

THE CONTENT OF CALCIUM, PHOSPHORUS AND MAGNESIUM IN THE BLOOD SERUM OF SHEEP DEPENDING ON THE SEASON AND PHYSIOLOGICAL STATE

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Abstract: The large number of sheep, especially the ones that spend the majority of time on pastures, is been fed with the meals, which do not contain all the necessary mineral substances. The level of calcium, phosphorus and magnesium on natural pastures is too low in relation to the needs of sheep. Therefore, the irregularity in the feeding of sheep occurs because of the absence of the other food sources. These irregularities are in the range from the acute mineral deficit or illness to the mild temporary forms, which could hardly diagnose, but they affect the level of production. The content of calcium, phosphorus and magnesium in the blood serum of sheep, as one of the indicators of the supplementation of sheep with these substances, has given in this paper. The average level of calcium in the blood serum of the control group of sheep was 2.61 mmol/l, phosphorus 1.04mmol/l and magnesium 1.31 mmol/l of serum. In the blood serum of the tested group, the average contents of calcium was 2.33 mmol/l, phosphorus 0.92 mmol/l and magnesium 1.37 mmol/l.

Key words: sheep, calcium, phosphorus, magnesium, serum

Introduction

The knowledge of the nutritive values of the grass mixture, with which sheep meet their needs during nearly one half of the year, is very important from the standpoint of the evaluation of the food quality (Kargin *et al.*, 2004; Khan *et al.*, 2006). Pasture is very important as the full-value source of food in the feeding of sheep, of which could not be easily substituted in the extensive breeding of ruminants. The quality of the pasture and its nutritional value depend on many factors but primarily on the quality of the soil and the climatic factors. In the

conditions of breeding sheep in the Šar Mountains, learning about the quality of pasture gives us the possibility of taking appropriately measures in its improving. It would be a starting point for resolving issues of proper nutrition of sheep during the summer, because that period represents an important phase of a changing in physiological states (lactation, and preparation for successful impregnation).

Certainly, these tests are not able to discover the genesis of their foundation, on which we can derive the conclusions for guidance of the direction of the further development by applying Hemo - Agro - Hido and other measures, which were dictated by the ecological factors. Because these researches do not have pretensions of such a capacity and profile, their goal is to show the picture of the chemical composition of the grass mass of tested pastures, as a very useful component of the main research, which related to the dynamics of calcium, phosphorus and magnesium in stages of vegetation, or the exploitation of the pastures. In many areas of the Šar Mountains, the content of stodgy cattle food is not enough tested. From the results obtained based on our previous researches of mineral substances in stodgy food, a low content of phosphorus is established in them, and appeared much more frequently than it is the case with calcium. This phenomenon is especially present during dry years and certain periods, which often occur at the Šar Mountains where the testing had performed. It should be added that in the Šar Mountains, there are soils in which the content of physiologically active phosphorus (P_2O_5) is extremely low, as was found by *Mirić (2000)*, and a low content of this element in the soil adversely affects its content in plants. To this, we should add the statement of *Stojković (2006)* that a substantial number of plant species from these areas has a low content of phosphorus.

Materials and Methods

The testing of the influence of the physiological conditions and the seasons on the dynamics of the content of calcium, magnesium and phosphorus in the blood serum of the crosses F_1 (Sharplaninska sheep x Wurttemberg sheep) was conducted during two years in farm of Štrpce. There were 60 sheep in the experiment, of which 30 were in the control group – the futile sheep (treatment 1, 2, 3, 4 and 5) and 30 in the experimental group - the fertile sheep (treatment 1, 2, 3, 4 and 5). At the end of each treatment, the blood has taken from the jugular vein of the sheep for the preparation of the serum. The testing of these mineral elements was carried out in the blood serum of the sheep and the food used in their meals. The sheep were staying in a cottage at the Šar Mountains started from the late May until the early October (2007). The main meal was their pasture with the addition of 200 g of corn grits. In addition, from October until May (2008), the sheep were in their home environment near Štrpce. The main meal was their meadow hay from 1.5 to 2.0 kg and corn grits from 200 to 300 g per day. The cattle salt in the form of the mineral plates was also available to the sheep.

Statistical analysis was performed using Statistica, version 6, SatSoft. Inc. (2003).

Table 1. The contents of nutrients in the feed for feeding sheep

Type of food	Chemical composition, (%)						
	Water	Protein	Cellulose	Ash	Ca/g	P/g	Mg/g
Meadow hay	11.15	8.37	30.67	7.80	0.76	0.26	0.20
Lucerne hay	11.90	9.80	28.90	8.58	0.98	0.15	0.34
Corn grits	11.51	8.51	1.80	1.56	0.10	0.35	0.19
Spring grass	20.71	9.67	24.90	6.79	0.76	0.15	0.05
Summer grass	18.68	6.80	30.22	5.56	0.77	0.16	0.06
Autumn grass	18.12	5.65	28.40	8.23	0.78	0.30	0.08

Results and Discussion

The data about the dynamic of the content of calcium in the blood serum of the sheep are shown in table 2.

Table 2. The contents of calcium in the blood serum of sheep (mmol/l)

	X±SD	SG
Control group – futile sheep		
Treatment 1 - August	2.47±0.18	0.47
Treatment 2 - November	2.59±0.18	0.47
Treatment 3 - January	2.58±0.13	0.35
Treatment 4 - July	2.57±0.24	0.64
Treatment 5 - September	2.60±0.17	0.44
Average	2.61±0.19	0.47
Tested group – the fertile sheep		
Treatment 1 – first half of the pregnancy	2.54±0.25	1.05
Treatment 2 – second half of he pregnancy	2.34±0.11	0.45
Treatment 3 - the beginning of lactation	2.02±0.13	0.54
Treatment 4 – the end of lactation	2.29±0.15	0.60
Treatment 5 – the period of becoming infertile	2.50±0.11	0.47
Average	2.33±0.15	0.62

After the fertilization of the sheep, the content of calcium in the blood serum was 2.54 mmol/l, while in the futile sheep at that time was 2.74 mmol/l. In the second half of the pregnancy, the content of calcium decreased to 2.34 mmol/l, while in the futile sheep this decrease was much lesser (2.59 mmol/l). In the phase of the most opulent secretion of the milk in the lactating sheep, the decrease in the

content of calcium is most pronounced, and it was slightly below the physiological limits (2.02 mmol/l) then. At that time, the content of calcium in the blood serum of the futile sheep was at the same level (2.58 mmol/l). In the phase of reduced secretion of the milk in the lactating sheep, the increase in the content of calcium of 4.00% was noted, and later by becoming infertile, the level of calcium in the blood serum climbed to 2.50 mmol/l. At that time, in the futile sheep, the content of calcium held at the same level (2.57 and 2.60 mmol/l). By the dynamics of calcium content in the blood serum of the fertile and futile sheep, general conclusion can be made, that, normocalcemia existed in the conception of the sheep and that the changes, that occurred in the fertile sheep, the result of normal moving, were caused by the different needs of the organism in accordance with the physiological conditions. Considering the content of calcium in the blood serum of the sheep it can be concluded that calcemia was normal and that there were no significant differences ($P > 0.05$) in physiological conditions, or season.

The dynamics of the content of calcium in the blood serum shows a regular rhythm when we correlate it with changes in the physiological status of the sheep, making a concave parabola with the lowest point in the time of the most opulent secretion of milk. At the same time, the contents of calcium in the serum in the futile sheep ranged slightly above average the normal values (between 2.57 and 2.74 mmol/ l) and despite the season it was not subjected to changes. The percentage of variation in the content of calcium was significantly lower in the futile sheep, which is understandable.

According to the data from *Underwood (1976)*, this value was lower for 5.15% of the average content of calcium in our previous experiments, *Stojković (2006, 2009)*. Although, in literature there are the data according to which the content of calcium in the blood serum of sheep can be much higher, *Pavličević et al., (1998)*, as was the case when the minerals with high calcium content were added to the meals. But there are also data on cases of the expressive hypocalcemia such are indicated by *Mirić et al., (2000)* in whose experiments the level of calcium in the blood reach the level of only 1.98 mmol /l.

The data about the content of phosphorus in the blood serum of the sheep are shown in the table 3.

Table 3. The content of phosphorus in the blood serum of the sheep (mmol/l)

	X±SD	SG
Control group – futile sheep		
Treatment 1 - August	0.98±0.16	0.43
Treatment 2 - November	1.01±0.15	0.39
Treatment 3 - January	1.02±0.08	0.22
Treatment 4 - July	1.07±0.06	0.15
Treatment 5 - September	1.13±0.04	0.11
Average	1.04±0.09	0.26
Tested group – the fertile sheep		
Treatment 1 – first half of the pregnancy	1.13±0.15	0.62
Treatment 2 – second half of he pregnancy	0.86±0.09	0.37
Treatment 3 - the beginning of lactation	0.74±0.08	0.33
Treatment 4 – the end of lactation	0.92±0.08	0.35
Treatment 5 – the period of becoming infertile	0.97±0.19	0.40
Average	0.92±0.10	0.41

In the first half of the pregnancy, the content of phosphorus in the blood serum of the sheep was 1.13 mmol/l. In the second trimester the content decreased to 0.86 mmol/l. During the period of full lactation, the maximum decrease was 0.74 mmol/l. From this, along with the decrease in the secretion of the milk, the level of phosphorus increased to 0.92 mmol/l, only to reach the level of 0.97 mmol/l in the infertile sheep in September. In the futile sheep, the content of phosphorus in the blood serum in August was 0.98 mmol/l and it was constantly increasing during the year (1.01 mmol/l, 1.02 mmol/l, 1.07 mmol/l and 1.133 mmol/l) to reach in the beginning of September 1.13 mmol/l. In the fertile sheep, the smallest deviation from the average values between the certain sheep was notice after the fertilization and they were constantly growing from the period of becoming infertile. In terms of percentage of the variations of the content phosphorus in the blood of the futile sheep, the flow was reverse. Varying is highest at the time when should be impregnated the sheep and it constantly declines until September.

From the data presented, it can be noticeable that the content of phosphorus in the blood serum of the sheep during the year is in all physiological states, under the physiological limits. Hypophosphatemia reached its peak in the fertile sheep in the phase of the most opulent secretion of the milk. Concerning that, hypophosphatemia was very pronounced in the fertile sheep, and the average varying of the inorganic serum of phosphorus during the year was higher than in the futile sheep. Differences in the content of phosphorus per season and physiological condition were in the level of the statistical significance ($P < 0.01$).

The content of phosphorus for all groups in the experiment was in the average 0.98 mmol/l of the serum. With these values it was located below the

physiological limits of the normal range of the content according to the data from the literature (1,45-2,00 mmol/l).

The lowest values recorded in the most opulent stage of lactation (0.74 mmol/l) and the highest in the first half of the pregnancy (1.13 mmol/l). With lactation passing off and the arrival of the non-productive lactation period (in autumn), the content increased to 0.97 mmol/l. In terms of variation, it is characteristic that, from the beginning to the end of pregnancy of the monitored period, the variety grew more and more (from 5.03 to 15.50%). Although, in the futile sheep, the average content of phosphorus in the serum was higher than in the fertile sheep by 11%, the general level was very low, and the changes were subordinate more to the factor of diet, which was not the same case and in the fertile sheep. The content of phosphorus in the futile sheep was constantly increasing during the year, starting from 0.98 mmol/l in August and to 1.13 mmol/l at the end of the observed period. Concerning the content of phosphorus in the serum of the fertile and the futile sheep, it can be terminated that hypophosphatemia is expressed during the whole year.

During the general hypophosphatemia, of both fertile and futile sheep, it is important to notice the appearance of a very different character of variation in the content of phosphorus per year, namely caused by the physiological state. In the futile sheep it can be seen a constant, although small increase in the content of phosphorus during the "vacation" until next season of fertilization, while at the same time narrowing of the variation span. Whereas, the pregnant sheep have a decrease in the phosphorus content, this goes along with the duration of stress of the organism (increased distress) in pregnancy in the first two months of lactation. In addition, it has increased with the arrival of the period of the physiological relief, with a permanent increase in the percentage of the variations in the content of phosphorus in the blood serum. These findings supports the assumption that in the states of hypophosphatemia in the temporary futile sheep, the balance of phosphorus is gradually improving to such an extent that they can, after a one-year break, manifest the sexual passion and be fertilized. In the "fertile" sheep, although in the second half of the lactation the balance of phosphorus in average improves, it cannot be improved in all animals, which is attested by the increased percentage of variation so that, slightly more than a half of the sheep came to a successful fertilization, while a smaller part remained unfertilized.

The displayed value of the content of phosphorus, it can be seen that there was virtually a low level of this element in the blood serum of the sheep with a minimal increase in the time of their pregnancy, and with the lowest value in the stage of lactation. A low content of phosphorus in the blood of the sheep has interpreted by its low content in food, especially hay, and reason for the low content in hay, by the interpretation of *Mirić (2000)*, lies in the fact that, in this region exist large areas of land on which the content of physiologically active phosphorus (P_2O_5) is extremely low. This is confirmed by the findings of *Stojković*

(1997, 2009) in which a large number of plant species from these areas have a low content of phosphorus. Its enhanced secretion in milk can interpret a particularly low content of phosphorus in the blood serum of the sheep in lactation.

The data about the dynamics of the content of magnesium in the blood serum of the sheep are shown in the table 4.

The content of magnesium in the blood serum of the sheep was within the normal range and it showed no significant differences with changes in the physiological state of the sheep, which is a characteristic of its behaviour. Similar or the same range of the levels of magnesium in the blood in the same intervals had established in the futile sheep, too. Differences in the content of magnesium in the blood serum of the sheep, both in season and in the physiological conditions were not significant ($P > 0.05$).

The content of magnesium in the blood serum of the sheep was within the normal physiological range of 1.34 mmol/l and showed no significant differences during the changes in the physiological state, which is the characteristic of its behaviour. Yet, although to a lesser extent, there was a reduction of its content in the blood of the sheep at the end of lactation (1.27 mmol/l). There were not any notable individual changes in the content of magnesium, or changes in groups of the sheep with different lactation. However, averages per groups indicate the existence of a reverse trend of a change in the content of this element in the blood serum in relation to the trend of a change in the level of calcium and phosphorus. This means that the content of magnesium was the highest in the blood serum of the sheep with the highest lactation, and the lowest in the infertile sheep.

Table 4. The content of magnesium in the blood serum of the sheep (mmol/l)

	X±SD	SG
Control group – futile sheep		
Treatment 1 - August	1.28±0.12	0.27
Treatment 2 - November	1.32±0.17	0.19
Treatment 3 - January	1.29±0.19	0.16
Treatment 4 - July	1.35±0.11	0.21
Treatment 5 - September	1.35±0.23	0.10
Average	1.31±0.16	0.18
Tested group – the fertile sheep		
Treatment 1 – first half of the pregnancy	1.33±0.15	0.62
Treatment 2 – second half of the pregnancy	1.31±0.21	0.37
Treatment 3 - the beginning of lactation	1.43±0.23	0.40
Treatment 4 – the end of lactation	1.27±0.10	0.59
Treatment 5 – the period of becoming infertile	1.55±0.18	0.61
Average	1.37±0.17	0.51

The data on the content of magnesium, which had obtained in these researches, correspond with the results of *Adamović and Pavličević (1990)* and *Pavličević and co. (1999)*. The explanation for the relatively small changes in the content of magnesium in the blood of the sheep, according to *Underwood (1976)* and *Stojković (2009)*, is that the magnesium in soft tissues is not reduced even when the sheep loose up to 30% of magnesium from the skeleton.

Conclusion

The content of calcium, phosphorus and magnesium in the blood serum of sheep, as one of the indicators of supplementation of animals in these materials, has investigated. The average content of calcium in the examined blood serum of the sheep was 2.47 mmol/l, phosphorus 0.98 mmol/l and magnesium 1.34 mmol/l of the serum. The values of calcium were at the upper limit of normal content referred to in the literature. The changes in the content of calcium were not significantly manifest in relation to the season and the physiological state of the sheep. The values of phosphorus were below the deficit, and these values decreased the most in the beginning of the lactation period of the sheep. The content of magnesium was within the normal physiological range. The changes in the content of magnesium were not significantly manifest in relation to the season and the physiological state of the sheep, but had a reverse trend compared to the trend of the change in calcium and phosphorus.

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Sadržaj kalcijuma, fosfora i magnezijuma u krvnom serumu ovaca po godišnjem dobu i fiziološkom stanju

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Rezime

Veliki broj ovaca, naročito onih koji dobar deo vremena provode na paši, hrani se obrocima koji ne sadrže sve potrebne mineralne materije. Nivo kalcijuma,

fosfora i magnezijuma na prirodnim pašnjacima je suviše nizak u odnosu na potrebe ovaca. Tako se nepravilnost u ishrani javlja kod ovaca u odsutnosti drugih izvora hrane. Ove nepravilnosti kreću se od akutnog mineralnog deficita ili bolesti, pa do blagih prelaznih formi koje se teško dijagnosticiraju, ali se odražavaju na nivo proizvodnje. U radu je dat sadržaj kalcijuma, fosfora i magnezijuma u krvnom serumu ovaca, kao jednog od indikatora obezbeđenosti ovaca ovim materijama. Prosečan sadržaj kalcijuma u krvnom serumu kontrolne grupe ovaca iznosio je 2,61 mmol/l, fosfora 1,04 mmol/l i magnezijuma 1,31 mmol/l seruma. Kod ogledne gupe prosečan sadržaj kalcijuma bio je 2,33 mmol/l, fosfora 0,92 mmol/ i magnezijuma 1,37 mmol/l seruma.

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