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ORIGINAL PAPER

CHANGES IN CONCENTRATIONS OF BIOCHEMICAL INDICATORS OF BLOOD OF POLISH HOLSTEIN-FRIESIAN COWS IN THE PERIPARTURIENT PERIOD AND AT THE LACTATION PEAK*

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Abstract

The experiment was conducted on 30 periparturient Polish Holstein-Friesian Black-and-White cows. Animals were kept in a loose-housing system and received TMR (total mixed ration) whose qualitative and quantitative composition varied with the stage of lactation and milk yield. Blood for biochemical analyses was collected by a veterinarian three times: 1-3 weeks prepartum (I), 1-3 weeks postpartum (II), and 4-8 weeks postpartum. Blood samples were analysed for energy metabolism (glucose concentration), protein metabolism (total protein and albumins), liver status (aspartate aminotransferase and alanine aminotransferase) lipid metabolism (cholesterol and triglycerides), and minerals (calcium, magnesium, phosphorus). The parameters were stable and generally within the physiologically normal range, except protein which in lactations 2-3 and 4 slightly exceeded the reference value at the third blood sampling. The reference values were also exceeded for albumins in lactations 1, 2-3 and 4 at the third blood sampling. Differences ($p \leq 0.01$) were observed only in the cholesterol content at the peak of lactation between the first and fourth lactation. The cows yielding more than 9,700 kg milk were characterized by higher blood glucose levels, but lower protein levels compared to the cows producing less than 9,700 kg milk. At the peak of lactation, the protein level exceeded normal values regardless of the milk production level. In our study, peak lactation cows with lower milk yield showed a higher level of AST, which exceeded normal values. In our study, the lipid indicators (cholesterol and triglycerides) were within the physiologically normal range. The highest cholesterol level was observed during the dry period in the blood of lower yielding cows. All blood parameters were stable and fell within the physiologically normal range except for protein, which slightly exceeded the reference value at the third sampling.

Keywords: cows, peripaturient, parameters in the blood, mineral economy.

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INTRODUCTION

Cows in high producing herds are often affected by health issues. Both clinical tests and laboratory analyses are essential to monitoring the health of animals. Recent years have seen intensive research to understand the physiology of dairy cows, especially during the periparturient period. Metabolic disorders in cattle herds are identified based on several diagnostic tests, a vital component of which is the determination of blood biochemical (Oetzel 2004, Mordak, Nicpoń 2006, Tracz et al. 2012) and mineral parameters (Nałęcz-Tarwacka et al. 2015). The health status of animals can be determined from the metabolic profile of their blood, which includes a number of parameters. Metabolic burdens have a direct effect on the reproductive performance and productivity of the cows, thus influencing the economic efficiency of dairy cattle breeding (Mordak, Nicpoń 2006). Because of increased incidence of hypocalcemia, hypomagnesemia and energy deficiencies during the periparturient period, the cow mobilizes body reserves for nutrients (Kurek, Stec 2005). Of particular importance in this context is the dry period, during which the mammary gland and the rumen regenerate in preparation for calving. The nutrients delivered during that period should meet the maintenance requirements of the cows, ensure normal fetal development, and prepare the cow for upcoming lactation (Mashek, Beede 2001). The common metabolic disorders during the periparturient period result in low weight and less resistant calves being born. Early detection of metabolic disorders is essential and hematological and biochemical analyses of blood parameters are helpful in this respect.

The aim of the study was to analyze the impact of the next lactation and milk yield for determination of some mineral and energetic parameters in the blood of cows of periparturient and at the lactation peak.

MATERIALS AND METHODS

The experiment was conducted from June to October 2014 on a farm located in the Kujawsko-Pomorskie province. Out of a herd of 269 Polish Holstein-Friesian Black-and-White cows, 30 periparturient, clinically healthy cows were investigated. Animals were kept in a loose-housing system and received TMR, which during the last 3 weeks of the dry period contained maize silage, 5 kg; grass haylage, 20 kg; wheat straw, 3 kg; and mineral supplements, 0.1 kg. Between calving and 100 days of lactation, the amount of feed was adjusted to ensure milk yield of 39-40 kg, and TMR contained maize silage, 7 kg; lucerne haylage, 6 kg; grass haylage, 4 kg; ensiled sugar--beet pulp, 9 kg; brewer's grains, 5 kg; straw, 1 kg; maize (from silage, 35% d.m.), 4.6 kg; cereals, 2.3 kg, protected fat, 0.3 kg, EKOPROT dried yeast, 1 kg; 37% protein mix, 3 kg; and mineral supplements, 0.4 kg. The rations were formulated according to the INRA (2009).

Blood for analyses was collected by a veterinarian three times: 1-3 weeks prepartum (I), 1-3 weeks postpartum (II), and 4-8 weeks postpartum (III).

Blood samples were analysed for energy metabolism (glucose concentration), protein metabolism (total protein and albumins), liver status (aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities), lipid metabolism (cholesterol and triglycerides), and minerals (calcium, magnesium, phosphorus). The determinations were performed with an Epoll 20 Bio photometer for blood analysis, equipped with a 20-point thermostat. The reagents were purchased from BioMaxima.

The effect of lactation number (1, 2-3, 4) and milk yield (<9,700 kg, >9,700 kg) on biochemical blood parameters at three consecutive samplings was analysed.

The results were compared with the reference values for healthy cattle, reported by Winnicka (2011).

The data were statistically analysed by SAS/STAT software (2014), using analysis of variance. The least square means and the CORR procedure were used.

RESULTS AND DISCUSSION

Table 1 shows the results of determinations of glucose, protein, albumins, AST, ALT, triglycerides, cholesterol, magnesium, phosphorus and calcium at three blood samplings from periparturient cows and from peak lactation cows depending on a lactation number. The parameters were stable and generally within the physiologically normal range, except protein, which in lactations 2-3 and 4 slightly exceeded the reference value at the third blood sampling. The reference values were also exceeded for albumins in lactations 1, 2-3 and 4 at the third blood sampling. Differences ($p \leq 0.01$) were observed only in cholesterol content at the peak of lactation between the first and fourth lactation. Glucose level (2.35 to 2.73 mmol L^{-1}) was the highest in cows within the first three weeks of calving regardless of a lactation number, and the highest between the fourth and eighth week of lactation (from 3.23 in the first lactation to 4.0 mmol L^{-1} in the fourth lactation). During the dry period, glucose level ranged from 2.51 in the second and third lactation to 3.81 mmol L^{-1} in the fourth.

The dry period and early lactation have an effect on the blood metabolic profile, with possible fluctuations in blood parameters during this time. The periparturient period is a time of great physiological stress for dairy cows (Rollin et al. 2010). The highest energy requirement occurs during early lactation (Radkowska 2015). A decrease in glucose is a common physio-

Table 1

Changes in the concentrations of biochemical indicators in three subsequent samples depending on consecutive lactations

Biochemical indicators	1 lactation (n = 3)			2-3 lactation (n = 21)			4 lactation (n = 6)			Standard
	1 download	2 download	3 download	1 download	2 download	3 download	1 download	2 download	3 download	
Glucose (mmol L^{-1})	2.75	2.35	3.23	3.80	2.51	3.52	3.81	2.73	4.05	2.2 - 4.5
Protein (g L^{-1})	71.00	59.80	66.80	63.30	71.40	80.70	54.40	65.40	74.40	51.0 - 71.0
Albumin (g L^{-1})	38.60	41.50	50.60	43.80 a	49.00	52.20	35.10*a*	42.3	55.70	32.0 - 49.0
ALT (U L^{-1})	69.20	74.12	68.56	54.70	42.20	39.87	41.23	38.70	58.90	25.0 - 74.0
AST (U L^{-1})	95.00	119.00	122.50	91.70	85.70	84.11	74.50	79.00	126.50	58.0 - 100.0
TG (mmol L^{-1})	0.08	0.12	0.15	0.16	0.21	0.19	0.20	0.18	0.20	0.1 - 0.3
Cholesterol (mmol L^{-1})	3.03	3.48	2.08*A*	4.30	3.42	3.53	3.15	3.01	4.37*A*	1.8 - 5.2
Phosforus (mmol L^{-1})	1.58	1.08	1.87	1.28	1.76	2.45	1.49	2.00	2.40	1.81 - 2.1
Calcium (mmol L^{-1})	2.40	2.40	2.43	2.22	2.12	2.35	2.05	2.12	2.19	2.25 - 3.03
Magnesium (mmol L^{-1})	1.00	0.75	0.86	0.95	0.87	1.22	0.91	0.84	1.01	0.78 - 1.23

ALT – alanine aminotransferase, AST – aspartate aminotrasnsferase, TG – triacylglycerol logical process during lactation. A decreased concentration of glucose during lactation is attributed to the increased demand for this sugar to support milk lactose synthesis (Mandebvu et al. 2003).

The level of protein increased with each consecutive sampling: 63.3 - 80.7 in the second, 71.4 - 65.4 in the third, and 80.7 - 74.4 mmol L^{-1} in the fourth lactation. In primiparous cows, the lowest protein level was observed within the first three weeks of calving. The lowest phosphorus level was found at the second blood sampling (1.08 mmol L^{-1}) in first lactation cows (within the first three weeks of calving), while in lactations 2-3 and 4, the phosphorus level was lowest at the first blood sampling.

Blood protein and albumin concentration is one of the indicators of nitrogen metabolism in the body, and depends on many factors such as the amount of dietary protein and energy, cow's age or stage of lactation (Kupczyński, Chudoba-Drozdowska 2002). The total protein and albumin level during lactation is higher than during the dry period, which is associated with the production of milk and higher immunoglobulin content of milk (Mohri et al. 2007). Similar to our study, many authors reported the blood protein content to decrease during the dry period and immediately after parturition, and to increase during lactation (Mordak, Nicpoń 2006, Radkowska 2015). The lowest phosphorus level was found at the second blood sampling (1.08 mmol L^{-1}) in first lactation cows (within the first three weeks of calving), while in lactations 2-3 and 4, the phosphorus level was the lowest at the first blood sampling. Likewise, Filipejowa and Kovacik (2009) found phosphorus levels to be significantly lower during the dry period compared to early lactation. In the study by Kupczyński and Chudoba-Drozdowska (2002), the average content of calcium and phosphorus was in the lower limit of normal values for dry cows.

Our results demonstrate that the cows showed normal calcium and phosphorus metabolism. Most of the analysed blood parameters increased with each sampling, but the differences were not statistically significant except for cholesterol. In our study, the lipid indices (cholesterol and triglycerides) were within the physiologically normal range. In the first lactation, the highest level of cholesterol was noted at the second sampling, after calving, and in subsequent lactations the cholesterol level was highest at the peak lactation, which supports the view of Brucka-Jastrzębska and Kawczuga (2007) that a high serum concentration of total cholesterol in periparturient cows is positively related to their reproductive potential. Čitek et al. (1997) and Rossato et al. (2001) showed the cholesterol content to differ between the first and subsequent lactations. They concluded that the cholesterol content increased with the next lactation. Skrzypek (2000) attributed this relationship to the fact that cholesterol is a substrate for synthesis of steroid hormones, which are essential for regulating reproductive function. Filipejowa and Kovacik (2009) observed markedly higher cholesterol concentrations in mid-lactation compared to early lactation. Serum triglyceride concentrations are mainly associated with the changes in the LDL concentration. The increased accumulation of triglycerides in the liver in late pregnancy and during lactation is a physiological condition and when moderate, it will not disrupt this organ (Pöso et al. 2000). In our study, these parameters were within the standard range, which is indicative, among others, of correct nutrition.

The liver parameters aspartate aminotransferase (AST) and alanine aminotransferase (ALT) fell within the normal range during the dry period, while AST exceeded the limit value for elements and in the 4-8th week after calving in the fourth lactation. Górski and Saba (2012) and Radkowska (2015) observed their level to increase in postpartum and early lactation cows, which was also shown in our study. The high level of aminotransferases in postpartum cows may be associated with intense physical exertion, increased metabolism in early lactation, and stress (Górski, Saba 2012).

The level of parameters such as AST, ALT, TG, Mg and Ca was very uniform in the blood sampled before calving and 1-3 weeks post-calving, and assumed slightly higher values during 4-8 weeks of lactation. In our study, we noted a higher blood calcium content in dry and early lactation primiparous cows, and in older cows during the lactation period.

In our study, we found blood calcium concentrations during the dry period and early lactation to be low, slightly below normal values. Similar findings were obtained by Kupczyński and Chudoba-Drozdowska (2002), Kurek and Stec (2005), and Radkowska (2015). Horst et al. (1997) report that daily calcium requirement increases around four-fold during the periparturient period. A small decrease in the blood calcium of postpartum cows is considered a physiological condition (Winnicka 2011). In the study by Nałęcz-Tarwacka et al. (2015), the highest magnesium concentration was observed in the eighth month of lactation and the results were within the normal range. Tetany occurs when magnesium concentrations fall below 1.7 mg dl^{-1}. In our study, the highest magnesium concentration was 1.14 mg dl^{-1} at the third blood sampling.

Table 2 lists the results of determinations of glucose, protein, albumins, AST, ALT, triglycerides, cholesterol, magnesium, phosphorus and calcium at three consecutive blood samplings from periparturient and peak lactation cows, depending on the milk production level. The cows yielding more than 9,700 kg milk were characterized by higher blood glucose levels but lower protein levels compared to the cows producing less than 9,700 kg milk. At the peak of lactation, the protein level exceeded normal values regardless of the milk production level. The glucose concentration generally remains stable or slightly increases during the dry period, but declines considerably after calving (Doepel et al. 2002, Górski, Saba 2012). The decrease in the blood glucose level observed in cows within 1-3 weeks of calving is supported by the results of Kupczyński and Chudoba-Drozdowska (2002), and Radkowska (2015). In the study by Winnicka (2011), serum glucose levels in cows fell within the normal range regardless of the milk yield. In our study, peak lac-

Table 2

Changes in the concentrations of biochemical indicators in three subsequent samples depending on milk yield

Biochemical indicators	Milk yield (kg)						Standard
	< 9700 (n = 16)			> 9700 (n = 16)			
	1 download	2 download	3 download	1 download	2 download	3 download	
Glucose (mmol L^{-1})	2.56	2.38	3.37	3.94	2.74	3.94	2.2 - 4.5
Protein (g L^{-1})	65.40	74.10	78.8	61.40	62.6	77.20	51.0 - 71.0
Albumin (g L^{-1})	41.60	44.60	52.60	41.50	46.10	55.10	32.0 - 49.0
ALT (U L^{-1})	67.14	59.80	62.18	49.87	44.20	46.31	25.0 - 74.0
AST (U L^{-1})	94.68	93.50	106.21	81.71	78.20	86.09	58.0 - 100.0
TG (mmol L^{-1})	0.16	0.18	0.16	0.16	0.22	0.23	0.1 - 0.3
Cholesterol (mmol L^{-1})	4.32	3.67	3.63	3.54	2.89	3.60	1.8 - 5.2
Phosphorus (mmol L^{-1})	1.49	1.73	2.46	1.19	2.06	2.32	1.81 - 2.1
Calcium (mmol L^{-1})	2.14	2.25	2.21	2.29	2.09	2.33	2.25 - 3.03
Magnesium (mmol L^{-1})	0.98	0.81	1.31	0.91	0.90	0.92	0.78 - 1.23

ALT, AST, TG – see Table 1

tation cows with lower milk yield showed a higher level of AST, which exceeded normal values. The changes in AST and ALT activity are associated with the physiological status, in particular with the liver function. In our study, the lipid indicators (cholesterol and triglycerides) were within the physiologically normal range. The highest cholesterol level was observed during the dry period in the blood of lower yielding cows. Different results were reported by Tracz et al. (2012), who showed the highest serum cholesterol concentration in high-yielding cows. The calcium content was within normal limits regardless of milk yield. According to Winnicka (2011), the analysis of the blood Ca content enables abnormal kidney, parathyroid and pancreas function to be diagnosed. According to Kowalski (2012), clinical hypocalcemia induced by blood Ca levels falling below 5 mg dl^{-1} is the main cause of downer cow syndrome. The results of our study indicate that the cows' body homeostasis was not threatened by the blood Ca content (Winnicka 2011). In both lower and higher yielding cows, the phosphorus level exceeded normal values at the peak of lactation. According to Winnicka (2011), blood phosphorus levels can be used to diagnose hyperparathyroidism and hyperthyroidism. In turn, Jamroz (2001) holds the view that this source of information is not accurate because the blood phosphorus concentration is dependent upon the dietary concentration of Mn, Mg and Cu. Our results demonstrate that the cows showed normal calcium and phosphorus metabolism.

Table 3 shows the coefficients of simple correlation for blood parameters between the first vs. second and third sampling, and between the second vs. third sampling. Negative correlations ($p \leq 0.05$) were found for the glucose content between the second (1-3 weeks after calving) and third sampling (4-8 weeks of lactation). In addition, correlations ($p \leq 0.01$) were observed for albumins, negative between the first and third sampling, and positive

Table 3

The values of coefficients of linear correlation between the parameters of blood in individual downloads

Indicators of blood in the first and second downloading	Indicators of blood in the second and third downloading							
	glucose 2	glucose 3	magnesium 2	magnesium 3	phosphorus 2	phosphorus 3	calcium 2	calcium 3
Glucose 1 Glucose 2	-0.04 -	-0.19 -0.53 x	-	-	-	-	-	-
Magnesium 1 Magnesium 2	-	-	0.04 -	-0.02 -0.21	-	-	-	-
Phosphorus 1 Phosphorus 2	-	-	-	-	-0.13 -	0.10 0.80 x		
Calcium 1 Calcium 2	-	-	-	-	-	-	-0.08 -	-0.18 -0.17

cont. Table 3

Indicators of blood in the first and second downloading	Indicators of blood in the second and third downloading									
	protein 2	protein 3	albumin 2	albumin 3	AST 2	AST 3	TG 2	TG 3	cholesterol 2	cholesterol 3
Protein 1 Protein 2	0.34 -	0.38 0.17	-	-	-	-	-	-	-	-
Albumin 1 Albumin 2	 -	 -	0.64 x -	-0.50 x -0.56 x	-	-	-	-	-	-
AST 1 AST 2	-	-	-	-	0.09 -	-0.04 0.17	-	-	-	-
TG 1 TG 2	-	-	-	-	-	-	0.06 -	0.20 0.68 x	-	-
Cholesterol 1 Cholesterol 2	-	-	-	-	-	-	-		0.27 -	0.06 0.18

AST, TG – see Table 1

between the first and second sampling. Significant positive correlations were found for the phosphorus content between the second and third sampling, and for triglycerides also between the second and third sampling. For the parameters such as protein, AST, ALT and cholesterol, the correlations between samplings were positive but low and not significant. For calcium and phosphorus, the correlation coefficients between blood samplings were negative and also low.

CONCLUSION

In our study, no major deviations from the physiologically normal values were established for the analysed blood parameters, which may suggest, among others, that the cows were fed well-balanced diets. Most of the analysed blood parametrers, except for ALT and AST, increased during lactation, and the level was highest at the peak lactation. At higher efficiency, the level of glucose increased with each consecutive sampling, but other blood parametrers were increased, while the efficiency was lower.

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