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# Features Of Eimeria-Strongyloid Invasion In Sheepsouth Kazakhstan

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## INTRODUCTION

Sheep breeding is one of the largest branches of agriculture, which has great economic importance in the Republic of Kazakhstan. According to the variety of products produced, sheep occupy the first place among farm animals. Sheep breeding meets the needs of the national economy in wool, rags, sheepskins, leather, and also supplies food for the population: milk, meat, cheese, and cheese.

In the conditions of the created multi-layered economy, the insufficiently high veterinary and sanitary level of animal husbandry and fragmented farms has led to an increase in invasive diseases. A special place among the diseases of sheep is occupied by protozooses and helminthiasis, which cause great economic damage due to a significant decrease in productivity, a decrease in the breeding value of young animals, resistance of the body and often the death of animals.

In Kazakhstan, until recently, eimerioses [1-5] and strongyliodoses [6] of sheep are considered and studied separately. At the same time, the authors took into account the influence of parasites on the host organism only from any one taxonomic group. However, in production conditions, pathogens of invasive diseases in animals in most cases parasitize in a mixed form [7] and cause associative diseases that develop in a more severe form and cause great economic damage [8]. In recent years, Kazakhstan began to appear in works devoted to the study of americna-esophagostomy [9], americna-strangulatory [10] and americna-marshalling [11] infestations of sheep and the development of measures to combat them.

Studies on parasitocenosis and associative diseases of sheep caused by eimeria and strongyloids have not been conducted in Kazakhstan[12-15]. The species composition of the joints of the parasitocenosis has not been established. The distribution and seasonal-age dynamics of parasite infestation in animals with combined infestations in various combinations have not been studied. The nature of the relationship between the joints of the parasitocenosis and the host organism is not determined. The reaction of the lambs ' body during their experimental infection with Eimeria and strongyloids is poorly studied. The reaction of the body of lambs when they are infected with the above pathogens in various combinations has not been studied. Methods of prevention of associative diseases of animals have not been developed[5,16].

It is known from the literature that in Kazakhstan epizootology, clinic, pathogenesis of eimeriosis, strongyloidosis of sheep were studied and measures were developed to combat them, means and ration terms of treatment of animals against these parasitoses were proposed. It should be noted that the above research and development did not take into account the presence of other parasites in the body of animals, although, as a rule, several parasites live there at the same time. So, in the South-Eastern regions of the Republic were recorded parasitocenosis of Strongyloides and Eimeria [17], nematodes and Eimeria [9,17], cestodes, nematodes, Eimeria and ticks [18,19], in the Eastern regions - nematodes, moniezia and Eimeria [9,20], nematodes and Eimeria [21-23], was tested a number of drugs when mixed infestations was investigated the relationship between Strongyloides and nematodirus, marshallagia and gamename [24] and proposed a rational scheme for pasture prevention of helminthiasis.

In the literature there is no information about the study of parasitocenoses and associative diseases of sheep caused by Eimeria, strongyloids in the south of Kazakhstan. The distribution and seasonal-age dynamics of infestation of animals with parasites during combined invasions have not been studied[25]. The nature of the relationship between the joints (eimeria, strongyloids) of the parasitocenosis and the host organism is not determined. Pathomorphological changes in the organs of lambs during experimental infection with eimeria and strongyloids, as well as changes in the organs of lambs during infection with eimeria and strongyloids at the same time, are poorly studied. Methods of prevention of mixed eimeriosis-strongyloidosis infestations of sheep have not been developed.

## MATERIALS AND METHODS

The collection of material in the mountainous zone of Southern Kazakhstan was carried out in 2015-2019 from 360 sheep in a peasant farm. The material was collected by the method of complete helminthological autopsies and in vivo studies. The matrices obtained by complete helminthological dissection were poured into cotton bags, labelled and preserved in a common jar with Barbagallo liquid. In vivo parasitological studies of sheep was performed by the method of O. Burkina. Fecal samples (3 g) and hip muscle biopsy with Popov's needle were taken from the rectum of the same animals. The feces were placed in paper bags and treated on site, or placed in penicillin vials and preserved with a 2.5% solution of potassium bicarbonate for further processing in the Institute's laboratory. The feces were thoroughly rubbed in a porcelain cup with 15-20 ml of water. The suspension was filtered through a metal sieve or gauze and centrifuged for 5 minutes at 1000-1500 revolutions per minute. Then the top layer of liquid was drained, and solutions of zinc chloride or lead nitrate with a specific gravity of 1.598 and 1,500, respectively, were added to the sediment. The precipitate was thoroughly mixed and centrifuged again for 1 minute at 1000 revolutions per minute. Then the top film was removed from the liquid with a wire loop, applied to a slide, added drops of distilled water, covered with a cover glass and microscopized.

Species belonging of eimeria was determined on the basis of morphological features of oocysts (shape, size, color, thickness and structure of the shell, presence of micropile, polar cap, residual body and light-refractive bodies), sporocysts (shape, size, presence of residual body and styd bodies), sporozoites (shape, size, presence of light-refractive bodies) and time of sporulation of oocysts.

In determining the eggs of strongyloids, the shape, size, color, thickness and structure of the shells were taken into account; corks at the poles, crushing balls or larvae in the center of nematodes.

The intensity of infection was determined by counting the number of eimeria oocysts and helminth eggs in 20 fields of view of the microscope. The intensity of infestation (AI) in a group (farm) or the arithmetic mean number of parasites per infected animal was determined by dividing the total number of parasites found by the number of infested animals.

The extent of infestation (EI) or the percentage of infected animals was calculated by the formula: EI = [Number of infested animals x 100]: the number of examined animals.

Experiments on experimental infection of lambs and the introduction of antiparasitic measures were carried out in the peasant farm "Aruzhan" of the Turkestan district of South Kazakhstan region. Experimental animals free of parasites were selected in two ways. The first is by raising parasite-free lambs. For this purpose, 3 weeks before lambing, sousyagny sheep with the same weight and a single fruit were determined. Then they were treated against helminths and protozoa, cleaned of dirt and kept in separate cages with separate feeders and drinkers on a lattice floor, which excluded the ingress of feces into food and water, and therefore, cases of repeated infection of animals with parasites. Lambing of queens and rearing of lambs was carried out in these cells. The second way is by screening clinically healthy and free from helminths and protozoa of animals. To do this, flocks of lambs were simultaneously examined for the detection of helminths and protozoa by in vivo methods: fecal samples, blood smears and a biopsy sample from the femoral muscles were taken. The selected parasite-free animals were kept under the conditions described above according to the first method.

Due to the fact that Amerie and Strongyloides in the body of the sheep occur as moneywise and extenuate have any questions about pathological changes in organs of infected lambs with their individual and combined invasions, and the relationship between the followers of parasitocenosis and the host organism. The answers to these questions could only be answered by experiments. From the listed parasites, the most common, pathogenic and model species studied in different laboratories, united by common clinical signs - disorders of the gastrointestinal tract, were selected for experiments. These are oocysts of Eimeriaovinoidalis, larvae of Strongyloidespapillosus (Figures 1 and 2).

To obtain the eimeria culture, the method of O. Berkinbaev and P. P. Osipov was used [204]. From lambs spontaneously infected with eimeria and secreting only monocultures, feces were taken for 5 days, placed in Petri dishes, poured with a 2.5% solution of potassium bicarbonate and cultivated in a thermostat at 22-23 ° C for 4 days. Excrement containing sporulated eimeria oocysts was filled with water, stirred, filtered and centrifuged at 1500 rpm for 5 minutes. The supernatant was drained, and an elevated solution of table salt was added to the sediment. After thorough mixing, it was re-centrifuged at 1500 rpm for 10 minutes. Then, eimeria oocysts were removed with a wire loop and diluted with distilled water 1: 10, again centrifuged at 1500 rpm for 10 minutes. 1 ml of a 2.5% solution of potassium dichromate was added to the precipitate. The number of oocysts was counted in the Goryaev chamber.

To obtain a culture of strongyloides, the method of V. P. Finnick and co-authors was used. The animals were infected orally. During the period of setting up the experiment, all lambs had their body temperature measured daily, their pulse and respiration counted, and feces taken for coprological studies.

Weighing, morphobiochemical blood tests were carried out before and after infection of animals according to the scheme of the experiment (details in each experiment); in animals that fell during the experiment and were forced to be slaughtered, pieces of intestines and internal organs were taken for histological examination.

Blood for a comprehensive study was taken from lambs from the jugular vein. Hemoglobin was determined by Sali, the number of erythrocytes and leukocytes in the camera Goryaeva, hematocrite index - microhematocrit microcentrifuge ITF-8 as well - the average volume of a single erythrocyte, the average amount of hemoglobin in the erythrocytes and the average concentration of hemoglobin in it.

For biochemical studies in order to more quickly separate the blood serum for one hour was kept in a thermostat at a temperature of 37°C. After that, blood clots were released from the walls of the test tube with a thin glass rod, and the liquid was centrifuged for 10-15 minutes at 1500-2000 revolutions per minute. The separated serum was transferred to a clean test tube and examined. Biochemical studies included the determination of total calcium in de Waard, chlorides according to Levinson, the acid capacity Nevodov, glucose Hultman modification, ionized calcium calculation method for Todorovo, inorganic phosphorus on Duwe, total protein in blood serum by spectrophotometry in the far ultraviolet as pohlman at a wavelength of 280 nm on the UV spectrophotometer.

The obtained digital data were processed by the statistical method according to N. L. Udolskaya. In total, 80 lambs were used in experiments on experimental reproduction of parasitoses.

**RESULTS**

General infestation of sheep with eimeria. When examining 360 sheep, 260 (72.2%) were infected with eimeria (Table 3.1), in which seven eimeria species were identified: Eimeria ahsata, E. crandallis, E. faurei, E. intracata, E. ovina, E. ovinoidalis, E. parva.

Our research has shown that the overall infestation of sheep with eimeria depends on age. Young animals are more infected than adult animals. The highest rates of extensiveness (90.0%) and intensity (159.8 oocysts) of infestation in animals were recorded at the age of 1 year (Table 2). However, with the age of sheep, their infestation decreases. Young animals under the age of 2 years are infected by 69.9%, with AI 23.0 oocysts, sheep older than two years-57.5%, with AI 8.5 oocysts.

The total infestation of sheep with eimeria also depends on the season of the year. The infection rate of animals in winter is 84.4% with AI 49.6 oocysts, in spring-63.3% with AI 17.8 oocysts, in summer-65.6% with AI 193.4 oocysts, in autumn-75.6% with AI 56.0 oocysts, that is, high EI is observed in winter, and AI-in summer. In young animals up to a year in all seasons of the year, EI is high: in autumn and winter it is 90.0%, in summer-93.3%, in spring-86.7%. The highest AI is observed in summer: 398.3 oocysts, the lowest-in spring 9.3 oocysts. In young animals up to two years of age, the lowest EI is observed in summer 50.0% with AI 8.8 oocysts, in autumn (76.7% with AI 21.1 oocysts) and in winter (86.7% with AI 32.4 oocysts), the infection of animals increases, and in spring slightly decreases (63.3% with AI 23.9 oocysts). In adult animals, the lowest infection rate is observed in spring (40.0% with AI 6.0 oocysts), then increases from season to season: in summer, EI is 53.3% with AI 7.8 oocysts, in autumn – 60.0% with AI 5.6 oocysts, in winter – 76.7% with AI 12.6 oocysts. Our data are consistent with those of a number of other researchers [1-4].

The infection of sheep with monenvasiaamerime. In a study of 360 sheep, 139 (38.6%) were infected with eimeriamonoinvasies (Table 3.1) with an AI of 79.9 oocysts. The infection of sheep with monenvasiaameriam depends on the age. Young animals are more infected than adult animals. High rates of extensiveness (51.7%) and intensity (171.9 oocysts) of infestation in animals were recorded at the age of 1 year (Table 3.2). However, with the age of sheep, their infestation decreases. Young animals under the age of 2 years are infected by 37.5%, with AI 6.7 oocysts, sheep older than two years – 26.7%, with AI 4.8 oocysts.

Infection of sheep with monoinvasiaeimeria also depends on the season of the year (Table 3.3). Infection of animals in winter is 47.8% with AI 17.4 oocysts, in spring-24.4% with AI 7.9 oocysts, in summer-38.9% with AI 233.4 oocysts, in autumn-43.3% with AI 51.9 oocysts, that is, high EI is observed in winter, and AI-in summer. In young animals up to a year, the highest EI (80.0%) and AI (338.9 oocysts) is in summer, then this indicator decreases in autumn to 43.3% with AI 146.5 oocysts, in winter-53.3% with AI 36.1 oocysts, in spring - to 30.0% with AI 5.3 oocysts. In young animals up to two years of age, the lowest EI is observed in summer 20.0% with AI 3.3 oocysts, in autumn (50.0% with AI 4.6 oocysts) and in winter (50.0% with AI 7.1 oocysts), the infection of animals increases, and in spring slightly decreases (30.0% with AI 11.6 oocysts). In adult animals, the lowest infection rate is observed in spring (13.3% with AI 5.5 oocysts), then increases from season to season: in summer, EI is 16.7% with AI 3.4 oocysts, in autumn – 36.7% with AI 4.6 oocysts, in winter – 40.0% with AI 5.3 oocysts.

General infection of sheep with strongyloids. In a study of 360 sheep, 144 (40.0%) were infected with strongyloids (Table 3.1) with an AI of 6.9 eggs, in which one strongyloid species was identified: Strongyloidespapillosus.

Infection of sheep with strongyloids depends on age. Young animals are more infected than adult animals. High rates of extensiveness (41.7%) and intensity (9.4%) of infestation in animals were recorded at the age of 1 year (Table 3.2). However, with the age of sheep, their infestation decreases. Animals older than one year are infected by 39.2%, with an AI of 7.4-3.7 eggs.

Infection of sheep with strongyloids also depends on the season of the year (Table 3.3). Infection of animals in winter is 37.8% with AI 6.0 eggs, in spring – 45.6% with AI 4.0 eggs, in summer – 37.8% with AI 6.6 eggs, in autumn – 38.9% with AI 11.3 eggs, that is, high EI is observed in spring, and AI – in autumn. In young animals up to a year, the lowest EI (13.3% with an AI of 7.5 eggs) is in the summer, then this indicator increases and reaches a maximum in the spring (66.7% with an AI of 4.2 eggs). In young animals up to two years of age, the highest EI of strongyloids is observed in the summer of 53.3% with an AI of 9.1 eggs, then this indicator gradually decreases reaching a minimum in the spring (30.0% with an AI of 3.8 eggs). In adult animals, the lowest infection rate is observed in autumn (33.3% with an AI of 3.7 eggs), then this indicator increases, reaching a maximum in summer (46.7% with an AI of 5.6 eggs).

**DISCUSSION**

Mixed eimeria-strongyloid infestation was found in all the examined areas. High extensiveness and intensity of sheep infestation in comparison with the average regional data is observed in the mountain zone.

The infestation of sheep with mixed infestations depends on the age and geographical zones. Young animals from 1 year to 2 years are more infected than young animals up to 1 year and adult animals (in all ages, eimeria outnumbered strongyloids by 2.0-9.6 times).

The infestation of sheep with mixed infestation depends on the season of the year. Some increase in the extensiveness of the invasion is observed in summer and autumn, in all seasons of the year, Eimeria outnumbered strongyloids by 2.8-26.5 times. In young animals up to a year, compared with the average data, high infection is noted in spring and autumn. In young animals up to two years old, compared with the average data, high infestation by mixed invasion is observed in winter and summer. In adult animals, high infection with mixed infestations, compared with the average data, is observed in summer and autumn. In all groups, in all seasons of the year, Eimeria outnumbered strongyloids by 1.6-237.6 times.

In the mountain zone, the high infestation of animals with mixed infestation is noted in winter and spring, in all seasons of the year, Eimeria outnumbered strongyloids by 1.1-100.6 times. In young animals up to a year, compared with the average data, high infection is noted in spring and autumn. In young animals up to two years old, compared with the average data, the infestation in all seasons of the year was low. In adult animals, high infection with mixed infestation, compared with the average data, is observed in the summer (36.7%). In all groups, in all seasons of the year, Eimeria outnumbered strongyloids by 1.1-100.6 times.

In the foothill zone, high infection of animals with mixed infestation is noted in winter and autumn; in winter and autumn, strongyloids outnumbered eimeria by 1.4 and -1.2 times, in spring and summer, on the contrary, eimeria by 1.5 and 1.2 times. In young animals up to a year, compared with the average data, high infection is noted in winter and autumn, in winter, spring and summer they exceeded eimeria (5.2, 3.4 and

1.6 times), and in autumn – strongyloids (1.4 times). In young animals up to two years old, compared with the average data, high infection is noted in winter, spring and summer, in winter they exceeded strongyloids (6 times), in spring and autumn – eimeria (1.1 times), and in summer the ratio of parasites was one to one. In adult animals, high infection with strongyloids, compared with the average data, high infection is noted in autumn, in spring and autumn they exceeded strongyloids (by 2.4 and 1.2 times), in summer – eimeria (by 1.6 times), and in winter the ratio of parasites was equal.

In the zone of deserts and semi-deserts, the central subzone, the high infestation of animals with mixed invasion is noted in spring, summer and autumn; in all seasons of the year, they exceeded eimeria (by 1.1, 1.8, 6.3 and 1.6 times). In young animals up to a year, compared with the average data, high infection is noted in the spring, they in all seasons of the year exceeded eimeria (2.4; 2.0; 155.8 and 2.0 times). In young animals up to two years old, compared with the average data, high infection is noted in winter and summer, in winter and spring they exceeded strongyloids (by 1.2 and 1.1 times), in summer and autumn – eimeria (by 1.9 and 1.4 times). In adult animals, high infection with strongyloids, compared with the average data, high infection is noted in summer and autumn, in winter and spring they exceeded strongyloids (by 1.2 and 1.4 times), in summer and autumn – eimeria (by 1.2 and 1.1 times).

## CONCLUSIONS

In the farms of Southern Kazakhstan, seven species of eimeria were found in sheep: Eimeriaahsata, E. crandallis, E. faurei, E. intracata, E. ovina, E. ovinoidalis, E. parva and 1 species of strongyloids: Strongyloidespapillosus. These parasites in animals occur as moneywise, and in the form of extenuate.The infestation of sheep with eimeria, strongyloid and eimeria-strongyloid infestations depends on the age, season and geographical zones.

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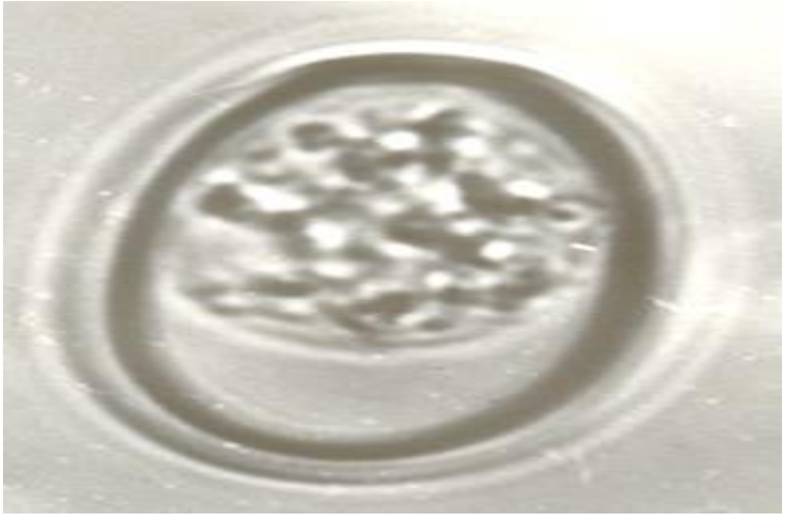

**Figure 1**. Eimeria ovinoidalis oocyst

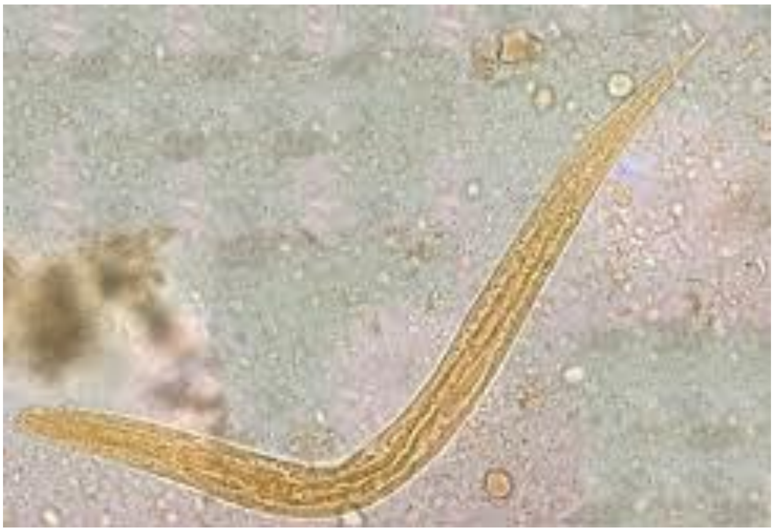

**Fig. 2.** Larva of Strongyloides papillosus

**Table 1-**Infestation of mountain sheep with eimeria, strongyloid and eimeria-strongyloid infestations

| Types of infestation | Number of sheep surveyed | Number of sheep surveyed | EI | II | |
|---|---|---|---|---|---|
| | | | | in abs. numbers | the ratio of parasites |
| General eimeria infestation | 360 | 260 | 72,2 | 75,9 | 0 |
| Monenvasia ameriam | 360 | 139 | 38,6 | 79,9 | 0 |
| General infection with Strongyloides | 360 | 144 | 40,0 | 6,9 | 0 |
| Monenvasia by Strongyloides | 360 | 23 | 6,4 | 4,5 | 0 |
| Mixed eimeria-strongyloid invasion | 360 | 117 | 32,5 | 73,9:7,4 | 10:1 |

**Table 2-**Infestation of mountain sheep with eimeria, strongyloid and eimeria-strongyloid infestations depending on age

| Types of infestation | Number of sheep surveyed | Number of sheep surveyed | EI | II | |
|---|---|---|---|---|---|
| | | | | in abs. numbers | the ratio of parasites |
| 1 | 2 | 3 | 4 | 5 | 6 |
| Young animals under the age of 1 year | | | | | |
| General eimeria infestation | 120 | 108 | 90,0 | 159,8 | 0 |
| Monenvasia ameriam | 120 | 62 | 51,7 | 171,9 | 0 |
| General infection with Strongyloides | 120 | 50 | 41,7 | 9,4 | 0 |
| Monenvasia by Strongyloides | 120 | 5 | 4,2 | 2,8 | 0 |
| Mixed eimeria-strongyloid invasion | 120 | 45 | 37,5 | 146,5:10,1 | 14,5:1 |
| Young animals aged from 1 to 2 years | | | | | |
| General eimeria infestation | 120 | 83 | 69,2 | 23,0 | 0 |
| Monenvasia ameriam | 120 | 45 | 37,5 | 6,7 | 0 |
| General infection with Strongyloides | 120 | 47 | 39,2 | 7,4 | 0 |
| Monenvasia by Strongyloides | 120 | 8 | 6,7 | 6,0 | 0 |
| Mixed eimeria-strongyloid invasion | 120 | 36 | 30,0 | 44,9:7,6 | 5,9:1 |
| Adult sheep | | | | | |
| Generaleimeria infestation | 120 | 69 | 57,5 | 8,5 | 0 |
| Monenvasia ameriam | 120 | 32 | 26,7 | 4,8 | 0 |
| General infection with Strongyloides | 120 | 47 | 39,2 | 3,7 | 0 |
| Monenvasiaby Strongyloides | 120 | 10 | 8,3 | 4,2 | 0 |
| Mixed eimeria-strongyloid invasion | 120 | 36 | 30,0 | 14,5:3,7 | 3,9:1 |