

## **DIFFERENCES IN THE LEVEL OF UREA IN MILK BETWEEN STANDARD AND EXTENDED LACTATION PERIOD AND THE IMPACT ON THE ENVIRONMENT**

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**Abstract.** The N concentration of milk urea is one of several factors that allow prediction of ammonia emissions from dairy cattle manure. The aim of the study was to investigate the effect of selected environmental factors on urea content in milk in the standard and extended lactation period in Polish Holstein-Friesian cows of the Black-and-White variety. The study has analysed 30 839 full lactations, which were at least one day longer than the standard 305-day lactation, but not longer than 600 days. The highest percentage of milk samples with optimal protein and urea content was found in extended lactation periods (>305 days). The percentage of milk tested during this period, containing urea from 150 to 270 mg · l<sup>-1</sup>, was 49.26%. The calculations also indicated that a significant influence on the differences in the milk urea content between the average standard and extended lactation period had an average daily production of milk in the first 305-day lactation. The higher the efficiency of the cow, the greater the difference in the level of urea in milk in the comparable periods of lactation. On the basis of these data it was estimated that the milk of cows producing on average more than 30 kg of milk per day contained approximately 12 mg · l<sup>-1</sup> more of urea in the standard lactation period than the milk of cows from the same group acquired during the extension. Overall, the results demonstrate that the urea content is higher in the milk from the second phase of lactation, in the milk of youngest cows (first to fourth parity), and in the milk of high yielding cows. It was found that the amount of urea in milk during extended lactation period was lower than in the first 10 months after calving. Due to the fact that the longer lactations resulted in lower levels of urea in milk and lower ammonia emission in last weeks of lactation, it was concluded that the full extended lactation had no negative impact on the environment.

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**Key words:** dairy cattle, milk urea, ammonia emission, extended lactation

## INTRODUCTION

Urea is the end product of nitrogen metabolism in cattle. Urea molecules regulate concentration of body fluids and serum by passive diffusion, and milk urea levels are positively and highly correlated [Butler et al. 1996, Castillo et al. 2003]. Urea concentration and protein content in milk are good indicators of a balanced diet in terms of energy and protein supply [Nagel 1994]. Therefore, if the protein content is within normal range (3.2–3.6%), and urea level is between 150–300 mg · l<sup>-1</sup> [Osten-Sacken 1999]. According to Jonker et al. [1999], the optimum value of this ratio in high yielding cows should be in the range of 100–180 mg · l<sup>-1</sup>. Diet optimization contributes considerably to increased nitrogen efficiency of dairy cattle, resulting in reduced nitrogen losses [Van Duinkerken 2011].

Nutrition, herd milk production level, age of cow and lactation period are the factors that have the greatest effect on the relationship between milk urea concentration and cow fertility parameters. Melendez et al. [2000] demonstrated that the cows with a high urea level (>16 mg · dl<sup>-1</sup>) during 0 to 30 days before the first insemination, and calving during the summer months, were characterized by a higher risk of infertility compared to cows with low milk urea levels that calved in the winter months. Nourozi et al. [2010] demonstrated that cows with MUN values of 12 to <14 and 14 to <16 mg · dl<sup>-1</sup> had higher fertility (15 and 8%, respectively) and cows with MUN values >18 mg · dl<sup>-1</sup> had lower fertility (10%) compared to cows with MUN value <12 mg · dl<sup>-1</sup>.

Dairy cattle barn can be a major source of ammonia emission to the atmosphere. Further aspects of this issue are related to feeding excess protein to dairy cattle, which is a growing concern for the environment in the last decade. Overfeeding of protein contributes to environmental pollution and higher costs of the feed [Burgos et al. 2007]. Under practical conditions, only 20 to 30% of the nitrogen (crude protein) fed to a dairy cow is converted into milk protein [Jonker et al. 2002, Powell et al. 2008]. Animal husbandry wastes can contribute to nitrogen (N) pollution of the environment as ammonia volatilizes to the air, nitrate leaches to ground water and N runs off to surface water. Nitrogen in milk urea may be a useful tool to predict the urinary N excretion (UN) and ultimately NH<sub>3</sub> emission from dairy cattle manure [Jonker et al. 1998, Burgos et al. 2007, Van Duinkerken et al. 2011].

The amount of NH<sub>3</sub> emission depends on many factors such as the type of animals, their weight and age, the composition of animal feed, time and type of animal housing and manure storage, method of waste application, the type and amount of used organic and inorganic fertilizer, meteorological conditions, soil

conditions or the regulation of agricultural practices and dietary crude protein (CP) [Broderick and Clayton 1997, Hutchings et al. 2001, Aguerre et al. 2010]. Major dietary strategies to mitigating N<sub>2</sub>O emission from cattle operations include reducing dietary N content or increasing energy content, and increasing dietary mineral content to increase urine volume. For further reduction of N<sub>2</sub>O emission, an integrated animal nutrition and excreta management approach is required [Dijkstra et al. 2013].

Milk production systems have traditionally been designed for maximisation of daily yield in the belief that this would increase economic efficiency. One consideration is the fact that high-yielding animals managed conventionally and calving at yearly intervals are increasingly being dried off while still producing significant quantities of milk. Yields of 10–20 kg per day at drying off are not uncommon. Another consideration is the perception that the high-yielding dairy cow is already under metabolic stress, and any further increase in milk yield would be at the expense of animal welfare.

The objectives of this study were to describe the influence of parity, calving season and the production level in Black and White cows on the concentration of milk urea obtained during the extended lactation period (from day 306 to the end of lactation) compared to the standard lactation. We also analysed, by assessing the level of urea in milk, whether the extension period of lactation has a negative impact on the environment.

## **MATERIAL AND METHODS**

The study was conducted on cows calving in 2005–2008, raised on private farms, and included in the program of milk production evaluation. Analyses of 30 839 full lactations were performed, which were at least one day longer than the standard 305-day lactation but not longer than 600 days. Data concerning daily milk performance were obtained from animal records kept by the Polish Federation of Cattle Breeders and Dairy Farmers. Physical and chemical milk evaluation was performed by laboratories accredited by the Polish Accreditation Centre that apply the quality system in accordance with the Standard PN-EN ISO/IEC 17 025 and operate under the ICAR guidelines.

The following definitions of the evaluated lactations, which constitute the main factor influencing milk urea level, are employed throughout the present work: standard lactation – 305-day lactation; extended lactation period – the period from day 306 to the end of lactation when a cow is dried off.

In the present work, the effect of the following factors was evaluated on the urea level in the milk of cows in the extended lactation: cow's parity (1; 2; 3–4; ≥5); calving season (September–November; December–February; March–May;

June–August); and average daily milk yield (in kg) in standard lactation ( $\leq 15$ ; 15.1–20.0; 20.1–30.0;  $\geq 30.1$  kg).

Statistical analysis included two-way variance analysis using the method of least squares according to the following linear model:

$$Y_{ijk} = \mu + a_i + b_j + (ab)_{ij} + e_{ijk};$$

where:

$Y_{ijk}$  – dependent variable (MU);

$\mu$  – overall mean;

$a_i$  – effect of lactation definition ( $i$  = standard lactation, extended lactation phase) or lactation stage ( $i$  = 1–100 days; 101–200 days; 201–305 days;  $>305$  days);

$b_j$  – effect of  $j$ -th parity ( $j$  = 1, 2, 3–4;  $\geq 5$ ), or  $j$ -th average daily milk yield in standard (305 days) lactation ( $j$  =  $\leq 15$ ; 15.1–20.0; 20.1–30.0;  $\geq 30.1$  kg); or  $j$ -th calving season ( $j$  = September–November; December–February; March–May; June–August);

$(ab)_{ij}$  – interaction of lactation definition (or lactation stage) with parity (or calving season or average daily milk yield in standard lactation);

$e_{ijk}$  – residual error.

The analysis included one-way and two-way ANOVA with interaction, using Tukey's honest significant difference test.

Wood's mathematical model was used to fit lactation curves of milk urea level [Wood 1976]. The model can be expressed by function:

$$Y_n = an^b \cdot e^{-cn};$$

where:

$Y_n$  – is the milk urea in the  $n$ th month;

$a, b, c$  – define the curve of milk urea of a character  $y$  in the month of lactation  $n$ .

The following equations were used to predict ammonia emission:

1. MUN (milk urea nitrogen,  $\text{mg} \cdot \text{dl}^{-1}$ ) = MU (milk urea,  $\text{g} \cdot \text{l}^{-1}$ )  $\times$  47 [Butler et al. 1996],
2. UN (urea nitrogen, g per day) for Holstein cows = 17.6  $\times$  MUN ( $\text{mg} \cdot \text{dl}^{-1}$ ) [Kauffman and St-Pierre 2001],
3. Ammonia emission (g per day per cow) = 25.0 + 5.03  $\times$  MUN ( $\text{mg} \cdot \text{dl}^{-1}$ ) [Burgos et al. 2010].

ANOVA models were used for statistical calculations and GLM of the SAS package [2012]. Results are presented as treatment means with an s.e.d. and significance was considered at  $P \leq 0.01$ .

## RESULTS

The average full lactation lasted 396 days and the average production of cows during this period amounted to 7727 kg of milk, containing 4.26% fat and 3.31% protein (Table 1). The highest productions during the extended lactation period (from day 306 to the end of lactation, up to a maximum of 600 days) had primiparous cows. Full lactation of these cows was longer than the standard one up to 98 days, and milk yield during this period averaged 1425 kg.

Table 1. Statistical characteristics (mean,  $\pm$ SD) of milk parameters for the population of cows which extended their standard lactation

Tabela 1. Charakterystyka statystyczna (średnia,  $\pm$ SD) parametrów mleka populacji krów, które przedłużyły laktację standardową

	Parity – Wycielenie			
	1	2	3, 4	$\geq 5$
No. of lactations – Liczba laktacji	8970	6926	8774	6169
Standard lactation (305-days) – Laktacja standardowa (305-dni)				
Milk yield – Wydajność mleka				
kg	5988 $\pm$ 1400	6707 $\pm$ 1603	6907 $\pm$ 1568	6422 $\pm$ 1478
kg per day – kg na dzień	19.6 $\pm$ 4.6	22.0 $\pm$ 5.3	22.6 $\pm$ 5.2	21.0 $\pm$ 4.9
Milk composition, % – Skład mleka, %				
Fat – Tłuszcz	4.16 $\pm$ 0.50	4.20 $\pm$ 0.52	4.22 $\pm$ 0.52	4.18 $\pm$ 0.52
Protein – Białko	3.20 $\pm$ 0.23	3.29 $\pm$ 0.25	3.24 $\pm$ 0.24	3.20 $\pm$ 0.23
Extended lactation period (from day 306 to the end of lactation) Okres przedłużenia laktacji (od 306 dnia do końca laktacji)				
Length, days – Długość, dni	98 $\pm$ 93	88 $\pm$ 87	84 $\pm$ 81	78 $\pm$ 75
Milk yield – Wydajność mleka				
kg	1425 $\pm$ 1410	1260 $\pm$ 1160	1175 $\pm$ 1025	992 $\pm$ 889
kg per day – kg na dzień	14.3 $\pm$ 4.3	13.8 $\pm$ 4.5	13.7 $\pm$ 4.6	12.3 $\pm$ 4.3
Milk composition, % – Skład mleka, %				
Fat – Tłuszcz	4.78 $\pm$ 0.78	4.80 $\pm$ 0.81	4.74 $\pm$ 0.83	4.62 $\pm$ 0.84
Protein – Białko	3.80 $\pm$ 0.46	3.94 $\pm$ 0.51	3.91 $\pm$ 0.48	3.83 $\pm$ 0.51
Complete lactation – Laktacja pełna				
Length, days – Długość, dni	403 $\pm$ 93	393 $\pm$ 87	389 $\pm$ 81	383 $\pm$ 75
Milk yield – Wydajność mleka				
kg	7413 $\pm$ 2348	7967 $\pm$ 2417	8082 $\pm$ 2247	7414 $\pm$ 2092
kg per day – kg na dzień	18.5 $\pm$ 4.3	20.3 $\pm$ 4.8	20.9 $\pm$ 4.8	19.5 $\pm$ 4.5
Milk composition, % – Skład mleka, %				
Fat – Tłuszcz	4.25 $\pm$ 0.51	4.28 $\pm$ 0.53	4.28 $\pm$ 0.53	4.23 $\pm$ 0.52
Protein – Białko	3.30 $\pm$ 0.25	3.36 $\pm$ 0.25	3.32 $\pm$ 0.25	3.27 $\pm$ 0.23

The study assessed the effect of environmental factors on the selected percentage of milk samples containing from 150 to 270 mg of urea in one liter, considered an optimal range, and the percentage of milk samples containing above 270

mg · l<sup>-1</sup> (Table 2). It was found that nearly all groups of animals, regardless of parity, calving season, and the average daily milk production in standard lactation samples had the highest percentage of the optimum urea content in milk in the last period of lactation exceeding 305 days. The percentage of the milk tested during this period, containing from 150–270 mg · l<sup>-1</sup>, ranged from 47.72% (calving season March–May) to 51.76% (average daily milk yields in 305-day ≥ 30.1 kg). The highest percentage of samples of milk urea content exceeding 270 mg · l<sup>-1</sup> was demonstrated in the second lactation period (101–200 day) – 27.72%.

Table 2. Proportion of milk samples with urea content between 150 to 270 mg · l and >270 mg · l<sup>-1</sup>\*

Tabela 2. Procent prób mleka o zawartości mocznika od 150 do 270 mg · l<sup>-1</sup> i >270 mg · l<sup>-1</sup>\*

Factors Czynniki	Proportion of milk samples with urea content Procent prób mleka o określonej zawartości mocznika							
	150–270 mg · l <sup>-1</sup>				>270 mg · l <sup>-1</sup>			
	Lactation stage, days Okres laktacji, dni				Lactation stage, days Okres laktacji, dni			
	1–100	101–200	201–305	> 305	1–100	101–200	201–305	> 305
Parity – Wycielenie								
1	46.14	45.07	46.25	48.33	19.31	28.73	26.02	22.60
2	42.94	44.21	47.48	49.41	22.82	29.15	26.01	25.17
3, 4	42.12	44.66	48.20	50.12	18.77	27.24	24.18	22.35
≥5	39.20	44.47	47.49	49.85	16.10	25.17	22.92	20.17
Calving season, month – Sezon wycielenia, miesiąc								
September–November Wrzesień–Listopad	45.49	48.36	48.71	49.18	20.80	22.61	22.85	24.05
December–February Grudzień–Luty	45.41	44.35	42.94	50.49	15.32	24.87	32.00	20.86
March–May Marzec–Maj	38.86	39.58	47.21	47.72	17.79	37.24	24.92	22.67
June–August Czerwiec–Sierpień	41.61	46.54	50.35	49.77	24.37	25.76	19.82	24.95
Average daily milk yields in the 305-day, kg – Średnia dobowa wydajność mleka w laktacji 305-dniowej, kg								
≤15.0	37.85	41.52	45.15	47.58	20.79	25.47	20.67	21.84
15.1–20.0	39.49	43.89	46.32	48.34	18.23	25.70	22.37	21.42
20.1–30.0	44.91	45.40	48.05	49.58	19.03	28.30	26.31	22.91
≥30.1	47.84	46.50	49.33	51.76	25.15	36.25	32.42	27.81
Average – Średnio	42.95	44.64	47.33	49.26	19.35	27.72	25.55	22.74

\*The complement to 100% corresponds to the <150 mg · l<sup>-1</sup>.

\*Uzupełnienie do 100% odpowiada zawartości mocznika <150 mg · l<sup>-1</sup>.

The main objective of the analysis was to compare the level of urea in the milk of lactating cows harvested in standard lactation (305-day) to the level of urea in the milk from extended lactation period (from 306 to the end of lactation) based on the designated non-genetic factors. The data in Table 3 indicate a statistically

significant ( $P \leq 0.01$ ) effect of calving season and the average daily milk production in 305-day lactation on the content of urea in milk and lactation standard extension period. These results comprise the average differences between milk urea level for standard and extended lactation phase, calculated using one-way analysis of variance (ANOVA) for all data. The largest difference was observed in two groups of animals, which calved in the months September–November and March–May. Feeding undoubtedly had the greatest impact on the amount of urea in milk in both of these groups. The extended lactation period (from day 306 to the end of lactation) for cows that calved in the autumn (September–November) fell in the summer months, the period of grass feeding. Thus, the amount of urea in the last period of lactation was higher when compared to the average content in standard lactation. However, cows that calved in the spring (March–May) had the period of extension in the winter months, and therefore, the level of urea in milk was higher during the first 305-day lactation.

Table 3. Milk urea levels (MU,  $\text{mg} \cdot \text{l}^{-1}$ ) in 305-day and extended lactation period

Tabela 3. Zawartość mocznika w mleku (MU,  $\text{mg} \cdot \text{l}^{-1}$ ) w laktacji 305-dniowej oraz w okresie przedłużenia laktacji

Factors Czynniki	MU ( $\text{mg} \cdot \text{l}^{-1}$ )			
	Standard lactation Laktacja standardowa		Extended lactation period Okres przedłużenia laktacji	
	Mean – Średnia	SD	Mean – Średnia	SD
Parity – Wycielenie				
1	213 <sup>Aa</sup>	67.9	208 <sup>Ab</sup>	81.7
2	214 <sup>Aa</sup>	68.2	209 <sup>Ab</sup>	82.2
3, 4	208 <sup>Ba</sup>	67.3	204 <sup>Bb</sup>	79.3
≥5	202 <sup>Ca</sup>	67.1	198 <sup>Cb</sup>	80.9
Calving season, month – Sezon wycielenia, miesiąc				
September–November – Wrzesień–Listopad	202 <sup>Cb</sup>	67.2	224 <sup>Aa</sup>	86.1
December–February – Grudzień–Luty	212 <sup>Ba</sup>	67.9	209 <sup>Ba</sup>	80.2
March–May – Marzec–Maj	218 <sup>Aa</sup>	69.1	196 <sup>Cb</sup>	76.4
June–August – Czerwiec–Sierpień	204 <sup>Ca</sup>	65.7	197 <sup>Cb</sup>	79.0
Average daily milk yields in the 305-day, kg – Średnia dobowo wydajność mleka w laktacji 305-dniowej, kg				
≤15.0	198 <sup>Da</sup>	67.6	194 <sup>Cb</sup>	82.7
15.1–20.0	203 <sup>Ca</sup>	67.6	198 <sup>Cb</sup>	81.8
20.1–30.0	214 <sup>Ba</sup>	66.7	209 <sup>Bb</sup>	79.1
≥30.1	235 <sup>Aa</sup>	79.8	229 <sup>Ab</sup>	82.3

A, B, C – means in columns, followed by different letters differ significantly at  $P \leq 0.01$ , a, b – means in rows, followed by different letters differ significantly at  $P \leq 0.05$ .

A, B, C – średnie w kolumnach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,01$ , a, b – średnie w wierszach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,05$ .

Average protein (Table 1) and urea (Table 3) content in milk during extended lactation period, depending on the parity, was 3.80% and  $208 \text{ mg} \cdot \text{l}^{-1}$  (first parity),

Table 4. Milk urea levels (MU,  $\text{mg} \cdot \text{l}^{-1}$ ) during different lactation stages of cows which extended their standard lactationTabela 4. Zawartość mocznika w mleku (MU,  $\text{mg} \cdot \text{l}^{-1}$ ) w różnych okresach laktacji krów, które przedłużały laktację standardową

Factors Czynniki	MU ( $\text{mg} \cdot \text{l}^{-1}$ )							
	1–100		101–200		201–305		>305	
	Mean Średnia	SD	Mean Średnia	SD	Mean Średnia	SD	Mean Średnia	SD
Parity – Wycielenie								
1	207 <sup>Ac</sup>	96.2	219 <sup>Aa</sup>	88.2	213 <sup>Ab</sup>	83.9	192 <sup>Bd</sup>	81.9
2	209 <sup>Ac</sup>	102.6	218 <sup>Aa</sup>	89.5	214 <sup>Ab</sup>	81.9	195 <sup>Ad</sup>	79.2
3, 4	202 <sup>Bb</sup>	100.0	213 <sup>Ba</sup>	88.4	210 <sup>Ba</sup>	81.4	189 <sup>Cc</sup>	78.5
≥5	187 <sup>Cb</sup>	96.3	206 <sup>Ca</sup>	87.4	205 <sup>Ca</sup>	82.8	185 <sup>Db</sup>	79.2
Calving season, month – Sezon wycielenia, miesiąc								
September–November Wrzesień–Listopad	201 <sup>Ba</sup>	91.6	203 <sup>Ca</sup>	80.1	201 <sup>Ca</sup>	81.1	202 <sup>Aa</sup>	85.3
December–February Grudzień–Luty	187 <sup>Cd</sup>	88.5	205 <sup>Cb</sup>	86.8	230 <sup>Aa</sup>	89.0	199 <sup>Bc</sup>	80.4
March–May Marzec–Maj	200 <sup>Bc</sup>	105.7	239 <sup>Aa</sup>	98.2	211 <sup>Bb</sup>	80.5	182 <sup>Cd</sup>	75.2
June–August Czerwiec–Sierpień	222 <sup>Aa</sup>	106.7	209 <sup>Bb</sup>	81.3	199 <sup>Cc</sup>	75.0	180 <sup>Cd</sup>	77.0
Average daily milk yields in the 305-day, kg – Średnia dobowa wydajność mleka w laktacji 305-dniowej, kg								
≤15.0	197 <sup>Cb</sup>	103.4	204 <sup>Da</sup>	92.4	197 <sup>Db</sup>	84.6	182 <sup>Cc</sup>	83.4
15.1–20.0	197 <sup>Cc</sup>	102.5	209 <sup>Ca</sup>	90.2	202 <sup>Cb</sup>	83.5	185 <sup>Cd</sup>	81.6
20.1–30.0	203 <sup>Bb</sup>	96.5	217 <sup>Ba</sup>	86.5	216 <sup>Ba</sup>	80.5	193 <sup>Bc</sup>	77.8
≥30.1	229 <sup>Ac</sup>	93.5	240 <sup>Aa</sup>	83.7	235 <sup>Ab</sup>	82.3	207 <sup>Ad</sup>	77.7

A, B, C – means in columns, followed by different letters differ significantly at  $P \leq 0.01$ , a, b, c, d – means in rows, followed by different letters differ significantly at  $P \leq 0.05$ .

A, B, C – średnie w kolumnach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,01$ , a, b, c, d – średnie w wierszach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,05$ .

3.94% and  $209 \text{ mg} \cdot \text{l}^{-1}$  (second parity), 3.91% and  $204 \text{ mg} \cdot \text{l}^{-1}$  (third and fourth parities), 3.83% and  $198 \text{ mg} \cdot \text{l}^{-1}$  (fifth and higher parities), respectively. The analysis of the content of milk components in the extended lactation period revealed that the feed rations of cows during this period contained too much energy. This can lead to cows' overweight and many health problems during the next lactation.

The calculations included in Table 3 also indicate that the average daily production of milk in the first 305-day lactation period had a significant influence on the differences in milk urea content between the standard lactation and the average extended lactation period. The higher the efficiency of the cow, the greater the difference in the level of urea in milk in the comparable periods of lactation. On the basis of these data it was estimated that the milk of cows producing on average more than 30 kg of milk per day contained approximately  $12 \text{ mg} \cdot \text{l}^{-1}$



Table 5. Urea nitrogen (UN, g per day) and ammonia emission (g per day per cow) in standard lactation and extended lactation period

Tabela 5. Poziom azotu mocznikowego (UN, g na dzień) i emisja amoniaku (g na dzień) od jednej krowy w laktacji standardowej i okresie przedłużenia laktacji

Factors Czynniki	UN, g per day Poziom azotu mocznikowego, g na dzień				Ammonia emission, g per day per cow Emisja amoniaku, g na dzień od jednej krowy				
	Standard lactation Laktacja standardowa		Extended lactation period Okres przedłużenia laktacji		Standard lactation Laktacja standardowa		Extended lactation period Okres przedłużenia laktacji		
	Mean Średnia	SD	Mean Średnia	SD	Mean Średnia	SD	Mean Średnia	SD	
Parity – Wycielenie									
1	177 <sup>Aa</sup>	57.76	159 <sup>Bb</sup>	67.72	75.6 <sup>Aa</sup>	16.51	70.3 <sup>Bb</sup>	19.35	
2	178 <sup>Aa</sup>	58.60	161 <sup>Ab</sup>	65.55	75.9 <sup>Aa</sup>	16.75	71.0 <sup>Ab</sup>	18.73	
3, 4	173 <sup>Ba</sup>	57.71	156 <sup>Cb</sup>	64.90	74.6 <sup>Ba</sup>	16.49	69.7 <sup>Cb</sup>	18.55	
≥5	167 <sup>Ca</sup>	56.75	153 <sup>Db</sup>	65.52	72.6 <sup>Ca</sup>	16.22	68.7 <sup>Db</sup>	18.72	
Calving season, month – Sezon wycielenia, miesiąc									
September–November Wrzesień–Listopad	169 <sup>Ba</sup>	57.65	167 <sup>Aa</sup>	70.55	73.2 <sup>Ba</sup>	16.48	72.7 <sup>Aa</sup>	20.16	
December–February Grudzień–Luty	172 <sup>Ca</sup>	56.38	164 <sup>Ba</sup>	66.48	74.1 <sup>Ca</sup>	16.11	71.9 <sup>Ba</sup>	19.00	
March–May Marzec–Maj	181 <sup>Aa</sup>	59.69	150 <sup>Cb</sup>	62.16	76.8 <sup>Aa</sup>	17.06	68.0 <sup>Cb</sup>	17.76	
June–August Czerwiec–Sierpień	176 <sup>Ba</sup>	57.18	149 <sup>Cb</sup>	63.70	75.2 <sup>Ba</sup>	16.34	67.6 <sup>Cb</sup>	18.20	
Average daily milk yields in the 305-day, kg – Średnia dobowo wydajność mleka w laktacji 305-dniowej, kg									
≤15.0	167 <sup>Ca</sup>	57.58	150 <sup>Cb</sup>	68.95	72.8 <sup>Ca</sup>	16.46	68.0 <sup>Cb</sup>	19.71	
15.1–20.0	169 <sup>Ca</sup>	58.39	153 <sup>Cb</sup>	67.54	73.4 <sup>Ca</sup>	16.69	68.8 <sup>Cb</sup>	19.30	
20.1–30.0	176 <sup>Ba</sup>	56.80	160 <sup>Bb</sup>	64.37	75.3 <sup>Ba</sup>	16.23	70.7 <sup>Bb</sup>	18.39	
≥30.1	195 <sup>Aa</sup>	59.79	171 <sup>Ab</sup>	64.26	80.6 <sup>Aa</sup>	17.09	74.0 <sup>Ab</sup>	18.36	
Average – Średnio	174 <sup>a</sup>	57.89	157 <sup>b</sup>	66.06	74.8 <sup>a</sup>	16.54	70.0 <sup>b</sup>	18.88	

A, B, C – means in columns followed by different letters differ significantly at  $P \leq 0.01$ , a, b – means in rows followed by different letters differ significantly at  $P \leq 0.05$ .

A, B, C – średnie w kolumnach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,01$ , a, b – średnie w wierszach oznaczone różnymi literami różnią się istotnie przy  $P \leq 0,05$ .

more of urea in the standard lactation period than the milk of cows from the same group acquired during the extension.

Detailed data on the average urea content in milk in different lactation stages in relation to the selected factors are shown in Table 4. The average level of urea in milk obtained from cows that calved in 2005–2008 ranged between  $180 \text{ mg} \cdot \text{l}^{-1}$  (lactation stage >305 days, calving season June–August) to  $240 \text{ mg} \cdot \text{l}^{-1}$  (101–200 days lactation stage, average daily milk yields in the 305 day  $\geq 30.1$  kg). The highest content of urea in the milk of cows was recorded in the second period of

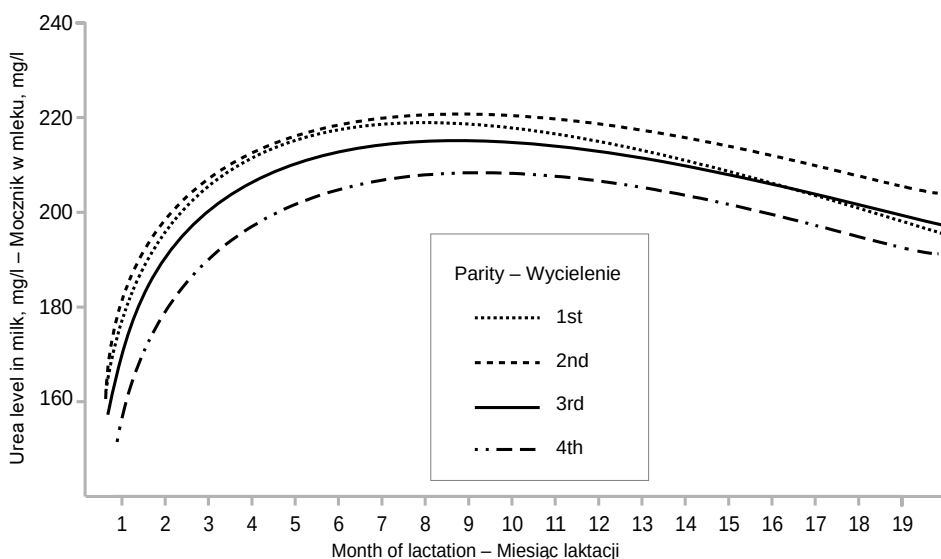


Fig. 1. Milk urea level ( $\text{mg} \cdot \text{l}^{-1}$ ) of cows in extended full lactation according to parity

Rys. 1. Zawartość mocznika w mleku ( $\text{mg} \cdot \text{l}^{-1}$ ) u krów w laktacji pełnej przedłużonej w zależności od wycielenia

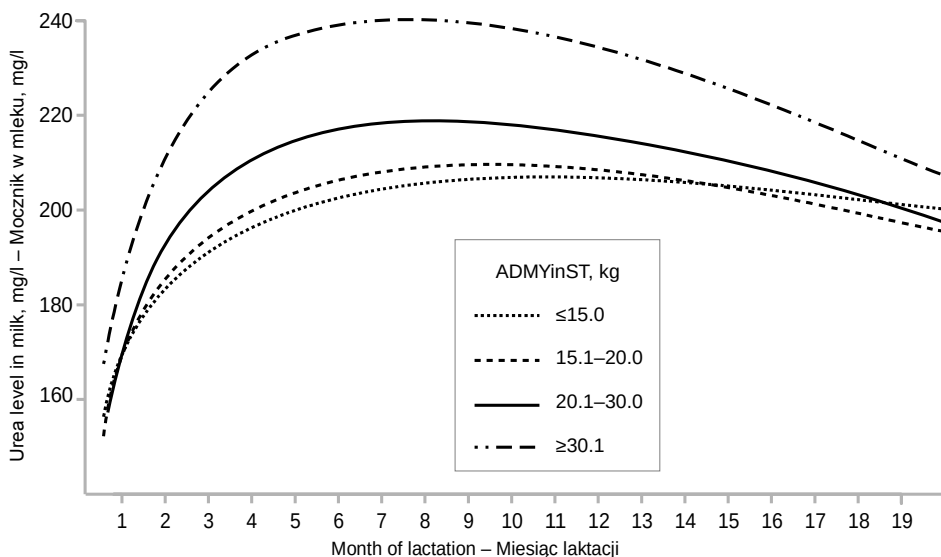


Fig. 2. Milk urea level ( $\text{mg} \cdot \text{l}^{-1}$ ) of cows in extended full lactation according to average daily milk yield in standard lactation (ADMYinST).

Rys. 2. Zawartość mocznika w mleku ( $\text{mg} \cdot \text{l}^{-1}$ ) u krów w laktacji pełnej przedłużonej w zależności od średniej dobowej wydajności mleka z laktacji standardowej (ADMYinST)

lactation, i.e., 101–200 ( $214 \text{ mg} \cdot \text{l}^{-1}$ ), while the lowest ( $190 \text{ mg} \cdot \text{l}^{-1}$ ) was found in the final stage from day 306 to the end of lactation. These differences were statistically significant at  $P \leq 0.05$ .

The next step of the analysis was to present the graphical change in the level of urea in the milk of cows from the population of Polish Holstein-Friesian black and white during the months of lactation. Due to their apparent disparity, it was decided to include the effects of two factors, i.e., the relationship of the parity and mean daily milk yield in lactating standard to the amount of urea in milk. This analysis confirmed higher urea content in milk in the second phase of lactation, in the milk of youngest cows (first to fourth parity), and in the milk of high yielding cows (mean daily milk production in lactating standard  $>30 \text{ kg}$ , Fig. 1, 2).

The final element of the study was to evaluate and compare the amount of emitted ammonia (g per day per cow) in the comparable periods of lactation, i.e., in the standard lactation and during the extended period of lactation. Higher emissions of ammonia were found in the first 305 days from calving ( $74.8 \text{ g per day}$ ) when compared to the extension of lactation ( $70.0 \text{ g per day}$ ). It should be noted that in both assessment periods, based on the separate groups of factors, the highest emission of ammonia showed the high-performance cows, i.e., cows, whose average daily milk yield in 305-day amounted to more than  $30.1 \text{ kg}$ .

## **DISCUSSION**

A properly balanced ration for dairy cows, in terms of protein and energy, is a prerequisite for the optimal course of lactation and proper composition of milk [Jonker et al. 1999]. Particularly vulnerable are the nutritional cows of high efficiency where the attention should be paid to the proportion in the feed ration of two key components, i.e., protein and energy and their inter-relationships. Many studies point to the negative effects of improperly balanced ration for cows in terms of protein and energy supply [Godden et al. 2001a, Jankowska et al. 2010, Rajala-Schultz et al. 2001, Szarkowski et al. 2009]. Poorly balanced feeding leads to a drop in production, reproduction disorders, loss of nutrients and pollution by excess nitrogen emission. According to Juszczak and Ziemiński [1997], shortages of these components, are especially dangerous at the peak of lactation, when the cow's nutritional requirements exceed the supply of the relevant components of the feed, and the resulting deficit must be covered with the reserves of the body. On average 47.6% of the milk samples tested had a proper supply of protein in the feed, while most such cases were recorded during the extended lactation period (49.26%). Studies conducted by Guliński et al. [2008] found that milk samples providing properly balanced rations in terms of protein and energy amounted to 17%. The milk collected from cows during the first weeks of lactation contained

less urea, due to altered physiology of cows after calving. The rapid growth of milk yield in early lactating cows accompanied by a loss of appetite, and thus reduced amount of feed consumption. Along with the duration of lactation, appetite improves, and milk yield also increases as well as the amount of urea. The current study also found that the highest amount of urea occurred in the second period of lactation (101–200 days). Peak of the MUN concentration ( $14.4 \text{ mg} \cdot \text{dl}^{-1}$ ) occurred at 78th day in milk. In contrast, peak of the milk production occurred at day 65 in milk [Jonker et al. 1999]. The level of milk urea was lowest during the first 60 days of lactation, and higher between days 60 and 150 [Godden et al. 2001]. The gradual increase in the urea content in milk from the first to the third month of lactation was also observed in a study of Winnicki et al. [2010], where the urea content in the milk of older cows (2nd to 7th parity) increased within successive months of lactation from 252 in the 1st, 275 in the 2nd, to  $297 \text{ mg} \cdot \text{l}^{-1}$  during the third month of lactation.

The third and fourth lactation period (201–305 days, and >305 day) were characterized by a reduction of the concentration of urea in milk. Similarly, in a study by Godden et al. [2001] approximately at day 150, the amount of urea in milk was decreasing. Osten-Sacken [2000] claims that the urea level in milk during lactation fluctuates to a certain degree, i.e., it is lower at the beginning of lactation, subsequently increases, and then decreases towards the end of lactation. A study by Nałęcz-Tarwacka and Grodzki [2004], showed that the highest amount of urea in milk is found most frequently in cows in the final stage of lactation (from the eighth month onwards). In turn, Antkowiak et al. [2007] found a systematic increase in the level of urea in milk during successive stages of lactation.

The evaluation of the impact of milk production on the amount of urea in different periods of lactation showed that higher performance during standard lactation is accompanied by higher urea content in milk. Genetic correlations studied by Yazgan et al. [2010] indicated similar relationship. Genetic correlations between the milk yield and milk urea nitrogen in the period from day 305 to 395 of lactation were approximately 0.7–0.8. The study of Litwińczuk et al. [2003] found the highest ( $205.3 \text{ mg} \cdot \text{l}^{-1}$ ) amount of urea in cows with daily performance ranging from 21 to 30 kg, while the work of Borkowska et al. [2006] found that the highest milk yield (> 25 kg) was accompanied by the lowest level of urea ( $228 \text{ mg} \cdot \text{l}^{-1}$ ), while the highest ( $275 \text{ mg} \cdot \text{l}^{-1}$ ) level was detected in cows producing less than 15.1 kg.

There have been a number of articles showing that MUN is a good indicator of ammonia emission [Burgos et al. 2007, Jonker et al. 1998, Powell et al. 2011, Van Daunkerken et al. 2005]. Ammonia is a gas toxic not only for animals but also for people, and it negatively affects the natural environment. Total emission of European countries is estimated at 8 mln Mg per year and 72% of this amount

comes from an animal farming [Sapek 1995]. In Poland, ammonia emission from dairy cows was estimated at 76.580 Mg in 2005, according to National Emission Center [Gorzka et al. 2012].

Ammonia emission ranged from 57.6 to 80.8 g per day per cow in standard lactation and from 67.6 to 74.0 g per day per cow in extended lactation period (Table 5). In the literature, values of ammonia emission per cow range from 5 to 35 g of N [Powell et al. 2008] and from 19 to 143 g of N [Cassel et al. 2005]. NH<sub>3</sub> release from cow manure was reduced when the protein level in the diet was lowered and that the concentration of MUN was positively correlated with protein level in the diet and NH<sub>3</sub> emission [Frank and Swensson 2002]. NH<sub>3</sub> emission rates per day per cow place depended on cowshed type, temperature and diet, too [Bleizgys and Baležentienė 2014, Van Duinkerken et al. 2011]. In order to reduce NH<sub>3</sub> pollution from cowsheds should be implemented: cows are kept in cold cowsheds, livestock manure removed as soon as possible and reducing dietary CP content resulted in less total N excretion and less urinary N excretion [Kebreab et al. 2002, Nousiainen et al. 2004].

## CONCLUSIONS

It was demonstrated that non-nutritional environmental factors, such as stage of lactation, parity, season of calving and average daily milk production in standard lactation, have an impact on milk urea (MU) level. It was found that the amount of urea in milk and ammonia emission during extended lactation period was lower than in the first 10 months after calving. However, the largest problem in the estimation of the population standard in prolonged lactating cows was the excess energy administered in the feed at the end of lactation. This phenomenon is dangerous due to the possibility of metabolic diseases and reproductive disorders during the next lactation. All these aspects require more research but they may prove to be useful incentives for future investigations on extended lactations.

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## **RÓŻNICE W ZAWARTOŚCI MOCZNIKA W MLEKU W LAKTACJI STANDARDOWEJ I OKRESIE PRZEDŁUŻENIA LAKTACJI ORAZ WPŁYW NA ŚRODOWISKO**

**Streszczenie.** Stężenie N mocznika w mleku jest jednym z wielu czynników, które umożliwiają przewidywanie emisji amoniaku z odchodów bydła mlecznego. Celem pracy było zbadanie wpływu wybranych czynników środowiskowych na zawartość mocznika w mleku w laktacji standardowej i okresie przedłużenia laktacji u krów rasy polskiej holsztyńsko-fryzyjskiej odmiany czarno-białej. W badaniach przeanalizowano 30 839 laktacji pełnych, trwających od 306 do 600 dni. Najwyższy odsetek prób mleka z optymalną zawartością białka i mocznika stwierdzono w okresie przedłużenia laktacji (>305 dni). Procent prób mleka, które zawierały od 150 do 270 mg · l<sup>-1</sup> mocznika wyniósł 49,26%. Badania wskazują również, na istotne różnice w zawartości mocznika w mleku między laktacją standardową i okresem przedłużenia laktacji u krów w zależności od średniej dobowej produkcji mleka w 305-dniowej laktacji. Oszacowano, że mleko krów produkujących średnio więcej niż 30 kg mleka dziennie zawierało około 12 mg · l<sup>-1</sup> więcej mocznika w laktacji standardowej niż mleko krów z tej samej grupy podczas okresu przedłużenia laktacji. Wyniki wskazują na wyższą zawartość mocznika w mleku w drugim okresie laktacji, w mleku krów najmłodszych (od 1 do 4 wycielenia) oraz w mleku krów wysokowydajnych. Stwierdzono, że zawartość mocznika w mleku w okresie przedłużenia laktacji była niższa niż w pierwszych 10 miesiącach po wycieleniu. Ze względu na fakt, że w okresie przedłużenia laktacji dochodzi do obniżenia stężenia mocznika w mleku i tym samym mniejszą emisję amoniaku stwierdzono, że laktacje pełne przedłużone nie mają tak negatywnego wpływu na środowisko.

**Słowa kluczowe:** bydło mleczne, mocznik w mleku, emisja amoniaku, laktacja przedłużona

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