

Epidemiological survey of helminths of goats in southern Punjab, Pakistan

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Abstract. The main purpose of this study was to investigate the prevalence of helminths of goats such as *Fasciola hepatica*, *Haemonchus contortus*, *Paramphistomum cervi*, *Oesophagostomum columbianum*, *Cotylophoron cotylophorum*, *Monezia expansa*, *Oestertagia oestertagi* and *Oestertagia circumcincta*. The overall prevalence of all species of helminths was 52% in goat. The study was designed to investigate the factors of helminths prevalence on the basis of sex and age of goat with the help of Chi-square. All the results obtained were non-significant due to some factors which directly affects the prevalence of helminths.

INTRODUCTION

In Pakistan, parasitism is one of the major problems for livestock population, causing obstacles in the development of profitable livestock industry, like in sheep (Iqbal *et al.*, 1984; Khan *et al.*, 1989) and goats (Iqbal *et al.*, 1984). Prevalence of gastrointestinal helminths in ruminants has been reported from 25.1 to 92% in different areas of Pakistan at different times (Sarwar, 1963; Durrani & Hayat, 1964; Siddiqi & Ashraf, 1980; Durrani *et al.*, 1981; Mohiuddin *et al.*, 1984; Khan, 1985; Murwat *et al.*, 1988; Khan *et al.*, 1989; Iqbal, 1993). However, there are certain geographical regions in which livestock population needs to be examined for the presence of gastrointestinal helminths. District Muzaffar Garh is sandwich in the form of strip between two rivers, Indus and Chenab River. This riverine belt is rich in livestock, due to its geographical location and availability of lush green pastures. These animals especially goats suffer from many unidentified helminthes problems, which lead to high economic losses to the poor people of the area having small semi-

commercial farms for their livelihood. Therefore, it was thought imperative to investigate the type of internal parasites present in this area in order to frame a treatment or control programme.

Keeping in view these considerations, this project was designed to investigate the prevalence of helminths in goats of District Muzaffar Garh, Southern Punjab, Pakistan.

MATERIALS AND METHODS

The present study was conducted to determine the prevalence of gastrointestinal helminths, in goats reared under normal field conditions in the District Muzaffar Garh, Southern Punjab, Pakistan.

Sample Collection

One hundred faecal samples of goats were randomly collected in sterile polythene bags directly from rectum of each animal (Ayaz, 2010). These faecal samples of 37 kids (3-12 months) and 63 adults (aged 1 yrs and above) grazing on natural conditions in field, water and food was *ad libitum*, were brought to

District Diagnostic Laboratory Muzaffar Garh and Department of Parasitology, Faculty of Veterinary Sciences, Bahauddin Zakariya University, Multan for identification of eggs/larvae or adult of helminths.

Parasitological Procedures

Fecal Examination

Fecal samples were examined for helminthes eggs/larvae by using direct and indirect techniques (Ayaz, 2010) and for identification of certain nematodes, copro-culture were performed to obtain larval stage. Both eggs and larvae from copro-culture were identified by using standard techniques as described by MAFF (1979) and Soulsby (1982). Briefly, one gram of fecal sample was mixed well in a drop of water and a relatively homogenous and transparent preparation was obtained and examined under microscope by placing a drop of suspension on slide with cover slip. At least three direct smears were examined from each sample. All the samples were also examined by concentration techniques, i.e. floatation and sedimentation.

For floatation technique, five grams of feces was mixed in 30-50 ml of water and strained through a sieve to remove the coarse material. The mixture was allowed to sediment for half an hour. The supernatant was poured off and sediment was mixed in a saturated solution of common salt. The suspension was centrifuged at 1000 rpm for two minutes. The upper 0.1 ml of centrifuged suspension was transferred to a glass slide and examined under microscope at 10 X for the presence of helminthes eggs. More over a relatively new technique for "a single slide positive sample" was developed as micro-floatation technique and described as The Floatation Solution like saturated solution of zinc sulphate was used. Dilution of the 0.5 gm. fecal samples was diluted by 2.0 ml of Zn SO₄ floatation solution in a petri dish. The whole diluted fecal sample was shifted into Sahli's Hemoglobin container (2 ml capacity) slowly and carefully by a small glass pipette (20 µl). Hemoglobin container was fixed in ESR stand for 30 minutes so that all the ova/eggs of nematodes could accumulate and float at the upper surface of the solution. One or two drops of diluted sample were taken

after floatation process on glass slide with a glass dropper, covered by cover slip and examined under the binocular microscope.

For sedimentation technique, five grams of faeces was mixed in 30-50 ml of water and strained through a sieve to remove the coarse material. The mixture was allowed to sediment for half an hour. After centrifugation, the supernatant was decanted and washing was continued until supernatant became clear. A drop was taken from sediment with Pasteur's pipette on slide and was examined under microscope at 10 X for the presence of helminths eggs.

Copro-culture

Faecal cultures provide an environment suitable for hatching and development of helminths eggs whereby the third stage larva is identified. Faeces found positive for nematode eggs were broken up finely, using either a large pestle and mortar or spatula and were placed in a glass jar or petri-dish which was closed and incubated at a temperature of about 26°C for 7 days. After incubation, samples were examined for the presence of larvae. Larvae were identified with the help of keys given by MAFF (1979) and the tapeworms were identified after staining with Giemsa stain the head and segments if obtained completely or partially and eggs were identified (Soulsby, 1982).

Statistical analyses

Data on the prevalence of helminthiasis was analyzed using Chi-square statistical design on the basis of sex and age. Graphical representation of data was also done.

RESULTS

The present study was undertaken from February 2012 to April 2012 to determine the prevalence of gastrointestinal helminthes in small ruminants especially goats. In goats, an overall prevalence of helminthes was 52% (52/100). The highest prevalence (24/100; 24%) was recorded for nematodes followed by trematodes (08/100; 8%), cestodes (02/100; 2%) and mixed helminthes infections (18/100; 18%). A total of eight

species of helminths including four nematodes, i.e. *Haemonchus contortus*, *Oestertagia oestertagi*, *Oestertagia circumcincta*, *Oesophagostomum columbianum*; two trematodes, i.e. *Fasciola hepatica*, *Paramphistomum cervi*; and two cestodes, i.e. *Moniezia expansa*, *Cotylophoron cotylophorum* were recorded. The prevalence of different species of helminthes was as; *H. contortus* was the most prevalent species of helminthes followed by *P. cervi*, *F. hepatica*, *O. circumcincta*, *M. expansa*, *O. columbianum*, *O. oestertagi* and *C. cotylophorum* (Table 1). The mixed helminthes infection (18/100; 18%) was often composed of 12 species including *F. hepatica*, *H. contortus*, *O. ostertagi*, *Trichuris globulosa*, *Chabertia ovina*, *C. cotylophorum*, *O. circumcincta*, *M. expansa*, *Monezia benedini*, *P. cervi*, *Fasciola gigantica* and *O. columbianum*.

The prevalence of helminthes was higher in young goats as compared to old animals. In goats (Table 2), kids were also found infected with five species of helminthes, i.e. *H. contortus*, *P. cervi*, *F. hepatica*, *O. circumcincta* and *O. oestertagi*; whereas, from adults, six species of helminthes were recorded, i.e. *H. contortus*, *P. cervi*, *F. hepatica*, *M. expansa*, *O. columbianum* and *C. cotylophorum* in the order of decreasing prevalence. A total of 18% goats (18/100) had mixed infection comprising 30% (11/37) in kids and 11% (7/63) in adults. The mixed infections were either with two, three or four species of helminthes.

In goats (Table 3), five species of helminthes i.e. *H. contortus*, *P. cervi*, *F. hepatica*, *O. columbianum* and *O. oestertagi* were recorded from males and seven species i.e. *H. contortus*, *P. cervi*, *F. hepatica*, *O. circumcincta*, *M. expansa* and *C.*

Table 1. Overall prevalence of different species of helminthes in all goats

Species of helminthes	Number of Fecal Samples Examined	Number of Fecal Samples Positive	Per cent Infected
<i>Haemonchus contortus</i>	100	20	20%
<i>Paramphistomum cervi</i>	100	5	5%
<i>Fasciola hepatica</i>	100	3	3%
<i>Oestertagia circumcincta</i>	100	2	2%
<i>Moniezia expansa</i>	100	1	1%
<i>Oesophagostomum columbianum</i>	100	1	1%
<i>Oestertagia oestertagi</i>	100	1	1%
<i>Cotylophoron cotylophorum</i>	100	1	1%

Table 2. Age-wise prevalence of different species of helminthes in goats in the Muzaffar Garh district, Southern Punjab, Pakistan

Species of helminth	Kids	Adult
	No. positive/ no. examined (%)	No. positive/ no. examined (%)
<i>Haemonchus contortus</i>	9/37; (24.32%)	11/63; (17.46%)
<i>Paramphistomum cervi</i>	3/37; (8.10%)	2/63; (3.17%)
<i>Fasciola hepatica</i>	1/37; (2.70%)	2/63; (3.17%)
<i>Oestertagia circumcincta</i>	2/37; (5.4%)	0/63; (0%)
<i>Moniezia expansa</i>	0/37; (0%)	1/63; (1.58%)
<i>Oesophagostomum columbianum</i>	0/37; (0%)	1/63; (1.58%)
<i>Oestertagia oestertagi</i>	1/37; (2.70%)	0/63; (0%)
<i>Cotylophoron cotylophorum</i>	0/37; (0%)	1/63; (1.58%)
Mixed infection	11/37; (30%)	7/63; (11%)
Overall prevalence	27/37; (73%)	25/63; (40%)

Table 3. Sex-wise prevalence of different species of helminthes in goats in the Muzaffar Garh district, Southern Punjab, Pakistan

Species of helminth	Male	Female
	No. positive/ no. examined (%)	No. positive/ no. examined (%)
<i>Haemonchus contortus</i>	9/43; (20.93%)	11/57; (19.29%)
<i>Paramphistomum cervi</i>	3/43; (6.97%)	2/57; (3.50%)
<i>Fasciola hepatica</i>	2/43; (4.65%)	1/57; (1.75%)
<i>Oestertagia circumcincta</i>	0/43; (0%)	2/57; (3.50%)
<i>Moniezia expansa</i>	0/43; (0%)	1/57; (1.75%)
<i>Oesophagostomum columbianum</i>	1/43; (2.32%)	0/57; (0%)
<i>Oestertagia oestertagi</i>	1/43; (2.32%)	0/57; (0%)
<i>Cotylophoron cotylophorum</i>	0/43; (0%)	1/57; (1.75%)
Mixed infection	8/43; (19%)	10/57; (18%)
Overall prevalence	24/43; (56%)	28/57; (49%)

cotylophorum from females were recorded in the order of decreasing prevalence. In goats, 56% male (24/43) and 49% females (28/57) were found infected.

In Table 2 and 3, The prevalence of helminthes was higher in young animals compared with the older ones and the prevalence of helminthes was higher in males compared with the females.

In Table 4, a total of eight species of helminthes (four nematodes, three trematodes and one cestodes) were recorded.

In Table 5, The prevalence of helminthes was higher in young animals/ kids (73%) compared with the older ones (40%) while sex-wise the prevalence of helminthes was higher in males (56%) as compared with the females (49%).

DISCUSSION

Helminthiasis is one of the major problems which affect the productivity of goats. Losses caused by helminthes invariably depend on the prevalence, nature and intensity of

Table 4. Prevalence of mixed species of helminthes in goats in the Muzaffar Garh district, Southern Punjab

Species of helminthes	Goats
Nematodes	
<i>Toxocara vitulorum</i>	-
<i>Oesophagostomum radiatum</i>	-
<i>Oesophagostomum columbianum</i>	+
<i>Bunostomum phlebotomum</i>	-
<i>Haemonchus placei</i>	-
<i>Haemonchus contortus</i>	+
<i>Cooperia</i> spp.	-
<i>Trichostrongylus</i> spp.	-
<i>Trichostrongylus axei</i>	-
<i>Oestertagia circumcincta</i>	+
<i>Oestertagia oestertagi</i>	+
<i>Chabertia ovina</i>	-
Trematodes	
<i>Fasciola hepatica</i>	+
<i>F. gigantica</i>	-
<i>Paramphistomum cervi</i>	+
<i>Cotylophoron cotylophorum</i>	+
Cestodes	
<i>Moniezia expansa</i>	+
<i>M. benedeni</i>	-
Total Number of Helminthes	8
Species recovered	

Table 5. Age-wise and sex-wise prevalence of different species of helminthes in goats in the Muzaffar Garh district

Animals	Kids	Adults	Overall prevalence
Goats	27/37; 73%	25/63; 40%	52/100; 52%
Animals	Males	Females	Overall prevalence
Goats	24/43; 56%	28/57; 49%	52/100; 52%

infection and the management practices. In this investigation, the prevalence of helminthes was higher in young animals compared with the older ones, and higher in males compared with the females in this study. In goats, a total of eight species of helminthes including *H. contortus*, *O. oestertagi*, *O. circumcincta*, *O. columbianum*; *F. hepatica*, *P. cervi*, *C. cotylophorum*; and *M. expansa* were recorded. These parasites were also recorded previously by Siddiqi & Ashraf (1980), Shah *et al.* (1980), Mohiuddin *et al.* (1984) and Khan *et al.* (1989) from different areas of Pakistan and while Vercruyssen (1983), Hunter & Heath (1984), Gupta *et al.* (1987), Charles (1989), Van Aken *et al.* (1990), Pandey *et al.* (1994), Jacquiet *et al.* (1995), Dorny *et al.* (1995) and many others workers reported from various parts of the world. However, these workers have also reported some other helminthes in addition to those recorded in the current study.

Such a regional variation in the record of various species has been widely reported. This variation may be attributed to different geographical distribution, host factors and climatic conditions required for the development of free-living stages of different nematodes. A variety of factors like age, sex and breed of the host, grazing habits, level of education and economic capacity of the farmers, standard of management and anthelmintic used may affect as discussed by Asanji & Williams (1987a), Pal & Qayyum (1992), Gulland & Fox (1992), Maqsood *et al.* (1996), Jorgensen *et al.* (1998), Komoin *et al.* (1999), Valcarcel & Romero (1999) and Ouattara & Dorchie (2001).

The most prevalent nematode recovered in this study can influence the prevalence of helminthes in goats. *Haemonchus contortus* was the highly prevalent nematode as reported earlier by Bali & Singh (1977), Grant (1981), Ahmed & Ansari (1987) and Gupta *et al.* (1987). Its higher prevalence could be attributed to the fact that this nematode has a relatively short generation interval and ability to take advantage of favorable environmental conditions (Grant, 1981). The mean monthly maximum temperature of 18°C

or above and total monthly rainfall of 50 mm are conducive for translation and transmission of *H. contortus* (Gordon, 1953). Therefore, climate of the study area is very conducive for the propagation of *H. contortus* larvae. Keeping in view the high pathogenicity of *H. contortus* compared with other nematode species, it is concluded that goat farmers of the study area are relatively at high risk of economic suffering due to lowered productivity of their animals.

This situation even can worsen by the fact that a majority of the infected goats harboured more than one species of nematode parasites, having minimum two and maximum three helminthes species in each of the host. The climatic conditions of the study area vary from tropical to sub-tropical, which are conducive for the development of larval forms or intermediate hosts of helminthes. A warm and moist summer is well suited to the development and survival of the free-living stages of nematodes (Grant, 1981). Moreover, the availability of green pastures in irrigated or water logged area also help in perpetuation of the life cycles of different species of helminthes. The pasture larval counts and fecundity of different species of parasites of helminthes also play a major role in the epidemiology of helminthes. The pasture contamination, therefore, have direct influence on the population dynamics of nematodes like that of *Trichostrongylus colubriformes* (Barnes & Dobson, 1990) due to mixed species grazing (Hoste *et al.*, 2011). This is particularly true for the nematodes, which are highly prolific like *H. contortus* laying up to 10,000 eggs per day for several months and under optimum climatic conditions, gross contamination of the pasture can occur in a very short time (Radostitis & Blood, 1994).

This study was based on random sampling for about two months (February to April 2011), therefore, seasonal effect on the prevalence of different helminthes could not be ascertained. Some species like *Ostertagia* species thrive better in cool moist conditions (Kates, 1950; Gordon, 1953). The findings of present study are, however, typical of helminthes infections reported in other

subtropical areas of the world (McCulloch & Kasimbala, 1968; Taylor & Cawthorne, 1972; Beveridge & Ford, 1982; Pino *et al.*, 1988; El-Sayed, 1997; Stear *et al.*, 1998). The higher infection in young animals than that in the older ones may be attributed to lesser resistance because of lesser exposure to different species of helminthes compared with the older animals. It was, however, interesting to note that prevalence was higher in males compared with females. Normally, females are assumed to be more infected due to stress of pregnancy and parturition. This may be due to the practice of stall feeding females around pregnancy and thus lesser exposure to pasture contamination. Most of the researchers Asanji & Williams (1987b), Pal & Qayyum (1992), Iqbal (1993), Maqsood *et al.* (1996), Komoin *et al.* (1999) and Valcarcel & Romero (1999), have observed higher rates of nematode infection/worm burden in female hosts as compared with the males.

Higher prevalence of nematode parasites in females compared with males may be because of lowered resistance of female animals due to their reproductive events and insufficient/unbalanced diet against higher needs. In contrast to the current results, Gulland & Fox (1992) reported that prevalence and intensity of infection FEC (fecal egg counts) were higher in males than females, except during the lambing periods, and decreased with age in both sexes. Effect of reproductive cycle has also been reported to affect the worm burdens in animals, which has an important epidemiological significance. For example, Lyons *et al.* (1987, 1992) reported a progressive increase in the EPG (egg per gram) and number of helminthes in ewes during and after the parturition period. This phenomenon has been attributed to a variety of reasons including seasonal variations, host factors, activation of hypo-biotic larvae, parturition stress, poor nutritional status, peri-parturient relaxation in immunity (PPRI), and hormonal changes around parturition, breed differences etc. In many parts of the world, parturition of grazing animals is synchronized to occur with the climate that is favorable to pasture growth and also suitable for

development and survival of free-living stages of most helminthes (Wedderburn, 1970). Morgan *et al.* (1951) found that the nematode FEC (fecal egg counts) of ewes were higher if they are subjected to excessive stress such as extremes of weather and poor nutrition. Khalafalla *et al.* (2011) discussed that increased EPG (eggs per gram) also occurred in lactating ewes from autumn-lambing flocks, and suggested that the increase was associated with parturition and lactation rather than season.

Likewise, Brunson & Vlassoff (1971) studied the relative generic composition of post-parturient Strongyles egg counts for lactating and non-lactating ewes. Mean egg counts (MEC) were similar until after the conclusion of lambing, when the egg counts for the non-lactating ewes declined rapidly to a negligible level; while egg counts for the lactating ewes rose to a normal post-parturient peak. In lactating ewes, *H. contortus* and *Ostertagia* species were the major contributors to the egg output but only negligible numbers of eggs of these genera were passed by non-lactating ewes. Gibbs & Barger (1986) monitored the level of fecal egg counts in pregnant and dry ewes. Peak egg counts (PEC) were seen in pregnant ewes just before lambing. Moreover, lactating ewes acquired greater burdens of *O. circumcincta* and that the impairment of immunity to helminthes infections was responsible for this rise while certain goats showed innate resistance as reported by Chiejina & Behnke (2011). The effect of lactation on peri-parturient rise has been considered due to lactogenic hormone "prolactin" (Kelly & Dineen, 1973; Rahman & Collins, 1992; Fleming, 1993a, b). The PPRI to nematode infection in ewes has also been associated with a rise in fecal egg counts (FEC) during the peri-parturient period (Etter *et al.*, 1999). The hypothesis of PPRI was proved by Armour (1967) by experimental administration of cortisone to sheep and cattle with nematode infections, resulting in elevated nematode fecal egg count (NFEC). The levels of cortisone are known to increase during periods of stress e.g. peri-parturient period.

In another study, Connan (1972) demonstrated that the host factors were responsible for immunological impairment around parturition and thus resulted in peri-parturient eggs rise. Jansen (1973a, b, c) reported that the combination of the immune and the endocrine state of the host are considered to be responsible for the appearance of the spring rise. The peri-parturient rise in fecal egg counts (FEC) of worms was also attributed to the breed differences by Courtney *et al.* (1984), who noticed that three exotic breeds (Florida Native, Barbados Blackbelly & St. Croix) showed no peri-parturient rise in fecal egg counts (FEC); while domestic breed ewes of Rambouillet and Finn Dorset showed a pronounced peri-parturient rise after 6-7 weeks of post-lambing period. Jansen (1987) monitored the level of Trichostrongylides egg output in normal and late lambing ewes and in this case host factor was also responsible for egg count rise.

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