Seasonal Prevalence and Pathological Changes of Ostertagiasis in Abomasum of Slaughtered Sheep in Khoy City in Iran

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ABSTRACT

Parasitic diseases are among the most important problems threatening animal’s lives. One of these diseases is Ostertagiasis caused by different species of Ostertagia in sheep. The main objective of this research is to investigate the prevalence of ovine Ostertagiasis and its pathological changes in Khoy city in Iran. Throughout this research, conducted within a year, it was specified that 39.4 % of slaughtered sheep were infected with Ostertagia. The rate of infection was higher in summer than the other seasons. The prevalent species were 92.64% and 7.36 % for Ostertagia circumcincta and Ostertagia ovisdalis respectively. The parasite caused anemia as a result of abomasal mucosa haemorrhagia and some white nodules generated in abomasal mucosa called morocco leather in severe infection macroscopically. Microscopic findings were reported as: infiltration of plasma cells, monocytes and eosinophils among gastric mucosal glands and granulomatous inflammation with necrotic foci. Furthermore, necrosis, destruction of gastric glands and abomasal congestion, mucosal glands hyperplasia and their cystic formation were observed. In mild infection only local congestion or hemorrhage in abomasal mucosa were seen. Results of this study indicated that the prevalence of ostertagiasis in slaughtered sheep in Khoy city, Iran, had significant difference (P<0.05) with season in contrast with sex, age and amount of pH (P> 0.05).

INTRODUCTION

Nematodes of the gastro-intestinal tract are known as a major factor in reduction of productivity in small ruminants (whether small scale or large amounts), causing vital economic losses [1]. Based on researches conducted in recent years [2, 3], Parasitic diseases of the digestive system are very deleterious in livestock [2, 3, 4]. Gastrointestinal nematodes are of high importance due to their insidious and severe pathological effects [5]. The development of control strategies aiming at improved managerial practices can be achieved through a better understanding of the characteristics of the bionomics of the free-living stages of these parasites [6]. Rainfall can be considered to be the main climatic factor for determining the availability of infective strongylid larvae and the transmission of infection in grazing animals [7]. Ostertagia Ostertagi is, in fact, a roundworm in ruminants called stomach worm originally. O. circumcincta is also another species of the genre Ostertagia, which is located in the abomasum of sheep and goats and has been adapted very well to life in this member [8]. The evolution of Ostertagia is direct and the infection happens by swallowing the third stage larvae (L3). Meanwhile, hypobiosis can be observed in the Ostertagia evolution. After ingestion, L3 enter parietal glands of abomasum and larvae stages develop in these glands. The development of larvae stages associate with local cellular reduction resulting in decrease of gastric acid secretion and increase of gastrin and plasma pepsinogen secretion. The abomasum will become alkaline and hypoplasia of abomasum epithelial will occur [9, 10]. The fifth stage larvae (L5) detach from parietal glands and become mature in mucosal surface of abomasum. Egression of L5 associated with release of waste- secretory products, which cause pathophysilogic changes in abomasum. These secretions have cytotoxic and inhibitory effects on parietal cells causing parietal cellular reduction. Abomasitis and destruction of immune system are other impacts of these secretions [9, 11]. O. ovisdalis, O. circumcincta and O. trifurcata are important species of Ostertagia in sheep all over the world. The treatment of this parasite, like...
major worms of the small intestine, is performed using anthelmintic drugs such as Fenbendazole, Levamizole and Eivermectin. On the other hand, parasites and their role in causing financial losses to the ranchers through infection of livestock, reducing productions or increasing the mortality should not be overlooked. Since, Pedigree took place in the traditional way, in most parts of Iran, sheep feeding on pasture leads animals to be infected with parasites [12]. Sheep and goats play a vital role in the rural economy by preparing the meat, milk, financial income, capital accumulation and manure. Moreover, these animals have a role in the national economy through getting exported in form of live animals, meat and skin [1]. Currently, one of the issues of control and treatment of gastrointestinal nematodes is the tolerance of these parasites against common anthelmintic drugs [13, 14]. Due to epidemiological features and high pathogenicity of Ostertagia and also economic importance of damages caused by this parasite in sheep the present paper has focused on ostertagiasis. Awareness of prevalence rate and predominant species of Ostertagia in the region is essential for control and treatment of Ostertagiasis. The findings of the present study would be useful in this manner. The location of this study was Khoy city in Iran. Khoy (38°33'01"N 44°57'08"E) is located in the northwest part of Iran. Although mainly influenced by moist air currents of the Atlantic and Mediterranean, in some winter months, cold air masses would be pulled from the north to this zone, which has a high impact on the reduction of temperature in this area. Totally, the average annual rainfall in Khoy is 400-300 mm. The highest seasonal rainfall is in spring. Rainfall period in Khoy starts from late December and ends in early May and early June. After that, the rate of rainfall will be decreased slowly. The most rainfall is in April and early May. Therefore, with investigation of the lesions in slaughtered sheep, the prevalence rate, kind and severity of lesions caused Ostertagia were examined.

![Geographical position of study](image)

**Fig. 1:** Geographical position of study has specified with a curved arrow.

**MATERIALS AND METHODS**

This study has been conducted on sheep of the abattoir in Khoy city. Since June 2011 to May 2012, 2430 abomasum were inspected to assess the prevalence of lesions caused by Ostertagia. Upon receipt of the abomasum, age and sex of sheep should be identified in order to find a relationship between these parameters with prevalence and lesions of Ostertagia. Both ends of each abomasum, after removing them from abdominal cavity were ligated. At first the abomasum opened longitudinally for macroscopic observations. The presence of parasites, Haemonchus and Ostertagia, was associated with symptoms such as hyperemia, hemorrhage and petechiae in the abomasum. In parasitology laboratory after depletion, washing and sifting of abomasum’s contents ostertagias in filtrated emulsion were separated. Then the parasites were put in 70% ethanol directly for one hour for fixation. After clearing of Ostertagia on glass slide with lactophenol solution, mounting with glycerin gel was performed. For identification of Ostertagia lens 10X and for differentiation of ostertagia’s species (on the basis of spicule’s shape) lens 40X were used [15]. Affected tissue samples were fixed in 10% buffered formalin. Then the pathologic sections were prepared on the basis of standard procedures including: dehydration, clearing, embedding and blocking respectively. Then sections were cut in 5 μm of thickness by rotary microtome (YL3-A). Finally the sections were stained by Haematoxylin and Eosin staining method [16]. Parasitological identification and Histopathological changes were evaluated by microscope (Olympus CX21). For statistical analysis of the data the software SPSS (VERSION 21) was used to determine the significant differences of prevalence of the parasite and the probable variables through ANOVA. Furthermore, to perform pairwise comparisons between group means, the Bonferroni test and paired sample t test were employed. In addition, Levene-test was performed to check the homogeneity of data.
RESULTS AND DISCUSSION

Focusing on the parasite Ostertagia, the present study came across another parasite Haemonchus. The total number and worm burden of each parasite (Ostertagia and Haemonchus) were measured. However, due to the important effects and changes of Ostertagia on abomasum’s tissue and lesser importance of lesions of Haemonchus, only species of Ostertagia were investigated. Out of 2430 abomasal samples inspected during a year, 957 cases were found to be infected with Ostertagia. In other words, 39.38% of whole inspected abomasum, were infested with Ostertagia. The highest rate of contamination was reported in July 2011, with 52.5% and the lowest rate was reported in June 2012, with 9.2%.

A number of 9834 Ostertagia circumcincta were collected from the infected abomasums. Throughout this study, only 781 of Ostertagia ovisconturtos were recorded. Therefore, a total of 10615 parasite were observed (92.64% O.circumcincta, 7.36% O.oxidentalis). In the current study, the average number of Ostertagia in each abomasum was 11, with the minimum and maximum number of 2 to 67 nematodes. In addition, the average worm burden of Ostertagia in sheep was highest in July 2011 with 20.2 worms per animal.

In the sheep with both parasite infection (Ostertagia and Haemonchus), macroscopic and microscopic lesions, in most cases, were originated from Ostertagia. About lesions generated in the infested abomasum with Ostertagia, macroscopically, multiple swelled nodules were observed on the mucosal surface. In addition, mild to severe mucosal hyperemia was observed. Macroscopically, in the abomasum with mild infections, hyperemia in blood vessels and rarely, mucous layer hyperplasia were observed. But in severe infections, lesions such as mucosal gland hyperplasia, edema, destruction of mucosal glands, an increase in the number of inflammatory cells and cystic formation of abomasal glands were identified. Moreover, granulomas due to the presence of parasites in the abomasum were also identified, which were symptom of chronic lesions. These granulomas had a necrotized center, and increased connective tissue in surrounding areas. In some pathological sections, infiltration of eosinophils, lymphocytes and plasma cells were also observed. In cases with mild ostertagiosis, intensity of the lesions were less compared with severe ostertagiosis. Signs such as congestion or hemorrhage were observed in mucosal surface of abomasum locally.

Table 1: Infection rate of Ostertagia in abomasum of sheep related to some variables.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Category</th>
<th>No. of examined</th>
<th>No. of infected</th>
<th>Infection mean (%)</th>
<th>SD</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>Male Female</td>
<td>1192 1238</td>
<td>477 479</td>
<td>40.01 38.69</td>
<td>0.49</td>
<td>0.48</td>
</tr>
<tr>
<td>Age</td>
<td>&lt;1≥1</td>
<td>1199 1231</td>
<td>460 497</td>
<td>38.36 40.37</td>
<td>0.48</td>
<td>0.49</td>
</tr>
<tr>
<td>Season</td>
<td>Spring Summer</td>
<td>848</td>
<td>250</td>
<td>30.18</td>
<td>0.35</td>
<td>0.038</td>
</tr>
<tr>
<td></td>
<td>Autumn Winter</td>
<td>636 553 393</td>
<td>332 286 86</td>
<td>52.19 51.71 21.88</td>
<td>0.23</td>
<td>0.24</td>
</tr>
<tr>
<td>AbomasalpH</td>
<td>5.2 ± 0.2</td>
<td>2430 957</td>
<td>41.3</td>
<td>0.49</td>
<td>0.146</td>
<td></td>
</tr>
</tbody>
</table>

SD= Standard Deviation

The present study revealed the fact that from the whole inspected abomasum, only 39.4% of them were infected with parasites of the genre Ostertagia. These values compared with the values obtained about prevalence of Ostertagia species in Jahangirabad city in Pakistan is very low and insignificant [17]. In contrast, in researches conducted in Nigeria in sheep [18] and in Pakistan in goats [19], incidence of Ostertagia was very low [18, 19]. However, based on the investigation performed in Jammu and Kashmir state in India no species of Ostertagia was found [20]. The differences in the prevalence of Ostertagia may be due to differences in environmental factors, differences in the season of investigations or differences in the animal resistance of various regions. On the other hand, in the present study, only two species of the genre Ostertagia including O. circumcincta and O.oxidentalis were found. In Studies performed in other countries of the world, other species
of this genre including *O. ovisintalis* and *O. trifurcata* have been found in sheep and goat [21, 22, 23, 24, 25]. In a research seasonal distribution of gastrointestinal helminthes was evaluated through coprological and post-mortem examination of small ruminant in Belgrade. The prevalence rate of infection to *Ostertagia circumcincta, Ostertagia trifurcata* and *Ostertagia ostertagi* was reported as 95.23%, 91.53% and 23.33% respectively. Also maximum prevalence of genus *Ostertagia* was recorded in March [26]. Also in other animals, in Finland, prevalence of *Ostertagia* was reported 98% in adult reindeer [27]. In a research on reindeer conducted in Spain, some species of the *Ostertagia* genre such as *O. leptospicularis* and *O. kuljida* were reported [28]. In another research on gastrointestinal helminthes of camels in Iran, the prevalence of this parasite was 18% [29].

In the present study, prevalence rate of *O. circumcincta* was 92.64%, which is considered significant, while the prevalence of this parasite in a region in Pakistan was only 6.65% [30]. This difference may be partly due to differences in environmental factors such as geographical situation or climate zone, or factors such as the type of examined animal, animal resistance or pharmacotherapy. In the present study, the incidence of *Ostertagia ovisintalis* was only 7.36%. These rates were much lower than the percentage obtained in other studies [12, 21, 22, 25]. In the other research conducted in Turkey, the most common parasite in goats was *O. circumcincta* (78%) [21]. As mentioned earlier, in this study, the prevalence of *Haemonchus* was 9.3%. In studies of Kumsa, the prevalence of *Haemonchus in* sheep was 91.2% which is much higher than the one reported in the present study [1]. Differences in the prevalence of the parasite may be related to the localization of the parasite in a particular region or animal resistance to the parasite due to the frequent use of the anthelmintic drugs. In the present study, maximum seasonal infection with *Ostertagia* happened in summer (52.1%). In the spring, the eggs which expelled along with faces turned into infective larvae gently. Following the increase in the temperatures in mid and late May, larval development also increased. Eating forage infested with the larvae by livestock could be the main cause of infection in summer. However, the impact of factors such as excessive rainfall in April and early May followed by an increase in the moisture which is vital for development of the larvae should not be ignored. On the other hand, the lowest infection rate was in winter (21.8%). According to the research conducted in northern region of Nile Delta, Egypt, the highest rate of infection with GI nematodes in sheep, was in autumn and the least infection was in spring [24]. Also in another research on sheep and goats conducted in Syrpus, maximum seasonal infection with *Ostertagia* was in autumn and winter [31]. In Turkey and Saudi Arabia, the least infestation in sheep and goats was observed in winter [21, 23]. The environmental temperature, humidity and pasture conditions play a significant role in the development and survival of free-living stages of gastrointestinal nematodes of small ruminants [32]. Perhaps these differences are either related to the rainfall in different areas or may be pertained to resumption of the larvae growth. In the present study, the highest monthly infection with *Ostertagia* happened in June, 2011 (52.5%). This may be due to several factors such as high prevalence of infective larvae on pasture, low resistance of newborn lambs or even incompetence in managerial performance about intermittent use of the pastures. On the other hand, the least infection was in May 2012 (9.2%). It may be due to absence of parasite eggs in the pasture, or lack of suitable conditions such as humidity and temperature for the development of eggs into infective larvae in late spring, 2012. Besides, an apparent increase was seen in the prevalence of *Ostertagia* in March and April. It can be attributed to a phenomenon called ‘Hypobiosis’. In other words, the fourth-stage larvae that have stopped their growth in early fall have resumed their growth in early spring.

**Fig. 2:** Seasonal infection rates of sheep’s abomasum with *Ostertagia*.

In the present research, it was found that age and gender do not affect the prevalence rate of *Ostertagia* in the abomasum of sheep. Similar results in another study in Spanish sheep were achieved [33]. In a study conducted in Pakistan, there was no correlation between age and gender of cattle with incidence of GI nematodes such as *Ostertagia* [30]. Also results of the research revealed that the age, gender and even weight of sheep may have implicated the incidence of *Ostertagia*. These differences between results of present study and the results of research conducted by Tasawar [17] is due to differences in managerial functions in research, nutritional status of the animals or natural factors such as climate and geographical situation.
Fig. 3: Infection rates of abomasum with *Ostertagia* in sheep’s abomasum during months of one year.

![Graph showing infection rates of abomasum with Ostertagia](image)

Fig. 4: A: Macroscopic appearance of infected abomasum with sever Ostertagiasis. White nodules related to *Ostertagia* have been specified by pointers. B: Cystic formation of gastric glands (arrow) compared normal glands (arrow head) H&E, X100. C: Pathologic section shows Ostertagia’s section (arrow head) with infiltration of many leukocytes it’s around. H&E, X400. D: Necrotic center (thick arrow) and infiltration of inflammatory cells (arrow head) of ostertagia’s nodule with cystic formation of gastric glands (thin arrow) H&E, X40.

Damages caused by *Ostertagia* was studied in two levels. In Macroscopic level, multiple nodules with various sizes were observed. Congestion widely reported in some areas too. The resulting lesions, at the microscopic level, were also reported. In this level, the damages including destruction of the mucosal gastric glands, the increase in the number of inflammatory cells, hyperplasia of the mucosal tissue and edema were also reported. Abomasal glands were partially cystic. However, penetration of inflammatory cells such as eosinophils, plasma cells and lymphocytes were observed in some sections. pH of healthy and infected abomasum were largely similar indicating that there was no significant difference between pH of the infected and healthy abomasum. Although a slight increase (3.5 to 5 percent) was reported in pH of contaminated abomasum, the difference was not significant (P>0.05). The lesions, reported by Blanchard [16] and Love [34], are similar to the lesions recorded in the present study. In another study, lesions caused by *Ostertagia* in sheep associated with an increase in abomasal pH, an increase in the concentration of serum gastrin and pepsinogen, reduction of the number of parietal cells, accumulation of inflammatory cells such as neutrophils and eosinophil, and mucosal hyperplasia [9]. The present study did not touch on the destruction of the gastric glands and cystic glands. Perhaps, the occurrence of this destruction can be attributed to the fact that the inspected sheep were infested with *Ostertagia* and *Haemonchus* synchronously. According to results of a research, the lesions caused by *Ostertagia* in deer and bison are similar with lesions in ruminants. The “Morocco leather” appearance has been mentioned as the characteristic of infections caused by *Ostertagia* [35]. In addition, in necropsy of infected sheep with *Ostertagia* inflamed abomasal mucosa associated with outstanding white nodules were observed [36]. The results of the current study reveal the epidemiological and pathological importance of gastrointestinal nematodes in sheep especially *Ostertagia* genre in Khoy city. In order to avoid economic loss it is necessary to pay special attention to the prevention and treatment of this disease.

**Conclusion:**

This study indicated that Ostertagiasis is an important factor in causing damages such as loss of production and increased mortality. In current study the highest infection with *Ostertagia* occurred in summer and the
lowest infection was in winter. Also there were no significant differences between prevalence of the parasite and sex, age and abomasal pH of sheep. Therefore, certain measures should be taken against endemic parasites in each area. As a first step, treatment with anthelmintic drugs such as Albendazole. Fenbendazole or ivermectin should be performed preferably thrice a year. One of these treatments should be performed in late winter to fight against inhibited larvae. Second treatment should be done in early summer to mid-summer to fight against parasite eggs that have been transformed in to the third stage larvae. The third treatment should be done in early autumn.

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REFERENCES


