INFLUENCE OF TREATMENT OF SPECIFIC FEED WITH FORMALDEHYDE ON INDICATORS OF RUMN METABOLISM AND BIOCHEMICAL INDICATORS OF BLOOD IN LAMB

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Abstract

The studies were carried out using the Latin square method on model fistula lambs with a live weight of 28-30 kg. All experimental animals were implanted with scar cannulas. The daily diet of group I consisted of 1 kg of alfalfa hay, 100 g of barley, 30 g of sunflower cake and 30 g of soybean meal (basic diet). Animals of the second group received the same diet, but the sunflower cake was treated with a 1% formaldehyde solution. The animals of the third group also received a basic diet and soybean meal treated with a 1% formaldehyde solution. It has been established that in animals that received sunflower cake and soybean meal treated with a 1% solution of formaldehyde in rumen fluid, the acidity decreases. This process most clearly occurs in lambs that received soybean meal treated with formaldehyde. VFAs are absorbed better in the body of lambs that received soybean meal treated with formaldehyde as part of the diet, the concentration of ammonia in the rumen contents was lower than in the groups consuming the main diet.

Treatment of feed with formaldehyde slightly reduces the number of rumen microflora. However, this decrease was insignificant and did not lead to statistically significant facts. A significant decrease in the level of urea in the blood of animals fed sunflower cake and soybean meal treated with formaldehyde was established. It was also noted that there was an increase in the concentration of total protein in the blood of animals fed with feed treated with formaldehyde, a significant increase in albumin, and a significant decrease in the concentration of urea and creatinine.

Keywords. Lambs, sunflower meal, soybean meal, formaldehyde, volatile fatty acids (VFA), ammonia, bacteria, protozoa and urea.

Introduction.

In feeding ruminants, one of the key factors is the breakdown of feed protein in the

rumen. Since in ruminants, part of the feed proteins is absorbed by the rumen microflora, and valuable microbial protein is synthesized. Another part of the feed protein avoids breakdown in the rumen. Thus, the protein nutrition of ruminants consists of microbial protein and undigested feed protein. The ratio of these proteins determines the metabolic processes in the body of ruminants, as well as their productivity. Research over the past 20-30 years shows that microbial protein can only meet ruminant needs by 30-40%. Therefore, feed protein not broken down in the rumen is of increasing interest (Aliev A.A., 1997; Ismailov I.S. et al., 2017; Kalnitsky B.V., Kharitonov E.L., 2002). Based on this, the concept of protecting feed protein from degradation in the rumen emerged. To do this, various feed processing methods are used (Haryanto B., 2014). One such method is treating feed with formaldehyde. Formaldehyde, a chemical used in animal feed, is environmentally friendly (EFSA Panel on Additives and Products or Substances used in Animal Feed, 2014) and can reduce protein degradation in the rumen (Mahima K.V. et.al., 2015). Mahima et al. (2015) reported that the use of mustard cake treated with 1.5% formaldehyde increases the in vitro protein digestibility of wheat straw. Formaldehyde may also reduce the in vitro breakdown of soybean meal into ruminal contents. Suhartanto et al (2014) reported that in in vitro studies, treatment of soybean flour with formaldehyde reduced dry matter breakdown by 89.9% to 52.3%. Although formaldehydetreated soybean meal is increasingly used in cattle (Perdhana P.W. et.al., 2013; Rochijan Widyobroto, B.P. and Ismaya, 2016) and sheep (Yörük, M.A. et.al., 2006; Abdel-Ghani A.A. et.al., 2011; Riyanto J. and Sudibya 2018), its use in goats is still limited. However, there is evidence of the use of sesame waste treated with formaldehyde when feeding goats (Tiwari M.R. et.al., 2015). Beigh Y.A. et.al. (2017) studied total mixed diets (concentrates) to improve nutrient intake and utilization in ruminants. The mixed diet included concentrated, roughage (Beigh Y.A. et.al., 2017), sources of protein, minerals, vitamins to form a balanced and economical diet (Beigh Y.A. et.al., 2017; Kmicikewycz A. and Heinrichs J., 2016; Linn J., 2016). Feeds with low nutritional quality can be mixed to increase intake (Kmicikewycz A. and Heinrichs J., 2016). Feed efficiency is increased by approximately 4% when using TMR compared to concentrate and roughage fed separately. Thus, these studies examined the effect of treatment of 1% formaldehyde in both single feeds and mixed feeds on the performance of ruminant animals. Based on the above, in our studies we studied the effect of formaldehyde treatment of individual diet feeds on some blood parameters and rumen metabolism in rams.

Materials and research methods.

The studies were carried out using the Latin square method in the vivarium of the Azerbaijan State Agrarian University on model fistula rams with a live weight of 28-30 kg. All experimental animals were implanted with scar cannulas according to the method of Aliev A.A. (Aliev A.A., 1998). The daily diet of group I consisted of 1 kg of alfalfa hay, 100 g of barley, 30 g of sunflower cake and 30 g of soybean meal (basic diet). Animals of the second group received the same diet, but the sunflower cake was treated with a 1% formaldehyde solution. Animals of the third group also received a basic diet and soybean meal treated with a 1% formaldehyde solution (Table 1).

Table 1. Experimental scheme and daily rations.

Feed	1st group	2nd group	3rd group
Alfalfa hay, kg	1	1	1
Barley, g	100	100	100
Sunflower cake, g	30	30 (treated with	30
		formaldehyde)	
Soybean meal, g	30	30	30 (treated with
			formaldehyde)
Salt licker	+	+	+

Soybean meal contained 48.02% protein and the following percentage amino acids: aspartic acid - 6.15; threonine - 2.03; serine-2.46; glutamic acid -9.52; proline - 2.5; glycine - 2.3; alanine - 2.42; cystine - 0.73; valine - 2.73; methionine - 0.75; isoleucine - 2.58; leucine - 4.18; tyrosine - 1.83; phenylalanine - 2.72; lysine - 3.38; histidine - 1.44; arginine - 3.94; tryptophan - 0.75.

In a comparative aspect, the processes of ruminal fermentation were studied in terms of pH, concentration of volatile fatty acids, ammonia nitrogen, and amylolytic activity of ruminal fluid. The pH in the rumen contents was determined using an Aquilon-410 device; the total amount of volatile vital acids - by steam distillation in a Markgamma apparatus; ammonia nitrogen using the Conway microdiffusion method; amylolytic activity, biomass of protozoa and bacteria by differential centrifugation for 20 minutes at 15,000 rpm. Feed analysis was carried out in the laboratory of the Azerbaijan Research Institute of Animal Husbandry.

Biochemical blood tests were performed on a Chem Well automatic biochemical analyzer. During statistical processing, the P value meant the level of significance, which was calculated using the Student's T-test. The data in the tables is presented as an average value and its error. The research results were considered highly reliable when p<0.001, significant when p<0.01 and p<0.05.

Results and Discussion

The efficiency of using energy and nutrients of feed in ruminants is directly dependent on the nature of microbial processes in the forestomach. Taking this fact into account, we determined pH, VFA and ammonia in the ruminal fluid of experimental rams.

Table 2. Indicators of rumen metabolism.

Group	Sampling time			
	1 hour before feeding	After feeding		
		After 3 hours	After 5 hours	
	pH of ruminal contents			
I	$6,64\pm0,03$	$6,45\pm0,05$	$6,38\pm0,06$	
II	$6.72\pm0,14$	$6,65\pm0,04$	$6,48\pm0,07$	
III	$6,95\pm0,04$	$6,59\pm0,06$	$6,57\pm0,02$	
	VFA in rumen fluid, Mmol/100ml			
I	7,46±0,39	10,78±0,30*	8,47±0,23	

II	8,27±0,21	11,96±0,46*	10,05±0,91
III	7,19±0,12	11,07±0,32**	$8,64\pm0,39$
	Ammonia in rumen fluid, mg%		
I	6,91±0,93	15,12±1,54	$10,19\pm 1,36$
II	6,59±0,97	12,96±1,27	$9,04\pm1,29$
III	6,04±0,45	11,32±0,69	$8,91\pm1,02$

Note: *-P<0,05; **-P<0,01.

An increase in the total acidity of the rumen contents was found in rams of all groups 3 and 5 hours after feeding. However, the highest level of acidity of the rumen contents is observed in group I rams 1 hour before feeding, which is 0.23 units. and by 0.31 units. respectively lower than those of groups II and III. There is also a tendency for the acidity of the rumen contents to decrease in animals of all groups 3 hours after feeding. The lowest level of rumen fluid acidity is also observed in group I rams, which is 6.45 units, versus 6.59 and 6.65 units. in groups II and III, respectively, with this trend maintaining 5 hours after feeding. As can be seen from Table 2, in animals that received sunflower cake and soybean meal treated with a 1% solution of formaldehyde in rumen fluid, the acidity decreases. This process most clearly occurs in lambs that received soybean meal treated with formaldehyde.

As for the formation of VFAs in the rumen fluid, rams of groups I and II predominated in terms of this indicator. The lowest values for VFA were observed in animals of group III 1 hour before feeding, which was significantly (P<0.01) lower than 3 hours after feeding. This suggests that VFAs are absorbed better in the body of lambs that received soybean meal treated with formaldehyde. In terms of the VFA content in the rumen fluid, another trend was observed: in all groups of animals, 3 hours after feeding, its concentration increased, and after 5 hours it decreased, almost reaching the initial level, i.e. an hour before feeding. This happened most clearly in animals of group I, since the content of VFAs in the rumen fluid in these animals 5 hours after feeding was significantly (P<0.05) lower than 3 hours after feeding.

With a general tendency to increase the level of ammonia formation after feeding, the highest intensity of this process was observed 3 hours after feeding in group I, which received the main diet, without treating feed with formaldehyde - 15.12 mg% versus 11.32 mg% in group III and 12. 96 mg% in group II. 5 hours after feeding, a general decrease in ammonia levels was noted in groups I, II and III by 48.4, 43.4 and 27.0%, respectively, relative to these values, which may characterize the efficiency of the use of protein nitrogen by their rumen microflora. At the same time, in animals that received sunflower meal and soybean meal treated with formaldehyde as part of the diet, the concentration of ammonia in the rumen contents was lower than in the groups that consumed the main diet. It is known that a high concentration of ammonia in rumen fluid determines the loss of protein in the form of ammonia and urea. Based on this, it can be assumed that treating feed with formaldehyde reduces excessive breakdown of protein in the rumen. Similar results were obtained by Yörük, M.A. et. al. (2006). In experiments on lambs with fistulas of the rumen and duodenum, it was found that when soybean meal is treated with formaldehyde, the flow of protein into the duodenum increases. These studies also found that treatment with formaldehyde reduced the concentration of ammonia in ruminal fluid.

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When determining the content of microbial mass in the rumen contents, it was found that the number of both bacteria and protozoa in all groups increases 3 hours after feeding.

Group	Bacteria		Protozoa		
	1 hour before	3 hours after	1 hour before	3 hours after	
	feeding	feeding	feeding	feeding	
	$0,44\pm0,04$	0,53±0,03	0,28±0,04	0,32±0,05	
I	$0,41\pm0,03$	$0,48\pm0,03$	$0,26\pm0,03$	0.31 ± 0.04	
II	$0,39\pm0,05$	$0,43\pm0,06$	$0,23\pm0,02$	$0,27\pm0,05$	
III					

Table 3. Content of microbial mass in ruminal contents, g/100ml.

As can be seen from Table 3, the lowest bacterial mass was observed in group I an hour before feeding 0.39 g/100 ml, versus 0.41 and 0.44 g/100 ml in groups II and III, respectively. As for the mass of protozoa, the lowest data for this indicator were also obtained one hour before feeding in group I rams - 0.23 g/100 ml, against 0.26 and 0.28 g/100 ml in groups II and III, respectively. This means that treating feed with formaldehyde somewhat reduces the number of rumen microflora. However, this decrease was insignificant and did not represent a statistically significant fact.

Table 4. Biochemical blood parameters.

Index	Group I	Group II	III group
Total protein, g/l	76,04±0,7	78,56±0,6	79,27±0,9
Albumins, g/l	31,24±0,5	33,45±0,3*	33,51±0,4*
Globulins, g/l	46,03±1,1	46,75±1,3	47,34±0,9
Urea, mmol/l	7,22±0,13	5,47±0,21*	5,43±0,31*
Creatinine, mmol/l	83,48±0,45	81,37±1,08	79,62±0,67*

Note: *- P<0.05.

It was revealed that feeding sunflower cake and soybean meal treated with formaldehyde as part of the diet has a more favorable effect on the course of nitrogen metabolism than use without treatment with formaldehyde. Thus, the concentration of total protein in sheep of groups II and III was higher relative to group I by 3.3 and 4.52%, respectively, mainly due to albumin. The concentration of albumin in the blood of rams of groups II and III was significantly (P<0.05) higher than that of group I.

A decrease in the level of urea in the blood of animals of groups II and III that received sunflower cake and soybean meal treated with formaldehyde was established by 31.99% (P<0.05) and by 32.96% (P<0.05) relative to group I, which may indicate a more rational use of nitrogen in the body of rams that consumed sunflower cake and soybean meal treated with formaldehyde as part of their diet.

Creatinine, like urea, is a product of protein metabolism, the content of which depends on both the level of protein and the intensity of metabolism. Its concentration in the blood of group III rams was lower by 4.85% (P<0.05) and 2.2%, respectively, than in animals of groups II and III, which may indicate a greater accumulation of creatine phosphate - in the form of loss of energy used during protein synthesis. The materials we obtained are in good agreement with the experimental data of Q. Z. Shams Al-dain (2019), H.M. El-Shabrawy and H.A. El-

Fadaly (2006) and Retno Adiwinarti et. al (2019).

Conclusion.

Thus, in our experiments it was established that in animals that received sunflower cake and soybean meal treated with a 1% solution of formaldehyde in rumen fluid, the acidity decreases. This process most clearly occurs in lambs that received soybean meal treated with formaldehyde. VFAs are absorbed better in the body of lambs that received soybean meal treated with formaldehyde. In animals fed sunflower meal and soybean meal treated with formaldehyde as part of the diet, the concentration of ammonia in the rumen contents was lower than in the groups consuming the main diet.

Treatment of feed with formaldehyde slightly reduces the number of rumen microflora. But this decrease was insignificant and did not represent statistically reliable facts. A significant decrease in the level of urea in the blood of animals fed sunflower cake and soybean meal treated with formaldehyde was established. It was also noted that there was an increase in the concentration of total protein in the blood of animals fed with feed treated with formaldehyde, a significant increase in albumin, and a significant decrease in the concentration of urea and creatinine.

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Author(s) hereby declare that generative AI technologies such as Large Language Models, etc have been used during writing or editing of manuscripts. This explanation will include the name, version, model, and source of the generative AI technology and as well as all input prompts provided to the generative AI technology

References

- 1. Aliev A.A. Experimental surgery. Moscow, Research Center Engineer, 1998. 445 p.
- 2. Aliev A. A. Metabolism in ruminants. Moscow, Research Center Engineer, 1997. 420 p.
- 3. Ismailov I.S., Tregubova N.V., Morgunova A.V. Features of amino acid metabolism in ruminants // Bulletin of the AIC of Stavropol. 2017. No. 2 (26). pp. 90–94.
- 4. Kalnitsky B. D., Kharitonov E. L. Physiological and biochemical approaches to assessing the nutritional value of feed and rationing the feeding of ruminant animals // Agricultural biology. 2002.№ 4. C. 3–10.
- 5. Perdhana, P.W., Riyanto, J., Ratriyanto, A., Widyawati, S.D. and Suprayogi, W.P.S. (2013) Pengaruh penggunaan tepung ikan dan menir kedelai terproteksi dalam ransum terhadap kecernaan nutrien pada sapi persilangan simmental peranakan ongole jantan. *Trop. Anim. Husbandry*, 2(1): 1-7.
- 6. Rochijan, Widyobroto, B.P. and Ismaya (2016) Effect of high rumen undegraded protein (HRUP) supplementation on estrous response and progesterone hormone profile in dairy cows raised under Indonesia tropical environmental conditions. *Asian J. Anim. Sci.*, 10(3): 175-181.
- 7. Yörük, M.A., Aksu, T., Gul, M. and Bolat, D. (2006) The effect of soybean meal treated with formaldehyde on amount of protected protein in the rumen and absorption of amino acid from small intestines. *Turk. J. Vet. Anim. Sci.*, 30(5): 457-463.

- 8. Abdel-Ghani, A.A., Solouma, G.M.A., Abd Elmoty, A.K.I., Kassab, A.Y. and Soliman, E.B. (2011) Productive performance and blood metabolites as affected by protected protein in sheep. *Open J. Anim. Sci.*, 1(2): 24-32.
- 9. Riyanto, J. and Sudibya (2018) Evaluation of feed for thin-tailed sheep fattening with supplemented protected and unprotected aldehyde. *IOP Conf. Ser. Earth Environ. Sci.*, 119(1): 012020.
- 10. Tiwari, M.R., Ghimire, R.P., Adhikari, D., Adhikari, D.P. and Ghimire, S.H. (2015) Effect of formaldehyde-treated sesame cake feeding on growth performance of growing female goats in fodder based basal diet effect of formaldehyde-treated sesame cake feeding on growth performance of growing female goats in fodder based basal diet. *Nepal. J. Agric. Sci.*, 13(1 Sept. 2015): 14-20.
- 11. Beigh, Y.A., Ganai, A.M. and Ahmad, H.A. (2017) Prospects of complete feed system in ruminant feeding: A review. *Vet. World*, 10(4): 424-437.
- 12. Kmicikewycz, A. and Heinrichs J. (2016) Total Mixed Rations for Dairy Cows: Advantages, Disadvantages, and Feeding Management. The Pennsylvania State University. Available from: http://www.extension.psu.edu. Last accessed on 7-10-2018.
- 13. Linn, J. (2016) Feeding Total Mixed Rations. The University of Minnesota Extension. Available from: http://www.extension.umn.edu/agriculture/dairy/feed-and-nutrition/feeding-total-mixed-rations. Last accessed on 13-10-2016.
- 14. Q Z Shams Al-dain, A K Nasser, N Y Abou and Aywad B Mahmood 2019 IOP Conf. Ser.: Earth Environ. Sci. 388 012031. Effect of feeding starter diets treated with formaldehyde, sex and age on some reproductive and blood parameters of the local calves until weaning.
- 15. H.M. El-Shabrawy and H.A. El-Fadaly. EFFECT OF FORMALDEHYDE TREATMENT AND FEEDING REGIME OF DIETS FOR CROSSBRED FRIESIAN COWS ON MILK PRODUCTION AND MICROBIOLOGICAL STATUS. Egyptian J. Anim. Prod., (2006) 43(1):25-39
- 16. European Food Safety Authority (EFSA) FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed). (2014) Scientific Opinion on the safety and efficacy of formaldehyde for all animal species based on a dossier submitted by Regal BV. *EFSA J.*, 12(2): 3561.
- 17. Haryanto, B. (2014) Manipulating protein degradability in the rumen to support higher ruminant production. *Wartazoa*, 24(3): 131-138.
- 18. Mahima, Kumar, V., Tomar, S.K., Roy, D. and Kumar, M. (2015) Effect of varying levels of formaldehyde treatment of mustard oil cake on rumen fermentation, digestibility in wheat straw based total mixed diets *in vitro*. *Vet. World*, 8(4): 551-555.
- 19. Suhartanto, B., Utomo, R., Kustantinah, Budisatria, I.G.S., Yusiati, L.M. and Widyobroto, B.P. (2014) The effect of formaldehyde inclusion on undegraded protein processing and its supplementation level on complete feed pellet on *in vitro* rumen microbial activities. *Bul. Peternakan*, 38(3): 141-149.