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EVALUATION OF NUTRITION ACCORDING TO MILK METABOLITES AND COMPONENTS IN SEASONAL DEPENDENCE IN DAIRY COWS

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ABSTRACT

The objective of this study was to evaluate the nutrition according to the milk metabolites and components during the seasonal dependence of the dairy cows. In the summer time, it was confirmed that significantly lower content of milk protein and fat ($P < 0.001$) and lower milk, protein, and fat yields ($P < 0.001$) occurred, compared to the autumn period. The content of milk lactose was not statistically significant ($P > 0.5$). The evaluation of nutrition according to protein in milk and milk urea in the summer period was confirmed in the 2nd group of milk samples which represented 53 % of the group's higher content of milk urea, higher content of urinary nitrogen excretion, and a lower nitrogen utilization efficiency compared to the 1st group of milk samples. The autumn period was confirmed in the 2nd group of milk samples which represented 53 % of the group with higher milk yield and lower content of milk proteins and milk fat compared to the 1st group of milk samples. The evaluation of nutrition using the analysed content of milk urea and proteins in milk represents a suitable tool for evaluating the impact of malnutrition or overfeeding of energy and protein on milk production and other components.

Key words: Holstein dairy cow; milk urea; milk yield; season

INTRODUCTION

Dairy cow's milk contains 12–14 % dry matter composed of proteins (2.5–4 %), fat (3–5 %), and lactose (~5 %) [18]. To assess the balance between the intake of proteins and energy in the feed ration, the evaluation of the content of milk urea (MU) is used, which is part of the non-protein nitrogen in milk dairy cows [9]. According to M a r e n j a k et al. [14] the optimal concentration of MU in cow's milk is 10–30 $mg.dl^{-1}$. Climate change poses a major threat to the sustainability of livestock production and the preservation of the ecosystem [7]. Dairy cows optimally tolerate an environmental temperature in the range of 5–25 °C, which represents their thermoneutral zone, when they do not have an increased need for energy to create or get rid of body temperature [10]. A large amount of metabolic heat is generated in dairy cows during lactation and they also accumulate heat from radiant energy. As a result of the higher temperature of the environment and the limited cooling capacity, there is an increase in body temperature [30]. At a temperature above 25 °C, dairy cows enter heat

stress, when the compensation of the metabolic load due to heat stress affects feed intake with an impact on the quantity and quality of production, deterioration of reproduction, and the health status of dairy cows, together with increased energy consumption to regulate body temperature and its release from the body by skin and inhalation. The result is increased respiratory and heart rates, impacting feed intake, physiological and metabolic functions, reproduction, and the immune system [7, 12, 29].

The aim of our study was to evaluate the nutrition according to analysed milk urea and protein content in milk Holstein dairy cows during the seasonal dependence.

MATERIALS AND METHODS

Data collection

This study included 140–160 Holstein cows in the first lactation phase from a Slovakian dairy farm from January 2018 to December 2018. The cows were selected according to season into two groups: summer (June – August) and autumn (September – November). Feed rations of dairy cows were predominantly based on corn and alfalfa silage, supplemented with different carbohydrate feeds (cereal grains and cereal grain by-products) and protein supplements (soybean and rapeseed meal) provided as the TMR (total mixed ration) *ad libitum*. The nutritional composition of the TMR and daily feed intake were intensively monitored and the dairy cows were kept in a free-stall housing system.

Analysis of total mixed rations

The samples of the TMR were taken from the feed manager and were analysed once per month for dry matter (DM), which consisted of crude protein (CP), fat, acid and neutral detergent fibre (ADF, NDF), and starch analysed by conventional methods according to the Commission Regulation (EC) No. 691/2013 [5]. The DM was determined by weight upon drying the sample at 105 °C under the prescribed conditions. The CP content was determined by the Kjeldahl method using a 2300 Kjeltec Analyzer Unit (Foss Tecator AB, Sweden). The fat was determined by the device Det-Gras (JP SELECTA, Spain). The ADF and NDF were determined using a Dosi-Fiber Analyser (JP SELECTA, Spain), and the content of starch was determined polarimetrically. The net energy for lactation (NEL) and non-fibre carbohydrates (NFC) were calculated according to the nutrient requirements of dairy cattle [17].

Analysis of production parameters

The farm was evaluated by the Breeding Services of the Slovak Republic using the Breeding Information System [20]. The cows were milked twice a day and individual milk samples were analyzed once per month for protein, fat, lactose content, and MU (milk urea) by spectrophotometric tests in the near-infrared region using MilkoScan FT+ (Foss Electric, Denmark) and BENTLEY FTS (Bentley Instruments Inc., USA) in the Central analytical laboratory for milk with accreditation under registration number 096/5878/2015/2. The total number of analysed samples of milk in one month was 140–160. Daily yields of protein, fat, and lactose were calculated according to S t o o p et al. [26]. Conversion of MU to milk urea nitrogen (MUN) was calculated according to O u d a h [19]. The nitrogen utilization efficiency (NUE) was calculated according to H u h t a n e n et al. [8] and the urinary nitrogen excretion (UNE) according to K a u f f m a n and S t - P i e r r e [11].

Environmental conditions during the summer and autumn periods

The influence of climatic conditions by measuring air temperatures during the summer (June – August) and autumn periods (September – November) were evaluated according to daily measurements by a climatological station located 13 km from the farm. Maximum and minimum daily temperatures were recorded in a climatological booth at a standard height of 2 m above the ground for 24 hours. The maximum daily temperature was usually recorded 2 hours after the culmination using a mercury thermometer, and the minimum temperature usually before sunrise was recorded using an alcohol thermometer. Information on the average, maximum, and minimum temperatures during the day was provided by the Slovak Hydrometeorological Institute of the Slovak Republic [23].

Ethical considerations

In this study we used data obtained with the consent of the farm in collaboration with The Breeding Services of Slovakia, using the Breeding Information System.

Statistical analysis

The data analysis was carried out via the Graph Pad Prism 8.3 (GraphPad Software, San Diego, CA, USA). The results of each variable were expressed as the means and

standard deviations (SD). One-way analysis of variance (ANOVA) was used to evaluate the statistical significance with Tukey's multiple comparisons between summer and autumn groups with a significance level set at $P < 0.001$.

RESULTS

The mean concentration of nutrients in the TMR and the production parameters of dairy cows are listed in Table 1. The nutrient content of TMR in the summer and autumn periods fluctuated according to the recommendation for the first lactation phase by standardized nutrient requirements [17]. In the summer, the dry matter intake (DMI) was reduced (23.8 ± 0.4 kg.d^{-1}) with the feed residues in the feed trough at 5–10 % compared to the calculated DMI (25.1 ± 0.4 kg.d^{-1}).

Table 1. Nutritional composition of the TMR, production and composition of milk in the summer and autumn periods

	Summer period	Autumn period
Nutrients in TMR (% Dry Matter)		
CP	14.6 ± 0.4	15.4 ± 0.3
Starch	24.1 ± 0.7	25.5 ± 0.4
Fat	4.3 ± 0.2	4.1 ± 0.1
NDF	34.4 ± 0.3	33.9 ± 0.9
ADF	21.6 ± 0.5	20.2 ± 0.3
NFC	37.4 ± 0.1	38.7 ± 0.7
NEL	6.7 ± 0.2	6.7 ± 0.2
Milk production and composition		
Milk yield (kg.d^{-1})	36.00 ± 5.3***	42.20 ± 6.6
Protein (%)	2.70 ± 0.1***	3.40 ± 0.2***
Fat (%)	3.20 ± 0.5***	3.48 ± 0.4***
Lactose (%)	4.85 ± 0.1	4.85 ± 0.1
Protein yield (kg.d^{-1})	0.95 ± 0.2***	1.43 ± 0.2***
Fat yield (kg.d-1)	1.12 ± 0.3***	1.69 ± 0.3***
Lactose yield (kg.d^{-1})	1.70 ± 0.4	2.04 ± 0.4

CP – crude protein; NDF – neutral detergent fibre; ADF – acid detergent fibre; NFC – non-fibre carbohydrates; NEL – net energy of lactation; TMR – total mixed ration; ***P < 0.001

Climatic summaries in the summer and autumn periods are shown in Table 2. In the summer period, the average daily temperatures were 20.4 ± 3.0 °C, with an average maximum temperature of 33.5 °C and a minimum value of 14.2 °C. In the evaluated period, the daily temperature above 25 °C with an average of 29.5 ± 2.8 °C (maximum 35.5 °C) was confirmed on 64 days out of a total of 92, which means that 70 % of the days during the summer period were dairy cows exposed to heat stress. In the autumn period, the daily average, maximum, and minimum ambient temperatures were 9.3 ± 5.0 °C, respectively, 21.0 °C, and -1.5 °C without the influence of heat stress during this period.

Table 2. Climate summaries in the summer and autumn

	Period	
	Summer	Autumn
Temperature, maximum (°C)	35.5	21.0
Average temperature (°C)	20.4 ± 3.0	9.3 ± 5.0
Days temperatures > 25 °C	64	-

During the summer period, it was confirmed that significantly lower content of milk protein and fat ($P < 0.001$) and lower milk, protein, and fat yields ($P < 0.001$) compared to the autumn period. The content of milk lactose was not statistically significant ($P > 0.5$).

The graphic representation proposed by S p o h r and W i e s n e r [24] is successfully used to diagnose of nutrition dairy cows according to the analysed content of protein and urea in the milk. Graphic representation of protein and urea content when inserting individual data of analysed protein samples (%) on the Y axis and MU (mg.dl^{-1}) on the X axis evaluates the breakdown of the analysed set of samples per group of dairy cows into 9 evaluated areas according to the reference, reduced or increased analysed urea and protein in milk.

The evaluation of nutrition according to protein in milk and MU in summer is presented in Fig. 1 and in Table 3. The 1st group consisted of milk samples with 15–30 mg.dl^{-1} MU and < 3.2 % proteins (represented 36.0 % of the group) and the 2nd group consisted of milk samples with MU > 30 mg.dl^{-1} and proteins < 3.2 % in milk (represented 53.0 % of the group). Milk production in both groups was very close with reduced protein content and fat in milk. In the 2nd group of milk samples were confirmed higher content of MU, higher content of UNE, and a lower NUE compared to the 1st group of the milk samples.

The evaluation of nutrition according to protein in milk and MU in the autumn period is presented in Fig. 2 and in Table 4. The 1st group consisted of milk samples with MU 15–30 mg.dl^{-1} and optimal proteins 3.2–3.8 % (represented 34 % of the group) and the 2nd group consisted of milk samples with MU 15–30 mg.dl^{-1} and low

proteins (< 3.2 %) in the milk (represented 53.0 % of the group). The 2nd group of milk samples confirmed higher milk yield and lower content of milk proteins and milk fat compared to the 1st group of the milk samples.

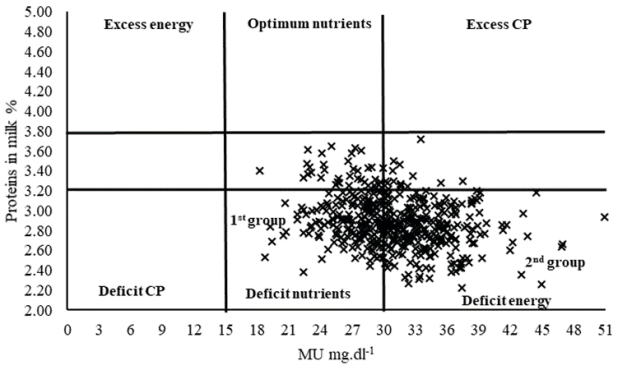


Fig. 1. Relationship of protein in milk and milk urea (MU) in the summer period

Table 3. Evaluation of milk in the summer period according to protein and urea in milk

	1st group Deficit nutrients	2nd group Deficit energy
	MU 15–30 mg.dl^{-1}	MU > 30 mg.dl^{-1}
	Proteins < 3.2 %	Proteins < 3.2 %
% of the group	36.0	53.0
Milk yield (kg.d^{-1})	39.5 ± 5.5	39.8 ± 7.3
Protein (%)	2.87 ± 0.2	2.80 ± 0.2
Fat (%)	3.12 ± 0.5	3.21 ± 0.8
Protein yield (kg.d^{-1})	1.13 ± 0.2	1.11 ± 0.2
Fat yield (kg.d^{-1})	1.23 ± 0.2	1.28 ± 0.3
MU (mg.dl^{-1})	27.00 ± 2.5	34.60 ± 4.0
UNE (%)	35.6	45.6
NUE (%)	31.5	30.6

MU – milk urea; UNE – urinary nitrogen excretion; NUE – nitrogen utilization efficiency

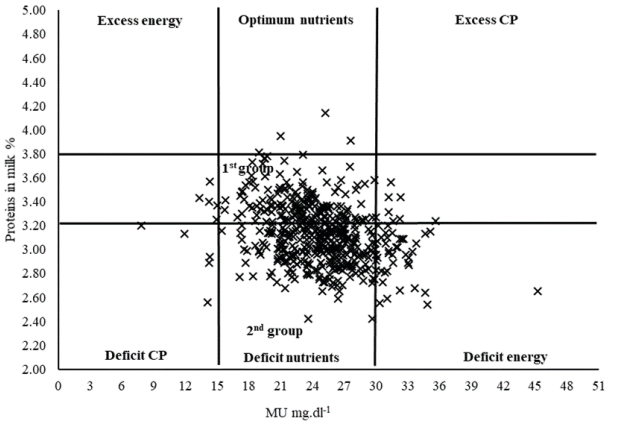


Fig. 2. Relationship of protein in milk and milk urea (MU) in the autumn period

Table 4. Evaluation of milk in the autumn period according to protein and urea in milk

	1st group Optimum nutrients	2nd group Deficit nutrients
	MU 15–30 mg.dl^{-1} Proteins 3.2-3.8 %	MU 15–30 mg.dl^{-1} Proteins < 3.2 %
% of the group	34.0	53.0
Milk yield (kg.d^{-1})	39.8 ± 6.9	42.6± 6.4
Protein (%)	3.38 ± 0.1	2.98± 0.1
Fat (%)	3.56 ± 0.6	3.28 ± 0.6
Protein yield (kg.d^{-1})	1.34 ± 0.2	1.27 ± 0.2
Fat yield (kg.d^{-1})	1.41 ± 0.3	1.40 ± 0.3
MU (mg.dl^{-1})	23.04 ± 3.2	24.65 ± 3.0
UNE (%)	29.0	31.0
NUE (%)	32.1	32.4

MU – milk urea; UNE – urinary nitrogen excretion; NUE – nitrogen utilization efficiency

DISCUSSION

In the summer, the influence of heat stress was confirmed by finding a high number of days with measured temperatures above 25 °C and with an impact on the decrease in DMI (5–10 % increase of residues on the feed trough compared to the calculated DMI).

The evaluation of production parameters (Table 1) confirmed reduced milk yield and synthesis of milk components with a decrease in fat and protein yield in the milk. Our conclusions agree with L i u et al. [13] who confirmed the effect of heat stress with a subsequent decrease in feed intake on reduced milk production and components. However, heat stress can also affect milk production directly through mechanisms that are independent of reduced DMI. The decrease in feed intake only contributed to approximately 35 % of the heat stress-induced decrease in milk production [21]. Rather, other feed intake-independent alterations induced by heat stress, particularly those related to nutrient partitioning, may be associated with changes in post-absorptive glucose and lipid metabolism changes [31]. The main precursor for the synthesis of lactose in the mammary gland is glucose, and the production of lactose is the main osmoregulator and therefore affects the production. During heat stress, the body tries to generate less metabolic heat (especially the skeletal muscles) and uses glucose to an increased extent. Subsequently, there is an insufficient supply of glucose to the mammary gland with a decrease in milk production [1]. Also, as a result of

the redirection of blood flow to peripheral tissues for heat removal, the supply of substrates to the mammary gland is limited [30].

The lactose content in the milk did not change due to heat stress, which agrees with the conclusions of Zheng et al. [34] because milk sugar is the most stable component of milk.

Heat stress reduces rumen motility, which affects the passage of digested in the gastrointestinal tract and reduces the production of volatile fatty acids [10, 15, 16, 32]. Heat stress with the impact of a decrease in feed intake (decreased roughage and increased concentrate) increases lactate production and decreases rumen acetate production with a decrease in rumen pH and milk fat [27, 33].

The confirmed reduced milk protein content during heat stress agrees with previous results [10, 22] when heat stress (rather than reduced amino acid intake) affects the synthesis process itself, especially α- and β-casein synthesis [2].

By evaluating the nutrition of dairy cows according to the analysed content of protein and MU using a graphic representation (Fig.1, Fig. 2), it points to the impact of disruption of the supply of nutrients and energy from feed ration on the parameters of protein metabolism to influence the components of milk. In the summer season, the feed ration was formed to produce 39.6 ± 1.1 kg.day^{-1} with a calculated protein content of 3.2 % and a fat content of 3.6 %. The production values in the divided groups (Table 3) confirm the same milk production as with the calculated feed ration, which was close in both groups but with a significantly reduced proportion of protein and fat in the milk. There was a significant difference in the composition of milk in terms of MU and the estimated daily amount of UNE and NUE. The Impact of heat stress on protein metabolism and amino acid balance such that the absorptive function of the rumen epithelium along with the amount of urea reabsorbed from the blood, causes urea to accumulate in the blood and transport to milk [4]. The confirmed higher content of MU in the second group points to the metabolic stimulation of body reserves in dairy cows due to thermal stress, when amino acids released by proteolysis from skeletal muscle are used for the synthesis of urea, which passes from the blood into the milk after deamination in the liver [3, 6]. The urea is filtered from the blood by the kidney and is excreted from the body in urine. Blood enters the kidneys through the renal artery and is filtered through the nephrons. This process concentrates the urea for excretion in the urine. Therefore, urea excretion is proportional to blood urea concentration. Because it is a small molecule, urea readily diffuses across cellular membranes. As milk is secreted in the mammary gland, urea diffuses into and out of the mammary gland, equilibrating with urea in the blood. Because of this process, MU equilibrates with and is proportional to blood urea. This process allows MU to be an excellent predictor of urea in urine. According to Huhtanen et al. [8] increases in MU were associated with a decrease in NUE.

In the autumn period, the feed ration was formed to produce 38.9 ± 1.7 kg.d^{-1} with a calculated protein content of 3.2 % and a fat content of 3.6 %. The production values in the divided groups (Table 4) confirm the reduced content of protein (2.98 ± 0.1 %) and fat (3.28 ± 0.6 %) in the 2nd group, which represented 53.0 % of the group with increased milk yield (42.6 ± 6.4 kg.d^{-1}) compared to the estimated daily production for the calculation of feed ration. The limited supply of nutrients and increased daily production of milk for the 1st stage of lactation in accordance with the genetic potential of the herd will not cover the needs for the synthesis of milk components (protein and fat), where the reduced supply of amino genic and lipogenic nutrients causes a decrease in milk components. The recommended procedure for feeding dairy cows with high genetic potential requires, when calculating the feed ration, to calculate the norm of the need for nutrients according to weight, production, and components in milk. The milk yield for individual dairy cows in the group according to the current milk yield for the 83rd percentile and according to the average production using the recalculation of the production stimulating factor (PSF) [25, 28].

CONCLUSIONS

The results from this study show a statistically significant ($P < 0.001$) impact of the season on milk yield and milk components. Our results confirmed that nutritional evaluation using the analysed milk urea and protein content in milk represents a suitable tool for evaluating the impact of malnutrition or overfeeding of dairy cows on farms. The assessment of milk urea, urinary nitrogen excretion, and nitrogen utilization efficiency is a useful

tool for the environment as well. Changing technologies through milk urea analysis, farms help to reduce the release of N into the environment.

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CONFLICT OF INTEREST

The authors declare there is no conflict of interest.

REFERENCES

1. **Baumgard, L. M., Rhoads, R. P., 2009:** The effects of heat stress on nutritional and management decisions. In *Proceedings of Western Dairy Management Conference*, Reno, Nevada, USA, March, 191–202.
2. **Bernabucci, U., 2002:** Effects of the hot season on milk protein fractions in Holstein cows. *Anim. Res.,* 51, 1, 25–33. DOI: 10.1051/animres:2002007.
3. **Bernabucci, U., Lacetera, N., Baumgard, L. H., Rhoads, R. P., Ronchi, B., Nardone, A., 2010:** Metabolic and hormonal acclimation to heat stress in domesticated ruminants. *Animal*, 4, 7, 1167–1183. DOI: 10.1017/S175173111000090X.
4. **Cai, L., Yu, J., Hartanto, R., Zhang, J., Yang, A., Qi, D., 2019:** Effects of heat challenge on growth performance, ruminal, blood and physiological parameters of Chinese crossbred goats. *Small Rumin. Res.,* 174, 125–130. DOI: 10.1016/j. smallrumres.2019.02.021.
5. **Commission Regulation (EC) No. 691/2013, 2013:** Amending Regulation (EC) No. 152/2009 as regards methods of sampling and analysis. *OJEU*, L 197/1—L197/12.
6. **Conte, G., Ciampolini, R., Cassandro, M., Lasagna, E., Calamari, L., Bernabucci, U., Abeni, F., 2018:** Feeding and nutrition management of heat-stressed dairy ruminants. *Ital. J. Anim. Sci.*, 17, 3, 604–620. DOI: 10.1080/1828051X.2017.1404944.
7. **Das, R., Sailo, L., Verma, N., Bharti, P., Saikia, J., Imtiwati and Kumar, R., 2016:** Impact of heat stress on health and performance of dairy animals: A review. *Vet. World*, 9, 3, 260. DOI: 10.14202/vetworld.2016.260-268.
8. **Huhtanen, P., Cabezas-Garcia, E. H., Krizsan, S. J., Shingfield, K. J., 2015**: Evaluation of between-cow variation in milk urea and rumen ammonia nitrogen concentrations and the association with nitrogen utilization and diet digestibility in lactating cows. *J. Dairy Sci*., 98, 5, 3182–3196. DOI: 10.3168/jds.2014-8215.
9. **Jílek, F., Řehák, D., Volek, J., Štípková, M., Němcová, E., Fiedlerová, M., Rajmon, R., Švestková, D., 2006:** Effect of herd, parity, stage of lactation and milk yield on urea concentration in milk. *Czech J. Anim. Sci.,* 51, 12, 510–517. DOI: 10.17221/3971-CJAS.
10. **Kadzere, C. T., Murphy, M. R., Silanikove, N., Maltz, E., 2002:** Heat stress in lactating dairy cows: a review. *Livest. Prod. Sci.,* 77, 1, 59–91. DOI: 10.1016/S0301-6226(01)00330-X.
11. **Kauffman, A. J., St-Pierre, N. R., 2001:** The relationship of milk urea nitrogen to urine nitrogen excretion in Holstein and Jersey cows. *J. Dairy Sci.,* 84, 10, 2284–2294. DOI: 10.3168/jds.S0022-0302(01)74675-9.
12. **Kumar, S., Mote, S., Singh, D., Chauhan, S. S., Ghosh, N., 2014:** Effects of environmental factors on lactation yield and lactation length of Holdeo crossbred cattle. *Indian J. Appl. Res.,* 4, 10, 4–7.
13. **Liu, J., Li, L., L., Chen, X., Lu, Y., Wang, D., 2019:** Effects of heat stress on body temperature, milk production, and reproduction in dairy cows: A novel idea for monitoring and evaluation of heat stress. A review. Asian-Australas. *J. Anim. Sci.*, 32, 9, 1332. DOI: 10.5713/ajas.18.0743.
14. **Marenjak, T., Poljičak-Milas, N., Stojević, Z., 2004:** The aim of urea determination in cows' milk (In Croatian, English abstract). *Praxis Veterinaria,* 52, 3, 233–241.
15. **Meneses, J. A. M., Lopes de Sá, O. A. A., Coelho, C. F., Pereira, R. N., Batista, E. D., Ladeira, M. M., et al., 2021:** Effect of heat stress on ingestive, digestive, ruminal and physiological parameters of Nellore cattle feeding low or high-energy diets. *Livest. Sci.* 252, 104676. DOI: 10.1016/j. livsci.2021.104676.
16. **Nonaka, I., Takusari, N., Tajima, K., Suzuki, T., Higuchi, K., Kurihara, M., 2008:** Effects of high environmental temperatures on physiological and nutritional status of prepubertal Holstein heifers. *Livest. Sci.* 113, 14–23. DOI: 10.1016/j. livsci.2007.02.010
17. **NRC, 2001:** *Nutrient Requirements of Dairy Cattle*. 7th rev. edn., 263–265.
18. **Osorio, J., S., Lohakare, J., Bionaz, M., 2016:** Biosynthesis of milk fat, protein, and lactose: roles of transcriptional

and posttranscriptional regulation. Physiol. Genomics, 48, 4, 231–256. DOI: 10.1152/physiolgenomics.00016.2015.

19. **Oudah, E. Z. M., 2009:** Non genetic factors affecting somatic cell count, milk urea content, test day milk yield and milk protein percent in dairy cattle of Czech Republic using individual test day records. *Livest. Res. Rural Dev*., 21, 5, 1–25. DOI: 10.21608/jappmu.2009.119192.
20. **Plemenárske služby Slovenskej republiky,** štátny **podnik (Breeding services of the Slovak Republic, state enterprise), 2018:** Available at https://www.pssr.sk/index.php/sk/o-nas/.
21. **Rhoads, M. L., Rhoads, R. P., VanBaale, M. J., Collier, R., J., Sanders, R., S., , Weber, W., J., Crooker, B., A., Baumgard, l. H., 2009:** Effects of heat stress and plane of nutrition on lactating Holstein cows: I. Production, metabolism, and aspects of circulating somatotropin. *J. Dairy Sci.,* 92, 5, 1986–1997. DOI: 10.3168/jds.2008-1641.
22. **Shwartz, G., Rhoads, M. L., VanBaale, M. J., Rhoads, R. P., Baumgard, L. H., 2009:** Effects of a supplemental yeast culture on heat stressed lactating Holstein cows. *J. Dairy Sci*., 92, 935–942. DOI: 10.3168/jds.2008-1496.
23. **Slovenský hydrometeorologický** ústav **SR, 2018 (Slovak hydrometeorological institute SR):** Available at http://www.shmu.sk/sk/?page=1.
24. **Spohr, M., Wiesner, H. U., 1991:** Monitoring herd health and milk production by means of the expected milk production performance. *Milchpraxis*, 1991, 29, 231–236.
25. **Stallings, C. C., Mcgilliard, M. L., 1984:** Lead factors for total mixed ration formulation. *J. Dairy Sci.*, 67, 4, 902–907. DOI: 10.3168/jds.S0022-0302(84)81386-7.
26. **Stoop, W. M., Bovenhuis, H., Van Arendonk, J. A. M., 2007**: Genetic parameters for milk urea nitrogen in relation to milk production traits. *J. Dairy Sci.*, 90, 4, 1981–1986. DOI: 10.3168/jds.2006-434.
27. **Uyeno, Y., Sekiguchi, Y., Tajima, K., Takenaka, A., Kurihara, M., Kamagata, Y., 2010:** An rRNA-based analysis for evaluating the effect of heat stress on the rumen microbial composition of Holstein heifers. *Anaerobe,* 16, 27–33. DOI: 10.1016/j.anaerobe.2009.04.006.
28. **Vajda, V., Maskaľová, I., 2016:** *Evaluation of Quality of Feed and Management of Production Health of Animals* (In Slovak). University of Veterinary Medicine and Pharmacy in Košice, Košice, Slovakia, 406 pp.
29. **Vale, W. G., 2007:** Effects of environment on buffalo reproduction. *Ital. J. Anim. Sci*., 6, 2, 130–142. DOI: 10.4081/ijas.2007.s2.130.
30. **West, J. W., Mullinix, B. G., Bernard, J. K., 2003:** Effects of hot, humid weather on milk temperature, dry matter intake, and milk yield of lactating dairy cows. *J. Dairy Sci.*, 2003, 86, 1, 232–242. DOI: 10.3168/jds.S0022-0302(03)73602-9.
31. **Wheelock, J. B., Rhoads, R., P., VanBaale, M., J., Sanders, S., R., Baumgard, L., H., 2010:** Effects of heat stress on energetic metabolism in lactating Holstein cows. *J. Dairy Sci.,* 2010, 93, 2, 644–655. DOI: 10.3168/jds.2009-2295.
32. **Yadav, B., Singh, G., Verma, A. K., Dutta, N., Sejian, V., 2013:** Impact of heat stress on rumen functions. *Vet. World,* 6, 992–996. DOI: 10.14202/vetworld.2013. 992-996.
33. **Zhao, S., Min, L., Zheng, N., Wang, J., 2019:** Effect of heat stress on bacterial composition and metabolism in the rumen of lactating dairy cows. *Animals*, 9, 925. DOI: 10.3390/ani9110925.
34. **Zheng, L., Chenh, M., Zhi-cheng, G., 2009:** Effects of heat stress on milk performance and fatty acids in milk fat of Holstein dairy cows. *J. China Dairy Industry,* 37, 9, 17–19.

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