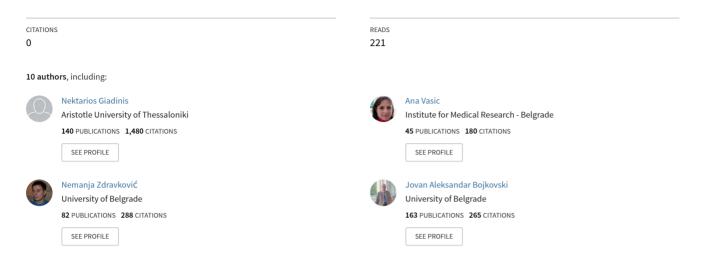
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Determination of Macroelement Parameters in Different Productive Stages of Simmental Cows

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ABSTRACT

The results of inorganic blood parameters are presented in the periparturient period and during lactation of Simmental dairy cows. Serum calcium and inorganic phosphorus in blood of dairy cows in puerperium were significantly lower (p<0.05) compared to the values in the blood of dairy cows in advanced pregnancy and during peak of lactation, probably indicating the increased use of these macro-elements by the Simmental cows' mammary gland at the early stages of lactation. When the cows were in the periparturient period serum magnesium levels were significantly lower (p<0.05) compared to the values of dairy cows during their maximal lactation probably indicating the increased use of magnesium by the Simmental dairy cow during the periparturient period.

Keywords: Simmental cows; Blood Serum; Calcium; Phosphorus; Magnesium.

INTRODUCTION

Deviation from the normal values of the inorganic blood parameter status of cows in early lactation, as well as their deficiency in the diet can lead to subclinical or clinical manifestations (puerperal paresis, tetany), which have a negative impact on health and fertility (1, 2, 3, 4, 5). Late pregnancy and the beginning of lactation present a burden for the cow's physiology, as it has increased demands in calcium, which is necessary for the construction of the fetal skeleton. Phosphorous represents the second most important macro-element for bone tissue production. In addition, it participation in the cellular process of phosphorylation and energy production, affects acid-base balance and plays a role in the detoxification process (6, 7).

Determination of calcium, magnesium and inorganic phosphorus in the blood serum is of important diagnostic

value in preventing puerperal paresis and other diseases (8, 9). However, their values can differ in cattle for many reasons, among which is the breed (10). The values of the above mentioned macro-minerals in Simmental cows particularly at different production stages are lacking in the literature. This study was aimed at determining and comparing levels of serum calcium, inorganic phosphorus and magnesium preparturienly, peri-parturiently and during the peak of lactation in Simmental dairy cattle.

MATERIALS AND METHODS Animal selection and study design

The study was conducted in a dairy cattle farm consisted of 150 Simmental cows. In total, 45 clinically healthy cows of 3 different productive stages were selected. The animals of the 1st group (15 cows - group A) were close-up cows, at the

last 15 days before the expected parturition. The animals of the 2nd group (15 cows - group B) were fresh cows 1 to 15 days after calving, while the 3rd group (15 cows - group C) consisted of high-production cows, from the 90th to 100th day of lactation. All the included cows were in their 3^{rd} lactation and their average milk production was $6,825\pm 305$ liters per lactation period.

Feeding and housing were in accordance with the intended use of the animals. Diet composition was adjusted to the requirements of cows during pregnancy and lactation. The feeding regime of the cows at different production stages is presented in Table 1.

Cows were judged to be clinically healthy for the duration of the study.

Table 1:	Feeding	cows	before	and	after	calving	
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Cows before the calving were fed daily	Cows after calving and during lactation consumed daily
3 kg alfalfa hay	1,800 Kg alfalfa hay
Wheat straw 3 kg, 10 kg	15 kg of corn silage (30% dry weight)
Corn silage (30% dry weight)	8 kg of alfalfa silage
4 kg alfalfa haylage	4 kg of maze ear silage (68% dry weight)
2 kg maize ear silage (68% dry weight)	2 kg of dry sugar beet pulp
0.5 kg dry sugar beet pulp	2 kg exstruded soybeans
1.5 kg of additional mixtures (30% of total protein)	4.5 kg additional mixtures (30% of total protein)

Blood sampling

Blood sampling of all the tested cows was conducted once, by jugular veni-puncture into 20 ml sterile vacuum glass tubes. All samples were drawn between 10:00-12:00 a.m. The samples were placed in ice and were forwarded to the laboratory within an hour; sera were separated by centrifugation at $2,500 \times g$ for 15 min. Thereafter the sera were placed in plastic tubes (1.5 ml) (Eppendorf, Germany) and stored at -20°C until analysed. The concentration of calcium and magnesium was determined by atomic absorption spectrophotometry (AAS), and inorganic phosphorus by a spectrophotometrical assay kit (Inorganic Phosphorus Assay Kit, Bioo Scientific Corporation, USA) using the spectrophotometer, UV-1800 (Shimadzu, Japan). Serum levels of calcium, magnesium and phosphorus in sera were determined by the standard method of by atomic absorption spectrophotometry (Serbian SRPS ISO 6869 2002 using UNICAM 969, USA).

Statistical analysis

The ANOVA procedure followed by LSD *post hoc* was used to analyze the difference between analyzed groups of Simmental cows. The differences with p-values of p<0.05 were considered significant (95% significance). Statistical analysis was carried out by Ver.5.0 Microsoft Statistical Stat.Soft.Inc.1995.

Ethical compliance

Statement of ethical compliance: The experiment was done in compliance with Serbian Law on Animal Welfare (Official Gazette of the Republic of Serbia No 41/09) and Ordinance on the conditions for registration for experimental animals and the keeping of such register, training programs on welfare on experimental animals, request forms for approval of conducting experiments on animals, standing, treatment and killing experimental animals and reproduction, circulation, or implementation experiments on animals (Official Gazette of the Republic of Serbia No 39/10).

RESULTS

Results of the concentration of inorganic phosphorus in serum of tested Simmental cows are shown in Table 2. Significantly higher (p<0.05) levels of phosphorus were in

 Table 2: Serum inorganic phosphorus concentrations (mmol/l) from all the sampled cows (groups A, B and C)

Productive stage	Group	Ν	Х	SD	CV	P<0.05
Close-up dry cows	А	15	2.09	0.43	32.55	10
Fresh cows	В	15	1.97	0.39	30.98	A:C B:C
High-production cows	С	15	2.40	0.38	18.93	

 Table 3: Serum magnesium concentration (mmol/l) from all the sampled cows (groups A, B and C)

Productive stage	Group	Ν	Х	SD	CV	P<0.05
Close-up dry cows	А	15	1.02	0.26	43.25	
Fresh cows	В	15	1.09	0.32	44.76	A:C
High-production cows	С	15	1.28	0.20	29.23	

 Table 4: Serum calcium concentrations (mmol/l) from all the sampled cows (groups A, B and C)

Productive stage	Group	Ν	Х	SD	CV	P<0.05
Close-up dry cows	А	15	2.35	0.24	21.98	
Fresh cows	В	15	2.17	0.22	23.56	B:C
High-production cows	С	15	2.53	0.26	19.23	
N = Number of cows X = Mean	SD = Standard deviation CV = Coefficient of variance					

group C compared to group B. The mean serum phosphorus concentration of close-up dry cows of group A was significantly higher (p<0.05) compared to fresh cows of group B. Variations in serum inorganic phosphorus concentration was not high within tested groups; the most pronounced variability was recorded in the fresh cows group (CV = 32.55%).

Results of serum magnesium concentration are shown in Table 3.The highest mean magnesium concentration was found in high-production lactating Simmental cows (1.28±0.20 mmol/l). Mean magnesium values were significantly higher (p<0.05) in high-production lactating cows (group C) compared to close-up dry cows (group A). Magnesium levels in the blood of tested cows had relatively high variations within tested groups: in range from 0.20 to 0.32 mmol/l and the coefficient of variation varied from 29.30% to 44.76%.

Results of serum calcium concentrations are shown in Table 4. Mean calcium concentration of fresh cows (group B) was significantly lower in comparison with those of high-production cows of group C (p<0.05). The variability of serum calcium concentrations among the three groups of cows was not high, being most pronounced in the fresh cows (CV=23.56%).

DISCUSSION

Lactating dairy cows use large quantities of calcium for milk synthesis (11). Hypocalcemia in intensively managed dairy cows causes periparturient paresis and contributes to "downer cow syndrome", while during lactation it can also cause fertility disorders (2,6). The results of the present study indicate that in a group of Simmental dairy cows in puerperium, lowest calcium levels were found, and these values were significantly lower than the level of calcium in the blood in Simmental cows in advanced pregnancy and during maximum lactation. These findings are similar to the findings of other researchers, who suggest that a sudden loss of calcium from the body occurs in the puerperal cows, as for every liter of colostrum 1.0-2.0 g of calcium is consumed (7), while a physiological decline of calcium in the blood is held steady for several days and on account of intensive production, serum calcium decrease is correlated with the appearance of puerperal paresis (7). Calcium metabolism is closely related with phosphorus metabolism and is responsible for the proper metabolism of vitamin D (12, 13).

Inorganic phosphorus levels in the blood of cows are at

low concentrations in the range of 2.1-3.0 mmol/l, while significantly more phosphorus is bound to organic compounds such as phospholipids (14, 15, 16). Similarly to serum calcium, inorganic phosphorus values were significantly lower in Simmental cows in puerperium compared to the values of inorganic phosphorus in the blood of cows in advanced pregnancy and during maximal lactation. Grünbergand coworkers (17) found that hypophosphatemia is present in the blood of highly productive Holstein-Friesian cows throughout the postpartum period which is associated with the entry of glucose in the glycolytic path of peripheral tissues, as well as to supply the necessary quantities of mammary gland phosphorus. The authors believe that hypophosphatemia allows proper glucose metabolism which prevents ketosis immediately after calving (6, 18). The sudden loss of phosphorus from the body occurs in the puerperal cows when 0.8-1.9 g phosphorus per liter of colostrum is spent. This physiological decline of phosphorus in the blood is held steady for several days and the intensity of phosphate fall is correlated with previous findings that phosphate blood level falls in older animals (19). The results of these tests show a decrease in serum calcium and inorganic phosphorus in the blood of Simmental cows in early lactation, suggesting increased use of these macroelements by the mammary glands. Our results show slightly higher calcium, phosphorus and magnesium blood serum levels than in Jersey cows but similar to levels of Holstein breed cows (12, 20), on the other hand calcium and magnesium blood sera levels were found to be lower than in beef cattle such as Angus, Charolais and Hereford, or Limousin (10).

Magnesium, like calcium reduces neuro-muscular irritability and a drop in its concentration in the blood results in spontaneous muscle contractions or tetany (1). The results of this study indicate that the lowest magnesium levels were detected in Simmental cows in advanced pregnancy and that greater values of magnesium in the blood were determined in cows in puerperium. However the differences of these values were not significant. The highest values of magnesium in the blood were detected in high production Simmental cows during the peak of lactation which were significantly higher compared to the values of the cows in advanced pregnancy and the puerperium. The results obtained in this study indicate that the magnesemia was within the normal range in all tested cows, and lower values were found in a group of cows in the periparturient period. This probably points to the increased use of magnesium during negative energy balance in early lactation.

CONCLUSION

Serum calcium and inorganic phosphorus in the blood of Simmental dairy cows in puerperium were significantly lower (p<0.05), compared to the values of cows in advanced pregnancy and during maximum lactation, indicating the increased use of these macro-elements by the mammary gland of cows in early lactation. For the cows in the periparturient period, values of magnesium in the blood were significantly lower (p<0.05), compared to the values during the periparturient period.

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