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EVALUATION OF SELECTED BIOCHEMICAL PARAMETERS IN BLOOD PLASMA, URINE AND MILK OF DAIRY COWS DURING THE LACTATION PERIOD

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ABSTRACT

The aim of our study was to determine selected biochemical parameters in the blood, urine and milk of dairy cows. Fifteen dairy cows from selected agricultural farms were divided into three groups as follow: group I- 3-4 weeks after calving (the beginning of lactation), group II - 3-4 months after calving (the middle of lactation), and group III - 2-3 weeks before calving (the dry period). Concentrations of selected biochemical parameters in the blood (aspartate aminotransferase – AST, urea, total proteins - TP, glucose, and cholesterol), urine (calcium - Ca, phosphorus - P, urea and pH), and milk (fats, proteins, lactose, solids, Somatic Cell Count, urea, milk freezing point and rennetability) were measured. Glucose concentration (3.22±0.21 mmol.l^{-1}) was significantly lower at the beginning of lactation ($p<0.05$) in comparison with the middle of lactation (3.69±0.08 mmol.l^{-1}) and the dry period (3.74±0.21 mmol.l^{-1}). However, concentration of AST (1.42±0.31μkat.l^{-1}) was significantly higher ($p<0.01$) at the beginning of lactation than in the dry period (0.88±0.31 μkat.l^{-1}). Concentration of cholesterol (6.22±0.76 mmol.l^{-1}) in the middle of lactation was significantly higher ($p<0.001$) than those at the beginning of lactation (3.82±0.67 mmol.l^{-1}) and dry period (2.842±0.67 mmol. l^{-1}). Cows in the middle of lactation had a higher freezing point of milk (526.20±1.92 -m.°C) ($p<0.01$) in comparison with dairy cows at the beginning of lactation (520.00±2.74-m°C). Some other symptoms leading to sub-clinical diseases, besides worsening the technological quality of the cow milk, were found.

Key words: biochemical parameters; metabolism; metabolic disorders; milk quality

INTRODUCTION

Various factors influence milk quantity and quality. The quality of milk and its nutritional value influenced by many factors (Tančin et al., 2006; Tančin et al., 2007). One of them is the milking process itself, in which the emphasis is to minimize the undesirable contaminant microflora, which significantly offset the technological quality of milk and may cause health injury (Kirchnerová and Foltys, 2005). Specifically, the nutrition and health status are major factors in determining cow milk characteristics. Metabolic perturbations, lack of nutrition and sub-clinical disorders could be detected by measuring some metabolic parameters in the blood, urine and milk (Slanina and Beseda, 1992). Such parameters are known to influence the composition and technological aspects of milk quality in dairy cows (Illek et al., 2002; Kováčik et al., 2004). Several factors like nutrition, metabolic status, production, health and the management affect dairy cow's fertility (Roxström et al., 2000; Clark et al., 2000; Mwaanga and Janowski, 2000; Mihm, 1999; Kruip et al., 1998). The changes that take place in the endocrine system of cows in the dry period and still more at early lactation intensify gluconeogenesis, lipolysis and ketogenesis (Ling et al., 2003).

Dairy cattle, like most lactating mammals, are usually in negative energy balance within the first few weeks of lactation (Nielsen, 1999). After the delivery, the cow requires high amount of energy from body reserves, but actually it is not able to cover the required nutrients consumed, what leads to the loss of body weight. The continuous increase in milk production has created new

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challenges for high-producing dairy cows, especially during the transition period. At the beginning of lactation, dairy cows have to cope with the high energy and protein demands for milk synthesis at the time when nutrient intake is low. Mobilizing energy and protein from body tissue stores and repartition of nutrients away from extra-mammary tissues are the primary alternatives to supply sufficient nutrients for milk production during the first weeks of lactation. Excessive body reserves, especially fat, can cause a series of metabolic disorders and consequent production losses (Fourichon et al., 1999).

Furthermore, physiological and pathological changes associated with negative energy balance are important factors related to development of ketosis, displaced abomasum, and retained placenta (Duffield *et al.*, 2002).

The aim of our study was a determination of selected biochemical parameters in the blood, urine and milk of dairy cows and their analysis.

MATERIAL AND METHODS

Fifteen dairy cows from selected agricultural farms were divided into three groups: group I - 3-4 weeks after calving (the beginning of lactation), group II - 3-4 months after calving (the middle of lactation) and group III - 2-3 weeks before calving (the dry period).

Blood samples for biochemical analysis were taken from *vena jugularis* 2 hours after morning feeding. The blood plasma was separated from whole blood by the centrifugation at 3000 rpm for 30 minutes and samples were stored at – 18 °C. Selected biochemical parameters in blood plasma (aspartate aminotransferase (AST), urea, total proteins (TP), glucose (GLU) and cholesterol (CHOL), in urine (calcium (Ca), phosphorus (P), urea, pH) were analyzed using semi–automated clinical chemistry analyzer Microlab 300 Vilat Scientific, Dieren, The Netherlands) (Massanyi et al., 1995; Massanyi et al., 2007).

Samples of milk were cooled down until 6 °C was reached. Samples were kept at the same temperature during the determination of milk quality parameters: Somatic Cell Counts (by the Fossomatic device), urea (photocolorimetric device with Ehrlich‘s reagent, 530 nm wavelength), Non-Fat-Solis (by the MilkoScan apparatus), acidity, rennetability, fermentace ability and thermostability. Significant differences between groups of dairy cows were evaluated by the Scheffe´s – test. Differences between the groups at $p<0.05$, $p<0.01$ or $p<0.001$ were considered as significant.

RESULTS AND DISCUSSION

Blood biochemical parameters of animals are affected by seasonal variations (Massányi et al., 2009) or food additives (Arpášová et al., 2009). Measurements of blood metabolites are associated with great variations, indicating large fluctuations in metabolite profiles during the peri-parturient period (Drackley, 1999).

Concentrations of biochemical parameters in blood serum, urine and milk during lactation period are shown in tables 1-3.

Table 1: Concentration of AST, urea, total proteins, glucose, cholesterol in blood plasma of dairy cows at the beginning of lactation, in the middle of lactation and the dry period

	Variable	Minimum	Maximum	x	s	cv
BL	AST (μkat.l^{-1})	1.15	1.90	1.42**	0.31	21.85
	Urea(mmol.l^{-1})	2.18	3.45	2.94	0.48	16.20
	TP (g.l)	72.00	88.00	79.00	6.44	8.15
	GLU(mmol.l$^{-1)}$	2.87	3.45	3.22*	0.21	6.64
	CHOL(mmol.l^{-1})	2.90	4.60	3.82	0.67	17.50
ML	AST (μkat/L)	1.04	1.53	1.28	0.18	14.18
	Urea(mmol.l^{-1})	2.87	5.53	4.82	1.17	24.17
	TP(g.l)	73.00	90.00	78.80	7.26	9.21
	GLU(mmol.l^{-1})	3.61	3.82	3.69	0.08	2.19
	CHOL(mmol.l$^{-1)}$	5.10	7.00	6.22***	0.76	12.21
D	AST (μkat.l)	0.77	1.05	0.88	0.31	21.85
	Urea(mmol.l^{-1})	0.19	0.40	0.30	0.08	25.50
	TP(g.l)	56.00	85.00	73.60	6.44	8.15
	GLU(mmol.l^{-1})	3.43	4.39	3.74	0.21	6.64
	CHOL(mmol.l^{-1})	2.00	3.90	2.84	0.67	17.50

BL-beginning of lactation, ML-middle of lactation, D-dry period, AST- aspartate aminotransferase, TP-total proteins, GLU-glucose, CHOL-cholesterol, x -mean, s-standard deviation, cv- coefficient of variation, * p <0.0 5, ** p<0.01, *** p<0.001

Table 2: Concentration of calcium, phosphorus, urea and pH in urine of dairy cows at the beginning of lactation, in the middle of lactation and the dry period

	Variable	Minimum	Maximum	x	s	cv
BL	Ca (mmol.l^{-1})	0.11	4.39	2.52	2.06	82.03
	P (mmol.l^{-1})	0.09	2.37	0.88	1.05	118.97
	Urea(mmol.l^{-1})	72.00	372.00	223.25	126.45	56.64
	pH (logmolc)	8.00	8.30	8.13	0.15	1.85
ML	Ca (mmol.l^{-1})	0.22	1.17	0.70	0.36	51.35
	P (mmol.l^{-1})	0.10	8.38	1.84	3.66	198.54
	Urea(mmol.l^{-1})	61.00	245.00	179.20	75.05	41.88
	pH (logmolc)	7.80	8.30	8.12	0.20	2.52
D	Ca (mmol.l^{-1})	0.96	3.50	1.85	1.13	61.32
	P (mmol.l^{-1})	0.09	0.86	0.35	0.35	101.24
	Urea(mmol.l^{-1})	135.00	468.00	293.25	136.76	46.64
	pH (logmolc)	8.00	8.30	8.15	0.13	1.58

BL-beginning of lactation, ML-middle lactation, D-dry period, Ca-calcium, P-phosphorus, x-mean, s-standard of deviation, cv- coefficient of variation

Pregnancy and lactation have a great impact on the intensity of metabolism and on metabolic parameters in the blood (Tab. 1). The activity of blood aminotransferases is very important because they act as a catalyst in connection to the metabolism of amino acids and carbohydrates. Changes in their activity in the blood can be a consequence of their increased activity in cells, or damages in cell structure (Milinković-Tur, 2005). The concentration of AST (1.42±0.31μkat.l^{-1}) in dairy cows at the beginning of lactation was significantly higher ($p<0.01$) in comparison with the dry period (0.88±0.31 μkat.l^{-1}). AST is a widely distributed enzyme, which is found in many tissues and organs, with high activity in the liver (Zimmerman et al., 1968). Increased AST activity in the serum is a sensitive marker of liver damage (Meyer and Harvey, 1998). The intensity of metabolic changes, mainly of proteins, is also reflected in the concentration of other biochemical indicators of the blood, such as total protein, urea or aspartate aminotransferase (Falkenberg et al., 1997; Kapelański

Table 3: Concentration of fats, proteins, lactose, somatic cell counts, urea, freezing point, rennetability, in milk of dairy cows at the beginning of lactation, and in the middle of lactation

	Variable	Minimum	Maximum	x	s	cv
BL	Fats (g.100g^{-1})	0.57	1.94	1.20	0.59	4950
	Proteins (g.100g^{-1})	2.93	3.58	3.17	0.26	8.07
	Lactose (g.100g^{-1})	4.73	5.01	4.93	0.12	2.47
	Solids (g.100g^{-1})	9.16	10.79	9.99	0.71	7.14
	Somatic Cell Counts (tis . ml^{-1})	9.00	366.00	134.20	15466	115.25
	Urea (mmol.l^{-1})	18.91	33.84	26.05	7.03	27.00
	Freezing point (-m.°C)	517.00	524.00	520.00	274	0.53
	Rennetability(s)	17.40	26.25	20.63	3.52	17.07
ML	Fats (g.100g^{-1})	0.66	3.27	1.61	1.20	74.34
	Proteins (g.100g^{-1})	2.69	3.85	3.05	0.47	15.26
	Lactose (g.100g^{-1})	4.02	4.99	4.59	0.44	56.64
	Solids (g.100g^{-1})	8.64	11.64	9.97	1.32	13.27
	Somatic Cell Counts (tis . ml^{-1})	20.00	6136.00	1525.40	2619.89	171.75
	Urea (mmol.l^{-1})	20.16	35.23	26.83	6.55	24.43
	Freezing point (-m.°C)	523.00	528.00	526.20**	1.92	0.37
	Rennetability(s)	20.62	46.12	34.17	11.79	34.50

BL-beginning of lactation, ML-middle of lactation, x - mean, s - standard of deviation, cv - coefficient of variation ** $p<0.01$

et al., 2000; Więcek and Skomiał, 2000). Measurements of blood metabolites require more frequent sampling to capture the dynamic changes in the peri-parturient period. The concentration of cholesterol (6.22±0.76 mmol.l^{-1}) in the middle of lactation was significantly higher ($p<0.001$) in comparison with concentration at the beginning of lactation (3.82±0.67 mmol.l^{-1}) and dry period (2.842±0.67 mmol.l^{-1}). Cholesterol increase in the course of lactation together with intensive steroid synthesis has been reported in several studies (Bösö et al., 2000; Pysera and Opalka, 2000).

The concentration of glucose (3.22±0.21 mmol. l^{-1}) in dairy cows at the start of lactation was significantly lower ($p<0.05$) in comparison with those in the middle of lactation (3.69±0.08 mmol.l^{-1}) and at the dry period (3.74±0.21 mmol.l^{-1}).

We did not observe any significant differences in urine indicators of dairy cows (Tab. 2).

Studies on influences of the milk yield increase on cow's health status and milk quality becoming more and more important (Sawa and Piwczynski, 2002; Ayadi et al., 2003; Remond et al., 2004; Junqueir, et al., 2005). However, it is possible only for a limited number of milk indicators, such as the milk yield, fat, protein, lactose and urea contents and somatic cell counts. The milk yield is an important economic and health factor closely connected with the health status of dairy cows, their reproduction performance, longevity and milk composition and properties (Janů et al., 2007). We found, that freezing point of milk (526.20±1.92 -m.°C) in the middle of lactation was significantly different ($p<0.01$) compared to dairy cows at the beginning of lactation (520.00±2.74-m.°C) (Tab. 3).

CONCLUSION

In conclusion, deficiency in dairy cow's nutrition may influence many biochemical and physiological processes. Consequently they disturb the relationship between metabolic capacities of animals required for milk production and, thereby, cause metabolic disorders.

In this study some significant differences related to the lactation period were found. Glucose concentration at the beginning of lactation was significantly lower ($p<0.05$) in comparison with the middle of lactation and the dry period. Concentration of AST at the beginning of lactation was significantly higher ($p<0.01$) in comparison with the dry period. Concentration of cholesterol in the middle of lactation was significantly higher ($p<0.001$) in comparison with the beginning of lactation and the dry period. Freezing point of milk in the middle of lactation was significantly higher ($p<0.01$) in comparison to dairy cows at the beginning of lactation. Obtained results could serve to better understanding of biochemical processes in dairy cows for estimating their physiological status.

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