






## Prevalence of *Cryptosporidium*, *Eimeria* Species and Gastrointestinal Helminths in Lambs and Sheeps in Erzurum Region

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### Keywords

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**Abstract:** This study was carried out in Pazaryolu, Pasinler and Ilıca districts of Erzurum from February to March 2009, and included 461 lambs (253 with diarrhea and 208 healthy, varying up to 1 month of age) and 397 sheeps (9 with diarrhea and 388 healthy), from 28 pens located in 11 focuses on the region. The fresh fecal samples taken from lambs and sheeps was examined microscopically. Of the 858 fecal samples examined, *Cryptosporidium* species were found in 397 (46.3%), *Eimeria* species in 313 (36.5%) and gastrointestinal helminth eggs were found in 21 (2.4%). Ten *Eimeria* species: *Eimeria ovinoidalis*, *E. parva*, *E. pallida*, *E. faurei*, *E. bakuensis*, *E. crandalis*, *E. ahsata*, *E. weybridgensis*, *E. intricata* and *E. granulosa* were detected in lamb and sheep feces. Additionally, in the samples examined, *Strongyloides papillosus*, *Nematodirus* spp. and *Trichuris* spp. eggs were found.

## Erzurum Yöresinde Kuzu ve Koyunlarda *Cryptosporidium*, *Eimeria* Türleri ve Gastrointestinal Helmintlerin Prevalansı

### Anahtar Kelimeler

*Cryptosporidium*  
*Eimeria*,  
 Gastrointestinal  
 helminths,  
 Yaygınlık,  
 Erzurum

**Öz:** Bu çalışma 2009 yılının Şubat ve Mart aylarında Erzurum'un Pazaryolu, Pasinler ve Ilıca ilçelerinde yürütülmüş olup, yöredeki 11 odakta bulunan 28 ağıldan yaşları 1 ayağa kadar değişen 461 kuzu (253 ishalleri, 208 sağlıklı) ve 397 (9 ishalleri, 388 sağlıklı) koyundan alınan dışkı örneği mikroskopik olarak incelenmiştir. İncelenen 858 dışkı örneğinin 397'sinde (%46.3) *Cryptosporidium* türlerine, 313'ünde (%36.5) *Eimeria* türlerine ve 21'inde ise (%2.4) gastrointestinal helmint yumurtalarına rastlanmıştır. Kuzu ve koyun dışkılarında *Eimeria ovinoidalis*, *E. parva*, *E. pallida*, *E. faurei*, *E. bakuensis*, *E. crandalis*, *E. ahsata*, *E. weybridgensis*, *E. intricata* ve *E. granulosa* olmak üzere 10 *Eimeria* türü tespit edilmiştir. Ayrıca incelenen örneklerde *Strongyloides papillosus*, *Nematodirus* spp. ve *Trichuris* spp. yumurtalarına rastlanılmıştır.

### 1. INTRODUCTION

Cryptosporidiosis is a zoonotic protozoan disease that is common all over the world. Disease-causing agents are Apicomplexa subphylum, protozoa of the *Cryptosporidium* genus, which is in the Sporozoa class. It can affect a wide variety of animal species, including reptiles, birds, fish, and mammals. Transmission occurs

through increased consumption of water or food contaminated with oocysts [1].

Cryptosporidiosis is considered to be an important disease, leading to important economic losses in calves and lambs. Mild or severe yellowish, malodorous, diarrhea, and weight loss, depression, abdominal pain are seen clinically in sheep, and in general, it can cause death

in animals in one month old animals [2, 3]. The clinical course of the disease in lambs is similar to that in calves, and the agent development takes 3-4 days. Outbreaks are generally seen in 7-10 day old lambs. Risk factors such as farm management, herd size, farm type and hygiene, bedding type, colostrum feeding, season, age and contamination sources (e.g. water) play a role in the spread of *Cryptosporidium* spp [4-6].

Due to the small size of *Cryptosporidium* spp. oocysts, it is difficult to be noticed by inexperienced people. The most widely used diagnostic test is acid-fast staining of smears prepared from feces. The most used are the modified Ziehl-Neelsen (mZN) or modified Kinyoun staining technique. Recently, ELISA and PCR diagnostic methods have been widely used in diagnosis [7-9].

*Cryptosporidium parvum*, *C. xiaoi* and *C. ubiquitum* are the most common of the *Cryptosporidium* species found in sheep, whereas *C. hominis*, *C. andersoni*, *C. bovis*, *C. scrofarum*, *C. suis*, *C. fayeri* and *C. canis* are seen as sporadic [10-12]. *C. parvum* is the main species found in humans and animals. Anthroponotic *C. parvum* type I (*C. hominis*) is observed in humans, while *C. parvum* type II (*C. bovis*) is found zoonotically in humans and animals (especially ruminants). *C. parvum* type II causes infection in lambs [2, 10-12].

Ovine cryptosporidiosis was first described in diarrheic lambs 1-3 weeks olds in Australia in 1974 [13]. However, its role as the primary etiological agent of diarrhea in lambs was defined by different studies in the early 1980 [14]. In subsequent studies, infection has also been reported from different geographical regions; Its prevalence varies between 4% and 85% in the USA, Canada, Iran, Trinidad Tobago, Spain and Italy [15].

Studies performed in Turkey have reported the prevalence of *Cryptosporidium* agents in diarrheic and nondiarrheic lambs as follows: 38.8% in Kars, 79.1% in Aydın, 2.97% in Konya, 12% in Elazığ, 23.3% in İzmir, and 13.17% in Van regions [6, 16-20].

Coccidiosis is a protozoan disease caused by protozoans belonging to the Eimeridae family [21, 22]. Coccidiosis, which is very common in many region of the world, is primarily seen in poultry, cattle, sheep, goats, dogs, cats, pigs and rabbits. Although it causes economic losses as it cause death in young animals, especially those who survived the disease play a carrier role because they become preimmune. Clinically, it may result in hemorrhagic diarrhea, depression, weight loss, and sometimes death, especially in young animals [21-23].

For the diagnosis of coccidiosis, the age of the animal, treatment and housing conditions and clinical findings are taken into consideration. Definitive diagnosis is made by detecting the presence of oocysts in fecal samples in line with clinical symptoms. For the diagnosis of *Eimeria* oocysts, simple flotation method is used for qualitative detection in fecal samples and Modified McMaster method is used for quantitative detection, while serological and molecular methods are also used for

diagnosis. The treatment phase in coccidiosis is based on killing the causative agent or preventing its development, eliminating fluid loss in animals and treating secondary infections. The most important factors in the prevention of coccidiosis are good business management, care-feeding and hygiene. Basic protection factors such as the number of animals in the barn or farm, the temperature in the barn, the presence of animals of different age groups in the same compartments, the diet and feed contents should be taken into account [21-25].

In the previous studies, it was detected that *Eimeria ovina*, *E. ahsata*, *E. crandalis*, *E. faurei*, *E. intricata*, *E. ovinoidalis*, *E. parva*, *E. granulosa*, *E. pallida*, *E. punctata*, *E. weybridgensis*, and *E. marsica* species caused coccidiosis in sheep [21-25]. It is commonly seen in sheep and 3-8 weeks old lambs almost all over the world. The prevalence of the disease in Turkey was detected between 12.6-100% (Kars, Aegean region, Elazığ, Bursa, Antakya, Van, Bitlis) [25-31]. Different prevalence rates have been reported in various studies conducted in Kenya, Australia, Spain, and Iran it has been found to be widespread all over the world. [32-35].

Gastrointestinal helminths in sheep cause a decrease in meat, milk, skin, and wool products and even death. Gastrointestinal nematodes are also neglected in terms of a treatment since it usually progresses subclinically without causing sudden deaths. Mixed infections with other helminths have also been reported to have worsened the disease and increased the death rates [21, 36-38].

In the studies performed to determine the nematodiasis-causing species and their prevalence rates in the gastrointestinal system in sheep in Turkey were determined: *Trichostrongylus*, *Ostertagia*, *Teladorsagia*, *Marshallagia*, *Haemonchus*, *Nematodirus*, *Cooperia*, *Strongyloides*, *Bunostomum*, *Oesopagostomum*, *Gonglonema*, *Chabertia*, *Trichuris* species at the prevalence rate of 0.2-89.3%. The parasitological examination revealed that the most prevalent species was *Ostertagia* spp., followed by *Nematodirus* spp. and *Trichostrongylus* spp. [39, 40-43].

This study was carried out to determine the prevalence of *Cryptosporidium*, *Eimeria* Species and Gastrointestinal Helminths in Lambs and Sheeps in Erzurum Region.

## 2. MATERIAL AND METHOD

This study was carried out in Erzurum province during the birthing season in February and March 2009. Fresh fecal samples were collected from the rectums of 461 lambs (253 with diarrhea, 208 healthy, ages varying up to 1 month) and 397 (9 with diarrhea, 388 healthy) sheep from 28 sheep pens in 11 foci in the Erzurum region.

Fecal samples were examined using the zinc sulfate flotation method. Gastrointestinal helminth eggs and *Eimeria* oocysts were detected using this method. Fecal samples containing detected *Eimeria* oocysts were placed in petri dishes and allowed to sporulate in a laboratory oven after being treated with 2.5% potassium dichromate

[22, 24]. Sporulated oocysts were examined using centrifugal flotation under a 10-40 lens microscope. The shape, color, size, and state of the wall of the oocysts reported in the literature, as well as the shape and size of sporocysts and sporozoites, sporocyst and oocyst residue, stidea body, polar granule, micropyle, and cap, were examined under the immersion lens [22, 24, 44].

Identification of *Cryptosporidium* oocysts was made as follows: The portioned fecal samples were first homogenized with tap water at 3000 rpm for 5 minutes, then centrifuged, and smears were prepared from the sediment obtained at the end of the centrifuge, stained with the modified acid-fast method (MAF), and examined under a microscope with a 40 lens [45].

### 3. RESULTS

In the Erzurum region, 397 (46.3%) *Cryptosporidium* spp. oocyst, 313 (36.5%) *Eimeria* spp. oocyst, and 21 (2.4%) gastrointestinal helminth eggs were found in 858 fecal samples.

*Cryptosporidium* was found in 34.8% (88/253) of lambs with diarrhea. When the relationship between the appearance of oocysts in the fecal is evaluated, *Cryptosporidium* sp. oocysts were found at a higher rate. In lambs with normal fecals, this rate was determined to be 19.2% (40/208). *Cryptosporidium* was found in 55.6% (5/9) of diarrheal sheep and 68% (264/388) of healthy sheep during the periparturient period.

*Eimeria* species identified in lambs and sheep and their prevalence are as follows; *Eimeria ovinoidalis* (22%), *E. parva* (12.2%), *E. pallida* (11.9%), *E. faurei* (10.7%), *E. bakuensis* (9.7%), *E. crandalis* (3.0%), *E. ahsata* (0.7%), *E. weybridgeensis* (0.3%), *E. intricata* (0.2%) and *E. granulosa* (0.1%). *Eimeria* was detected in 41/78 (52.6%) of sheep during the periparturient period. *Strongyloides papillosus* 12/21 (57.1%), *Nematodirus* spp. 7/21 (33.3%), and *Trichuris* spp. 2/21 (9.5%) species were detected in sheep, and no gastrointestinal helminth eggs were found in lambs.

Prevalence rates in lambs and sheep and their distribution according to age groups are given in Tables 1-4.

**Table 1.** Prevalence of *Cryptosporidium*, *Eimeria* and Gastrointestinal helminths in lambs.

	Diarrheic lamb	Healthy lamb
<i>Cryptosporidium</i> spp.	88/253 (34.8%)	40/208 (19.2%)
<i>Eimeria</i> spp.	75/253 (29.6%)	94/208 (45.29%)
Gastrointestinal helminth	0/253	0/208

**Table 2.** Prevalence of *Cryptosporidium*, *Eimeria* and Gastrointestinal helminths in sheep.

	Preparturient Sheep	Postparturient Sheep
<i>Cryptosporidium</i> spp.	74/78 (94.9%)	195/319 (61.1%)
<i>Eimeria</i> spp.	41/78 (52.6%)	103/319 (32.3%)
Gastrointestinal helminth	12/78 (15.4%)	9/319 (2.8%)

**Table 3.** According to age groups in lambs *Cryptosporidium* spp. prevalence.

	Diarrheic lamb	Healthy lamb	Total
1 Week	12/45(26.7%)	1/7 (14.3%)	13/52 (25%)
2Week	14/46(30.4%)	8/55(14.5%)	22/101(21.8%)
3 Week	23/75(30.7%)	16/72(22.2%)	39/147 (26.5%)
4 Week	39/87(44.8%)	15/74(20.3%)	54/161 (33.5%)

**Table 4** According to age groups in lambs *Eimeria* spp. prevalence.

	Diarrheic lamb	Healthy lamb	Total
0-2 Week	10/91 (11%)	24/62 (38.7%)	34/153 (22.2%)
3-4 Week	66/162 (40.7%)	70/146 (47.9%)	136/308(44.2%)

### 4. DISCUSSION AND CONCLUSION

*Cryptosporidium* species are highly important in domestic animals and are recognized as the principal etiological agent in the diarrhea cases of newborn lambs. It progresses with high morbidity and mortality in inadequate care and feeding. The postpartum period is the time when animals are most susceptible to natural infection [46]. The disease is controlled by the immune system, and it can be self-limiting in those with a developed immune system. It has been reported that risk factors such as herd size, rearing type, birth time and weaning are effective in the occurrence of oocysts. Farm management and hygiene are important in the prevention of infection [5, 6].

In a study performed on lamb diseases in the neonatal period in Turkey, it was reported that diarrhea cases ranked top with 15.4%. It has been observed that *Cryptosporidium* infections are one of the most important causes of neonatal lamb diarrhea. It has been reported that *Cryptosporidium* oocyst excretion has been observed to be high in animals affected with diarrhea [47].

In a study performed on the prevalence of *Cryptosporidium* was found a rate of 17.2% in sheep and lambs in Iran [5]. In studies performed in Spain, prevalence rates ranging from 14.7% to 45% in lambs with diarrhea and 59% in sheep were reported [48]. Another research revealed a prevalence of 2.6% with microscopic testing and 26.2% with PCR in lambs in Australia [49]. Other studies found that the prevalence of *Cryptosporidium* infection in lambs was 12.5% in

Zambia, 3.7% in Brazil, 10.1% in Poland, 42.1% in Serbia, 11.2% in Tunisia, and 23% in Canada [50-55].

According to studies done in various geographical and climatic regions of Turkey, the prevalence of *Cryptosporidium* agents in lambs with diarrhea is as follows: 12% in Elazığ, 23.3% in İzmir, 2.97% in Konya, 79.1% in Aydın and 3.63 percent in Van area, while in Kars region 21.05% and 38.8% were found in two different studies [6, 16-20]. In this study, the prevalence was found to be 46.3%. The difference in prevalence between studies is assumed to be caused by the number of materials used, the number of animals with diarrhea, and the age of the animals. The prevalence of *Cryptosporidium* infection has been reported to be between 4% and 85% in sheep worldwide [56]. Studies on the prevalence of *Cryptosporidium* have found prevalences ranging from 13.6% to 46.5% in Turkey [16, 57]. These studies indicate that *Cryptosporidium* infections in sheep and lambs are common in many areas across the world, including Turkey.

The age of the host animals is the most important risk factor influencing *Cryptosporidium* prevalence. Cryptosporidiosis is widespread in companies that keep a high number of calves, lambs, and kids, and the clinical picture is most common in lambs under one month old. *Cryptosporidium* oocysts are more common in 1-3 week old animals. It has been reported that the incidence of oocysts decreases with age [6, 18]. In this study, however, the prevalence was higher (33.5%) in 4-week-old lambs. The reason is based on the fact that the number of 4-week-old animals used is higher than the animals of other species.

The prevalence of *Eimeria* in sheep is high in all age groups. However, the disease is important in lambs from two weeks to three months old. It shows a latent course in the elderly. Among the key risk factors for the disease are hygiene, care, and feeding [24-25, 30].

Coccidiosis in sheep and lambs is common infection in Turkey and many other countries across the world. The prevalence of coccidiosis in Turkey ranged from 12.6 to 100% (Antakya, Bitlis, Aegean region, Bursa, Elazığ, Kars, Van). It was determined that nine different *Eimeria* species infect 29.9% of the sheep in Bursa. Nine *Eimeria* species of 94.8% of lambs in Elazığ, seven different *Eimeria* species of 37.26% of sheep in Elazığ, nine different *Eimeria* species of 100% of sheep in Van and 10 different *Eimeria* species of 93.9% of lambs and sheep in Kars province were detected [25-31]. In this study, the prevalence of *Eimeria* was found to be 36.5%, and it was determined that the animals were infected with 10 different *Eimeria* species. In the world, this rate was found to be between 12.7% and 97% [32-35]. The differences and prevalences among *Eimeria* species vary depending on climate, vegetation, immunity, age, species, farm management, other diseases and stress factors [29-30, 34].

Periparturient period is very important because of parasitic diseases. The immune system is suppressed during this period due to an increase in glucocorticoid and

estrogen levels. Hormonal changes and stress increase the prevalence of parasite factors. Therefore, there is an increase in the excretion of *Cryptosporidium* and *Eimeria* oocysts in the periparturient period [46, 58]. In this study, the infection rate in sheep during the periparturient period, also known as the transition time in animals, was determined to be higher than in those during the postparturient period.

A prevalence of 0.2-89.3% was observed in studies done in Turkey to investigate the species causing gastrointestinal nematodiosis and their prevalence rates in sheep [39-43]. This rate was determined to be 2.5% in present study. The prevalence of helminth infections varies according to several factors, including parasite (biological characteristics), host (genetic resistance, age, physiological state), and environmental factors (climate, altitude, breeding system). Factors such as pasture composition, animal diversity, the proportion of herds or herds in the same pasture, and pasture pollution play a very important role in the spread of helminths. In addition, antiparasitic drug usage, sample size, and coinfections impact the prevalence of parasitic infections. Age is another factor that affects the prevalence of gastrointestinal parasites. In general, adults are more resistant to parasitic infections than young animals due to age-related changes in host immunity [59-61]. However, infection was observed in sheep in this study.

The increased infection rate in adults can be attributed to the disease spreading across large areas during grazing, overfeeding in contaminated grassland, and exposure to stress factors [32, 59, 61].

In this study, *Cryptosporidium* spp. was found in 397 (46.3%) of 461 lambs (253 with diarrhea, 208 healthy) and 397 (9 with diarrhea, 388 healthy) sheep feces samples ranging from 28 sheep pens up to 1 month old in 11 foci in Erzurum region. *Eimeria* spp. (36.5%) and gastrointestinal helminth eggs (2.4%) were found. The prevalence of *Cryptosporidium* was determined as 34.8% (88/253) in lambs with diarrhea and 19.2% (40/208) in healthy lambs, and the prevalence of *Cryptosporidium* in sheep was 55.6% (5/9) in diarrheal lambs and 68% (264/388) in healthy ones. Furthermore, different *Eimeria* species were identified in this study. These species are as follows; *Eimeria* *ovinoidealis*, *E. parva*, *E. pallida*, *E. faurei*, *E. bakuensis*, *E. crandalis*, *E. ahsata*, *E. weybridgeensis*, *E. intricata* and *E. granulosa*. *Strongyloides papillosus*, *Nematodirus* spp., and *Trichuris* spp. were found among the gastro-intestinal nematod eggs. These nematod eggs were not detected in lambs.

Care-hygiene, rearing type, pen condition, season, water resources, breeding season, long winter conditions, and prolonged stay of young animals in the barn were determined as risk factors that determine the frequency of parasites in the region (Field Observations). Parasite density was reported especially in areas where the physical conditions of the pen were poor.



As a result, *Cryptosporidium* and *Eimeria* species have been identified as common in local lambs. It was determined that gastrointestinal helminth eggs were found at a low rate. The reason for this is the use of drugs against helminths to sheep following the return from grassland in the area.

It has been determined that *Cryptosporidium* and *Eimeria* species have been found to be common in asymptomatic sheep, and *Cryptosporidium* oocyst is excreted at least as much as diarrheal lambs with sheep feces throughout the periparturient period. For this reason, separating the lambs from their mothers and keeping them in separate compartments after birth will limit diarrhea cases and hence clinical cryptosporidiosis, preventing the disease from spreading.

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