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ANTHELMINTIC-SALT MIXTURES – AN EFFECTIVE MEANS IN PREVENTION OF HELMINTHIASES IN SHEEP

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Abstract. *The article presents results of studying the effectiveness of the developed anthelmintic-salt mixture enriched with microelements (Co, I, Zn, Mn, Mg, and Cu) against sheep helminths and its effect on hematological parameters of animals. Experimental studies were performed under laboratory and farm conditions. The results of laboratory studies have shown that one animal consumed an average of 6.48 g of an anthelmintic-salt mixture per day. It was found that in sheep of the experimental group the extensiveness of infection by Marshallagia was 18.75%, Nematodirus – 6.25%, other gastrointestinal Strongyloides – 43.75%, Moniezia – 12.5%, Fasciola – 37.5%, Dictyocaulus – 25.0%, while in sheep of the control group, these figures were much higher and constituted 87.5%, 50.0, 50.0, 37.5, 62.5, and 50.0%, respectively. The results of hematological studies have shown that long-term feeding of an anthelmintic-salt mixture enriched with microelements does not have a negative effect on the body of sheep. When conducting studies directly under sheep farm conditions, in sheep fed an anthelmintic-salt mixture enriched with microelements for 8 months, the extensiveness of infection was 28.6%, while among sheep in the control group, this figure was 70.0%. Therefore, the results of studies indicate the feasibility of application the anthelmintic-salt mixture enriched with microelements for the treatment and prevention of helminthiases in sheep.*

Keywords: *anthelmintic-salt mixture, microelements, helminths, helminthiases, extensiveness of infection, intensity of infection*

Introduction

Sheep breeding is one of the important areas of animal husbandry. In the Republic of Uzbekistan, the sheep population amounts to more than 20 million, of which more than half are Karakul sheep (Aripov et al., 2020). The domestic sheep provide humans with milk, meat, wool, and sheepskins. Besides that, the by-products obtained from the sheep, like tallow, intestine, wool wax, horns, and hooves, are also valuable. The main challenges that the sheep industry faces with in desert and semi-desert areas are water shortage, complicated grazing problems, and a prevalence of diseases, including helminthiases. On the territory of the Republic of

Uzbekistan, the spread of helminthiases among sheep is facilitated by extensive sheep breeding, year-round grazing in case of insufficient compliance with the terms and frequency of deworming. The infection of sheep by helminths causes complex pathological processes in the body, which may decrease the growth rate in young animals, reduce the qualitative and quantitative performance indicators (wool, meat, and dairy), increase feed conversion ratio. Therefore, parasitic diseases cause significant economic damage to the sheep industry, and the development of modern methods and tools for the treatment and prevention of helminthiases in sheep is important.

Analysis of recent researches and publications

Among helminth infections in sheep, the lungworms and gastrointestinal parasites are dominated. The impacts of parasitism on livestock can be severe, arising from pathological changes to the gastrointestinal tract. However, many impacts are sub-clinical, acting through suppression of appetite and impaired assimilation of forage (Coulson et al., 2018).

The levels of infection by parasites is highly dependent on the season and have annual variations. For example, the incidence of the liver fluke, Fasciola hepatica, is inextricably linked to high rainfall and is particularly prevalent in high rainfall years. Even climate change may have profound effects on parasite epidemiology, especially for those parasitic diseases where the weather has a direct effect on the development of free-living stages (Taylor, 2012).

According to Karlsson & Greeff (2012), there is evidence of genetically determined host resistance mechanisms for most of the sheep parasites. These mechanisms vary; from no or reduced establishment, early expulsion, to suppression of parasites resulting in reduced size and fecundity.

There are different methods for identifying species of helminth parasites in ruminants. The development of molecular markers for the identification of helminths has given successful results as far as precision is concerned. The molecular methods include amplified fragment-length polymorphism analysis, a parasite-specific DNA probe, restriction fragment length polymorphism analysis, a polymerase chain reaction (Dallas et al., 2000; Pyziel et al., 2015; Biswal, 2016). The association of light and scanning electron microscopy allowed a detailed analysis of the morphology and ultrastructure of nematodes (Bashtar et al., 2011; Borji et al., 2011). However, the fecal egg count reduction test (FECRT) is the recommended method to monitor anthelmintic drug efficacy in ruminants. There is a large variation in fecal egg count methods applied to determine FECRT (Levecke et al., 2012).

Prophylactic treatment with anthelmintics is the primary control measure for internal parasite infection in sheep. Since their first introduction into the market in the late 1930s (phenothiazine) and advances in the 1960s (thiabendazole and levamisole), these chemicals have had a revolutionary effect on sheep husbandry practices, in particular facilitating single enterprise intensive sheep farming. Anthelmintic use is now so widespread and conventional that it is believed that without anthelmintic drugs the sheep industry could not exist in its current form (Sayers & Sweeney, 2005).

It is known that helminthiases of sheep, such as gastrointestinal strongylatoses, dictiocaulosis, anoplocephalatoses, especially monieziosis, are widespread in all climatic and geographical

zones all over the world and cause colossal damage to sheep breeding. Against the widespread occurrence of these helminthiases, in the 50–60s of the past century, a prophylactic drug was developed and widely introduced – an anthelmintic-salt mixture, consisting of 10% phenothiazine, 1% copper sulfate, and 89% sodium chloride (NaCl). However, with the cessation of phenothiazine production in the 90s, the use of an anthelmintic-salt mixture became impossible.

Due to the importance and effectiveness of this prophylactic drug in the Republic of Uzbekistan, studies were carried out to develop a new composition of the anthelmintic-salt mixture using modern anthelmintic drugs – albendazole, fenbendazole, tetramisole instead of phenothiazine (Oripov et al., 2007, 2007a, 2007b). These anthelmintic drugs have shown fairly high efficiency in the prevention of clinically manifested cases of the above-mentioned helminthiases in sheep.

However, in order to improve these means, as well as taking into account the enrichment of the composition of the anthelmintic-salt mixture with microelements, which are usually lacking in the soil and plants of desert and semi-desert regions – the main areas of sheep breeding, we conducted research on the development of an anthelmintic-salt mixture enriched with microelements. So, the purpose of the study was to test the developed anthelmintic-salt mixture enriched with microelements for its anthelminthic properties in laboratory and farm conditions.

Materials and methods of research

The experiments were carried out in laboratory conditions as well as directly under conditions of Karakul farms. The sheep of experimental groups were fed with anthelmintic-salt mixture containing 0.02% albendazole (by active substance), 1.0% copper sulfate (CuSO4), 0.1% zinc sulfate (ZnSO4), 0.05% magnesium sulfate (MgSO4), 0.05% manganese sulfate (MnSo4), 0.005% sodium iodine (NaI), 0.025% cobalt chloride (CoCl), and 98.5% sodium chloride (NaCl) as the filler. The experiments conducted on sheep in the laboratory conditions lasted for 630 days, and the experiments under farm conditions in accordance with the recommended terms of feeding the anthelmintic-salt mixture – 8 months, from October 1 to May 31.

Before the beginning of feeding sheep with anthelmintic-salt mixture and during the experiment, helminth-ovoscopic and larvoscopic examinations were carried out in order to determine the infection of animals by the causative agents of helminthiases – gastrointestinal and pulmonary nematodoses, anoplocephalatoses (monieziosis), fascioliasis, and other helminthiases.

Hematological examinations were carried out to determine the blood formed elements, the amount of hemoglobin, and erythrocyte sedimentation rate by conventional methods.

Results of the research and their discussion

The results of experiments carried out in the laboratory conditions have shown that the average amount of consumed anthelmintic-salt mixture per one animal was 6.476 g per day.

The level of infection in sheep of the 1st group, i.e. the experimental, fed with the anthelmintic-salt mixture enriched with microelements for 630 days, was significantly lower than in sheep of the control group (Table 1).

1. Results of laboratory testing of anthelmintic-salt mixture enriched with microelements on sheep

Group	Extensiveness of infection, %						
	Marshallagia	Nematodirus	Other gastrointestinal Strongyloides	Moniezia	Fasciola	Dictyocaulus	All helminths
1st Experimental, n=16	18.75	6.25	43.75	12.50	37.50	25.00	90.00
2nd Control, n=8	87.50	50.00	50.00	37.50	62.50	50.00	100.00

So, the sheep of the 1st (experimental) group were infected by different helminths by 90.0%, their extensiveness of infection by Marshallagia was 18.75%, Nematodirus – 6.25%, other gastrointestinal Strongyloides – 43.75%, Moniezia – 12.5%, Fasciola – 37.5%, and Dictyocaulus – 25.0%. These indicators in sheep of the 2nd (control) group were 100.0%, 87.5, 50.0, 50.0, 37.5, 62.5, and 50.0%, respectively, i.e. were significantly higher than indicators in sheep of the experimental group.

Our results have shown that long-term, i.e. within 630 days, feeding sheep with an anthelmintic-salt mixture enriched by microelements does not have a negative effect on the body, as evidenced by the results of hematological studies given in Table 2. So, the amount of hemoglobin in the blood of sheep of the experimental group was on average 10.82%, the number of erythrocytes – 5.9 million/mm3, leukocytes – 8.9 thousand/mm3, the erythrocyte sedimentation rate in 24 hours – 8.9 mm. These indicators in sheep of the control group were 11.82%, 5.8 million/mm3, 7.4 thousand/mm^3, and 11.9 mm/hour, respectively.

Consequently, the blood parameters in sheep of both the experimental and control groups did not undergo significant changes and were within the physiological norm during the experimental period.

The results of testing anthelmintic-salt mixture enriched with microelements directly in sheep breeding farms (Table 3) showed that the extensiveness of infection in sheep of the experimental flock that received anthelmintic-salt mixture for 8 months with all helminths, i.e. the total extensiveness of infection by helminths was 28.6%, and the extensiveness of infection by Marshallagia was 2.8%, by Nematodirus – 17.14%, by other gastrointestinal Strongyloides and Moniezia – 2.8%. The sheep of the control flock that did not receive anthelmintic-salt mixture were infected with all helminths by 70.0%, Marshallagi

2. Hematological parameters in sheep treated with anthelmintic-salt mixture and control animals

Group	Hemoglobin, g%	Erythrocyte sedimentation rate, mm/hour		Erythrocytes, million/mm^3	Leukocytes, thousand/mm^3
		1 hour	24 hours		
1st Experimental	10.8 ± 0.56	1.9	8.4	5.9 ± 0.38	8.9 ± 0.26
2nd Control	11.0 ± 0.71	1.5	11.9	6.2 ± 0.44	7.4 ± 0.41

3. Results on testing the anthelmintic effectiveness of anthelmintic-salt mixture enriched with microelements in sheep farms

Group	Content of active substance	Extensiveness of infection in relation to the control group, %				
		Marshallagia	*Nematodirus*	*Other gastrointestinal Strongyloides*	*Moniezia*	Total
1st Experimental, n = 961	Albendazole (0.02%), $CuSO_4$ (1%), $ZnSO_4$ (0.1%), $MgSO_4$ (0.05%), $MnSO_4$ (0.05%), NaI (0.005%), CoCl (0.025%)	2.8 16.8	17.14 57.1	2.8 16.8	2.8 16.8	28.6 40.8
2nd Control	None	16.6 100.0	30.0 100.0	16.6 100.0	26.7 100.0	70.0 100.0

and other Strongyloides – 16.6%, Nematodirus – 30.0%, Moniezia –26.7%.

It should be noted that despite the definitely high extensiveness of infection indices of sheep in the experimental group by some helminths (Nematodirus – 17.1%) and total infection (40.6%), the intensity of infection by helminths was significantly lower than those in sheep of the control group.

Consequently, the application of the anthelmintic-salt mixture enriched with microelements for sheep in farm conditions causes a significant (by 60–80%) decrease in the level of infection by helminths, which leads to the conclusion that it is advisable to use the developed anthelmintic-salt mixture in sheep farming.

The helminth-ovoscopic and larvoscopic examinations allowed determining the species of helminths that infected sheep. These results have shown that sheep in Karakul farms were infected with Marshallagia, Nematodirus, gastrointestinal Strongyloides, Moniezia, Fasciola, and Dictyocaulus.

The intestinal cestode Moniezia spp. is of global occurrence, and its infection leads to economic losses in livestock, especially in calves and lambs (Obanda et al., 2019). Cestodes are common gastrointestinal parasites of humans and livestock. They attach to the host gut and, without a mouth or intestinal system, absorb nutrients through their epidermis (Mair et al., 2020).

Species of Marshallagia are abomasal parasites in free-ranging and domesticated ungulates in temperate climatic zones throughout the world. The development of Marshallagia marshalli in the abomasal glands of ruminants causes pathophysiological changes, which include a reduced acidity of the abomasal contents, increased abomasal pH, and increased serum pepsinogen concentrations. The reduced acid secretion is explained by a replacement of functional parietal cells by undifferentiated cells (Moradpour et al., 2013).

The presence of Strongyloides spp. and in the host community is of public health interest because of the zoonotic nature of these nematodes (Obanda et al., 2019). The pathogenic impact of the parasites on the host derives from the pres-

ence of the larvae during migration and/or the adult forms in the intestines, which through their mechanical and secretory/excretory products are harmful to the host's tissues. The presence of Strongyloides papillosus causes disturbances in animals' health, quite frequently inducing the sudden death syndrome in lambs due to heart failure (Dimitrijević et al., 2012).

Fasciola hepatica and Fasciola gigantica are the major causative agents of liver fluke disease (fascioliasis) in domestic animals in regions with temperate and tropical climates, respectively. Although traditionally regarded as a disease of livestock, fascioliasis is now recognized as an important emerging zoonotic disease of humans. Transmission occurs where rural farming communities regularly share the same water source as their animals or consume water-based vegetation growing in endemic areas (Robinson & Dalton, 2009).

Nematodirus spp. are among the most common nematodes of ruminants, mainly inhabiting the small intestines in sheep. Nematodirosis is a major cause of severe diarrhea and mortality in young lambs (Melville et al., 2020). Acute disease is the consequence of very heavy larval challenge and the effects of the developing larvae (Morrison et al., 2014).

Dictyocaulus lungworms are the causative agents of parasitic bronchitis (dictyocaulosis) in ruminants. Clinical signs include coughing, nasal discharge, emphysema, and pneumonia, which may lead to death of heavily infected individuals (Pyziel et al., 2015). Lungworms cause the development of bacterial pneumonia due to secondary complications in the lungs, as well as a decrease in milk production and liveweight loss, especially in young animals. Dictyocaulus filaria is the most pathogenic lungworm in sheep. The adult worms live in the trachea, bronchi, and bronchioles of small ruminants (Sevimli et al., 2011).

Thus, the high prevalence of mixed parasitic infections among sheep in Karakul farms is a very serious problem for owners and requires urgent control measures. There are several ways for effective control and prevention of described helminth infections. Novel developments for the management of nematode parasites such as vaccines, biological anthelmintics, genetic markers, and selective breeding of sheep may, in the future, provide additional or alternative means of parasite control. However, such alternative control methods are likely to be more dependent on a sound understanding of the lifecycle and population dynamics of the parasites involved and the epidemiology of disease they cause than current methods that rely heavily on broad-spectrum anthelmintics (Vlassoff et al., 2001).

The application of anthelmintic treatment of sheep on farms and pasture management are the main tools to provide effective control measures of gastrointestinal and respiratory tract helminths. However, the emergence of anthelmintic-resistant strains of parasitic nematodes and the increasing reliance placed on anthelmintics for their control can exert profound changes on the epidemiology of these helminths (Taylor, 2012).

This proves the necessity to develop new effective anthelmintics for sheep husbandry. The application of anthelmintic-salt mixture enriched with microelements have shown sufficient anthelmintic efficacy in testing on sheep under laboratory and farm conditions, didn't have a negative impact on hematological parameters and might be recommended to use for control and prevention of parasitic infections among sheep.

Conclusions and future perspectives

The long-term ad libitum feeding of sheep with anthelmintic-salt mixture consisting of 0.02% albendazole (by active substance), 1.0% copper sulfate (CuSO4), 0.1% zinc sulfate (ZnSO4), 0.05% magnesium sulfate (MnSO4), 0.05% manganese sulfate (MnSo4), 0.005% sodium iodine (NaI), and 0.025% cobalt chloride (CoCl) does not have a negative effect on the body and significantly reduces the degree of infection by gastrointestinal Strongyloides and Moniezia in animals, and also prevents the clinical manifestation of the diseases caused by these helminths.

It is advisable to use an anthelmintic-salt mixture in sheep-breeding farms, which includes microelements missing in the soil and plants of a particular region.

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Анотація. *У статті представлені результати дослідження ефективності застосування розробленої антгельмінтно-сольової суміші збагаченої мікроелементами (Co, I, Zn, Mn, Mg і Cu) проти гельмінтів овець та її вплив на гематологічні показники тварин. Експериментальні дослідження проводили в лабораторних та виробничих умовах. За результатами досліджень проведених у лабораторних умовах встановлено, що одна*

тварина в середньому споживала 6,48 г антгельмінтно-сольової суміші на добу. З'ясовано, що в овець дослідної групи екстенсивність інвазії маршалагіями складала 18,75%, нематодірусами – 6,25%, іншими стронгілятами травного каналу – 43,75%, монієзіями – 12,5%, фасціолами – 37,5%, диктіокаулами – 25,0%, тоді як у овець контрольної групи ці показники були значно вищими і відповідно складали: 100,0%, 87,5, 50,0, 50,0, 37,5, 62,5 і 50,0%. Результати гематологічних досліджень показали, що тривале згодовування антгельмінтно-сольової суміші збагаченої мікроелементами не має негативного впливу на організм овець. Під час проведення досліджень безпосередньо у вівчарських господарствах у овець, яким упродовж 8 місяців згодовували антгельмінтно-сольову суміш збагачену мікроелементами, екстенсивність інвазії гельмінтами складала 28,6%, тоді як поміж овець контрольної групи цей показник становив 70,0%. Отже, результати досліджень вказують на доцільність застосування антгельмінтно-сольової суміші збагаченої мікроелементами для лікування та профілактики гельмінтозів у овець.

Ключові слова: *антгельмінтно-сольова суміш, мікроелементи, гельмінти, гельмінтози, екстенсивність інвазії, інтенсивність інвазії*