both are included in the models. This is confirmed by *Narushin et al.* (*2002*) who affirmed that the weight, volume and surface area of eggs were the

best predictors of hatchlings' weight with coefficients of determination ranging between 0.26 and 0.63 according to the line.

Similar predictions are important elements of selection work and therefore, for improvement of productivity of poultry. To this end, we aimed to develop and evaluate optimum models for weight prediction in two White Plymouth Rock lines – line L and line K on the basis of the incubation egg weight and egg geometry characteristics.

Materials and Methods

The experiment was performed in the Experimental base of the Agricultural Institute – Stara Zagora with 280 eggs of two White Plymouth Rock lines: L and K (140 from each line) laid by 40-week-old hens. The hens were reared in boxes in groups of 10 hens and 1 rooster on deep permanent litter of wooden shavings. Restricted feeding with daily ration compliant to the age and egg production of layers was used: metabolisable energy 1810.005 kcal/kg, crude protein – 16.012 %, crude fat – 6.836 %, crude fibre 5.889 %, lysine 0.75 %, methionine 0.38 %, calcium 3.2 %, phosphorus 0.81%.

Incubation eggs were randomly selected, and those with irregular shape or shell cracks were removed. Before the incubation, eggs were disinfected through fumigation with formaldehyde vapours. Every egg was numbered on the blunt end, weighed on a balance with precision up to 0.01 g, and its breadth and length were measured with digital caliper with precision up to 0.01 mm. On the basis of these dimensions, the geometric mean diameter (Dg), egg volume (V) and egg surface area (S) were calculated as followed:

$$Dg = (LB^2)^{1/3}$$ *Mohsenin (1970),*

$$V = (0.6057 - 0.0018B)LB^2$$ *Narushin (2005)*

$$S = (3.155 - 0.013L + 0.0115B)L,$$ *Narushin (2005),* where
B = egg maximum breadth; L is the egg length in mm

Egg incubation took place under optimum conditions. On the 19th day, eggs were transferred from the incubator to the hatcher and placed in wooden frames with partitions for individual hatching. After the hatch, all chicks were individually weighed with precision of 0.01 g.

Statistical methods

The means, standard deviations and coefficients of variation of studied traits were calculated for each line and evaluated by paired samples t-test. Pearson's correlation coefficients (r) between independent and dependent variables were determined. Initially, the egg weight and egg geometry traits were included to predict hatchling weight individually, and then a step-wise multiple regression was run with statistically significant predictors only in order to eliminate collinearity. problem. Collinearity was established according to VIF values, obtained as $VIF=1/1-R_i^2$, which should not exceed 10. The regression curve was determined as linear and therefore, linear models were found most appropriate to predict hatchlings' weights:

$\hat{Y} = a + b.X + \varepsilon$ (1) Simple regression model

$\hat{Y} = a + b_1.X_1 + b_2.X_2 + \ldots\ldots + b_k.X_k + \varepsilon$ (2) Multiple regression model,

where $\hat{Y}$ - dependent variable (chick weight), a – intercept, b_k – regression coefficients, X_k independent variables (egg weight, egg geometry parameters), ε - residual (error).

The significance of the regression coefficients was tested with a t-statistic while the goodness-of-fit of the regression was assessed using the coefficient of determination (R^2).

The best models were validated by incubation of 60 randomly selected eggs from each line, with preliminary determined weight and geometry traits using the described methods. The data were used for calculation of predicted weights of chickens after hatching. After the individual hatching, the weight of chicks was determined on digital balance. The differences between observed and predicted values of dependent variable were established.

Statistical analyses were performed with software SPSS (version 19.0 for Windows).

Results and Discussion

The means, standard deviations and coefficients of variations of egg weight and geometry traits in both studied lines (L and K) are shown in Table 1. For all traits, mean values were higher for line L, that could be attributed to genetic differences ($p<0.001$). The differences in egg weight and hatchling weight were 6.79 g (10.34 %) and 5.18 g (11.70 %). respectively. Our data were somewhat

comparable to the results reported by *Alabi et al. (2012)*, that egg weight had an effect on egg length and volume, but not on its breadth. The presented results indicated that egg weight determined studied geometry traits (length, breadth, geometric mean diameter, volume, surface area). Unlike us, *Narushin et al. (2002)* did not established differences with regard to egg weight in three egg-laying chicken lines while egg volume, surface area, density and hatchlings' weight differed substantially. In this study, the variation of weight of day-old chicks from both lines was higher as compared to incubation eggs' weight – 6.85-7.34 vs 5.18-5.55 %, which in the view of *Shalev and Pasternak (1995)* could probably result from incubation conditions and hatchery management. Unlike us, *Tahir et al. (2011)* reported that the weight of chicks varied at a lower extent than the weight of eggs, and according to *Wolanski et al. (2007)* coefficients of variations of both were similar. They are considered to be parameters of uniformity (*Shalev and Pasternak, 1995).* The least changeable parameter was the geometric mean diameter of eggs with coefficients of variation 1.79 and 1.98 %, followed by egg breadth– 2.06 and 2.44 % for line L and K respectively. The egg volume exhibited higher coefficients of variation: 5.06 % in line L and 5.64 % for line K. In a study by *Narushin et al. (2002)*, egg volume and weight were outlined as most variable parameters, as confirmed by the present study as well.

Table 1. Descriptive statistics of egg weight, egg geometry parameters and chick weight at hatch used to determine models in line L and line K

Traits	Line L			Line K		
	Mean	SD	CV	Mean	SD	CV
Chick weight (ChW), g	44.26***	3.03	6.85	39.08	2.87	7.34
Egg weight (EW), g	65.68***	3.40	5.18	58.89	3.27	5.55
Egg length (L), mm	57.81***	1.52	2.63	56.12	1.92	3.42
Egg maximum breadth (B), mm	44.61***	0.92	2.06	43.06	1.05	2.44
Geometric mean diameter (D*g*), mm	48.63***	0.87	1.79	47.03	0.93	1.98
Egg volume (V), cm^3	60.47***	3.06	5.06	54.99	3.10	5.64
Egg surface area (S), cm^2	74.32***	2.52	3.39	69.75	2.64	3.78

SD – standard deviation, CV – coefficient of variation, %
Significant at ***-$p<0.001$

Before the regression analysis, a correlation matrix was composed with linear coefficients of correlations between the dependent and independent variables. Table 2 shows that all predictors included in the analysis had significant correlation coefficients with the weight of hatchlings ranging from moderate (with egg length and breadth: r=0.418-0.695*)* to strong (mean geometric diameter, volume, surface area and weight of eggs: r=0.731-0.819), $p<0.001$. The presence of significant correlations with the dependent variable indicated their suitability for inclusion in regression models. The data of *Narushin et al. (2002)* demonstrated

slightly higher correlation between the hatchlings' weight and egg weight (r=0.56), than with egg volume (r=0.50) and egg surface area (r=0.50) in three egg-laying chicken lines; this was confirmed in our experiments. *Sahin et al.* (*2009*) also reported higher correlation coefficients - 0.87 (hatchling weight vs egg weight) and 0.81 (hatchling weight vs egg volume). High positive relationship (0.82) was present between hatchling weight and egg surface area (0.74 for line L and 0.80 for line K) as also reported by *El-Safty* (*2011*).

Table 2. Phenotypic correlations among chick weight at hatch, egg weight and egg geometry parameters

Traits	EW	L	B	Dg	V	S	ChW
EW		0.624 ***	0.729 ***	0.961 ***	0.965 ***	0.966 ***	0.819 ***
L	0.629 ***		-0.013	0.564 ***	0.596 ***	0.652 ***	0.418 ***
B	0.722 ***	0.211 **		0.818 ***	0.794 ***	0.749 ***	0.695 ***
Dg	0.868 ***	0.655 ***	0.877 ***		0.999 ***	0.994 ***	0.812 ***
V	0.870 ***	0.679 ***	0.861 ***	0.999 ***		0.997 ***	0.811 ***
S	0.872 ***	0.714 ***	0.835 ***	0.997 ***	0.999 ***		0.802 ***
ChW	0.779 ***	0.539 ***	0.602 ***	0.731 ***	0.733 ***	0.735 ***	

EW- egg weight, L- egg length, B- egg maximum breadth, Dg- geometric mean diameter, V- egg volume, S- egg surface area, , Ch W- chick weight
Significant at **-p<0.01, ***-p<0.001
Upper matrix: Line K
Lower matrix: Line L

Furthermore, independent variables correlate statistically significantly among, except for egg breadth and length for line K that exhibited insignificant relationship (r= -0.01), while for line L eggs, the relationship was weak but significant (0.21, p<0.01). Unlike us, *Yakubu et al.* (*2008*) reported a strong positive correlation between egg length and breadth (r=0.71).

The predictors egg volume, surface area and mean geometric diameter were very closely related in both lines (0.994-0.999). High linear relationship was reported by *Nedomova and Buchar* (*2014*) between egg volume and surface area in geese with R^2 = 0.996. A probable reason could be the involvement of the same parameters e.g. egg breadth and length in their formulas. At the same time, egg volume, surface area and mean geometric diameter correlated strongly with egg weight (0.868-0.966). The substantial relationships between egg volume and eggs in this study agreed with finding of *Malago and Baitilwake* (*2009*), *Kabir et al***.** (*2012*). The latter researchers reported coefficient of phenotypic correlation

between ISA Brown and local chickens of r=0.72 and r=0.88. Strong interrelationships between egg weight, volume and surface area were communicated by *Narushin* (*1997*). A high correlation coefficient (0.99) was found out between ostrich egg weight and surface area (*El-Safty 2011*) and this was confirmed in our study as well.

Table 3 presents regression coefficients, coefficients of determination and levels of statistical significance of models predicting the weight of hatchlings on the basis of egg weight and geometry parameters in both lines. Data showed that all linear regression models were adequate as could be seen from high level of statistical significance ($p<0.001$). The comparison of models demonstrated that the coefficient of determination was useful parameters of variation of the dependent variable, explained with regression. The highest coefficients of determination in both lines were those of simple linear models which used egg weight as predictor - 0.606 for line L and 0.671 for line K, e.g 61-67 % of hatchlings' weight depended on egg weight (model 1). According to *Tserveni-Gousi and Yannakopoulos* (*1990*) 70m% of variation in the weight of pheasant chicks was attributable to egg weight which was a better predictor than shape index and shell deformity. *Tahir et al.* (*2011*) and *Ramaphala and Mbajiorgu* (*2013*) also predicted the hatching weight of chickens but reported higher coefficients of determination R^2, 0.856 and 0.995 respectively, while *Olutunmogun et al. (2017)* reported a much lower value (R^2=0.15) than our data.

Table 3. Regression coefficients, coefficients of determination and level of significance of models in lines L and K

№	Regression model	Predictors	Line	Regression coefficients			R^2	SE	F-value
				Const.	b_1	b_2			
1	Simple	EW	L	-1.360	0.695	-	0.606	1.909	229.509***
			K	-3.327	0.720	-	0.671	1.653	284.093***
2	Simple	B	L	-44.226	1.984	-	0.363	2.429	84.768***
			K	-42.467	1.894	-	0.483	2.075	129.626***
3	Simple	L	L	-17.751	1.073	-	0.291	2.562	61.166***
			K	3.999	0.625	-	0.175	2.620	29.420***
4	Simple	Dg	L	-80.183	2.559	-	0.534	2.076	170.948***
			K	-78.930	2.509	-	0.660	1.683	269.274 ***
5	Simple	V	L	0.268	0.728	-	0.537	2.070	173.025***
			K	-2.227	0.751	-	0.658	1.686	267.956***
6	Simple	S	L	-21.460	0.884	-	0.540	2.063	175.066***
			K	-21.878	0.874	-	0.643	1.724	250.191***
7	Multiple	B, L	L	-80.461	1.684	0.858	0.541	2.069	87.090***
			K	-78.969	1.909	0.639	0.665	1.675	136.930***
8	Multiple	EW, S	L	-10.345	0.513	0.282	0.620	1.883	120.502***
9	Multiple	EW, B	K	-19.853	0.587	0.566	0.692	1.607	154.75***

EW - egg weight, B- egg maximum breadth, L- egg length, Dg- geometric mean diameter, V- egg volume, S- egg surface area, R^2 – coefficient of determination, SE- standard error of estimate, ***- Significant at $p<0.001$

Linear parametric equations associating hatchlings' weight and egg length (model 3) had the lowest coefficients of determination - 0.175 (line K) and 0.291 (line L) followed by those using egg breadth as predictor (model 2). However, when both dimensions were simultaneously included in the model, coefficients of determination increased considerably to 0.541 and 0.665 for lines L and K respectively (model 7). In line K the values were comparable with those of model 1, where the independent variable was egg weight. When the geometric mean diameter (Dg), egg volume (V) and surface area (S) in both lines were used as independent predictors (models 4, 5 and 6) the values of R^2 were lower than respective values in model 1 including also egg weight, which is more pronounced in line L. Our data confirmed the findings from a previous study of *Narushin et al. (2002)*, that linear equations using as predictor egg weight were more accurate that those using egg volume and surface area independently.

The high correlation coefficients between predictors egg weight, volume, surface area and mean geometric diameter (Table 2) presumed multicollinearity as confirmed by VIF values, significantly higher than allowed ones. It is acknowledged that models based on multicollinear variables could influence the accuracy of the prognosis (*Chatterjee et al., 2000*). An option for elimination of the negative impact of multicollinearity is the elimination of some strongly correlating predictors from the model through application of stepwise regression. The calculated coefficients of determination in multiple regression models by means of stepwise regression were 0.620 for line L and 0.692 for line K (models 8 and 9). The comparison with model 1, that uses one independent variable (egg weight), shows increase in the coefficient of determination when a second predictor was included, in other words, the addition of egg surface area (model 8) and egg breadth (model 9) contributed to a greater extent for explication of the dependent variable (hatchling weight) for line L and line K.

After evaluation of regression models, the most accurate (those with highest R^2 values) were selected – models 1 and 7 for both lines, model 8 (line L) and model 9 (line K) for validation of their prediction power. They served for calculation of predicted weights of day-old chicks for 60 randomly selected eggs from each line set for incubation. Table 4 presents the expected and observed values for weights of hatchlings. The differences between predicted ($\hat{Y}$) and observed (Y) values were small and for line L ranged between 0.04-0.48 g, while for line K - between 0.24-0.34 g, corresponding to 0.09-1.09 % and 0.63-0.86 % from respective real values.

Conclusion

On the basis of data, it could be concluded that the best prediction was obtained with an additional predictor when apart egg weight as followed: egg

surface area for line L (*ChW*=0.513*EW*+0.282*S*-10.345) and egg maximum breadth for line K (*ChW*=0.587*EW*+0.566B -19.853). Models on the basis of main egg dimensions – breadth and length could be also used for tentative determination of hatchlings'' weight from both lines. According to the study, multiple regression models were more efficient than single linear models.

Table 4. Mean of actual and predicted values, difference and percent difference for the models generated to predict the chick weight at hatch in line L and line K

Line	Chick weight (g)					Difference							
	Actual (Y) *	Predicted				(Y-$\hat{Y}_1$)		(Y-$\hat{Y}_7$)		(Y-$\hat{Y}_8$)		(Y-$\hat{Y}_9$)	
		Model 1 ($\hat{Y}_1$)	Model 7 ($\hat{Y}_7$)	Model 8 ($\hat{Y}_8$)	Model 9 ($\hat{Y}_9$)	g	%	g	%	g	%	g	%
L	44.18	43.70	44.14	43.83	-	0.48	1.09	0.04	0.09	0.35	0.79	-	-
K	39.52	39.27	39.18	-	39.28	0.25	0.63	0.34	0.86	-	-	0.24	0.61

*- chick weight was measured by digital balance

Regresijski modeli za procenu mase pilića na izleganju na osnovu određenih geometrijskih karakteristika jajeta

Nadya Mincheva, Mitko Lalev, Magdalena Oblakova, Pavlina Hristakieva

Rezime

Predviđanje telesne mase pre izleganja je važan element selekcije, čiji je cilj poboljšanje uniformnosti i produktivnosti živine. S tim u vezi, naš cilj bio je da razvijemo i procenimo optimalne modele za slično predviđanje u dve linije pilića: White Plymouth Rock - linija L i linija K, na osnovu težine jaja u inkubaciji i geometrijskih karakteristika jajeta - maksimalna širina jajeta (B), dužina jajeta (L), geometrijski srednji prečnik (Dg), volumen (V) i površina jajeta (S). Ukupno 280 jaja (140 iz svake linije) koja su izlegle kokoši uzrasta od 40 nedelja su odabrana nasumično. Za svaku liniju su određene aritmetičke vrednosti, standardna odstupanja i koeficijenti varijacije proučavanih parametara. Korelacioni koeficijenti između mase izleglih pilića i prediktora bili su najviši za težinu jajeta, geometrijski srednji prečnik, zapreminu i površinu jajeta (r=0,731-0,779 za liniju L; r=0,802-0,819 za liniju K).

Izrađeno je devet modela linearne regresije i procenjena njihova tačnost. Regresijske jednačine težine izleglih pilića prema dužini jajeta imale su najmanji koeficijent determinacije (0,175 za liniju K i 0,291 za liniju L), ali kada su dužina i širina jajeta ušla u model zajedno, vrednost koeficijenta se značajno povećala na 0,541 i 0,665 za linije L i K, respektivno. Masa jednodnevnih pilića iz linije L mogla se predvideti sa većom preciznošću sa modelom koji uključuje površinu

jajeta u odnosu na masu jajeta (*ChW*=0,513*EW*+0,282*S* – 10,345; R^2=0,620). U liniji K, postignuta je preciznija prognoza dodavanjem širine jajeta kao dodatnog prediktora za masu u modelu (*ChW*=0,587*EW*+0,566*V* -19,853; R^2=0,692). Studija je pokazala da su višestruki linearni regresioni modeli bili precizniji od pojedinačnih linearnih modela.

Ključne reči: masa pilića, masa jajeta, geometrijski parametri jajeta, regresijski modeli

References

ALABI O.J., NG`AMBI J.W., NORRIS D. (2012): Effect of Egg Weight on Physical Egg Parameters and Hatchability of Indigenous Venda Chickens. Asian Journal of Animal and Veterinary Advances, 7: 166-172.

ALTUNTAŞ E., SEKEROGLU A. (2008): Effect of egg shape index on mechanical properties of chicken eggs. Journal of Food Engineering 85(4):606-612.

CHATTERJEE S., HADI A.S., PRICE B. (2000): Regression analysis by example. John Wiley and Sons, New York, USA.

EL-SAFTY S.A. (2011): Using stepwise regression analysis to determine the factors affecting chick weight at hatch in ostrich (Struthio camelus). Egyptian Poultry Science 31 (4): (695-704).

FAROOQ M., DURRANI F.R., ALEEM M., CHAND N., MUQARRAB A.K. (2001): Egg traits and hatching performance of Desi, Fayumi and Rhode Island Red chicken. Pakistan Journal of Biological Sciences, 4 (7): 909-911.

IQBAL J., MUKHTAR N., REHMAN Z., KHAN S.H., AHMAD T., ANJUM M.S., PASHA R. H., UMAR S. (2017): Effects of egg weight on the egg quality, chick quality, and broiler performance at the later stages of production (week 60) in broiler breeders. Journal of Applied Poultry Research , 26 (2): 183-191.

IQBAL J., KHAN S.H., MUKHTAR N., AHMED T., PASHA R.A. (2016): Effects of egg size (weight) and age on hatching performance and chick quality of broiler breeder. Journal of Applied Animal Research Vol. 44 , 1, 54-64.

KABIR M.A., ISLAM M.S., DUTTA R.K. (2012): Egg morphometric analyses in chickens and some selected birds. University Journal of Zoology, Rajshahi University, 31:85-87

KHURSHID A., M. FAROOQ, F.R. DURRANI, K. SARBILAND CHAND, N. (2003): Predicting egg weight, shell weight, shell thickness and hatching chick weight of Japanese quails using various egg traits as regressors. International Journal of Poultry Science 2: 164-167

LOTFI A., SHADRYAR H.A., MAHERI-SIS N., ABEDI A.S., NAHAND M.K. (2011): Hatching characterizes of Japanese quail (Coturnix coturnix japonica) eggs

with different egg shape indexes. American-Eurasian Journal Agricultural & Environmental Sciences, 10 (3): 475-477.
MALAGO J.J., BAITILWAKE M.A. (2009): Egg traits, fertility, hatchability and chick survivability of Rhode Island Red, local and crossbred chickens. Tanzania Veterinary Journal, 26 (1): 24-36.
MBAJIORGU C. A., RAMAPHALA N.O. (2014): Insight into egg weight and its impact on chick hatch weight, hatchability and subsequent growth indices in chicken- a review. Indian Journal of Animal Research, 48 (3): 209-213.
MITROVIĆ S., DJERMANOVIĆ V., NIKOLOVA N. (2011): Phenotype correlation between age and major production and reproductive traits of heavy hybrid parental flock Ross 308. Macedonian Journal of Animal Science, 1, 2, 327–334.
MOHSENIN N. N. (1970): Physical properties of plant and animal material. Gordon and Breach, New York.
MUKHTAR N., KHAN S.H., ANJUM M.S. (2013): Hatchling length is a potential chick quality parameter in meat type chickens. World's Poultry Science Journal 69(4): 889-896.
NARUSHIN V.G. (1997): Non-destructive measurements of egg parameters and quality characteristics. World's Poultry Science Journal, 53, 141–153.
NARUSHIN V.G. (2005): Production, modeling and education: Egg geometry calculation using the measurements of length and breadth. Poultry Science, 84: 482-484.
NARUSHIN V.G., ROAMANOV M.N., BOGATYR V.P. (2002): Relationship between pre-incubation egg parameters and chick weight after hatching in layer breeds. Biosystems Engineering, 83 (3): 373–381.
NARUSHIN V.G., ROMANOV M.N. (2002): Egg physical characteristics and hatchability. World's Poultry Science Journal, 58: 297-303.
NEDOMOVÁ, Š., BUCHAR, J. (2014): Goose eggshell geometry. Research in Agricultural Engineering, Vol. 60, (3):100-106.
NG'AMBI J.W., THAMAGA M.W., NORRIS D., MABELEBELE M,, ALABI O.J. (2013): Effects of egg weight on hatchability, chick hatch-weight and subsequent productivity of indigenous Venda chickens in Polokwane, South Africa. South African Journal of Animal Science, 43 (Suppl. 1): 67-69.
OLUTUNMOGUN A.K., UMAR A.U., ADEYINKA I.A., ADEJOH-UBANI E.O., NWAGU B.I., ABDULAHI I., ABEKE F.O., SEKONI A.A., IBRAHIM M.A., OTU M.O., HARUNA H.M. (2017): Fertility, body weight and survivability of two lines of broiler stock under selection. Nigerian Journal of Animal Production, 19-27.
RAMAPHALA N.O., MBAJIORGU C.A. (2013): Effect of Egg Weight on Hatchability and Chick Hatch-weight of COBB 500 Broiler Chickens. Asian Journal of Animal and Veterinary Advances, 8: 885-892.

RASHID A., KHAN S.H., ABBAS G., AMER M.Y., KHAN M.J.A., NAVEED I. (2013): Effect of egg weight on hatchability and hatchling weight in Fayoumi, Desi and crossbred (Rhode Island Red X Fayoumi) chickens. Veterinary World, 6 9, 592.

RASHIDI M., GHOLAMI M. (2011): Modelling of egg mass based on some geometrical attributes. American-Eurasian Journal of Agricultural & Environmental Science 10 (1): 9-15.

SAATCI M., KIRMIZIBAYRAK T., AKSOY A.R., TILKI M. (2005): Egg weight, egg shape index and hatching weight and interrelationships among these traits in native Turkish geese with different coloured feathers. Turkish Journal of Veterinary and Animal Sciences, 29: 353-357.

SAHIN E., SENGOR E. YARDIMCI M., CETINGUL I.S. (2009): Relationship Between Pre-Incubation Egg Parameters from Old Breeder Hens, Egg Hatchability and Chick Weight. Journal of Animal and Veterinary Advances, 8: 115-119.

SHAFEY T.M., MAHMOUD A.H., MOHAMED A.A. (2014); Dealing with multicollinearity in predicting egg components from egg weight and egg dimension. Italian Journal of Animal Science, 13: 4, 3408.

SHALEV B.A., PASTERNAK H. (1995): Incremental changes in and distribution of chick weight with hen age in four poultry species. British Poultry Science, 36: 415–424.

TAHIR M., CERVANTES H., FARMER C.W., SHIM M.Y., PESTI G.M. (2011): Broiler performance, hatching egg, and age relationships of progeny from standard and dwarf broiler dams. Poultry Science 90: 1364–1370.

TRALDI A.B., MENTEN J.F.M., SILVA C.S., RIZZO P.V., PEREIRA P.W.Z., SANTAROSA J. (2011): What determines hatchling weight: breeder age or incubated egg weight? Revista Brasileira de Ciência Avícola, 13 (4): 283-285

TSERVENI-GOUSI A.S., YANNAKOPOULOS A.L. (1990): Quality characteristics of pheasant eggs and effect of egg weight and shell quality on chick weight. Archiv fur Geflugelkunde, 54: 54–56.

WOLANSKI N.J., RENEMA R.A., ROBINSON F.E., CARNEY V.L., FANCHER B.I. (2007): Relationships among egg characteristics, chick measurements and early growth traits in ten broiler breeder strains. Poultry Science., 86 (8): 1784-92.

YAKUBU A., OGAH D.M., BARDE R.E. (2008): Productivity and egg quality characteristics of free range naked neck and normal feathered Nigerian indigenous chicken. International Journal of Poultry Science, 7: 579-585.

YILMAZ A., CAGLAYAN T. (2008): Egg weight, egg shape index, hatching weight and correlations among these traits in Japanese quail (Coturnix coturnix japonica) with different colored plumages. Fırat University Veterinary Journal of Health Sciences; 22 (1): 5-8.

Received 4 July 2018; accepted for publication 8 August 2018

Biotechnology in Animal Husbandry 34 (3), p 335-344 , 2018 ISSN 1450-9156
Publisher: Institute for Animal Husbandry, Belgrade-Zemun UDC 637.04/.06'637.4
https://doi.org/10.2298/BAH1803335R

QUALITY AND CHEMICAL COMPOSITION OF EGGS AFFECTED BY REARING SYSTEM AND HEN'S AGE

Simeon Rakonjac[1], Snežana Bogosavljević-Bošković[1], Zdenka Škrbić[2], Miloš Lukić[2], Vladimir Dosković[1], Veselin Petričević[2], Milun D. Petrović D.[1]

[1]University of Kragujevac, Faculty of Agronomy Čacak, Cara Dušana 34, 32 000 Čačak, Serbia
[2]Institute of Animal Husbandry, Belgrade-Zemun, Autoput 16, 11080 Zemun, Serbia
Corresponding author: Rakonjac Simeon e-mail: simcepb@yahoo.com
Original scientific paper

Abstract: The aim of this study was to evaluate the effect of rearing system and hen's age on quality and chemical composition of eggs. The tested rearing systems were: cage, floor and organic (30 birds Isa Brown hybrid per group). Fifteen eggs per group were collected for analyses in each of three phases of the productive cycle (32, 48 and 72 weeks hen's age). In these samples were investigated external (egg weight and egg shape index), internal (albumen height, Haugh unit and yolk colour) and chemical composition of eggs (dry matter, minerals, protein and lipids content). The general conclusion is that the egg weight and Roche values were increased, while egg shape index, albumen height and Haugh unit were decreased with hen's age. Organic eggs had higher albumen height and more Haugh units in the 48 and 64 weeks compared to the floor and cage eggs. Yolk colour of the floor and cage hens most dependent on the hen's age, on the other hand - yolk colour of organic hens most dependent on the grass availability at the outlet. Eggs from the organic rearing system had a lower dry matter, proteins and lipid content as compared to the cage system.

Keywords: Laying hens, rearing systems, egg quality, chemical composition, hen's age.

Introduction

Although producers around the world are increasingly converting their facilities to cage-free, data from the *International Egg Commission (2016)* show that the majority of laying hens (89.7%) are housed in a cage system. The main reason why the cage system of laying hens is the most widespread in the world is that it is the most economical - it provides that the price of eggs is low and that product be accessible to all segments of the population. Egg consumption in Hungary, USA

and Argentina are more than 250, in Japan more than 300 and in Mexico more than 350 eggs per person per year *(International Egg Commission, 2015).*
However, in many countries, consumers demands are more focused on quality rather than price. In research of *Pavlovski et al*. (2011) states that the number od of consumers which were willing to pay a higher price by 10% for eggs of guaranteed and controlled quality or from free-range system increased from 46% in 1981. to 71.5% in 2011. In the report of *Committee for the Common Organisation of the Agricultural Markets (2018)*, it is stated that in the EU countries 18.4% of the hens have access to an outlet (free range and organic together).
Due to all these facts, the aim of this study was to evaluate the effect of rearing system and hen's age on quality and chemical composition of eggs.

Material and methods

The study was carried out in three rearing systems: cage, floor and organic. In each system, the same genotype of laying hens (Isa Brown hybrid) was housed - 30 birds per group.
Each bird in battery system had at its disposal 750 cm² floor area. In floor and organic system the stocking density was 2.5 birds/m^2. The organic layers also had about 5 m^2 per birds available outdoor area which was covered with grass and bushes and these hens were able to supplement their diets using vegetation and small creatures living in an outlet.
Cage and the floor laying hens were fed with the same standard commercial diets whose average chemical composition is shown in Table 1. In organic system, except in the facility, the feeders and drinkers were located in the outlet. It is important to note that the diet from organic hens was complete without additions of synthetic amino acids, vitamins and minerals, with the use of more 80% organically grown components. Its chemical composition is also shown in Table 1. In all of three experimental groups feed and water were available *ad libitum.*

Table 1. The chemical composition of diet from laying hens

	Cage and floor system	**Organic system**
Chemical composition	%	%
Dry matter	88.38	89.82
Crude proteins	16.79	16.82
Crude fats	5.15	4.31
Cellulose	4.82	4.29
Ash	12.52	12.68
Ca	3.72	3.43
Total P	0.71	0.81
Na	0.17	0.18
Lysine	0.79	0.80
Methionine+cystine	0.68	0.48
Metabolisable energy, MJ	11.5 MJ	11.3 MJ

Fifteen eggs per group were collected for analyses in each of three phases of the productive cycle (32, 48 and 72 weeks hen's age). In these samples (eggs were one day old) were investigated external, internal and chemical composition of eggs.
- Egg weight was measured on an electronic scale with accuracy of 10^{-2} g.
- Egg shape index, or short-axis to long-axis length ratio (%), was determined using a special device (B.V. Apparatenfabriek van Doorn, Holland).
- Albumen height was determined tripod micrometer (AMES, USA)
- Haugh units were calculated according to following formula: HU=100log ($H+7.57-1.7M^{0.37}$) where H = albumen height, mm, M = egg weight, g.
- Egg yolk colour was determined according to Roche yolk colour fan.

Analysis of the basic chemical composition of the eggs was performed based on SRPS ISO procedures (dry matter SRPS ISO 1442/1998, mineral materials SRPS ISO 936/1999, proteins SRPS ISO 937/1992, lipids 1444/1998).
Quality of eggs data were analysed by ANOVA and LSD test (*Stat Soft Inc Statistica For Windows, Version 7.0., 2006*).

Results and discussion

Egg quality parameters are presented in Table 2 shows that the egg weight was significantly influenced by hen's age ($p \leq 0.05$) while the effect of the rearing system was not significant ($p \geq 0.05$). Generally, egg weight was increased with hen's age, which is consistent with the results published by *Zita et al. (2009)* and *Rakonjac et al. (2017)*. It is important to note that the egg weight between the 32 and 48 weeks old hens differed significantly ($p \leq 0.05$), while in the later period (48-72 weeks) there were no differences ($p \geq 0.05$). That indicated that in an early period of laying there was increasing in egg weight but in later phases of egg production egg weight remain static (*Padhi et al., 2013*). On the other hand, the effect of rearing system on egg weight was not significant, similar to the results reported by *the Mugnai et al. (2009)* and *Rakonjac et al. (2017)*. Contrary to this, *Lolli et al. (2013)* and *Kralik et al. (2013)* found higher egg weight in free range than the cage system, while *Ferrante et al. (2009)* and *Lewko and Gornowicz (2011)* found the opposite - a higher egg weight in the cage than in the rearing systems with the outlet.

Egg shape index decreased with hen's age, and eggs from 32 week age hens had higher values of the index form of eggs from the 48 and 72 week age hens. As well as the weight of the eggs, the rearing system did not affect differences in this parameter ($p \geq 0.05$). *Škrbić et al. (2011)* also found that the egg shape index value decreased with hen's age ($r = - 0.15$). These results *Nikolova and Kocevski (2006)* were explained by the fact that the higher shape index of eggs from younger hens showed a presence of more eggs of rounded shape, while lower shape index of eggs from older layers showed more percentage of eggs of an

elongated shape which is typical for hens in deep age. Similar results were published and by *Zita et al. (2009)*. The rearing system did not influence differences in egg shape index, which is in agreement with the findings by *Đukić-Stojčić et al. (2009), Lewko and Gornowicz (2011), Ahhamed et al. (2014) and Dikmen et al. (2017).*

Table 2. Effect of the rearing systems and laying hen's age on egg quality parameters

Rearing systems	Hen's age		Egg weight (g)	Egg shape index	Albumen height (mm)	Haugh unit	Yolk colour (Roche)
Cage	32 week	$\bar{x}$	61.30 c	78.13 ab	7.42 ab	85.20 abc	9.07 f
		Sd	3.91	2.88	0.87	5.45	1.33
	48 week	$\bar{x}$	63.31 bc	75.93 cde	6.91 b	81.07 bc	10.60 de
		Sd	4.89	2.31	0.81	6.16	0.51
	72 week	$\bar{x}$	67.39 a	75.07 de	4.93 d	62.67 e	13.40 a
		Sd	5.75	1.71	1.04	12.08	0.51
Floor	32 week	$\bar{x}$	61.19 c	78.93 a	7.85 a	87.73 a	8.87 f
		Sd	3.15	2.63	1.12	5.97	0.92
	48 week	$\bar{x}$	66.86 ab	76.53 bcd	7.03 b	80.13 c	10.27 e
		Sd	5.78	2.90	1.26	9.09	0.96
	72 week	$\bar{x}$	67.03 a	75.93 cde	4.87 d	62.67 e	13.00 a
		Sd	5.45	2.31	1.07	10.31	0.65
Organic	32 week	$\bar{x}$	60.03 c	77.20 abc	7.62 ab	87.00 ab	11.47 bc
		Sd	4.53	2.01	0.91	6.00	0.64
	48 week	$\bar{x}$	65.54 ab	76.13 cde	8.07 a	88.07 a	11.13 cd
		Sd	5.84	1.92	1.39	8.42	0.64
	72 week	$\bar{x}$	65.63 ab	74.67 e	5.71 c	70.67 d	11.73 b
		Sd	4.44	2.64	1.03	9.62	0.46
P value							
Rearing system			0.427	0.078	0.005	0.003	≤0.001
Hen's age			≤0.001	≤0.001	≤0.001	≤0.001	≤0.001
Rearing system x Hen's age			0.421	0.846	0.159	0.240	≤0.001

a-f: Values within columns with different superscripts are significantly different (P≤0.05)

Haugh unit was decreased throughout the experimental period, due to the decreasing albumen height with hen's age. A significant effect of the age of hens on albumen height determined also *Ojedapo (2013)*. Our results that Haugh unit decreases with hen's age (p≤0.05) are consistent with the findings by *Škrbić et al. (2011)* and *Padhi et al. (2013)*. Effect of rearing systems on Haugh units was not significant in the 32 weeks (p≥0.05), but eggs from organic rearing system had more Haugh units in the 48 and 64 weeks compared to the floor and cage (p≤0.05). There are a numerous reasons why the organic eggs have a more Haugh units compared to eggs from the floor and the cage system: less stress in the oviduct tract

(*Castellini et al., 2006*), effect of the higher concentration of ammonia in cage and floor systems that enhances the albumen pH affecting thus its consistency (*Minelli et al., 2007*), the high vitamin C content in albumen influenced intake fresh grass from outlet (*Mugnai et al., 2009*).

Both studied factors and their interactions had a significant effect on the yolk colour ($p \leq 0.05$). Generally, Roche values increased in floor and cage system with the hen's age. On the other hand, the yolk colour of organic eggs was relatively constant throughout the entire experimental period. Increasing Roche value with the hen's age in our experiment is in accordance to research *Rizzi and Chiericato (2005)* and *Škrbić et al. (2011)*. Both groups - cage and floor consumed constant amount synthetic carotenoids in feed throughout the entire production cycle, and increasing intensity of yolk colour was a result of their greater synthesis in the body with hen's age. On the other hand, in organic laying hens, the intensity of yolks colour was much more dependent on season and availability of grass at the outlet (no synthetic colours in their feed).

Organic hens had available a significant amount of grass rich in carotenoids in an outlet in week 32, which caused a higher Roche values compared with a floor and cage laying hens ($p \leq 0.05$). At week 48, grass amount at the outlet was reduced, which caused that there were no significant differences between Roche values in all three investigated rearing systems ($p \geq 0.05$). Finally, at week 72, all the amount of the grass at the outlet was spent, and the floor and the cage produced eggs had a significant higher Roche values then organic ($p \leq 0.05$) - organic hens no longer had grass available as an additional source of carotenoids, while the floor and the cage hens received synthetic carotenoids through feed.

Table 3. Effect of the rearing systems and laying hen's age on the chemical composition of eggs

Rearing systems	Hen's age		Dry matter (%)	Minerals (%)	Protein (%)	Lipids (%)
Cage	32 week	$\bar{x}$	25.08 a	0.91 ab	12.70 a	9.82 ab
		Sd	1.37	0.07	0.53	1.46
	48 week	$\bar{x}$	24.27 abc	0.88 c	12.23 bc	9.45 abc
		Sd	0.37	0.04	0.29	0.44
	72 week	$\bar{x}$	24.67 ab	0.92 a	12.38 ab	10.02 a
		Sd	0.71	0.03	0.19	0.76
Floor	32 week	$\bar{x}$	23.75 bc	0.85 d	12.33 ab	8.61 c
		Sd	1.11	0.10	0.70	0.79
	48 week	$\bar{x}$	24.29 abc	0.88 c	12.11 bc	9.54 abc
		Sd	1.21	0.06	0.51	1.07
	72 week	$\bar{x}$	24.63 ab	0.90 abc	12.23 bc	9.83 ab
		Sd	1.62	0.04	0.43	1.69
Organic	32 week	$\bar{x}$	23.40 c	0.85 d	12.13 bc	8.62 c
		Sd	0.60	0.06	0.26	0.68
	48 week	$\bar{x}$	23.41 c	0.91 ab	11.82 c	9.12 abc
		Sd	1.17	0.04	0.39	1.32
	72 week	$\bar{x}$	23.82 bc	0.89 bc	12.31 ab	8.94 bc
		Sd	0.64	0.12	0.44	0.59
P value						
Rearing system			≤0.001	≤0.001	0.015	0.014
Hen's age			0.378	≤0.001	0.020	0.134
Rearing system x Hen's age			0.312	≤0.001	0.460	0.313

a-d: Values within columns with different superscripts are significantly different (P≤0.05)

Table 3 shows that the eggs from the organic rearing system had a lower dry matter content as compared to the other two systems (P≤0.05). The results similar to these are given by *Matt et al. (2009)*, which found the higher content of dry matter in cage eggs compared to organic.

Ash content was significantly influenced by studied factors and their interactions (P≤0.05). These results are in agreement with the results published by *Zhu et al. (2015)*, which found a significant difference in the content of a large number of minerals in eggs between the cage and the free-range layers.

Cage produced eggs had a higher content of protein and lipids compared to the organic eggs (P≤0.05). A significant effect of the rearing system on the content of protein and lipids in eggs was determined by numerous authors. *Mat et al. (2009)* obtained the similar results in their research, the cage eggs had higher

protein and lipids content compared to free range eggs. Also, *Pavlovski et al. (2011)* determined higher protein content, and *Radu-Rusu et al. (2014)* higher lipids content in cage eggs compared to free range.

Conclusion

Based on the results of these investigations it can be concluded that the rearing system and hen's age had a significant effect on egg shape index, albumen height, Haugh unit and yolk colour, while egg weight was influenced only by the hen's age. The interaction of the observed factors significantly influenced yolk colour. On the other hand, the rearing system had a significant effect in all four parameters of the chemical composition of eggs, while the significant effect of the hen's age on these parameters was established for mineral and lipids content. Rearing system x hen's age interaction significantly influenced only the mineral content in eggs.

Kvalitet i hemijski sastav jaja uzrokovan sistemom gajenja i starošću kokoši nosilja

Simeon Rakonjac, Snežana Bogosavljević-Bošković, Zdenka Škrbić, Miloš Lukić, Vladimir Dosković, Veselin Petričević, Milun D.Petrović

Rezime

Cilj ovog rada je bio da se ispita uticaj sistema gajenja i starosti na kvalitet i hemijski sastav jaja kokoši nosilja. Ispitivani sistemi gajenja su bili: kavezni, podni i organski (30 jedinki Isa Brown hibrida po grupi). Petnaest jaja po grupi je sakupljeno u tri faze proizvodnog ciklusa (32, 48 i 72 nedelje starosti nosilja). Na ovim uzorcima su ispitivani parametri spoljašnjeg (masa i indeks oblika) i unutrašnjeg kvaliteta jaja (visina belanca, Hogove jedinice i boja žumanca) kao i osnovni hemijski sastav jaja (sadržaj suve materije, minerala, proteina i masti). Generalno se može zaključiti da su se masa jaja i intenzitet obojenosti žumanca povećavali, a indeks oblika, visna belanca i Hogove jednice smanjivale sa starošću nosilja. Organska jaja su imala veću visinu belanca i više Hogovih jedinica u 48. i 64. nedelji u poređenju sa jajima proizvedenim u podnom i kaveznom sistemu. Boja žumanca je kod podno i kavezno gajenih jedniki najviše zavisla od starosti nosilja, dok je sa druge strane kod organskih kokoši boja žumanca najviše zavisila od dostupnosti trave na ispustu. Organsaka jaja su imala manji sadržaj suve materije, proteina i masti u poređenju sa jajima iz kaveznog sistema gajenja.

Ključne reči: kokoši nosilje, sistem gajenja, kvalitet jaja, hemijski sastav, starost.

Acknowledgement

This study was financed from funds of the Ministry of Education, Science and Technological Development of the Republic of Serbia, project No. TR-31033.

References

AHAMMED M., CHAE B.J., LOHAKARE J., KEOHAVONG B., LEE M.H., LEE S.J., KIM D.M., LEE J.Y., OHH S.J. (2014): Comparison of Aviary, Barn and Conventional Cage Raising of Chickens on Laying Performance and Egg Quality. Asian-Australas Journal of Animal Science, 27 (8), 1196-1203.

CASTELLINI C., PERELLA F., MUGNAI C., DAL BOSCO A. (2006): Welfare, productivity and qualitative traits of egg in laying hens reared under different rearing systems. Poster at: XII European Poultry Conference, 10-14 September 2006, Verona, Italy.

COMMITTEE FOR THE COMMON ORGANISATION OF THE AGRICULTURAL MARKETS (2018): EU Market Situation for Eggs.

DIKMEN B.Y., İPEK A., ŞAHAN Ü., SÖZCÜ A., BAYCAN S.C. (2017): Impact of different housing systems and age of layers on egg quality characteristics. Turkish Journal of Veterinary and Animal Sciences, 41, 77-84.

FERRANTE V., LOLLI S., VEZZOLI G., CAVALCHINI L.G. (2009): Effects of two different rearing systems (organic and barn) on production performance, animal welfare traits and egg quality characteristics in laying hens. Italian Journal of Animal Science, 8, 165-174.

ĐUKIĆ-STOJČIĆ M., PERIĆ L., BJEDOV S., MILOŠEVIĆ N. (2009): The quality of table eggs produced in different housing system. Biotechnology in Animal Husbandry, 25 (5-6), 1103-1108.

INTERNATIONAL EGG COMMISSION (2015): Annual Review.

INTERNATIONAL EGG COMMISSION (2016): Annual Review.

KRALIK Z., RADIŠIĆ Ž., GRČEVIĆ M., KRALIK G. (2013): Comparison of table eggs quality originating from hens kept in different housing systems. Proceedings of XV European Symposium on the Quality of Eggs and Egg Products and XXI European Symposium on the Quality of Poultry Meat / Petracci, Massimiliano (ed). - Bergamo: The Italian Branch of The World´s Poultry Science Association, 2013, 1-5.

LEWKO L., GORNOWICZ E. (2011): Effect of housing system on egg quality in laying hens. Annals of Animal Science, 11 (4), 607-616.

LOLLI A., HIDALGO C., ALAMPRESSE C., FERRANTE V., ROSSI M. (2013): Layer performances, eggshell characteristics and bone strength in three different housing systems. Biotechnology in Animal Husbandry, 29 (4), 591-606.
MATT D., VEROMANN E., LUIK A. (2009): Effect of housing systems on biochemical composition of chicken eggs. Agronomy Research, 7 (Special issue II), 662–667.
MINELLI G., SIRRI E., FOLEGATTI A., MELLUZZI A., FRANCHINI A. (2007): Egg quality traits of laying hens reared in organic and conventional systems. Italian Journal of Animal Science 6 (SUPPL. 1), 728-730.
MUGNAI C., DAL BOSCO A., CASTELLINI C. (2009): Effect of rearing system and season on the performance and egg characteristics of Ancona laying hens. Italian Journal of Animal Science, 88, 175-188.
NIKOLOVA N., KOCEVSKI D. (2006): Forming egg shape index as influenced by ambient temperatures and age of hens. Biotechnology in Animal Husbandry, 22 (1-2), 119-125.
OJEDAPO L.O. (2013): Effect of Age and Season on Egg Quality Traits of Isa Brown Layer Strain Reared in Derived Savanna Zone of Nigeria. Transnational Journal of Science and Technology, 3 (7), 48-60.
PADHI M.K., CHATTERJEE R.N., HAUNSHI S., RAJKUMAR U. (2013): Effect of age on egg quality in chicken. Indian Journal of Poultry Science, 48 (1), 122-125.
PAVLOVSKI Z., ŠKRBIĆ Z., LUKIĆ M. (2011): Free systems of rearing of chickens and layer hens: quality of meat and eggs. International 56th Meat Industry Conference, Tara, June 12-15th, 2011, 160-166.
RADU-RUSU R.M., USTROI, M.G., LEAHU A., AMARIEI S., RADU-RUSU C.G., VACARU-OPRIS I. (2014): Chemical features, cholesterol and energy content of table hen eggs from conventional and alternative farming systems. South African Journal of Animal Science, 44 (1), 33-42.
RAKONJAC S., BOGOSAVLJEVIĆ-BOŠKOVIĆ S., ŠKRBIĆ Z., PERIĆ L., DOSKOVIĆ V., PETROVIĆ M.D., PETRIČEVIĆ V. (2017): The effect of the rearing system, genotype and laying hens age on the egg weight and share of main parts of eggs. Acta Agriculturae Serbica, Vol. XXII, 44, 185-192.
RIZZI C., CHIERICATO G.M. (2005): Organic farming production. Effect of age on the productive yield and egg quality of hens of two commercial hybrid lines and two local breeds. Italian Journal of Animal Science, 4 (SUPPL. 3), 160-162.
STAT SOFT INC STATISTICA FOR WINDOWS, VERSION 7.0. (2006). Computer program manual Tulsa.
ŠKRBIĆ Z., PAVLOVSKI Z., LUKIĆ M., VITOROVIĆ D., PETRIČEVIĆ V., STOJANOVIĆ LJ. (2011): Changes of egg quality properties with the age of layer hens in traditional and conventional production. Biotechnology in Animal Husbandry 27 (3), 659-667.

ZITA L., TUMOVA E., ŠTOLC L. (2009): Effects of Genotype, Age and Their Interaction on Egg Quality in Brown-Egg Laying Hens. Acta Veterinaria Brno, 78, 85–91.
ZHU S.B., ZHAO Q.Y., LIU B.L., WANG L., LIU S.J. (2015): Variations in Yolk Mineral Element Contents from Different Chicken Rearing Systems: Eggs Analyzed by Inductively Coupled Plasma Mass Spectrometry. Advance Journal of Food Science and Technology, 7 (7), 530-533.

Received 23 August 2018; accepted for publication 17 September 2018

Biotechnology in Animal Husbandry 34 (3), p 345-353 , 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 631.554'633.15
https://doi.org/10.2298/BAH1803345M

EFFECT OF HARVESTING TIME ON FORAGE YIELD AND QUALITY OF MAIZE

Violeta Mandić[1*], Zorica Bijelić[1], Vesna Krnjaja[1], Aleksandar Simić[2], Maja Petričević[1], Nenad Mićić[1], Violeta Caro Petrović[1]

[1]Institute for Animal Husbandry, Department of Feed Science, Belgrade, Republic of Serbia
[2]Faculty of Agriculture, University of Belgrade, Crop science, Republic of Serbia
Corresponding author: violeta_randjelovic@yahoo.com
Original scientific paper

Abstract: Maize is the very important silage source in the world. Timely harvesting ensure high maize forage yield and quality. Therefore, the study focused on the effects of four harvesting times (starting at the 12 August every 7 days) on yield and qualitative parameters of forage green mass of maize hybrid ZP 677. The experiment was set in Vojvodina Province, Serbia, during the 2013 and 2014 growing seasons. Plant height, stem diameter, number of leaves per plant, ear percentage, forage yield, dry matter content and crude protein content were higher, while stem percentage was lower in 2014 with favorable climatic condition. Forage yield, crude protein content, ADF and NDF decreased, while dry matter content significantly increased with delay in harvesting. The maize hybrid should be harvested when the milk line is three-quarter of the way down the grain that is in the third decade of August.

Introduction

The maize harvested for silage is an important feed crop on livestock farms in the world. Maize silage is the most used for animal feed in order to increase productivity and animal performance. Large use of maize for silage stems from large production of green forage mass, high energy content of dry matter content and quality of the forage biomass *(Mandić et al. 2013),* low buffering power and high content of soluble carbohydrates *(Nussio et al., 2001).* Maize silage is an important source of energy and fiber for livestock *(Bernardes, 2012).* The forage harvested at the right time and well conserved is fundamental for high efficiency of cows and reducing concentrate feed consumption *(Kohoutek et al., 2010)*. However, nutritive quality of maize silage depends on the many factors, such as genotype, plant density, growing conditions, maturity and moisture of the crop at harvest *(Satter and Reis, 2012). Kung (2008)* stated that the optimum maturity of harvesting forages when the whole plant is at 32 to 35% dry matter content. In generally, the wet maize silage results in produce high concentrations of acetic acid

and nutrient losses caused by runoff. Contrary, dry maize silage (> 38-40% dry matter content) restricts fermentation, material is difficult to pack and fiber and starch digestibility are low.

In Serbia, maize silage production increased by 11% between 2010 and 2016 (from 27503 ha to 30524 ha), with the increase by 17.6% in the Vojvodina. However, maize for silage does not always achieve its potential due to unfavorable environment conditions, especially the amount and distribution of rainfall during growing season *(Mandić et al., 2017)*. The maize genotypes were strongly influence by silage nutritive value where crude fibre and NDF contents of the early maturing hybrids were lower than the mid-early and mid-late maturing hybrids *(Zeller and Schwarz, 2010)*. Also, these authors concluded that crude fibre and NDF contents increase with later harvest dates. *Latre et al. (2010)* found that the maize hybrids and harvesting date did not have effect on dry matter and sugar contents and density at harvest, as well as on dry matter, crude protein, starch content, crude fibre, sugar content, ash, pH, ammonia, lactic acid, volatile acids and alcohols at 50 days after ensiling. On another hand, *Nadeau et al. (2010)* reported that the chemical composition of the maize for silage varied according to maturity stage at harvest. Delaying of harvested of maize for silage increases dry matter content due to increase leaf dry matter content, and adversely affect quality because reduces sugar content and increases ADF, NDF and lignin content *(Kwabiah, 2005)*. The advance of maturity of maize reduces the crude protein content, NDF and ADF and increased in vitro true digestibility *(Darby and Lauer, 2002)*. *Shehzad et al. (2012)* concluded that the plant height, stem diameter, leaf area per plant, dry matter content, dry matter yield, green fodder yield and crude fiber content were increased, while crude fat, crude protein and ash contents were decreased with delayed harvesting time of maize. *Hatew et al. (2015)* found that the apparent total tract digestibility of dry matter, organic matter, crude protein, crude fat, starch, NDF and gross energy for growing dairy cows significantly decreased with maturity of maize crop.

The objective of our research was to determine forage yield and nutritive value of maize hybrid at four maturity stages under dry land farming in the Vojvodina region.

Materials and Methods

Field experiments were conducted during 2013 and 2014 years in dryland farming Pannonian region of Serbia, located at 45° 01′ N and 19° 33′ E. Maize hybrid ZP 677 (FAO maturity group 600) was tested. Preceding crop was winter wheat. Maize was planted with 70 cm inter-row spacing at 13 April in both years. The plant density was 59.000 plant ha^{-1}. Subplot size was 16.8 m^2. Plots were arranged in randomized complete blocks design in three replications. A standard agrotechnical measures were applied. NPK fertilizer 10:30:20 applied at the rate of

300 kg ha^{-1} after harvested of wheat in October. Nitrogen fertilizer KAN (27% N) was applied in May at the V6-V7 stage at a rate of 334 kg ha^{-1}.

Soil of experimental area was calcareous chernozem with pH in water of 7.12, having 16.45% $CaCO_3$, 3.64% of humus, 0.18% of total N, 17.81 mg 100 g^{-1} of P_2O_5 and 21% of K_2O at the depth of 30 cm.

Monthly average temperature in 2013 was higher for 0.8 °C, while monthly total rainfall was lower for 153 mm than in 2014 (429.0 mm and 18.3 °C, respectively), Figure 1. Dry period was in 2013 from mid of July to the harvested time.

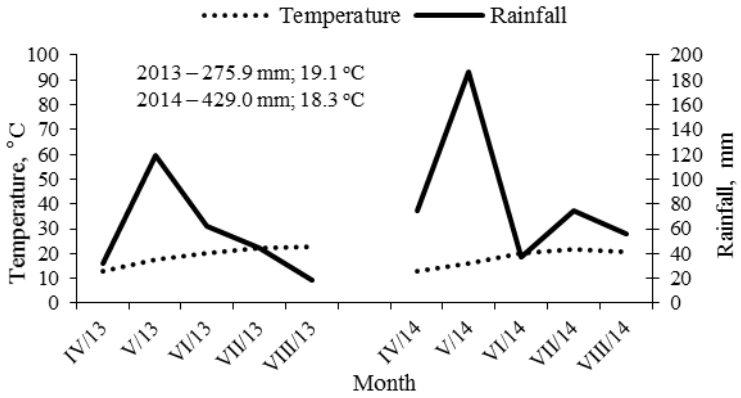


Figure 1. Climate diagram according to *Walter and Lieth (1967)* for Sremska Mitrovica, Serbia.

Plant morphological traits (plant height, stem diameter and number of leaves per plant) were recorded before cutting on 10 plants per subplot. After manual cutting on these plants the stem, leaf and ear ratio were determined. Forage yield was determined by cutting and chopping of plants using maize forage combine harvester, harvesting it in from two central rows from subplot and converted into kg ha^{-1}. Maize hybrids were harvested during four harvesting times, starting at the 12 August every 7 days. Dry matter content was determined by drying of 1 kg of forage mass from each subplot at 105°C and converted into kg ha^{-1}. The crude protein content was determined by the methods of Kjeldahl *(AOAC, 1990)*, while neutral detergent fibre (NDF) and acid detergent fibre (ADF) by methods of *Van Soest et al. (1991)*.

The experimental data were subjected to ANOVA using STATISTICA (version 10; StatSoft, Tulsa, Oklahoma, USA) at significance levels at $p \leq 0.01$ and $p \leq 0.05$. Significance between means were tested using Duncan Multiple Range Test at $P \leq 0.05$ level.

Results

The year affect on plant height, stem diameter, number of leaves per plant, stem percentage and ear percentage (Table 1). Plant height (265.2 cm), stem diameter (2.36 cm), number of leaves per plant (14.5) and ear percentage (25.4%) were higher in 2014 with favorable climatic condition compared to 2013 with unfavorable climatic condition (255.8 cm, 2.25 cm, 14.0 and 19.2%, respectively). Contrary, stem percentage (52.6%) was higher in 2013.

The harvesting time and year × harvested time interaction did not significantly affect on mentioned parameters.

Table 1. Year and harvesting time effect on agronomic performance of maize hybrid

Factor	Plant height (cm)	Stem diameter (cm)	Number of leaves per plant	Stem percentage (%)	Leaf percentage (%)	Ear percentage (%)
Year (Y)						
2013	255.8^{b}	2.25^{b}	14.0^{b}	52.6^{a}	28.2	19.2^{b}
2014	265.2^{a}	2.36^{a}	14.5^{a}	44.8^{b}	29.8	25.4^{a}
F test	**	*	**	**	ns	**
Harvesting time (HT)						
12.08.	260.8	2.28	14.3	49.6	28.9	21.5
19.08.	262.3	2.30	14.1	49.8	29.0	21.2
26.08.	260.9	2.34	14.4	48.7	28.9	22.4
02.09.	258.0	2.31	14.1	46.8	29.2	24.0
F test	ns	ns	ns	ns	ns	ns
Interactions						
Y × HT	ns	ns	ns	ns	ns	ns
M	260.5	2.31	14.2	48.7	29.0	22.3

Means followed by the same letter within a column are not significantly different according to Duncan's Multiple Range test ($p \leq 0.05$); *, ** - Significant at the 0.05 and 0.01 probability levels, respectively; ns - non-significant.

The year affect on forage yield, dry matter and crude protein contents (Table 2). Forage yield (58913 kg ha^{-1}), dry matter content (33.66%) and crude protein content (9.23%) were higher in 2014 compared to 2013 (52884 kg ha^{-1}, 32.26% and 8.82%, respectively).

The harvesting time significantly affect on yield and quality of forage mass. Forage yield, crude protein content, ADF and NDF significantly decreased with delayed harvesting time. Dry matter content significantly increased with delayed harvesting time.

Year × harvested time interactions were significant for forage yield, dry mater content, crude protein content and ADF.

Table 2. Year and harvesting time effect on forage yield and quality of maize hybrids

Factor	Forage yield (kg ha^{-1})	Dry mater content (%)	Crude protein content (%)	ADF (%)	NDF (%)
Year (Y)					
2013	52844^{b}	32.26^{b}	8.82^{b}	28.00	57.34
2014	58913^{a}	33.66^{a}	9.23^{a}	27.83	55.75
F test	**	**	**	ns	ns
Harvesting time (HT)					
12.08.	67448^{a}	24.65^{d}	9.68^{a}	30.63^{a}	61.83^{a}
19.08.	58637^{b}	29.38^{c}	9.70^{a}	28.90^{b}	58.87^{b}
26.08.	52175^{c}	36.20^{b}	8.97^{b}	27.89^{c}	55.85^{c}
02.09.	45254^{d}	41.62^{a}	7.74^{c}	24.24^{d}	49.63^{d}
F test	**	**	**	**	**
Interactions					
Y × HT	**	**	**	**	ns
M	55879	32.96	9.02	27.91	56.55

Means followed by the same letter within a column are not significantly different according to Duncan's Multiple Range test ($p \leq 0.05$); ** - significant at the 0.01 probability level; ns - non-significant.

Discussion

Forage yield, dry matter and quality characteristics are important parameters in choosing the maize hybrid to be ensiled. In generally, the chemical composition and fermentation quality of maize silage depends on the nutritional value of hybrid. Our results showed that the hybrid ZP 677 had higher plant height, stem diameter, number of leaves per plant, ear percentage, forage yield, dry mater and crude protein content in 2014 than in 2013. Favorable climatic condition in 2014 resulted in a high values of these parameters. Climate diagram according to *Walter and Lieth (1967)* showed that dry period was not in 2014. The amount of rainfall was higher for 153 mm and monthly average temperature was lower for 0.8 ^{o}C in 2014 than in 2013 (275.9 mm and 18.3 ^{o}C). Contrary, in 2013 dry period were in July at the stage of flowering and in August at the stage of grain filling (from July to harvested time), which explains a smaller percentage of ear in whole-plant forage yield. Also, study of *Mandić et al. (2015)* showed that the dry period, high temperature and low rainfall, from June to August reduced forage yield and dry matter yield of maize. However, stem percentage was lower in 2014, although are plant height and stem diameter were significantly higher. This trend can be explained because of negative correlation between stem ratio and plant height and stem diameter, as stated by study of *Carpici and Celik (2010)*. It is recommended that farmers are engage in hybrid maize forage production with high share of grain in whole plant forage yield because of increases the palatability, energy level, and

digestibility of fodder maize *(Wolf et al., 1993)*. According to *Gaafar et al. (2018)* hybrids should have ear percentage over 35% in order to farmers produce maize on a profitable basis. In our study, the hybrid had lower ear percentage than 35%. Similar, *Gaafar et al. (2018) and Saiyad and Kumar (2018)* reported that maize fodder yield and quality traits depends on the genetic variability and interaction of genotypes with environment conditions.

The farmers have a goal to produce high yield of maize silage hybrids with good quality. Therefore it is very important to determine the optimum harvest date. Also, this process is crucial factor for minimize losses during silage storage and feed-out phases. Our results showed that the yield and nutritive value of maize for silage depends on the degree of crop maturity at harvest time. Thus, maize hybrid had lower forage yield, crude protein content, ADF and NDF and higher dry matter content with the harvest delay in September. The process of nutrient translocation from stem to grain with the maturation of plants is intensified. Also, leaf and husk become dry and brittle. According to *Milašinović-Šeremešić et al. (2017)*, the Serbian maize hybrids for ensiling have dry matter content between 32.40% and 38.23%. In our case, dry matter content in average for years varied between 24.65% (first harvested time) and 41.62% (fourth harvested time). Accordingly, the silage harvested in first time would be susceptible to effluent losses because biomass is too wet. In addition, there is smaller grains formed on the ear which are a source of starch and energy. Contrary, maize harvested in fourth time had high dry matter content and are more difficult to compress. For this reason, the third harvesting time was optimal. *Shinners (2007)* pointed that delayed harvesting time of forage maize results in a significant loss of leaves, husk and upper stem and reduce moisture of maize stover biomass. The study of *Gaile (2008)* showed that the maize forage yield significantly differ among years and that delayed harvest resulted in reductions in NDF, ADF and crude protein, and in increasing of dry matter content and ear percentage.

Conclusion

The hybrid ZP 677 had good nutritional and production characteristics and can be are recommended for silage. The agronomic performance of maize is more dependent on the environmental conditions during growing season. Contrary, yield and quality of forage is more dependent on the harvested time. The advance of maturity reduces the forage yield and quality of the whole plant. Optimum harvesting time of maize hybrids is in the third decade of August at three-quarter milk line stage.

Acknowledgment

The research was supported by the Ministry of Education, Science and Technological Development of Republic of Serbia, project TR 31053.

Uticaj vremena žetve na prinos i kvalitet silokrme kukuruza

Violeta Mandić, Zorica Bijelić, Vesna Krnjaja, Aleksandar Simić, Maja Petričević, Nenad Mičić, Violeta Caro Petrović

Rezime

Kukuruz je veoma važan izvor silaže u svetu. Pravovremenom žetvom kukuruza obezbeđuje se visok prinos i kvalitet krme za silažu. Stoga je istraživanje fokusirano na uticaj četiri datuma žetve (počev od 12. avgusta svakih sedam dana) na prinos i kvalitativne parametre silokrme hibrida kukuruza ZP 677. Eksperiment je postavljen u Vojvodini, u Srbiji, tokom 2013. i 2014. godine. Visina biljke, prečnik stabla, broj listova po biljci, udeo stabla, prinos krme, sadržaj suve materije i sadržaj sirovih proteina bili su veći, dok je udeo stabla bio manji u 2014. godini sa povoljnim klimatskim uslovima. Prinos krme, sadržaj sirovih proteina, ADF i NDF su se smanjivali, dok se sadržaj suve materije povećavao sa kašnjenjem u žetvi. Hibrid kukuruza za krmu treba sakupljati kada je mlečna linija tri četvrtine visine zrna što je u trećoj dekadi avgusta.

References

AOAC (1990): Association of Official Analytical Chemists (AOAC), 1990. Official methods of analysis, Washington DC, USA, 1, 14, 684.

BERNARDES T.F. (2012): Levantamento das práticas de produçã o e usode silagens em fazendas leiteiras no Brasil. Universidade Federal de Lavras, Lavras (E-book).

CARPICI E.B., CELIK N. (2010): Determining possible relationships between yield and yield-related components in forage maize (*Zea mays* L.) using correlation and path analysis. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 38, 280-285.

DARBY H.M., LAUER J.G. (2002): Harvest date and hybrid influence on corn forage yield, quality, and preservation. Agronomy Journal, 94, 559-566.

GAAFAR H.M.A., RIAD W.A., EL-ESAWY G.S. (2018): Nutritional and economical evaluation of Watania corn hybrids silages. Journal of Animal and Poultry Production Mansoura University, 9, 4, 211-218.
GAILE Z. (2008): Harvest time effect on yield and quality of maize (*Zea Mays* L.) grown for silage. Latvian Journal of Agronomy, 10, LLU, 104-111.
HATEW B., BANNINK A., VAN LAAR H., DE JONGE L. H., DIJKSTRA J. (2016): Increasing harvest maturity of whole-plant corn silage reduces methane emission of lactating dairy cows. Journal of Dairy Science, 99, 1, 354-368.
KOHOUTEK A., ČERNOCH V., KOMÁREK P., NERUŠIL P., ODSTRČILOVÁ V. (2010): Production ability impact of temporary grasslands on forage quality. 14th International symposium forage conservation, Brno, Czech Republic, 17-19 March 2010, 28-30.
KUNG L. (2008): Update on forage management. Four-state dairy nutrition and management conference, Dubuque, Iowa, 11 – 12 June 2008, 15-18.
KWABIAH A. B. (2005): Frost and harvest date effects on yield and nutritive value of silage maize (*Zea mays* L.) in a short-season environment. Journal of New Seeds, 7, 3, 15-29.
LATRE J., WAMBACQ E., DE BOEVER J., DE BRABANDER D., MAENHOUT S., DEROO B., COOPMAN F., HAESAERT G. (2010): Effects of variety type and maturity at harvest on whole crop maize silage characteristics. 14th International symposium forage conservation, Brno, Czech Republic, 17-19 March 2010, 89-91.
MANDIĆ V., BIJELIĆ Z., KRNJAJA V., RUŽIĆ MUSLIĆ D., CARO PETROVIĆ V., OSTOJIĆ ANDRIĆ D., PETRIČEVIĆ M. (2017): Forage maize yield in function of rainfall in climatic conditions of Vojvodina (Republic of Serbia). Scientific Papers. Series A. Agronomy, LX, 491-494.
MANDIĆ V., KRNJAJA V., BIJELIĆ Z., TOMIĆ Z., SIMIĆ A., STANOJKOVIĆ A., PETRIČEVIĆ M., CARO-PETROVIĆ V. (2015): The effect of crop density on yield of forage maize. Biotechnology in Animal Husbandry, 31, 4, 567-575.
MANDIĆ V., SIMIĆ A., TOMIĆ Z., KRNJAJA V., BIJELIĆ Z., MARINKOV G., STOJANOVIĆ LJ. (2013): Effect of drought and foliar fertilization on maize production. Proceedings of the 10th international symposium modern trends in livestock production, Belgrade, Serbia, 2-4 October 2013, 416-429.
MILAŠINOVIĆ ŠEREMEŠIĆ M., RADOSAVLJEVIĆ M., TERZIĆ D., NIKOLIĆ V. (2017): The utilisable value of the maize plant (biomass) for silage. Journal on Processing and Energy in Agriculture 21, 2, 86-90.
NADEAU E., RUSTAS B-O., ARNESSON A., SWENSSON C. (2010): Maize silage quality on swedish dairy and beef farms. 14th International symposium forage conservation, Brno, Czech Republic, March 17-19, 2010, 195-197.
NUSSIO, L.G.; CAMPOS, F.P.; DIAS, F.N. (2001): Importância da qualidade da porção vegetativa no valor alimentício da silagem de milho. Simpósio sobre produção e utilização de forragens conservadas, Maringá, 127-145.

SAIYAD M. M., KUMAR, S. J. (2018): Evaluation of maize genotypes for fodder quality traits and SSR diversity. Journal of Plant Biochemistry and Biotechnology, 27, 78-89.
SATTER L.D., REIS R.B. (2012): Milk production under confinement conditions. Accessed on: Sept 24, 2018.
SHEHZAD M.A., MAQSOOD M., BHATTI M.A., AHMAD W., SHAHID M.R. (2012): Effects of nitrogen fertilization rate and harvest time on maize (*Zea mays* L.) fodder yield and its quality attributes. Asian Journal of Pharmaceutical and Biological Research, 2, 19-26.
SHINNERS K.J., BINVERSIE B.N. (2007): Fractional yield and moisture of corn stover biomass produced in the Northern US Corn Belt. Biomass Bioenergy, 31, 8, 576-584.
VAN SOEST P.J., ROBERTSON J.B., LEWIS B.A. (1991): Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science, 74, 3583-3597.
WALTER H., LIETH H. (1967): Klimadiagram-Weltatlas. VEB Gustav Fischer Verlag, Jena, Germany.
WOLF D.P., COORS J.G., ALBRECHT K.A., UNDERSANDER D.J., CARTER P.R. (1993): Agronomic evaluations of maize genotypes selected for extreme fiber concentrations. Crop Science, 33, 1359-1365.
ZELLER F., SCHWARZ F.J (2010): Chemical composition and feeding value of maize residual plants in different varieties. 14th International symposium forage conservation, Brno, Czech Republic, 17-19 March 2010, 12-14.

Received 25 August 2018; accepted for publication 26 September 2018

Biotechnology in Animal Husbandry 34 (3), p 355-367 , 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 636.085.1/.2'633.2.03/.31
https://doi.org/10.2298/BAH1803355B

BASIC CHEMICAL COMPOSITION AND ENERGY NUTRITIONAL VALUE OF FODDER BIOMASS FROM ARTIFICIAL ECOSYSTEMS

Tatyana Bozhanska, Boryana Churkova, Teodora Mihova

Research Institute of Mountain Stockbreeding and Agriculture, 5600 Troyan, Bulgaria
Corresponding author: Tatyana Bozhanska, tbozhanska@mail.bg
Original scientific paper

Abstract: In 2014-2016 at the Research Institute of Mountain Stockbreeding and Agriculture- Troyan, we followed the change in the qualitative composition of dry biomass from two-component grass mixtures. The grasslands with the mixture of *Trifolium repens* L. - *Poa pratensis* L. have the highest crude protein content (151.6 g kg^{-1} DM), mineral substances (75.6 g kg^{-1} DM) and crude fat (33.9 g kg^{-1} DM). The legume grass associated with perennial ryegrass forms forage matter with the highest carbohydrate amount (434.9 g kg^{-1} DM) and the lowest amount of crude fiber (269.8 g kg^{-1} DM). The biomass of *Lotus corniculatus* L. and *Festuca rubra* L. is the richest in Ca (19.1 g kg^{-1} DM), and the mixture of blue hybrid alfalfa - cock's foot has the highest dry matter content (909.7 g kg^{-1} DM). The biomass of blue hybrid alfalfa - cock's foot (16.99 MJ/kg DM) and red clover - meadow fescue (16.96 MJ/kg DM) is with the highest caloric values. Both mixtures have almost identical values regardless of the different botanical composition and the predominant component in the grassland. The mixture of *Trifolium pratense* L. with *Phleum pratense* L. has the highest energy nutritional value (FUM - 0.71 kg DM and FUG - 0.66 kg DM), and the biomass of the blue hybrid alfalfa - cock's foot registered the lowest amount of exchange energy (7.30 MJ/kg DM), as well as the smallest number of feed units (FUM – 0.67 kg DM and FUG – 0.61 kg DM) in the dry matter. For the experimental period with the lowest values of gross energy (16.70 MJ/kg DM) are the mixed grasslands of *Trifolium repens* L.

Key words: legume and grass mixtures, fodder quality

Introduction

In mountainous and foothill areas of Bulgaria, artificial hay and pasture ecosystems are an important feed for ruminants. The sowing of two-component mixtures, consisting of medium perennial and grass meadows species *(Churkova, 2007; Naydenova, 2008; Mitev and Naydenova, 2008; Mihovski and Sabeva, 2011)*

are a factor for obtaining high yield and quality biomass *(Stosic et al., 2003; Lazarevic et al., 2004; Vasileva, 2011)*. An important advantage in mixed cultivation of forage grasses is species diversity and balanced grassland quality. Alfalfa, bird's-foot-trefoil, red and white clover are the legume components used to create temporary meadow and grazing grasslands in Bulgaria *(Mihovsky, 1995)*. Meadow grasses are a major resource for feeding animals in the natural or artificial ecosystem. Therefore, the exact analysis of the main chemical composition *(Mihovski and Goranova, 2006)* is essential for the evaluation of nutritional value and effective use *(Naydenova and Katova, 2013; Bozhanska, 2017a)* of the obtained feed. In this connection, *Slavkova et al. (2017)* report a direct relationship between the general body state of ruminants and the protein, calcium and phosphorus content of grazing grassland.

The composition of dry matter in mixed crops is characterized by a lower content of fiber components compared to individual grass and legume monocultures *(Naydenova et al., 2015)*, and the energy value of grass biomass gives a real estimate of the productive potential and a balance between the quantity and the quality of components in artificial grass associations *(Bozhanska, 2017b)*.

The purpose of this study is to assess the quality of feed biomass derived from artificial ecosystems, including basic chemical composition and energy nutritional value of two-component grasslands.

Material and Methods

The experiment was set in the spring of 2014 at the Research Institute of Mountain Stockbreeding and Agriculture - Troyan, using the block method in four replications with the size of the harvest plot of 5 m^2. The study period is three years. Experimental variants are:

1. Bird's-foot-trefoil (*Lotus corniculatus* L.) + Red fescue (*Festuca rubra* L.)
2. White clover (*Trifolium repens* L.) + Perennial ryegrass (*Lolium perenne* L.)
3. White clover (*Trifolium repens* L.) + Kenthucky bluegrass *(Poa pratensis* L.)
4. Red clover (*Trifolium pratense* L.) + Timothy-grass (*Phleum pratense* L.)
5. Blue hybrid alfalfa (*Medicago sativa* L.) + Cock's foot (*Dactylis glomerata* L.)
6. Red clover (*Trifolium pratense* L.) + Meadow fescue (*Festuca pratensis* L.)

Soil preparation includes autumn plowing of self-sown fallow ground at a depth of 22-25 cm and repeated spring disc, milling and rolling before and after sowing.

The sowing of the field experiment was carried out manually, scattered, at the optimum agro-technical period (middle of March) on a pre-created solid bed at a depth of 0.5-1.0 cm. Sowing rates were calculated on the basis of 100% seed germination as for bird's-foot-trefoil and white clover is 1.2 kg/da, for red clover - 1.5 kg/da, and for blue hybrid alfalfa - 2.5 kg/da. Grasses in the grass mixtures are sown with a 2.5 kg/da sowing rate. The ratio of components in mixtures is 50:50. Immediately after sowing, sown areas are rolled for better seed contact between

soil and seeds and their joint germination.
Seeds of the most common cultivars of legumes and meadow grasses were used: Bird's-foot-trefoil – cv. *'Leo'*, White clover – cv. *Huia*, Red clover – cv. *'Altaswede'*, Blue hybrid alfalfa, a local population of Troyan origin and Red fescue – cv. *'Ryder'*, Meadow fescue – cv. *'Laura'*, Cock's foot – cv. *'Loke'*, Kenthucky bluegrass – cv. *'Sobra'*, Timothy-grass – cv. *'Erecta'* and Perennial ryegrass – cv. *'Belida'*.
Fighting weeds during vegetation is mechanical in order to avoid further chemical interference with plants.
The grasslands were cut in the beginning of the blossoming phase of the legumes and tasseling/ear formation of grass crops.
The chemical composition of the dry feed is analyzed according to *Weende* analysis: Crude protein (CP, g kg^{-1}) according to *Kjeldahl* (according to BDS/ISO-5983); Crude fiber (CF, kg kg^{-1}); Crude fat (Cft, g kg^{-1}) (according to BDS/ISO-6492) - by extraction into a *Soxhlet* extractor; Ash (g kg^{-1}) - (according to BDS/ISO-5984) degradation of the organic matter by gradual burning of the sample in a muffle furnace at 550°C; Dry matter (DM, g kg^{-1}) - empirically calculated from % moisture; Nitrogen-free extractable substances (NFE)=100-(CP, %+CF, %+Cft, %+Ash, %+Moisture, %) converted to g kg^{-1}; Calcium (Ca, g kg^{-1}) - Stotz (Complexometric) and Phosphorus (P, g kg^{-1}) - with vanadate-molybdate reactive according to Gericke and Kurmis - spectophotometer (agilent 8453 UV - visible Spectroscopy System) measuring in the area of 425 ηm.
The nutritional value of the feed was assessed by the Bulgarian system as Feed Unit for Milk (FUM) and Feed Unit for Growth (FUG) and calculated on the basis of equations according to the experimental values of CP, CF, Cft and NFE, recalculated by the coefficients for digestibility by *Todorov (2010)*: Gross energy (GE, MJ/kg DM)=0,0242*CP+0,0366*Cft+0,0209*CF+0,017*NFE-0,0007*Zx and Exchangeable energy (EE, MJ/kg DM)=0.0152*DP (Digestible protein)+0.0342*Dft (Digestible fat)+0.0128*DF (Digestible fibers)+0.0159*DNFE (Digestible Nitrogen-free extractable substances)-0.0007*Zx.
Data was analyzed with ANOVA (Anova: Two-Factor Without Replication) and means comparison with a Least Significant Difference at 5% level of probability (LSD at $P<0.05$). The studied sources of variation included: the composition of the components in the mixtures, the conditions of the year and the age of the grass.

Results and discussion

Basic chemical composition of perennial grass-legume mixtures.
The chemical composition of the feed gives a real idea of its nutritional value. The most prominent in this regard is the crude protein content in the dry matter composition. The data from the chemical analysis (Table 1) in the year of sowing

indicate that the values of this quality index are highest in the feed of the mixture of bird's-foot-trefoil - red fescue (165.2 g kg^{-1} DM) and the lowest in the grassland of red clover with timothy-grass (137.9 g kg^{-1} DM).

Table 1. Basic chemical composition of perennial grass-legume mixtures by years and average for the period 2014-2016 (g kg^{-1} DM)

Variants	DM	CP	Cft	CF	Ash	NFE	Ca	P
2014								
Bird's-foot-trefoil+Red fescue	874.8	165.2	39.3	287.8	64.0	318.5	20.6	2.0
White clover+Perennial ryegrass	871.4	162.5	40.4	279.1	65.5	323.9	16.1	0.9
White clover+Kenthucky bluegrass	855.7	139.0	41.8	260.8	71.0	343.1	14.3	2.4
Red fescue+Ttimothy-grass	863.3	137.9	37.8	255.3	53.0	379.3	15.3	2.2
Blue hybrid alfalfa+Cock's foot	879.2	145.2	49.5	282.4	77.7	324.4	18.2	1.3
Red fescue+Meadow fescue	872.3	141.0	54.0	262.0	90.3	325.0	20.2	1.6
2015								
Bird's-foot-trefoil+Red fescue	929.3	127.8	22.0	310.1	74.5	400.3	9.0	1.9
White clover+Perennial ryegrass	913.8	106.8	25.4	313.6	78.0	396.7	4.8	4.1
White clover+Kenthucky bluegrass	905.9	156.5	27.8	227.5	89.0	413.5	11.5	2.7
Red fescue+Ttimothy-grass	921.9	109.3	20.4	296.2	72.1	429.6	4.8	4.3
Blue hybrid alfalfa+Cock's foot	925.1	99.6	15.2	391.3	64.9	359.0	4.8	3.3
Red fescue+Meadow fescue	918.7	124.2	14.8	331.1	62.6	391.1	9.1	3.5
2016								
Bird's-foot-trefoil+Red fescue	910.9	119.3	22.2	322.0	76.4	371.1	27.7	4.7
White clover+Perennial ryegrass	910.1	163.8	17.4	216.6	77.4	434.9	25.5	5.1
White clover+Kenthucky bluegrass	903.4	159.4	32.2	340.9	66.7	304.2	16.8	4.5
Red fescue+Ttimothy-grass	916.5	121.1	8.8	296.9	65.2	424.5	14.4	5.3
Blue hybrid alfalfa+Cock's foot	924.7	99.8	14.6	365.2	68.7	376.4	9.9	4.5
Red fescue+Meadow fescue	906.2	138.7	32.2	350.6	65.1	319.5	13.5	5.5
2014-2016								
Bird's-foot-trefoil+Red fescue	905.0	137.4	27.8	306.6	71.6	371.1	19.1	2.8
White clover+Perennial ryegrass	898.4	144.4	27.7	269.8	73.6	434.9	15.5	3.4
White clover+Kenthucky bluegrass	888.3	151.6	33.9	276.4	75.6	304.2	14.2	3.2
Red fescue+Ttimothy-grass	900.6	122.8	22.3	282.8	63.4	424.5	11.5	3.9
Blue hybrid alfalfa+Cock's foot	909.7	114.9	26.4	346.3	70.4	376.4	11.0	3.1
Red fescue+Meadow fescue	899.1	134.6	33.7	314.6	72.7	319.5	14.3	3.5
Mean	***900.2***	***134.3***	***28.6***	***299.4***	***71.2***	***371.8***	***14.3***	***3.3***
SD	***7.2***	***13.6***	***4.5***	***28.9***	***4.2***	***53.1***	***2.9***	***0.4***
$LSD_{0,05}$	***6.2***	***23.0***	***9.1***	***53.0***	***13.9***	***41.0***	***5.7***	***0.9***

The relatively high protein values in the first vegetation can be explained by the

fact that the yield is mainly formed from high protein legumes. In a second vegetation, the indicator levels range from 99.6 g kg^{-1} DM to 156.5 g kg^{-1} DM and are significantly lower for all variants (exception makes the mixture of white clover - Kenthucky bluegrass) because of the higher participation of the grasses in the grassland and respectively in the cut forage mass. In the third experimental year, the limited participation of alfalfa (4.6%) in the cock's foot variant determined the grassland with the lowest crude protein (99.8 g kg^{-1} DM), and the legume crop share (81.5%) in dry white clover and perennial ryegrass biomass increased the protein content of the mixture to 163.8 g kg^{-1} DM.

In the year of sowing, the protein content of the mixture of white clover with perennial ryegrass (162.5 g kg^{-1} DM) is 23.5 g kg^{-1} DM higher than in its mixture with Kenthucky bluegrass (139.0 g kg^{-1} DM). The values obtained relate to the type of the grass component and its contribution to grassland, as well as to the dry matter content of the reduced biomass from both mixtures.

The perennial ryegrass is a grass forage crop with great competitive ability and fast development. In the first experimental year, the plant biomass prevails over the legume representative. Quite the opposite is the ratio between grasses: legumes in the mixture of white clover - Kenthucky bluegrass, where the share of *Trifolium repens* L. is 21.43% above that of the Kenthucky bluegrass. The levels for crude protein content in both grasses, which were found in foreign studies *(Lemežienė et al., 2007; Bostan et al., 2010)*, also differ for ryegrass is about 9% and for Kenthucky bluegrass is over 14%. Interestingly, the high protein content in the mixture of white clover - perennial ryegrasss blend is associated with a higher percentage of crude fiber in the dry matter, and in the mixture of clover with Kenthucky bluegrass, the amount of crude protein and crude fiber is lower by 16.9 and 7.0%. This trend is the opposite of the experimental year 2015, when the mixture of white clover - Kenthucky bluegrass recorded the highest values of crude protein and lowest crude fiber content (227.5 g kg^{-1} DM). The crude protein amount in dry biomass of white clover and perennial ryegrass is lower by 49.7 g kg^{-1} DM and the crude fiber and dry matter index values are higher by 86.1 and 7.9 g kg^{-1} DM versus its mixture with Kenthucky bluegrass. In a third vegetation, the participation of grass crops in white clover grasslands was below 10%, which affected the percentage of protein in the feed mass. Its mixtures with *Lolium perenne* L. has a higher dry matter content, richer in protein and poorer in crude fiber than those of *Poa pratensis* L.

In the year of sowing, the difference in dry matter content and crude protein in the forage matter of red clover is minimal (1-2%). *Trifolium pratense* L. exhibits better adaptability and resistance with meadow fescue compared to timothy-grass. Legume culture dominates in spring and summer regrowth of the crop and ensures higher values of the protein fraction, as well as better take-up and absorption of animal feed. Here too, as in the case of white clover mixtures, the following regularity is observed: the higher protein content (141.0 g kg^{-1} DM) is combined

with a higher fiber content of 262.0 g kg^{-1} DM (red clover - meadow fescue) and lower protein values (137.9 g kg^{-1} DM) are combined with a lower percentage of fibers - 255.3 g kg^{-1} DM (red clover – timothy-grass). The differences between the values of the indicators are statistically unproven, which gives little significance to the established ones. The low protein trend - low fiber and vice versa - remains in the second and third years of the plant life cycle, but the percentage between 13.6 and 14.5% (CP) and 11.8 to 18.1% (CF). This atypical manifestation is due to the botanical peculiarities of crops in the grass association, in this case the uniform response of legume crop to wheat and grassland use.

The blue hybrid alfalfa - cock's foot is the only grasshopper whose components in the year of sowing shows almost equal share participation in the grassland, and in the second and third year alfalfa, which is also a major source of protein, records an insignificant presence (2-4%) in plant biomass. The resulting crude protein content in the dry matter is 145.2 g kg^{-1} DM (2014), 99.6 g kg^{-1} DM (2015) and 99.8 g kg^{-1} DM (2016). The comparatively dry climate in 2015 determined a higher content of dry matter and crude fiber (925.1 and 391.3 g kg^{-1} DM) in the feed of the mixture versus the first (879.2 and 282.4 g kg^{-1} DM) and a third (924.7 and 365.3 g kg^{-1} DM) vegetation. Given the data from the analysis, the expected nutritional value of the grass is low.

The dry matter, crude fats and fibers, the mineral and nitrogen-free extractable substances, the macro-elements phosphor and calcium are indicators characterizing the main chemical composition of the herb mixtures included in the study, whose values are regularly increased from first to third vegetation.

Lipids are the most concentrated source of energy for the living organism, improving the flavor of the feed, facilitating the dissolution and absorption of fat-soluble vitamins A, D, E and K. In a first vegetation their dry matter content ranges from 37.8 g kg^{-1} DM (red clover - timothy-grass) to 54.0 g kg^{-1} DM (red clover - meadow fescue). In the second experimental year the feed from the red clover mixture - meadow fescue leaves for all variants of this indicator (14.8 g kg^{-1} DM), and third, together with the white clover grass - Kenthucky bluegrass registers the highest values (32.2 g kg^{-1} DM).

In the sowing year, the mineral content of the dry matter is 53.0 g kg^{-1} DM (red clover - timothy-grass) to 90.3 g kg^{-1} DM (red clover - meadow fescue). The amount of ash in the red clover and fescue variant decreases with the age of the grass in the second and third vegetation and the values of the indicator are the lowest (62.6 and 65.1 g kg^{-1} DM). For comparison, in the plant biomass of white clover mixtures, we observe a backward dependence associated with increasing the amount of mineral substances.

In the first and second experimental year the mixed grassland of red clover and timothy-grass has the highest content of nitrogen-free extractable substances in the dry matter (379.3 and 429.6 g kg^{-1} DM) and in third the mixture of white clover - pasture ryegrass (434.9 g kg^{-1} DM) In the second and fifth variants we observe an

increasing regularity of the indicator from first to third vegetation.
The difference between the maximum (20.6 g kg^{-1} DM - bird's-foot-trefoil + red fescue) and minimum (14.3 g kg^{-1} DM - white clover + Kenthucky bluegrass) value of the macro-element Ca in the grasslands of grass and legume mixtures in 2014 is 41.3%. In a second vegetation, only in the plant biomass of the mixture of white clover - Kenthucky bluegrass we observe an insignificant increase in the amount of calcium. In the other variants, the trend is decreasing and totally opposite to the third vegetation when the content of this element in the feed of all experimental mixtures is increased, while the grassland of bird's-foot-trefoil - red fescue has a maximum value of 27.7 g kg^{-1} DM.
Spectrophotometric measurement showed a significant increase in phosphorus content in the dry matter of two-component grass mixtures from first to third vegetation. The maximum values for the indicator are the variants with white clover with a Kenthucky bluegrass (2.4 g kg^{-1} DM for 2014) and perennial ryegrass (4.1 g kg^{-1} DM for 2015), as well as the grassland with red clover - meadow fescue (5.5 g kg^{-1} DM for 2016).
On average, during the study period with the highest protein content (151.6 g kg^{-1} DM), the amount of mineral substances (75.6 g kg^{-1} DM) and crude fat (33.9 g kg^{-1} DM) white clover - Kenthucky bluegrass. This is also the grassland with the lowest dry matter content (888.3 g kg^{-1} DM) and nitrogen-free extractable substances (304.2 g kg^{-1} DM). *Trifolium repens L.* associated with grazing ryegrass forms a feedstock with the highest carbohydrate mass (434.9 g kg^{-1} DM) and the lowest amount of raw fiber (269.8 g kg^{-1} DM). The plant biomass of bird's-foot-trefoil and red fescue is the richest in the macro-element Ca (19.1 g kg^{-1} DM) and the poorest P (2.8 g kg^{-1} DM) than the other mixtures included in the experiment.
The plants of *Dactylis glomerata* L. predominate by over 75% of the blue hybrid alfalfa feed, and the herbaceous herb is determined as the highest fibrous (346.3 g kg^{-1} DM) respectively corresponding to the lowest amount of crude protein (114.9 g kg^{-1} DM) and with the highest dry matter content. On average, for 2014-2016, this blend is also the poorest element of Ca (11.0 g kg^{-1} DM).

Potential energy nutrition value of mixtures of grasses and legumes.
The current biomass quality assessment is mainly based on the energy nutrition value and is determined by the feed unit of milk and growth. The feed value is determined based on the chemical composition - crude protein content, fat, fiber, nitrogen-free extractable substances and digestibility ratios, reported by *Todorov (2010)*. The total energy value of mixtures varies from 16.18 MJ/kg DM to 16.87 MJ/kg DM - Table 2.

Table 2. Energy nutrition value of mixtures of grasses and legumes by year and average for the period 2014-2016

Variants	GE	EE	FUM	FUG
2014				
Bird's-foot-trefoil+Red fescue	16.87	7.37	0.68	0.62
White clover+Perennial ryegrass	16.75	7.37	0.68	0.62
White clover+Kenthucky bluegrass	16.18	7.23	0.67	0.61
Red fescue+Ttimothy-grass	16.50	7.52	0.70	0.64
Blue hybrid alfalfa+Cock's foot	16.74	7.32	0.67	0.61
Red fescue+Meadow fescue	16.39	7.24	0.67	0.61
2015				
Bird's-foot-trefoil+Red fescue	17.18	7.70	0.71	0.65
White clover+Perennial ryegrass	16.81	7.49	0.69	0.63
White clover+Kenthucky bluegrass	16.59	7.80	0.73	0.68
Red fescue+Ttimothy-grass	16.89	7.70	0.71	0.66
Blue hybrid alfalfa+Cock's foot	17.25	7.28	0.67	0.60
Red fescue+Meadow fescue	17.12	7.57	0.70	0.64
2016				
Bird's-foot-trefoil+Red fescue	16.74	7.36	0.68	0.62
White clover+Perennial ryegrass	16.52	7.91	0.74	0.69
White clover+Kenthucky bluegrass	17.33	7.34	0.67	0.60
Red fescue+Ttimothy-grass	17.10	7.82	0.73	0.67
Blue hybrid alfalfa+Cock's foot	16.98	7.32	0.67	0.61
Red fescue+Meadow fescue	17.38	7.44	0.68	0.62
2014-2016				
Bird's-foot-trefoil+Red fescue	16.93	7.47	0.69	0.63
White clover+Perennial ryegrass	16.70	7.59	0.70	0.65
White clover+Kenthucky bluegrass	16.70	7.46	0.69	0.63
Red fescue+Ttimothy-grass	16.83	7.68	0.71	0.66
Blue hybrid alfalfa+Cock's foot	16.99	7.30	0.67	0.61
Red fescue+Meadow fescue	16.96	7.42	0.68	0.62
Mean	***16.9***	***7.5***	***0.7***	***0.6***
SD	***0.1***	***0.1***	***0.0***	***0.0***

GE - gross energy - MJ/kg DM; EE - exchange energy - MJ/kg DM; FUM - feed unit for milk - number in kg of dry matter; FUG - feed units for growth - number in kg of dry matter.

In the year of sowing, the share of cereal crops in the grass: blue hybrid alfalfa - cock's foot, predominates over legumes (34.6%) by about 3%. Data are opposite for the second and third experimental years. Traces of alfalfa are minimal, whereas *Dactylis* L. species occupy 93-95% of the volume of plant biomass in the mixture.

Botanical composition of grassland is a factor that affects the content of fiber fractions in the composition of the forage vegetation, the degradation of which becomes a major source of energy for ruminants. Cock's foot is a high-energy forage grass *(Naydenova et al., 2015; Naydenova and Vasileva, 2016)*. Its mixtures with blue hybrid alfalfa are distinguished with the highest gross energy in the second (17.25 MJ/kg DM) vegetation from the experimental period when the excess over the mean value of the indicator is 0.26 MJ/kg DM. In a third vegetation, gross energy values (16.98 MJ/kg DM) of the variant are identical to the average for the period. On average for the period, the legume and grass grassland of blue hybrid alfalfa - cock's foot has the highest energy.

In the year of experiment establishment, fodder biomass from the mixture of red clover - timothy-grass has the highest energy content that regulates the metabolic processes in the animal organism (7.52 MJ/kg DM) and the highest values of feed units for milk (FUM - 0.70 in kg DM) and growth (FUG - 0.64 in kg DM). In the second and third experimental years, the maximum values of these indices are respectively the mixtures of white clover with Kenthucky bluegrass (EE - 7,80 MJ/kg DM; FUM – 0.73 in kg DM, FUG – 0.68 in kg DM) and perennial ryegrass (EE - 7.91 MJ/kg DM, FUM - 0.74 in kg DM, FUG - 0.69 in kg DM). The excess over the average for the experimental period is minimal.

The nutritional value of feed depends on the predominant botanical composition of the grassland. The good distribution of legume and grass component in the mixture of red clover - meadow fescue - influenced the energy nutrition value of the grassland, by increasing in direct ratio its basic indicators from the first to the third vegetation. Red clover as a legume component dominates in that variant for the entire study period, and the high energy nutritional value of the mixture is influenced by the higher average crude fiber content in the dry matter of the meadow fescue (4.4%) compared to timothy grass *(Kostov and Pavlov, 1999)*. The forage mass in the variants with *Trifolium pratense* L. is characterized by an increasing trend of GE and EE amount from the first to the third vegetation. The values of both indicators in the mixture of red clover - meadow fescue in the second and third experimental years show higher exceed compared to the average for the period: 0.16 MJ/kg DM (2015) and 0.42 MJ/kg DM (2016) - GE and 0.15 MJ/kg DM (2015) and 0.02 MJ/kg DM (2016) - EE. As opposed to gross and exchange energy, the harvested forage mass of the mixture of red clover - meadow timothy has a higher net energy (except for the first vegetation of the plant development), and the values of FUM and FUG in the second and third vegetation are close to the average for the experimental period.

The total energy value of plant matter of bird's-foot-trefoil and red fescue marks a maximum (17.18 MJ/kg DM) in a second vegetation. The excess over the average for 2014-2016 is 0.25 MJ/kg DM. In the third vegetation, the indicators: GE, EE, FUM and FUG of the variant have lower values than the average for the period.

The higher temperatures combined with lower rainfall rates in 2015 and 2016

affected the process of becoming wooden and the fiber content of mixed grasslands. Fiber composition is determinant for the digestibility, quality and nutritional value of the feed *(Fahey and Hussein, 1999; Brink et al., 2007)* and the main source for the observed increase in the amount of gross and exchange energy in the feed of all grass mixtures in the second and third experimental year.
Against the background of data characterizing energy nutrition of feed in the period 2014-2016 year, the harvested biomass of blue hybrid alfalfa - cock's foot (16.99 MJ/kg DM) and red clover - meadow fescue (16.96 MJ/kg DM) have the highest calorie. Both mixtures have almost identical values regardless of the different botanical composition and the predominant component in the grassland. The energy nutrition value of grass mixtures is a major factor determining the quality of the feed to satisfy the animals' need for food and the production of products of animal origin. The forage from the other observed grasslands is characterized by lower overall energy and high protein content. For the same period, the average value of energy useful for the normal course of physiological needs in the animal organism is highest in the grasslands of red clover - timothy-grass (7.68 MJ/kg DM) and white clover - perennial ryegrass (7.59 MJ/kg DM). This unidirectional data is also maintained in terms of the number of feed unit for milk (0.71 in kg DM for variant 4 and 0.70 in kg DM for variant 2) and growth (0.66 in kg DM for variant 4 and 0.65 in kg DM for variant 2).
For the experimental period, the mixed grassland of *Trifolium repens* L. has the lowest values of gross energy (16.70 MJ/kg DM) energy, and the forage biomass of blue hybrid alfalfa - cock's foot has registered the lowest amount of exchange energy (7.30 MJ/kg DM) as well as the smallest number of feed units (FUM - 0.67 in kg DM and FUG - 0.61 in kg DM) in the dry matter.

Conclusions

The analyzed dry matter of the mixture of white clover - Kenthucky bluegrass has the highest content of crude protein (151.6 g kg^{-1} DM), mineral substances (75.6 g kg^{-1} DM) and crude fat (33.9 g kg^{-1} DM). *Trifolium repens L.* associated with perennial ryegrass forms a forage mass with the highest carbohydrate amount (434.9 g kg^{-1} DM) and the lowest amount of crude fiber (269.8 g kg^{-1} DM). The plant biomass of bird's-foot-trefoil and red fescue is the richest in the macro-element Ca (19.1 g kg^{-1} DM), and the grassland with blue hybrid alfalfa - cock's foot has the highest dry matter content (909.7 g kg^{-1} DM).
In the period of study, the harvested biomass of blue hybrid alfalfa - cock's foot (16.99 MJ/kg DM) and red clover - meadow fescue (16.96 MJ/kg DM) has highest caloric values. Both mixtures have almost identical values regardless of the different botanical composition and the predominant component in the grassland. The feed of the mixture of *Trifolium pratense* L. with *Phleum pratense* L. has the

highest energy nutritional value (FUM - 0.71 in kg DM and FUG - 0.66 in kg DM), while the forage biomass in the variant of blue hybrid alfalfa - cock's foot register the lowest amount of exchange energy (7.30 MJ/kg DM), as well as the smallest number of feed units (FUM – 0.67 in kg DM and FUG – 0.61 in kg DM) in dry matter.
The mixed grasslands of *Trifolium repens* L. have the lowest gross energy (16.70 MJ/kg DM).

Osnovni hemijski sastav i energetska nutritivna vrednost krmne biomase iz veštačkih ekosistema

Tatyana Bozhanska, Boryana Churkova, Teodora Mihova

Rezime

U periodu od 2014. do 2016. godine, u Istraživačkom institutu za planinsko stočarstvo i poljoprivredu - Trojan, pratili smo promenu kvalitativnog sastava suve biomase iz dvokomponentnih travnatih smeša. Pašnjaci sa mešavinom *Trifolium repens* L. - *Poa pratensis* L. su imali najviši sadržaj sirovog proteina (151,6 g kg^{-1} SM), mineralnih supstanci (75,6 g kg^{-1} SM) i sirove masti (33,9 g kg^{-1} SM). Krmna trava koja se povezuje sa višegodišnjim ljuljom stvara krmu sa najvišom količinom ugljenih hidrata (434,9 g kg^{-1} SM) i najmanjom količinom sirovih vlakana (269,8 g kg^{-1} SM). Biomasa biljaka *Lotus corniculatus* L. i *Festuca rubra* L. je najbogatija u Ca (19,1 g kg^{-1} SM), a smeša plave hibridne lucerke - ježevice ima najveći sadržaj suve materije (909,7 g kg^{-1} SM) . Biomasa plave hibridne lucerke - ježevice (16,99 MJ / kg SM) i crvena detelina – livadski vijuk (16,96 MJ / kg SM) je imala najviše kalorijske vrednosti. Obe mešavine imaju gotovo identične vrednosti bez obzira na različiti botaničke sastav i pretežnu komponentu u travnjacima. Mešavina *Trifolium pratense* L. sa *Phleum pratense* L. ima najvišu energetsku hranljivu vrednost (FUM - 0,71 kg SM i FUG - 0,66 kg SM), a biomasa plave hibridne lucerke – ježevice imala je najmanju količinu energije za razmenu (7,30 MJ / kg SM), kao i najmanji broj jedinica za ishranu (FUM - 0,67 kg SM i FUG - 0,61 kg SM) u suvoj materiji. U navedenom eksperimentalnom periodu, sa najnižim vrednostima bruto energije (16,70 MJ / kg SM) bila je smeša travnjaka *Trifolium repens* L.

Ključne reči: mešavine krmnih biljaka i trava, kvalitet krme

References

BOSTAN C., MOISUC A., RADU F., COJOCARIU L., SĂRĂŢEANU V. (2010): Study of the action of poa pratensis l. vegetal extract on the chemical composition of some perennial grasses. Research Journal of Agricultural Science, 42, 1, 367-371.

BOZHANSKA T. (2017a): Study on perennial legume-grass mixtures in the conditions of the Central Balkan mountain. Banat's Journal of Biotechnology, VIII, 15, 34-42.

BOZHANSKA T. (2017b): Productivity and quality characteristic of forage grasses and grass mixtures for the conditions of the Central Balkan mountain. Dissertation, Research Institute of Mountain Stockbreeding and Agriculture – Troyan, Bulgaria.

BRINK G. E., CASLER M. D., HALL M. B. (2007): Canopy structure and neutral detergent fiber differences amond temperate perennial grasses. Crop Science, 47, 5: 2182-2189.

CHURKOVA B. (2007): Seed yield of birdsfoot trefoil grown in mixture with meadow grasses. Bulgarian Journal of Agricultural Science, 13, 515-520.

FAHEY G. C., HUSSEIN H. (1999): Forty years of forage quality research: Accomplishmend and impact from an animal nutrition perspective. Crop Science, 39, 4-12.

KOSTOV K., PAVLOV D. (1999): Fodder production. Academic Publishing House of Agricultural Institute - Plovdiv.

LAZAREVIC D, STOSIC M., DINIC B., LUGIC Z., TERZIC D. (2004): Production and proportion of spesies in grass-leguminous mixtures in hilly-mountainous region of Serbia. Journal of Mountain Agricultural on the Balkans, 7, 5, 518-525.

LEMEŽIENĖ N., KANAPECKAS J., TARAKANOVAS P. (2007): Evaluation of smooth-stalked meadow grass (*Poa pratensis* L.) genetic resources for breeding purposes. Biologija 53, 3, 56-58.

MIHOVSKI TS., SABEVA M. (2011): New technological approaches in the creation of mixed crop of white clover and perennial ryegrass. Journal of Mountain Agriculture on the Balkans, 14, 3, 541-547.

MIHOVSKI TS., GORANOVA G. (2006): Comparative testing of cultivars of white clover (*Trifolium repens* L.) in the pre-mountainous conditions of Central Balkan Mountain. Bulgarian Journal of Crop Science, 43, 1, 57-61.

MIHOVSKY TS. (1995): Les recherches sur le trefle blanc en Bulgarie: Production et composition chimique. Fourrages, 141, 57-62.

MITEV D., NAYDENOVA G. (2008): Persistency of artificial swards with participation of Red fescue on the slopes of the Central Balkan mountains. V State of mixed swards of red fescue and birdsfoot trefoil. Journal of Mountain Agriculture on the Balkans, 11, 7, 1342-1352.

NAYDENOVA Y., VASILEVA V. 2016. Analysis of Forage Quality of Grass Mixtures – Perennial Grasses with Subterranean Clover. Journal of Basic And Applied Research, 2, 4, 534-540.
NAYDENOVA G. (2008): Productivity, biological and morphological characteristics of red clover selection populations. Bulgarian Journal of Crop Science, 45, 230-235.
NAYDENOVA Y., KATOVA A. (2013): Forage quality evaluation of perennial grass species in breeding process. Journal of Mountain Agriculture on the Balkans, 13, 6, 1519-1538.
NAYDENOVA Y., VASILEV E., KIRILOV A. (2015): Plant cell wall fiber components content and digestibility of orchardgrass (*Dactylis glomerata* L.) and legume forage species in pure stands and mixtures. Journal of Mountain Agriculture on the Balkans, 18, 1, 61-76.
SLAVKOVA S., DIMITROV D., SHINDARSKA Z., MARKOV N. (2017): Study on the relation among some economic and biochemical indicators with the qualitative composition of grazing in beef cattle. Journal of Mountain Agriculture on the Balkans, 20, 1, 92-101.
STOSIC M., LAZAREVIC D., DINIC B. (2003): Inprovement of production of fodder on grassiands in function of livestock production development in hilly mountainous region. Biotechnology in animal husbandry, 19, 5-6, 413-421.
TODOROV N. (2010): Practice in Animal Nutrition. Sofia, Bulgaria.
VASILEVA V. (2011): Study on productivity of perennial legume crops in mixtures. Journal of Mountain Agriculture on the Balkans, 14, 2, 296-307.

Received 9 August 2018; accepted for publication 18 September 2018

Biotechnology in Animal Husbandry 34 (3), p 369-369 , 2018
Publisher: Institute for Animal Husbandry, Belgrade-Zemun
ISSN 1450-9156
UDC 636.2:575.113
https://doi.org/10.2298/BAH1803369E

Article Retraction

The Editor's office of the scientific journal **Biotechnology in Animal Husbandry,** is issuing the Note of retraction of the following article - **PROTEINS SEQUENCE ANALYSIS OF CONTAGIOUS CAPRINE PLEUROPNEUMONIA** by authors *Ayuba Dauda, Abdulmojeed Yakubu, Ihe Ndu Dim, Deeve Sebastian Gwaza*, published in the journal Biotechnology in Animal Husbandry 33 (3), p 309-319, 2017 (Details Full text https://doi.org/10.2298/BAH1703309Y)

The decision of the Editor's office is based on the information received from CEON (the Centre for Evaluation and Education in Science), no. 35-07/18, from July 20th, 2018, showing major overlapping of content with already published paper, indicating this article as highly suspect of (auto)plagiarism.

The fore mentioned article has been processed according to the Editorial procedure, and received a positive review. The author, however, has submitted the same article to another journal, where it was also published, without previously withdrawing it from the **Biotechnology in Animal Husbandry** or informing our Editor's office.

Therefore, the Editor's office has decided to retract the fore mentioned article, by publishing the Note of retraction in this issue of our journal, also in the electronic version, original article is retained unchanged save for a watermark on the .pdf indicating on each page that it is "retracted, with a link to the original article.

As a result of this decision of the Editor's office, quoting and stating of this article in literature references and bibliographies is NOT ALLOWED.

Editorial

Link to the retracted article: https://doi.org/10.2298/BAH1703309Y)

Manuscript submission

By submitting a manuscript authors warrant that their contribution to the Journal is their original work, that it has not been published before, that it is not under consideration for publication elsewhere, and that its publication has been approved by all co-authors, if any, and tacitly or explicitly by the responsible authorities at the institution where the work was carried out.

Authors are exclusively responsible for the contents of their submissions, the validity of the experimental results and must make sure that they have permission from all involved parties to make the data public.

Authors wishing to include figures or text passages that have already been published elsewhere are required to obtain permission from the copyright holder(s) and to include evidence that such permission has been granted when submitting their papers. Any material received without such evidence will be assumed to originate from the authors.

Authors must make sure that all only contributors who have significantly contributed to the submission are listed as authors and, conversely, that all contributors who have significantly contributed to the submission are listed as authors.

The manuscripts should be submitted in English (with a summary in English or Serbian language – translation of Summaries into Serbian language for non-domestic authors will be performed by the Editor's office) by email to: biotechnology.izs@gmail.com

Manuscripts are be pre-evaluated at the Editorial Office in order to check whether they meet the basic publishing requirements and quality standards. They are also screened for plagiarism.

Authors will be notified by email upon receiving their submission. Only those contributions which conform to the following instructions can be accepted for peer-review. Otherwise, the manuscripts shall be returned to the authors with observations, comments and annotations.

Manuscript preparation

Authors must follow the instructions for authors strictly, failing which the manuscripts would be rejected without review.

The manuscript should be prepared in Microsoft Word for Windows, maximum 8 pages of typed text using, Paper size: Custom size, Width 17 cm, Height 24 cm; format (Portrait), normal spacing (Single Space). Margins: Top 2.0 cm, 2.0 cm Left, Bottom 2.0 cm, 2.0 cm Right, no pagination.

Use font Times New Roman, size 11 (except where it is stated otherwise), single space, justify

Title of the paper should be Times New Roman, font size 14, bold, capital letters, justify

Authors – Times New Roman, font size 12, bold, specify the full names of all authors on the paper. Use 1,2, … numbers in suffix to refer to addresses of authors, only in the case of different affiliations (institution)

Affiliations of authors – Times New Roman, font size 9, normal, under affiliations of authors should be mentioned e-mail of corresponding author and after that category of paper.

Example 1

POTENTIALS OF SERBIAN LIVESTOCK PRODUCTION – OUTLOOK AND FUTURE

Milan M. Petrović[1], Stevica Aleksić[1], Milan P. Petrović[1], Milica Petrović[2], Vlada Pantelić[1], Željko Novaković[1], Dragana Ružić-Muslić[1]

[1]Institute for Animal Husbandry, Belgrade – Zemun, 11080 Zemun, Serbia
[2]University of Belgrade, Faculty of Agriculture, Nemanjina 6, 11080 Zemun, Serbia
Corresponding author: Milan M.Petrović, **e-mail address**
Review paper

Example 2

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

Zdenka Škrbić, Zlatica Pavlovski, Miloš Lukić, Veselin Petričević

Institute for Animal Husbandry, Autoput 16, 11080 Belgrade, Serbia
Corresponding author: Zdenka Škrbić, **e-mail address**
Original scientific paper

Original scientific paper should contain following paragraphs with single spacing (title of paragraphs should be in Times New Roman 14 **bold**, except for **Abstract** and **Key words** where font size is 11 **bold**):

Abstract: up to 250 words, Times New Roman, font size 11, justify. Abstract should contain a brief overview of the methods and the most important results of the work without giving reference. Abstract submitted in English language.

Key words: not more than 6. The selection carried out by relying on widely accepted international source such as a list of keywords Web of Science.

Introduction – present the review of previous research and objective of the paper.

Materials and Methods – state methods applied in the paper; experimental research design. Use SI system of measurement units.

Results and Discussion – present investigation results separately from discussion or together in one paragraph. Presentation of the results should be precise and without repetitions, and include the evaluation of significant differences and other parameters.

Text and titles of tables, figures and graphs, Times New Roman, font size 9, **bold**, in the following form:

Table 1. Least square means for the reproductive traits of cows

Tables and figures should be numbered and with adequate title and legend, width and height not exceeding 12 cm and 17 cm, respectively. Tables should be prepared according to instruction for forming of tables in Office Word. Each column in table must have heading and, when necessary, abbreviations should be explained in the legend/footnote.

Conclusion – containing the most important issues of the paper

After Conclusion the title of the paper in Serbian in Times New Roman 14 **bold**, is stated, followed by authors in Times New Roman 11 *italic*, example:

Potencijali srpske stočarske proizvodnje – izgledi i budućnost

Milan M. Petrović, Stevica Aleksić, Milan P.Petrović, Milica Petrović, Vlada Pantelić, Željko Novaković, Dragana Ružić-Muslić

Summary – in Serbian language, 250 max. words (non-Serbian authors should provide Summary in English language that will be translated to Serbian by Editor's office)

Key words: not more than 6 (in Serbian language)

Acknowledgment – for example:

Research was financed by the Ministry of Science and Technological Development, Republic of Serbia, project TR 6885.

References – should be in alphabetical order. Names of the authors must be given in capital letters followed by the year of publication in brackets, titles in the language of the original. Use only the full name of the journal.

In scientific journals:

PETROVIĆ M. M., SRETENOVIĆ LJ., BOGDANOVIĆ V., PERIŠIĆ P., ALEKSIĆ S., PANTELIĆ V., PETROVIĆ D. M., NOVAKOVIĆ Ž. (2009): Quantitative analysis of genetic improvement of milk production phenotypes in Simmental cows. Biotechnology in Animal Husbandry, 25,1-2, 45-51.

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.

WEBB E., O'NEILL H. (2008): The animal fat paradox and meat quality. Meat Science, 80, 28-36.

PhD Thesis:

RUŽIĆ-MUSLIĆ D. (2006): Uticaj različitih izvora proteina u obroku na proizvodne rezultate jagnjadi u tovu. Doktorska disertacija. Univerzitet u Beogradu, Poljoprivredni fakultet.

CAETANO A.R. (1999): Comparative mapping of the horse (*Equss caballus*) genome by synteny assignment of type-I genes with a horse-mouse somatic cell hybrid panel. Ph.D. Dissertation, University of California, Davis.

In Scientific Books:

PETROVIĆ P.M (2000): Genetika i oplemenjivanje ovaca. Naučna knjiga, Beograd, pp365.

FITZGERALD M. (1994): Neurobiology of Fetal and Neonatal Pain. In: Textbook of Pain. 3rd edition. Eds Wall P. and Melzack R. Churchchill Livingstone, London, UK, 153-163.

At Scientific Meetings:

ŠKRBIĆ Z., LUKIĆ M., BOGOSAVLJEVIĆ-BOŠKOVIĆ S., RAKONJAC S., PETRIČEVIĆ V., DOSKOVIĆ V., STANOJKOVIĆ A. (2015): Importance of farm management in reducing broilers skin lesions. Proceedings of the 4th International Congress "New Perspectives and Challenges of Sustainable Livestock Production", October 7 – 9, Belgrade, 145-158.

Citations in the text are presented in italic form, examples: …results of *Petrović (2009)*; *Petrović et al. (2009)*; *Webb and O'Neill (2008)….*; *(Škrbić et al., 2015); (Ružić-Muslić, 2006); (Webb and O'Neill, 2008)*

Editor's office