



## Identification of Internal Parasites in Sheep and Goats in Garmiyan Province/Kurdistan Region/Iraq

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### Abstract

A study was conducted to identify different kinds of internal parasites that are distributed in ruminants in Garmiyan province, southeast Kurdistan region/ Iraq. Fecal samples from 460 animals (360 sheep and 100 goats) were examined from April to December, 2015 that included applying different parasitological techniques. Mini-flotac flotation method was used for the detection of nematode and cestode helminths; the sedimentation technique used for trematodes whereas for recovering lung worms and infective larvae Baermann technique was applied and culturing of fecal samples was implemented for the differentiation of free living stages. The study revealed that overall 46.52% of tested animals had been infected with several gastrointestinal helminths that were distributed between 48.05% in sheep and 41% in goats respectively. *Marshallagia marshalli* was by far the most prevalent parasite in the area with about 74% of positive samples followed by Nematodes like eggs including (*Trichostrongylus* spp., *Teledorsagia circumcincta*, *Haemonchus contortus* and *Cooperia* spp.) then lung worms. The majority of samples were mixed infection by nearly 76% and the rest were single infection. According to the age category, adult animals recorded more rates of infection (49.09%) than young (39.84%), while in regards to the sex factor; females were relatively more than males (47.13% compared to 42.37%). The study suggested that susceptibility of animals, environmental conditions in addition to managing trials implemented like husbandry and density of stocking may play a role in increasing contamination on pasture and increasing abundance of parasites. Other aspects like testing anthelmintic resistance and existing control measures used by people in this area should be taken into account when research is undertaken in the future.

### Introduction

Animals are mainly infected with different kinds of helminths whenever they are fed on green pasture and drunk from river banks where intermediate hosts and infective stages exist [1]. Helminths, especially gastrointestinal worms, represent a major effect on animal production; they cause huge economic losses and influence the income of farming communities [2] and [3], and can cause variety of health disorders like anemia, weight loss, and protein loss [4] and [5]. Susceptibility to parasitic diseases appears in some species of animals more than others, for example infection in sheep is more than goats and cattle, particularly in ages between 2 months to 2 years [6], [1] and [7]. The most important genera among gastrointestinal parasites that affect ruminants and *Haemonchus*, *Teledorsagia* (*Ostertagia*), *Trichostrongylus*, *Cooperia*, *Nematodirus* [6] and *Marshallagia* [8]. Ability of parasites to produce severe infections and diseases is usually related to combinations of the different species [9].

One of the difficulties that influence the measures of prevention and control of parasites is the generations of overuse or incorrect usage of anthelmintic de-wormers which lead to raise resistance of these organisms against common anthelmintic trials [7]. Surveillance on gastrointestinal helminths in a given study area regarding associated risk factors can help in implementing an effective control regime of several kinds of helminths [3]. Risk factors like age, weather condition and management practices can influence the prevalence of gastrointestinal worms and their control processes [10] and [3]. The presence of right conditions like humidity and warmth can act on the severity of infections, abundance of worm eggs on pasture, hatching of eggs and the survival of infective stages L3 [10] and [7]. Research conducted on parasites around the world suggested possible limitation measures to reduce the prevalence of these parasites depending on anthelmintic usage and some immunological and protecting systems [1].

In Iraq and Kurdistan region, there is a large population of sheep and goats that can be affected by a wide range of gastrointestinal parasites (4) and [1]. Garmiyani province is a semi-governmental area in the south-eastern part of Kurdistan region which represents a large geographic zone and contains different kinds of livestock like sheep, goats, cattle, buffalos, horses and poultry. People in this area mainly depend on grazing of these kinds of animals as the main source of income for their life and any disorders belonging to these animals directly affect them ending with serious economic damage. Parasitic diseases infect most of the animal flocks and herds in Garmiyani province; where appropriate studies have not been carried out on the detection and examination of parasites especially gastrointestinal helminths. Therefore, this study aims to perform a general survey to identify different kinds of internal parasites in two most populated domestic animals in this region, sheep & goats, in order to examine the size of infection and put forward suitable control strategies to be implemented against them.

## **Materials and Methods**

### ***A. Study Design***

The study targeted sheep and goat flocks distributed in different parts of Garmiyani province starting from Kalar to Kifri towns and including eight surrounding sub-districts. 460 fecal samples were randomly collected from both male and female animals at different ages. Samples collected from 84 flocks (8-10% of each flock selected) located in 35 villages in the region from April to December, 2015. An excel sheet was used for recording data about every particular aspect within the study including samples addresses, date of sample collection, village name, owner's name, type of animal, sex of animal, age, case history, clinical signs and results. Fecal samples for parasitological examination were collected from the rectum of animals using disposable gloves and transferred in plastic containers in cool boxes to the laboratory for diagnostic examinations.

### ***B. Parasitological Techniques***

Helminths egg (ova) counting is useful from the pathological, epidemiological and clinical aspects for the diagnosis of several parasitological infections and should be established for climatic areas as the pathogenicity of parasite populations may differ between them [11] and [12]. Fecal egg count is useful to determine the level of contamination of pasture with parasites, but can be varied between flocks and from one animal to another [7]. For detection of nematodes and cestodes eggs, the method used in the study was Mini-flotac floatation which is a new, more sensitive, simple and less time consuming method. It has two parts, the mini-flotac apparatus which is used for interpretation via using a reading disc of two 1ml chambers to be multiplied by 5 (EPG = eggs in both chambers multiply by 5) and the fill-flotac apparatus for mixing, homogenizing and centering samples with the floatation solution (sodium chloride). This method has been used recently in the detection of fecal egg counts in both human and animal samples and measured as an alternative to other techniques like McMaster, Kato-Katz, and other fecal count measures [13] and [18]. In addition, it is useful in the identification of light or lower helminth infections when present with a higher sensitivity of 5 e.p.g compared to 50 e.p.g for McMaster [13].

For trematode eggs, the sedimentation method was used to examine heavy eggs through three aperture sieves (there diameters range from: 710µm, 150µ m and 53µ m). The Baermann technique was applied for the recovery of lung worms and isolation of larvae of different parasites after culturing infected fecal samples as the morphology of most nematode eggs are very similar and cannot be clearly identified via microscopic detection and it is necessary to cultivate fecal samples to reach the free living infective stage larvae to differentiate between genera [14 ]and [11].

### C. Analysis of Data

Data collected from the study about different categories were added to an excel sheet, then data tools used for analysis of the number of eggs found, percentiles of different kinds of parasites in comparison to eggs established and morphology of larvae recovered, prevalence and rates of infection according to seasons and environmental conditions.

### Results:

Results of this study from several parasitological tests showed that animals in Garmiyan province were infected by different kinds of parasites. According to the number of eggs found in fecal samples from (460) sheep and goats, the proportion of positive samples recorded was 46.52% (214 samples) whereas the rest were negative with about 53.48% (246 samples). In the study, fecal samples from 360 sheep and 100 goats were examined, percentage of positive samples in sheep in relation with the total sheep samples recorded 48.05% (no=173) despite the fact that it was lower in goat in comparison to sheep with about 41% (no=41) as shown in (Table 1).

Table-1: Number and percentages of positive samples in sheep and goats in the study

<u>Kind of Animal</u>	<u>No of Positive Samples</u>	<u>No. of Negative Samples</u>
Sheep	360 173 (48.05%)	187 (51.95%)
Goats	100 41 (41%)	59 (59%)
Total	460 214 (46.52%)	246(53.48%)

Infected animals examined in the survey demonstrated several kinds of clinical disorders and signs; however some animals were recorded positive despite their healthy status. Clinical signs recorded were emaciation, weight loss, diarrhea (watery, bloody or profuse), loss of appetite, nasal discharge and coughing, anemia and submandibular edema.

*Marshallagia marshalli* recorded the highest number between all helminths found which was repeated in (160) from total (214) positive samples with about 74.76%. Nematodes like eggs including (*Trichostrongylus* spp., *Teledorsagia circumcincta*, *Haemonchus contortus* and *Cooperia* spp.) altogether found in 131(61.21%) positive samples followed by lung worms which were repeated in 69(32.24%) samples. Percentage of infection with *Eimeria* spp. was (31.30%) which were found in 67 cases, while about 43 (20.09%) of samples were infected with the tape worm *Moniezia* spp. (*Moniezia expansa* and *M.benedeni*). The nematode *Chabertia ovina* was found only in (9) samples with a proportion of 4.20% of infected animals while the other nematode *Oesophagostomum* spp. was repeated in (7) of those infected animals representing about 3.27%. Both *Bunostomum trigonocephalum* and the trematode *Paramphistomum cervi* recorded the same results with 4(1.86%). Less animals' cases were found with *Gongylonema* spp. and *Strongyloides papillosus* in 3 samples only which were (1.40%) for both whereas the lowest number recorded was for *Toxocara vitulorum* with about (0.46%) (Table: 2)

Table- 2: Types of ova and larvae of internal parasites

<u>Types of Parasites</u>	<u>No. of Infected Animals</u>	<u>Percentages</u>
<i>Marshallagia marshalli</i>	160	74.76%
Nematodes Like eggs	131	61.21%
Lung worms	69	32.24%
<i>Eimeria</i> spp.	67	31.30%
<i>Moniezia</i> spp.	43	20.09%
<i>Nematodirus</i> spp.	40	18.68%
<i>Chabertia ovina</i>	9	4.20%

<i>Oesophagastomum</i> spp.	7	3.27%
<i>Bunostomum trigonocephalum</i>	4	1.86%
<i>Paramohistomum cervi</i>	4	1.86%
<i>Gongylonema</i> spp.	3	1.40%
<i>Strongyloides papillosus</i>	3	1.40%
<i>Toxocara vitulorum</i>	1	0.46%

Positive samples were recorded during the whole period of the study but were different from one month to another, while about 75% of samples collected in April and November were positive, the records in August was only 9% and the rates of infection fluctuated in the other months (Figure: 1). Mixed infection of nematodes like eggs approached 1700 e.p.g in April then started to decrease in summer months until the late of Autumn when they raised again in numbers. Similarly, *Marshallagia marshalli* showed an increase in number of eggs per gram in April (about 125 e.p.g) then started to decline steeply in summer and then increased from October to reach nearly (530 e.p.g) in November. Images of some eggs found can be seen in (Figure: 2).

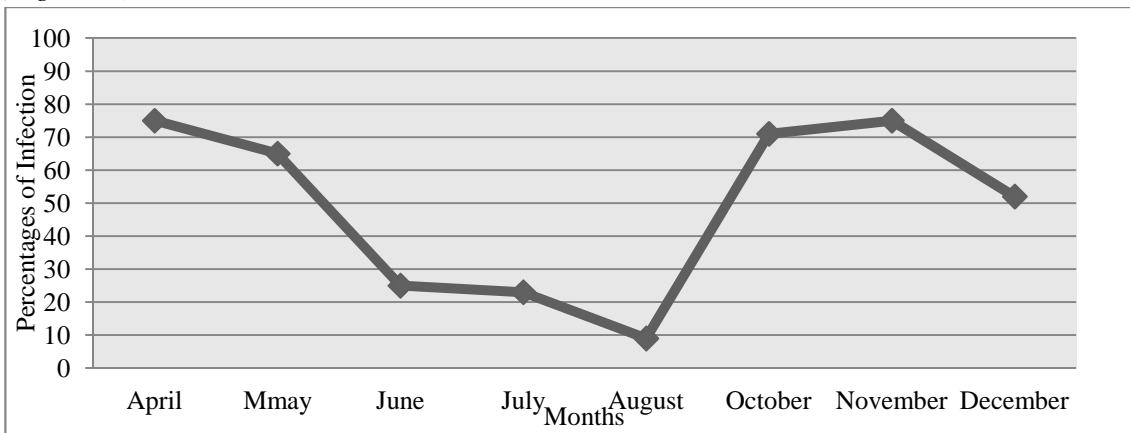


Figure-1: Percentage of infected samples throughout the time period of the study

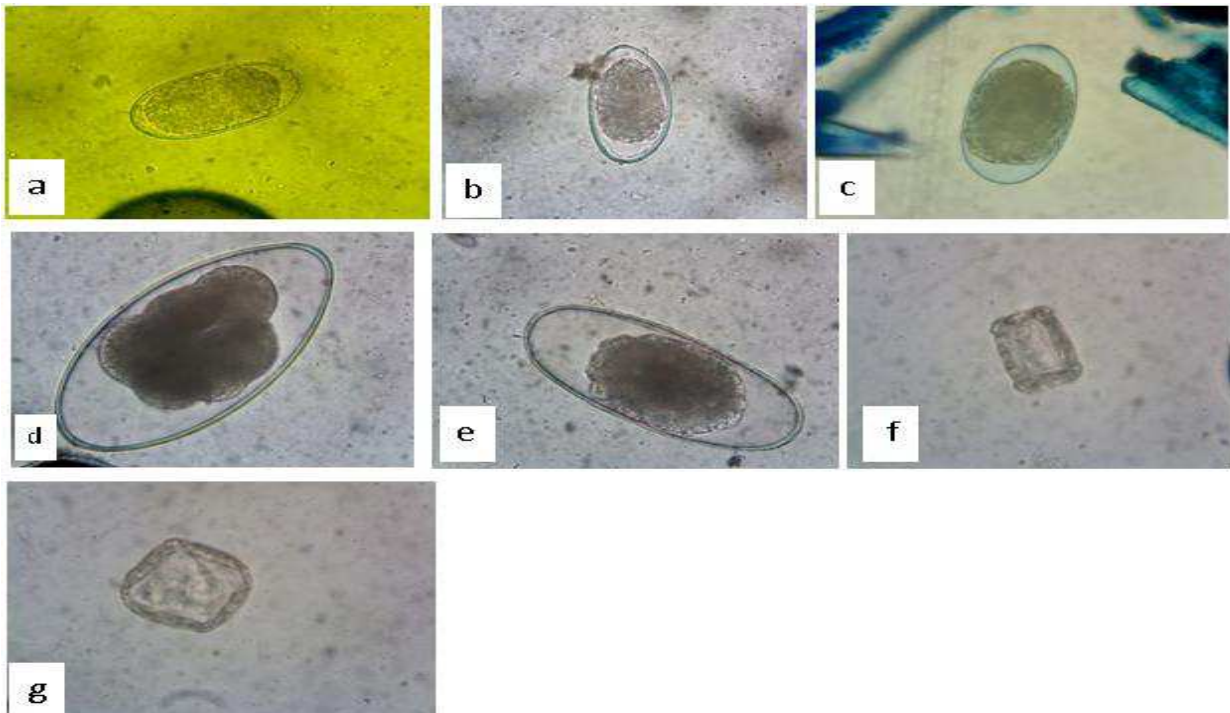


Figure-2: Images of some eggs (ova) found better showing; (a), (b) and (c) Nematodes like eggs (40X), (d) *Nemaodirus* spp. egg (40X), (e) *Marshallagia marshalli* egg (40X), (f) *Moniezia benedeni* egg (100X), (g) *Moniezia expansa* egg (100X).

Animals in this study were divided into two age categories: young animals lower than 2years (<2years) and adults more than (>2years), the minority of samples 128 of total 460 samples were young animals and the rest of samples 332 were from adult animals. The percentage of infection in adults was more than that of young animals as it was nearly 49.09% in adults and 39.84% in young animals respectively. Throughout the study, number and rates of mixed infections by two or more parasites were higher than single infections. Only (51) samples from the whole positive samples were single infections about 23.83% and the rest of cases (163) samples and 76.17% were mixed infections. Results of infection in animals according to sex category were relatively more in females in comparison to males, out of 59 male examined 25 samples were tested positive (42.37%) and of 401 female tested 189 samples were positive(47.13%) (Table: 3).

Table-3:Prevalence of small ruminant helminthiasis in sex category in selected study area

<u>Sex of Animal</u>	<u>No of samples</u>	<u>Positive samples</u>
Male	59(12.9%)	25 (42.37%)
Female	401(87.1%)	189(47.13%)
Total	460	214 (46.25%)

## Discussion

The study on several numbers of faecal samples from different parts of Garmiyan province showed infection of sheep and goats with a wide range of gastro-intestinal helminths (GIH). These results are similar to other research like [1] but differ from [4] which recorded only 3 species. Environmental and seasonal conditions, management and husbandry systems and number of samples collected can be the most important factors behind the variance in results. In this study, *Mrashallagia marshalli* was by far the most circulated and recorded species between other parasites, this result is similar to other studies like [1] in Mosul/Iraq, and [15]in Iran which refers to this worm as the major cause of parasitic infection in ruminant among several species of ostertagiinae in Iran. *Marshallagia* species are typical abomasal parasites that spread around the world which reported different prevalence rates from 0.72% to 84% in domestic animals [15].

The rates of infection in this survey were relatively more in sheep (48%) than goats (41%) which agree with a study conducted by [2] and disagree with some researchers that detected higher rates of GI helminths in goats compared to sheep [3]. These results support the idea that parasites might cause severe diseases in some species of animals more than others as the susceptibility to these diseases are not the same between ruminants [6] and [7]. Correspondingly, some breeds of animals especially sheep are more susceptible to some kinds of parasites while others demonstrate some resistance to these worms [14].

Result in adults was nearly 49% and was higher than those in young animals with nearly 40%, these records were similar to a study conducted by [16] on gastrointestinal helminths, but differs from other researches done by [2] and [3] which reported more infection in young animals than adults. Presumably, even though adults show immunity relatively more than young to parasites, but still at risk when moved into an endemic area [17]. In this study there were more rates of infection in females (47.13%) compared to (42.43%) in males which are nearly the same of that found by [3] as the prevalence in females was (48.8%) and (42.42%) in males respectively. These records support the phenomenon that said females are more susceptible than males due to the presence of some conditions in females like pregnant, pre-parturient period and stress which can affect immune status and increase rates of infection [2] and [3].

The prevalence of worms varied within the period of the study, for example in late spring the level of cases were higher than the summer months in which levels dropped gradually until the beginning of winter when rates of infection raised steeply. These differences may be interpreted as consequences of the diversity in host susceptibilities as well as the influence of climatic conditions as some helminths are found to be more susceptible to climatic conditions than others [10] and [3]. Climate change in some regions leads to the existence of periods of drought after a few days of rainfall which can affect parasitic transmission during

driest months and reversely leading to severe increase in larval emergence in the period of rain days [10]. This phenomenon might explain the continued presence of parasites particularly in small animals even in less rainfall or dry seasons as parasitic infections can be carried from season to another in the host [16] and [8].

In this study species of *Eimeria* were found in nearly 31% of infected samples (mainly in young animals >2years) which supported the fact that this protozoa is able to cause diseases in lambs and kids under intensive husbandry conditions [17] and [18] and some species has been considered as pathogenic and can cause morbidity and mortality in large numbers especially in lambs when exist in high population rates [16] and [17]. The most important GI nematodes (*Trichostrongylus* spp., *Teledorsagia circumcincta*, *Haemonchus contortus* and at less rates *Cooperia* spp.) were identified but could not separate in this study as they need more intensive tests like cultures, measurement of tail and type of oesophagus to differentiate between them surely, instead we used the term nematodes like eggs for all. Faecal egg counts of these nematodes were fluctuated from one period time to another, this can best describe to be driven by environmental and management conditions, for example, *Haemonchus contortus* is more susceptible to cold and desiccation while *Trichostrongylus* spp. and *Teledorsagia circumcincta* have lower susceptibility to desiccation [6]. Additionally, high number of egg per grams of these nematodes were recorded in October and November which are represent most favourable months for the development of some nematodes like *Teledorsagia* [10] and [17], this parasite also known to became established in huge numbers when they were present in mixed infections whereas *Haemonchus* and *Trichostrongylus* can be established in large numbers while exist in single infections [9].

Other kinds of gastrointestinal helminths like (*Chabertia ovina*, *Oesophagostomum* spp. *Bunostomum trigonocephalum*, *Strongyloides papillosus* and *Toxocara vitulorum*) were at low levels which are alike in the result from other studies [1]. The nematode *Gongylonema* spp. which can also affect human and almost all kinds of ruminants was found in this study while the trematode *Paramphistomum cervi* was the only trematode found. Trematodes has a very complex life cycle and required fresh water snail as an intermediate host as a part in their life cycle. Snails are highly affected by increased temperature and this can act on the abundance of trematodes followed by reducing their population [10].

Throughout the study clinical signs and case histories of every particular animal were established. Respiratory signs like coughing; nasal discharge and sometimes dyspnoea were recorded in the study which can be a result of the presence of lung worms which repeated in out of 69 (32.24%) cases of the whole positive samples. In some severe cases signs of dyspnoea and sometimes diarrhoea and anaemia may be observed especially in the existence of other trichostrongyloides helminths [17]. Another reason of the presence of respiratory signs could be the migration of species of *Toxocara vitulorum* and *Strongyloides papillosus* which can cause similar clinical signs while passing the lungs in order to complete their life cycle [1].

Applying sustainable control strategies like well-planned programs for the proper use of de-wormers, evaluation of the cost of parasitism and implementing appropriate management procedures can lead to reduce level of contamination on pasture, decrease of production losses and more significantly declining the possibility of anthelmintic resistance [5 ]and [18]. Management procedures like density of stocking and gathering of animals around feeding troughs is particularly important in nematodes and cestodes infections and can increase the level of contamination [17]. Relatively, several studies refer to anthelmintic resistance as one of the main issues that has a great effect on sheep industry [5] and [18]. Additionally, anthelmintic resistance is one of the major issues that affect the process of deworming which mainly produces by overuse and incorrect usage of anthelminthic that leads to raise resistance of parasites against common anthelmintic [7 ]and [19]. However, repeating the) dosing and using of anthelminthic can kill wide range of gastrointestinal helminths, but genetically resistant populations of parasites will lay eggs continuously and contaminate pasture. Drugs within the same class generally shared resistance by all kinds within that class, for example if one parasite resists fenbendazole, it also resists albendazole [19].

## Conclusion

Internal parasites play a great role in the production and economic losses to the animal sector, and the results from this study approved the high prevalence rates of different kinds of helminths in Garmiyan province. Presence of parasites with fluctuating levels of infections in domestic animals need a widespread range of treatment and prevention measures in addition to apply well developed managing systems which represent a continuous threat on animal industry. Implementing operative prevention measures and treatment programs should be based on seasonal activities of parasites. Two main recommendations we suggest are:

1. Conducting studies on drug resistance patterns of anthelmintic that are widely used by animal owners and veterinarians in this area.
2. Studying the influence of temperature, humidity and seasonality on the survival and abundance of gastrointestinal parasites and free living infective stages which can produce high rates of infection

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