

An Epidemiological Study to Investigate Intestinal Protozoal Infection (*Giardia* & *Cryptosporidium*) in Cattle, Sheep and Goats at Kerbala City

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Abstract— The ruminant output is significantly hampered by gastrointestinal parasite infection, which can result in production losses and, in extreme situations, even death. *Giardia* spp. and *Cryptosporidium* spp. are currently regarded in veterinary medicine as significant pathogens in the aetiology of diarrhea in confined calves. The aim of study was to detect the Gastro-intestinal parasites (*Giardia* spp. and *Cryptosporidium* spp.) in farm animals and risk factor to transport the infection with effect the infection on Hematological parameter. The aim of study was to detect the Gastro-intestinal parasites (*Giardia* spp. and *Cryptosporidium* spp.) in farm animals and risk factor to transport the infection with effect the infection on Hematological parameter.

In this study, 300 blood and fecal samples (100 sheep, 100 goats and 100 cattle) of ages (<1 year and >1 year) and sexes from the Karbala province were collected between August and March. All sampled animals were maintained under an extensive open-field grazing system. Microscopic analyses Protozoa were the parasites that were discovered in the fecal samples of sheep, goats, and cattle during the study period. The overall prevalence of intestinal parasites was 28.66%, or almost 41%. In sheep, 27% of the goats and 18% of the cattle were infected. The study shows a clear dominance of *Cryptosporidium* spp. over *Giardia* spp. across all farm animals. For both parasites, the infection rates followed an identical trend: Sheep > Goats > Cattle. *Cryptosporidium* spp. showed higher infection rates than *Giardia* spp. across all species. *Cryptosporidium* spp. the data showed higher infection rates in animals older than one year, the female higher infection rates than male

The study demonstrates that protozoan parasites significantly compromise the health and hematological status of livestock, with males and older animals showing unexpected prominence in infection rates, necessitating improved environmental management and targeted veterinary intervention.

Keywords — *Giardia* spp., *Cryptosporidium* spp., diarrhea, emaciation Intestinal Protozoal Infection.

INTRODUCTION

The ruminant output is significantly hampered by gastrointestinal parasite infection, which can result in production losses and, in extreme situations, even death (1). *Giardia* spp. and *Cryptosporidium* spp. are currently regarded in veterinary medicine as significant pathogens in the aetiology of diarrhea in confined calves. However, it wasn't until the end of the previous century that the pathogenicity of these parasites was acknowledged. *Cryptosporidium* spp. was linked to diarrhea in calves in 1971 (2). has a straightforward life cycle that consists of two stages: a trophozoite stage that colonizes the host's intestinal lumen and causes the clinical symptoms, and an infectious cyst that is resistant to numerous environmental stressors (3).

The parasite *Giardia* spp. lamblia is a flagellated binucleated parasite protozoa from the Mastigophora class that inhabits the duodenum down to the upper part of the ileum in humans and a wide range of vertebrates (4). It is typically regarded as an epidemic and zoonotic illness that spreads from humans to animals, including farm animals, dogs, cats, birds, and rodents (5).

Giardia spp. infection can be contracted through the fecal-oral route (due to poor hygiene practices). In warm water, the *Giardia* cyst can live for weeks or months. The trophozoite uses a ventral sticky disk to adhere to the epithelium and reproduces through binary fission (6). Regarding *Giardia*, the successful spread of *Cryptosporidium* infections in calves is attributed to a number of parasite traits. In contrast to the small amount of oocysts required for infection, calves with patent cryptosporidiosis defecate a large quantity of infectious oocysts. Additionally, at temperatures between -4°C and 20°C, oocysts can endure for over 12 weeks in soil and up to 24 weeks in water. Oocysts are degraded in bovine feces due to faecal microorganisms, however they are still contagious in cattle manure for at least 70 days (7).

Cryptosporidium spp. is highly zoonotic and more aggressive in its destruction of intestinal epithelial cells. It causes significant fluid loss and inflammation (8). Despite the high frequency in farm animals and the fact that the first instance of bovine giardiasis was documented in 1921 (9), the clinical

significance of giardiasis in cattle was only recently characterized (10).

The breeding of farm animals, such as sheep, goats, and cattle, is a significant part of the economy, particularly in Iraq, where it is regarded as a major source of food products, such as meat, milk, dairy products, and leather (11). Enteritis, anemia, emaciation, dehydration, and mortality are clinical signs of intestinal parasite infection (12). Tropical and subtropical areas of the world have higher rates of intestinal parasitism (13).

Inflammation of the gut wall caused by intestinal parasites alters how certain vitamins and minerals are absorbed (14).

The aim of study was to detect the Gastro-intestinal parasites (*Giardia* spp. and *Cryptosporidium* spp.) in farm animals and risk factor to transport the infection with effect the infection on Hematological parameter.

MATERIALS AND MTHODS

In this study, 300 blood and fecal samples (100 sheep, 100 goats, and 100 cattle) of ages (<1 year and >1 year) and sexes from the Karbala province were collected between August and March. All sampled animals were maintained under an extensive open-field grazing system.

The research received ethical approval from the College of Veterinary Medicine, University of Karbala No. (UOK.VET.ME.2025.162)

A sample of blood collected using an anticoagulant (EDTA, 1 mg/5 ml blood) to estimate various hematological parameters (total WBC count, PCV, RBCs, and hemoglobin) (15).

Parasitological examination:

Using disposable gloves, a single rectal fecal sample was taken from each animal with non-contamination from plants, bedding, or dirt. The animals were then labeled and transported to a lab for fecal analysis (16). The presence of parasite infection was then detected by examining fecal samples using the straightforward flotation approach as outlined by (17). Following collection, the samples were kept in an ice chest to preserve the material until laboratory processing. Flotation method in saturated sucrose solution was used to concentrate fecal samples (18). Using a disposable culture loop, the top surface film was moved onto a microscope slide and then covered with a glass slip. Under high power (magnification x 40), the entire covered area was inspected. Since the modified Ziehl-Neelsen technique is a specialist staining method, it was utilized for confirmation. To create a uniform and translucent layer, fresh feces and isotonic saline were combined and applied to the microscope slide. Slides were allowed to air dry before being fixed for three minutes in absolute methanol, stained for ten minutes in cold carbol fuchsin, and then decolorized for one minute in 3% hydrochloric acid. Then, for 30 seconds, 1% methylene blue was administered. Rinsed with running water and allowed to air dry. In order to identify parasite infection, the fecal smears were stained and then examined using an optical microscope, first at a magnification of 40 and subsequently at a magnification of 100 (19). *Giardia* spp. cysts and trophozoites were identified by either floating the sample in zinc sulfate, a solution that has been shown to be more effective in causing *Giardia* spp. cysts to float (20), or by looking at the deposit of a formol-ether concentrate of a fecal

preparation (21). To make the *Giardia* easier to see under a microscope, stain the sample with iodine of some kind.

Evaluation of Hematological Parameters

The BCC-3600-Duruy-South Korea automated hematology analyzer was used to measure leukocyte counts, erythrocyte counts, hemoglobin concentration (Hb), hematocrit (HCT/PCV), and leukocyte differential counts, among other hematological parameters.

Statistical analysis:

Version 17 of the SPSS computer program was used to analyze the findings (22).

RESULT AND DISCUSSION

Microscopic analyses Protozoa were the parasites that were discovered in the fecal samples of sheep, goats, and cattle during the study period. The overall prevalence of intestinal parasites was 28.66%, or almost 41%. In sheep, 27% of the goats and 18% of the cattle were infected, show as Table (1).

Table 1. Total percentage of intestinal parasite infection in farm animal (feces samples).

species	Number of examined	Number of infected	Percentage%
sheep	100	41	41 %
goat	100	27	27 %
cattle	100	18	18 %
Total	300	86	28.66 %

Total infestation rate of *Giardia* spp. according to sex In contrast, 14% of the 77 female goats under investigation had an infection. the 38 goat males analyzed, 13% had a *Giardia* spp. infection, whereas 5% of the 77 goat females under investigation had an infection. In contrast, 9% of the 73 female cattle under investigation had an infection. The infection rate for *Giardia* spp. infection was 11% in the 27 male cattle that were inspected and 4% in the 73 female cattle that were studied.

Table 2 displayed the proportion of farm animals infected with intestinal parasites based on the parasite species, *Cryptosporidium* spp. Infection rates in sheep were 32%, goats were 20%, and cattle were 12%. About 9% of sheep, 7% of goats, and 6% of cattle are infected with *Giardia* spp.

Table 2. Percentage of intestinal parasite infection in farm animal according the parasite species

parasite	Number of examined animal			Number of infected animal			Infection rate		
	sheep	goat	Cattle	sheep	goat	Cattle	sheep	goat	Cattle
<i>Cryptosporidium</i> spp.	100	100	100	32	20	12	32%	20%	12%
<i>Giardia</i> spp.				9	7	6	9%	7%	6%
Total	300			86			28.66 %		

According to the results, 39% of the 38 male sheep that were analyzed had a *Cryptosporidium* spp. infection. In contrast, 27% of the 62 female sheep under investigation had an infection. 39% of the 23 male goats analyzed had a *Cryptosporidium* spp. infection, and 10% of the 38 male sheep evaluated had a *Giardia* spp. infection, compared to 8% of the

62 female sheep under investigation. In contrast, 14% of the 77 female goats under investigation had an infection. The 38 goat males analyzed, 13% had a *Giardia* spp. infection, whereas 5% of the 77 goat females under investigation had an infection. In contrast, 18% of the 27 male cattle that were analyzed exhibited a *Cryptosporidium* spp. infection. In contrast, 9% of the 73 female cattle under investigation had an infection. The infection rate for *Giardia* spp. infection was 11% in the 27 male cattle that were inspected and 4% in the 73 female cattle that were studied

Table 3. percentage of intestinal parasite infection in farm animal according sex

sex	Number of examined animals			Number of infected animals					
	sheep	goat	Cattle	sheep		goat		Cattle	
				<i>Crypto.s</i> pp.	<i>Giardi</i> a spp.	<i>Crypto.s</i> pp.	<i>Giardi</i> a spp.	<i>Crypto.s</i> pp.	<i>Giardi</i> a spp.
Male	38	23	27	15 (39%)	4 (10%)	9 (39%)	3 (13%)	5 (18%)	3 (11%)
Female	62	77	73	17 (27%)	5 (8%)	11 (14%)	4 (5%)	7 (9%)	3 (4%)

Nonsignificant values (P<0.05)

Table 4 revealed that 23 % of the 43 sheep under a year old that were analyzed had a *Cryptosporidium* spp. infection. Of the 57 sheep older than a year that were examined, the incidence of infection was 38%. The infection rate for *Giardia* spp. was 6% of the 43 sheep that were inspected within a year. However, among the 57 sheep older than years that were examined, the frequency of infection was 11%. 15% of the 46 goats under a year old that were analyzed had a *Cryptosporidium* spp. infection. However, among the 54 goats older than one year that were examined, the frequency of infection was 24%. Four out of forty-six goats under a year old that were evaluated had a *Giardia* spp. infection. However, among the 54 goats older than one year that were examined, the frequency of infection was 9%. However, 8% of the 48 cattle who were inspected within a year had a *Cryptosporidium* spp. infection. In contrast, 15% of the 52 cattle who were examined for more than a year had an infection. Four percent of the 48 cattle that were inspected within a year acquired an infection from *Giardia* spp.. In contrast, 7% of the 52 cattle older than one year that were examined had an infection.

Table 4. Percentage of intestinal parasite infection in farm animal according age

Age	Number of examined animal			Number of infected animal					
	sheep	goat	Cattle	sheep		goat		Cattle	
				<i>Crypto.s</i> spp.	<i>Giardia</i> spp.	<i>Crypto.s</i> spp.	<i>Giardia</i> spp.	<i>Crypto.s</i> spp.	<i>Giardia</i> spp.
<1 year	43	46	48	10 (23%)	3 (6%)	7 (15%)	2 (4%)	4 (8%)	2 (4%)
>1 year	57	54	52	22 (38%)	6 (11%)	13 (24%)	5 (9%)	8 (15%)	4 (7%)

significant values (P<0.05)

The total leucocyte count significantly increased in infested animals compared to healthy normal ones, while Hb, total erythrocyte count, and PCV values significantly decreased in infested animals.

Nine parameters were substantially different between infected and uninfected sheep (at p < 0.05) when blood parameters and parasite infections were evaluated using independent sample T-tests. Were much lower in another parameters and considerably higher in WBCs, Eosinophils, and Lymphocytes in infected sheep as compared to uninfected ones (Table 5).

Table 5. Some hematological parameters in healthy and infested sheep with Intestinal parasites.

Parameter	Normal animal	infested animal
	Mean ± SEM	Mean ± SEM
Basophils (%)	0.33±5.88	0.29±0.06
Eosinophil's %	2.44±0.06	5.03 ± 0.03
Hb (g/dL)	10.22±0.44	7.30±0.20
Lymphocytes %	67.55 ± 0.80	71.14 ±2.30
Monocytes (%)	3.77±4.11	2.10±3.60
Neutrophils %	24.33± 0.20	23.04±1.30
PCV (%)	34.50±0.80	24.03±1.80
RBC 106 /µl	9.50±0.45	4.90±0.33
WBCs (x103/cumm)	8.34±0.11	9.22±0.16

significant values (P<0.05)

Using independent sample T-tests to evaluate blood parameters and parasite infections, it was found that nine parameters differed substantially between infected and uninfected goat (at p < 0.05). WBCs, eosinophils, and lymphocytes were considerably higher in infected goat than in uninfected ones, but another parameters were lower (Table 6).

Table 6. Some hematological parameters in healthy and infested goat with Intestinal parasites.

Parameter	Normal animal	infested animal
	Mean ± SEM	Mean ± SEM
Basophil	0.67 ± 0.12	0.62 ± 0.11
Eosinophils	1.23 ± 0.22	2.23 ± 0.24
Hb (g%)	10.77±0.40	8.66±0.21
Lymphocytes %	1.34 ± 0.30	2.29 ± 0.21
Monocyte	2.12 ± 0.20	1.06 ± 0.11
Neutrophils	47.60 ± 0.26	44.74 ± 0.28
PCV (%)	37.33±1.03	31.04±0.50
RBCs (x106/cumm)	9.55±0.33	7.88±0.22
WBCs (x103/cumm)	8.33±0.13	9.27±0.13

significant values (P<0.05)

The use of independent sample T-tests to evaluate parasite infections and blood parameters revealed that nine parameters differed substantially between infected and uninfected cattle (at p < 0.05). Showed a substantial drop in another parameters and

a significant rise in WBCs, Eosinophils, and lymphocytes in infected cattle as compared to uninfected ones (Table 7).

Table 7. Some hematological parameters in healthy and infested cattle with Intestinal parasites

Parameter	Normal animal Mean ± SEM	Infected Mean ± SEM
Basophil	0.66 ± 0.18	0.62 ± 0.11
Eosinophils	3.44 ±0.09	6.01 ± 0.04
HGB g/dL	12.70 ±0.15	10.54 ±0.13
Lymphocytes%	55.34 ±2.55	57.60 ±1.89
Monocytes%	11.58 ±2.42	8.20 ±1.50
Neutrophils	44.56 ± 0.23	43.11 ± 0.13
PVC fL	9.55 ±0.27	7.31 ±0.54
RBC 10e3/ul	8.13 ±0.12	6.06 ±0.11
WBC 10e3/ul	14.7 ± 1.34	16.15 ±.99
significant values (P<0.05)		

The ovoid oocysts of *Cryptosporidium* spp. (Figure 1), which were 4-6 µm in diameter and tinted red to pink with a granular appearance on a blue background, were encircled by a halo (*Cryptosporidium* spp. parvum) when examined under a microscope. Giardia cyst was discovered by microscopically examining the iodine-stained smears (Figure 2). is an oval cyst with dimensions of 7–11 µm in length and 7-9 µm in width. Its outer wall is 0.3–µm thick. Two or four nuclei, flagella, ribosomes, and pieces of the ventral disk (*Giardia duodenalis* cysts) are found inside. *Giardia duodenalis* trophozoite multiply by longitudinal binary fission and remain in the lumen of the proximal small bowel, where they can either be free or attached to the mucosa by a ventral sucking disk (Figure 3).

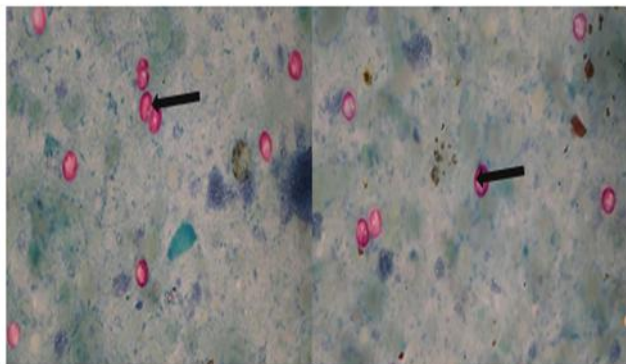


Figure 1. Under a microscope, the oocyst of *cryptosporidium* spp. (40 X).

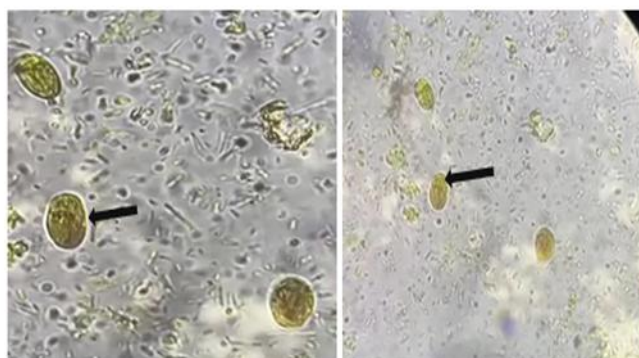


Figure 2. Under a microscope, the *Giardia duodenalis* cysts.(40 X).

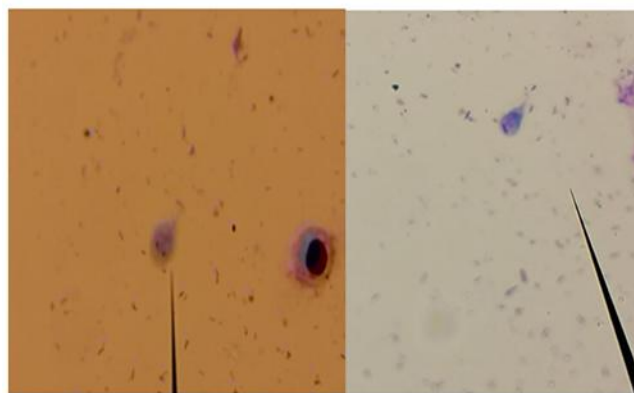


Figure 3. Under a microscope, the *Giardia duodenalis* trophozoite.(40 X)

The study revealed an overall prevalence of 28.66% for intestinal parasites, with sheep showing the highest susceptibility (41%). The clinical signs observed—*anemia*, *diarrhea*, and *emaciation*—are classic manifestations of protozoan malabsorption. Both *Cryptosporidium* spp. and *Giardia* spp. attach to the intestinal villi, causing villous atrophy and crypt hyperplasia. This reduces the surface area available for nutrient absorption, leading to the "poor growth" and "emaciation" The majority of the animals with the infestation had *anemia*, *diarrhea*, *emaciation*, *pale mucous membranes*, and *poor growth and reproductive function*, according to a clinical examination. that agree with (23). According (24), the most significant pathogen responsible for neonatal calf diarrhea is thought to be *Cryptosporidium* spp.. According to (25), hematological analysis has been found to be a reliable indicator for evaluating the health state of animals and may provide an assessment of the severity of infection as well as the extent of host tissue damage.

Cryptosporidium spp. showed higher infection rates than *Giardia* spp. across all species. *Cryptosporidium* spp. oocysts are highly resistant to environmental stressors and are immediately infective when passed in feces. This "autoinfection" cycle and high environmental persistence often lead to higher prevalence in communal farming environments compared to *Giardia* spp (26).

Interestingly, the data showed higher infection rates in animals older than one year (e.g., 38% in older sheep vs. 23% in younger sheep).

While neonates are physiologically more vulnerable, older animals in a farm setting have had a longer duration of exposure to contaminated pastures and water sources. These older animals often act as asymptomatic "carriers" or reservoirs, shedding oocysts into the environment, which maintains the cycle of infection within the herd (27).

The significant changes in blood parameters provide a clear picture of the host's systemic response to infection:

Leukocytosis, Eosinophilia and Lymphocytosis: The increase in WBCs and Lymphocytes signifies an active immune response as the body attempts to clear the protozoa.

Eosinophilia is a hallmark of parasitic infections; eosinophils release cytotoxic granules (like major basic protein) specifically designed to combat parasitic organisms (28).

The significant drop in these values indicates microcytic or normocytic anemia.

This is likely due to "Anemia of Chronic Disease" and nutrient malabsorption (especially Vitamin B12 and Iron). The intestinal damage caused by these parasites prevents the absorption of the essential building blocks required for erythropoiesis (red blood cell production) (29).

The identification of *Cryptosporidium* spp. oocysts (4-6 µm) and *Giardia* spp. cysts (7-11 µm) confirms the etiologic agents.

The use of Modified Acid-Fast staining (resulting in the red/pink hue) is essential for *Cryptosporidium* spp. because their small size and transparency make them nearly invisible under standard saline mounts. The presence of the ventral sucking disk in *Giardia* spp. trophozoites explains the mechanical damage to the intestinal mucosa, as this disk physically prevents nutrient uptake by the host (30).

The significantly higher prevalence observed in sheep compared to goats and cattle could be attributed to several epidemiological factors. Sheep generally graze closer to the ground, which increases their likelihood of ingesting infective parasitic oocysts or cysts from contaminated pastures. Additionally, flocking behaviors, overstocking, or differences in host immune responses among these ruminant species might contribute to this variation (31).

The study shows a clear dominance of *Cryptosporidium* spp. over *Giardia* spp. across all farm animals. For both parasites, the infection rates followed an identical trend: Sheep > Goats > Cattle. Sheep exhibited the highest vulnerability (32% for *Cryptosporidium* spp. and 9% for *Giardia* spp.), indicating they serve as the primary environmental reservoir for these protozoa in the studied region (32).

While *Cryptosporidium* is traditionally known to cause acute infections in newborns, the higher prevalence in older animals here indicates chronic or subclinical persistence. Older animals act as asymptomatic carriers, continually shedding oocysts into the environment due to prolonged exposure and accumulation over time on contaminated pastures. The higher rate of *Giardia* in young goats and cattle aligns with standard veterinary findings, as younger animals lack fully developed immune systems, making them highly susceptible to colonization. The

inverse trend in sheep might be linked to specific management practices or persistent reinfection in adult flocks (33).

CONCLUSION

The study demonstrates that protozoan parasites significantly compromise the health and hematological status of livestock, with males and older animals showing unexpected prominence in infection rates, necessitating improved environmental management and targeted veterinary intervention.

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