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Original Contribution

EFFECT OF THE MULTIENZYME PROTOZIN-A ON THE LEVEL OF SOME HEMATOLOGICAL PARAMETERS OF SHEEP

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ABSTRACT

The effect of the multienzyme Protozin-A on the hematological parameters of sheep was studied. These parameters included total protein, albumin and globulin, urea and blood sugar, hemoglobin level, total number of erythrocytes and leukocytes. The study was done on 6 sheep of the Tsigai breed. The multienzyme Protozin–A, dose 1g/kg of food, was added to the mixture concentrate that served as food for the animals. Blood was collected from animals before and 2.5 h after feeding. Analysis of this blood showed that protozin-A did not change the total blood protein. Albumin level dropped by 50% while globulin level increased by the same amount. There was significant increase in erythrocyte count. Urea level was unchanged, while glucose level was reduced.

Key words: Sheep, Diet, Enzyme, Blood

INTRODUCTION

Most of nutritive antibiotics and other chemical stimulators in animal food are banned in a number of European countries. The use of natural products, obtained by modern technologies, can solve the problem of production of cheap agricultural bio-products. Because of the ban on the use of nutritive antibiotics as stimulators in animal feeding in a number of countries, including Bulgaria, there is need for research on the effect of enzymes. The enzymes are proteins that act as catalysts in metabolism, and are involved in almost every biochemical process in animals. There are numerous studies on the effect of enzymes in single-stomach animals and birds. The effect of enzyme supplements on fattening abilities (1, 2) and meat (3, 4) and milk (5) quality, as well as on fermentation processes (6) has been studied. On the other hand, the effect of enzymes on blood constituents has not been studied.

Protozin-A is manufactured by *Plastchim*, Botevgrad, Bulgaria. According to the manufacturer protozin-A is a substance with wide range of enzyme activity with the following enzyme constituents: amylase, cellulase, xylanase, β-glucanase, protease, lipase, phytase. Protozin-A improves digestion of forages and increases the assimilation of the basic food substances, which are: starch, carbohydrates, proteins, fats and cellulose from 6 to 15 %. There are no data on the effect of the substance on blood components.

The objective of the present study is to investigate the effect of the multienzyme Protozin-A, at dose 1 g/kg of mixture concentrate, on some blood components in sheep.

MATERIAL AND METHOD

The experiment was conducted using the two-period method, Control and Experimental, on 6 Tsigai breed sheep, aged 3 years and weighing 45±3 kg. The animals were reared in separate cages in a barn. The animals were fed diet consisting of 0.850 kg meadow hay 0.400 kg ground wheat. The experimental animals were given the multienzyme Protozin-A, in a dosage 1 g/1 kg of concentrate. The feed was given twice a day – at 8 a.m. and 1 p.m.

Blood test samples were taken from the jugular veins twice a day, before feeding in the morning and 2.5 h after feeding. Total protein, albumins and globulins, urea level, blood sugar and hemoglobin, the total number of erythrocytes and leukocytes in blood were determined. Blood components were determined by means of standard tests and the blood cells by means of Thoma and Bürker count chambers. All data were biometrically

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processed with a computer software for statistic analysis - Microsoft Excel. The graphic presentations were made with the same software.

RESULTS AND DISCUSION

Data on the tested parameters during the two periods are given in the **Table 1**. Total protein was 96.33 g/l before feeding and 90.11 g/l 2.5 h after feeding during the Control period. During the experimental period these values were 91.45 and 85.10 g/l respectively. All differences were not significant.

Table 1. Effect of the enzyme Protozin-A on blood components in sheep

Parameters	Control period				
	Before feeding			2.5 h after feeding	
	n	x±Sx	C	x±Sx	C
Total protein (g/l)	12	96.33±2.15	7.42	90.11±2.53	9.32
Albumins (g/l)	12	48.12±5.30	36.55	46.98±5.37	37.95
Globulins (g/l)	12	48.22±5.95	40.94	43.12±4.73	36.43
Urea (mmol/l)	12	19.96±1.78	29.66	21.27±3.08	48.05
Blood sugar (mmol/l)	12	4.61±0.10	7.37	4.44±0.20	15.32
Hemoglobin (g/l)	6	128.67±7.13	12.41	100.17±5.02^{aa}	11.23
Erythrocytes 10^{12}/l	12	10.08±0.82	26.88	11.31±0.58	16.96
Leucocytes 10^{9}/l	12	9.43±0.64	22.38	9.65±053	18.34

Parameters	Experimental period				
	Before feeding			2.5 h after feeding	
	n	x±Sx	C	x±Sx	C
Total protein (g/l)	12	91.45±3.02	10.96	85.10±1.83	7.13
Albumins (g/l)	12	22.52±1.52***	22.38	22.04±0.89***	13.38
Globulins (g/l)	12	68.92±3.60**	17.34	63.07±2.29**	12.10
Urea (mmol/l)	12	18.19±1.17	20.12	17.69±1.21	22.67
Blood sugar (mmol/l)	12	3.41±0.09***	8.79	3.87±0.10*^{aa}	8.79
Hemoglobin (g/l)	12	117.5±9.16	25.88	112.42±7.67	22.65
Erythrocytes numb.x10^{12}/l	12	14.80±0.77**	17.36	13.71±045**	10.94
Leucocytes numb.x 10^{9}/l.	12	5.72±0.50***	28.85	7.02±0.46	21.65

- reliability of differences between Control and experimental period -*- p<0.05, ** - p< 0.01, * < 0.001*
a - reliability of differences before and after feeding – a < 0.05, aa< 0.01

Feeding time did not influence the amount of blood urea in experimental animals. The addition of the multienzyme Protozin–A in sheep diets did not bring about any significant decrease in the amount of blood urea.

Change in blood sugar before and after feeding during the Control period was quite minimal– 4.61 mmol/l and 4.44 mmol/l. Two and half hours after feeding during the experimental period blood sugar increased compared to time before feeding (p<0.01). Compared to the control period the amount of glucose was lower at both points of testing (p<0.001), (p<0.05). As a result of the quicker metabolism when supplementing Protozin A, the reduced blood sugar parameters were probably due to its participation in a number of metabolic chains (glycolysis, pentosophosphate cycle, etc.), and vitally important energy and intermediate metabolites were produced.

During the Control period the amount of albumins was approximately the same before feeding and 2,5 hours after feeding: 48.12 and 46.98 g /l. Feeding time did not influence the albumin content during the second period, too. During the experimental period, as opposed to the Control one, the amount of albumins decreased very reliably both before feeding – 22.52 g /l (p<0.001), and 22.04 g/l 2.5 h after feeding (p<0.001).

Feeding time did not influence the amount of globulins, both during the Control period and experimental one. Globulins during the Control period were 48.212 g /l and 43.12 g /l. On the other hand, there was an increase in globulins by approximately 50% both before feeding – 68.92 g /l (p<0.01) and 2.5 h after feeding – 63.07 g /l (p<0.01).

The ratio of albumins to globulins is known as "protein coefficient". During the Control period the protein coefficient was 1.00 and 1.10, while during the experimental

period it decreased and reached 0.33 and 0.35. The data showed that total protein was neither influenced by the feeding time nor by the enzyme supplementation; only the ratio between albumins and globulins was changed (**Figure 1** and **Figure 2**). Similar increase of the amount of blood globulins with protozin has been established in other studies conducted by us in rabbits (7).

The amount of hemoglobin in both test periods varied from 100.17 g/l to 128.67 g/l. The supplementation of Protozin-A in the sheep diet reduced without significance the amount of blood hemoglobin.

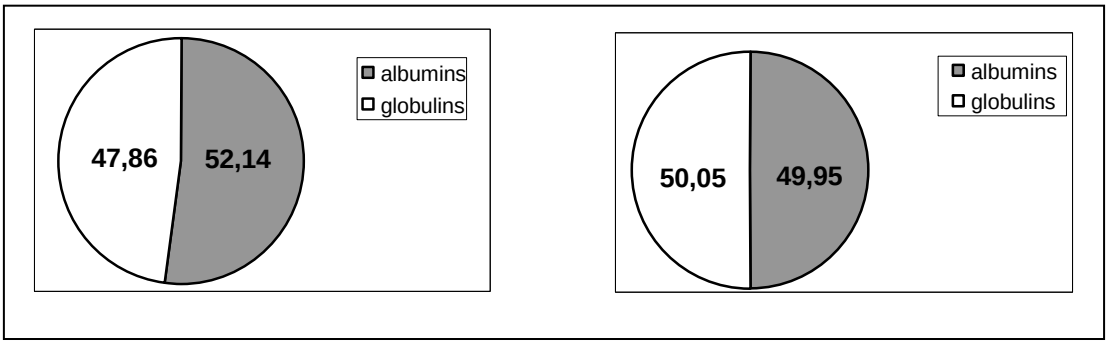

Figure 1. Ratio between albumins and globulins (%) Control period

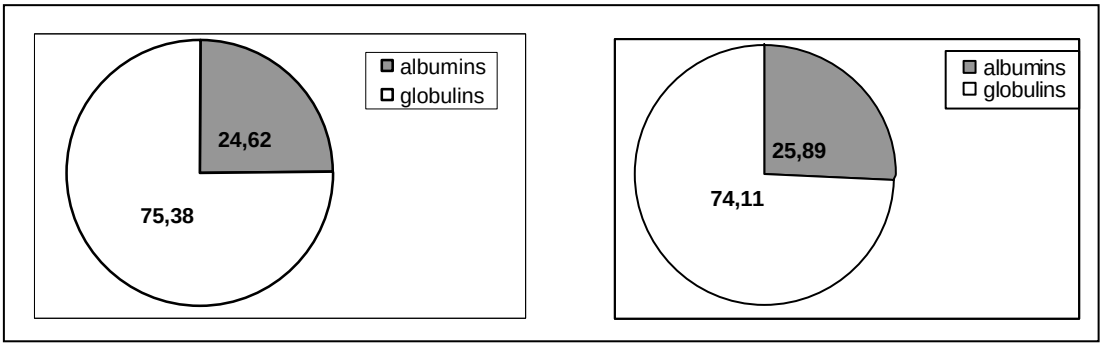

Figure 2. Ratio between albumins and globulins (%)experimental period

Erythrocyte number was not influenced by the feeding time in both test periods. During the experimental period the number of erythrocytes was greater both before feeding ($p<0.01$) and 2.5 h after feeding ($p<0.01$). Unlike leukocytes, the number varied greatly depending on the time in the twenty-four-hour period and the functional condition of the body, while the number of erythrocytes remained comparatively constant. The increase in the number of erythrocytes during the experimental period was most probably due to the enzyme included in the sheep diet.

The number of leukocytes was approximately the same during the two test periods before and 2.5 h after feeding, although there was a tendency towards alimentary leukocytosis. There was a decrease in the number of leukocytes but the difference was reliable only at the time before feeding ($p<0.01$).

CONCLUSIONS

The results so far obtained showed some inconclusive findings that should provoke the necessity for further studies in order to elaborate on the detailed mechanism of the metabolic processes in the animal.

While there was no change in the amount of the total blood protein, there was a reduction in the amount of albumins by 50%. There was an increase in the amount of globulins by the same percentage.

The supplementation of the enzyme in the sheep diet resulted in reliable increase in the number of erythrocytes.

The multienzyme Protozin-A did not change blood urea but reduced the amount of glucose in the blood of the experimental animals.

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