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Prevalence and Identification of Gastrointestinal Nematodes in Bovine in and Around Hawassa, Ethiopia

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Abstract: A cross sectional study was conducted from November 2017 to April 2018 in and around Hawassa, Ethiopia; to determine the prevalence and identification of gastrointestinal nematodes (GI) in cattle. A total of 384 faecal samples were collected from randomly selected cattle at different kebelles. Out of the examined samples, 183 (47.7%) were positive for gastrointestinal nematodes. The relative prevalence of gastrointestinal nematodes with different explanatory variables shows that age of animals have significant effect ($P = 0.031$) on the occurrence of gastro-intestinal nematodes with a relatively higher prevalence recorded in young age (57.4%) than adult (46.2%) and calf (36.7%). There was also a strong statistical association ($P<0.001$) between occurrence of GI nematode in cattle among different kebeles in and around Hawassa. However, the difference in occurrence of GI nematode within other explanatory factors such as sex, breed, body condition and management system of the study cattle were not statistically different ($p>0.05$). Coproculture of strongyle positive samples and recovery of nematode larvae stage three (L_3) revealed six different genera namely; *Trichostrongylus* 108 (28.1%), *Haemonchus* 89 (23.2%), *Strongyloides* 64 (16.7%), *Oesophagostomum* 44 (11.5%), *Bunostomum* 13 (3.4%) and *Nematodirus* 3 (0.8%). The study also identified higher occurrence of mixed genera infection (68.5%) as compared to single genus infection (31.5%). In conclusion, gastrointestinal nematodes are still great enough to compromise cattle productivity in and around Hawassa. Therefore, strategic parasite prevention and control measures should be implemented in the study area.

Key words: Coproscopy · Third Stage Larva · Occurrence

INTRODUCTION

Livestock sector in Ethiopia has a significant contribution to the national economy. The country endowed with abundant livestock resources of varied and diversified genetic roles with specific adaption to its wide range of agro ecologies. Due to these it have claimed to the largest livestock population of 47.5 million cattle Ethiopia 26.1 million sheep, 21.7 million goat, 7.8 million equines, 1 million camel and 39.6 million chickens [1] and But currently it has Africa's largest livestock record with an estimated total cattle population of about 57.83 million [2]. However, productivity per animal is very low because of different constraints of which parasitism represents a major drawback to livestock development in the country [3].

Gastrointestinal parasite infections are a world-wide problem for both small and large scale farmers, but their impact is greater in sub-Saharan Africa in general and Ethiopia in particular due to the availability of a wide range of agro-ecological factors suitable for diversified hosts and parasite species. Economic impacts are caused by Gastro intestinal parasites in a variety of ways: they cause losses through lowered fertility, reduced work capacity, involuntary culling, a Reduction in food intake and lower weight gains, lower milk production, treatment costs and mortality in heavily parasitized animals [4].

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In addition, the diverse agro climatic conditions, animal husbandry practice and pasture management largely determine the incidence and severity of various parasitic diseases in certain area. Furthermore, the prevalence of gastrointestinal parasites, the genera of helminth parasites involved, species and the severity of infection also vary considerably depending on local environmental conditions such as humidity, temperature, rainfall, vegetation and management practices [5, 6].

Some of the major nematodes responsible for gastrointestinal tract parasitosis in ruminants under tropical environment are: *Haemonchus* spp., *Trichostrongylus* spp., *Nematodirus* spp. and *Ostertagia* spp. (Family: Trichostrongylidae); *Bunostomum* spp. (Ancylostomatidae) *Oesophagostomum* spp. (Strongylidae) and *Trichuris* spp. and *Strongyloides* sp. [7]. Despite the immense progress made to control parasitosis, farmers in Ethiopia continue to incur significant losses due to insufficient availability of information in the epidemiology of the parasites. Furthermore, parasites appear to be a major factor for lowered productivity of Ethiopian livestock sector [8]. To take the control measures; assessment and epidemiological surveillance of nematode parasite by different diagnostic methods like fecal examination, EPG determination and identification of specific nematode is important [9, 10].

Haemonchosis is one of highly pathogenic blood feeding nematode disease of sheep, goats and cattle which causes major damage to the livestock industry and its sustainability. Haemonchosis is mainly caused by three species *Haemonchus contortus, H. placei* and *H. similis.* The adult worm and L4 larval stage of the *Haemonchus* adhere to the abomasa of the host, cause severe anemia which can lead to host mortality. Due to the huge economic threats of haemonchosis on livestock production, accurate identification is critical to their management and control [11]. The anthelmintics are used against GI nematode infections as prophylactic measures. Due to emergence of resistant strain of *Haemonchus* worm and over use of anthelmintics for treatment and control purposes has greatly reduced its efficacy across the world in sheep, goats and cattle [12, 13].

There is still scarcity of data pertaining to prevalence and identification of gastrointestinal nematodes in and around Hawassa town. Thus, this study was carried out to determine the prevalence, identifications of gastrointestinal nematode and to determine the different risk factors for parasite occurrence. Therefore, the objectives of this study were to determine the prevalence of gastrointestinal nematode occurrence in cattle; identify the major GIT nematodes of cattle in and around Hawassa; to determine different risk factors of parasite occurrence.

MATERIALS AND METHODS

Description of Study Area: The study was conducted in and around Hawassa in Southern Ethiopia situated 275 km South of Addis Ababa at a latitude of 7°04'N and a longitude 38°31'E on the escarpment of the Great Rift Valley. The altitude ranges from 1650 to 1700 m above sea level. The mean annual rainfall and temperature are 900-1100 mm and 27°C, respectively. The total livestock population of Sidama zone is estimated to constitute, 2,053, 6 25 cattle, 300,716 goats, 531,132 sheep, 71,128 donkeys, 5,729 mules, 1,760, 300 poultry and 117,248 beehives [14]. The study area shown on Figure 1.

Study Population: The study was conducted in cattle which had been reared under intensive, semi-intensive and extensive management system in and around Hawassa. All cattle in the study area which were not recently treated with anthelminthic drug were a focus of the study irrespective of their age, sex, breed, body condition and management system.

Study Design and Study Period: A cross-sectional study method was conducted to determine the prevalence and identification of bovine gastro intestinal nematodes in and around Hawassa Administration. Faecal samples were collected from study animals in each management system. The study was conducted from October 2017 to April 2018.

Sampling Technique and Sample Size: Simple random sampling method was employed to select the animals and faeces were collected from the individual animals for coprological examination. In this study, the sample size was determined based on the expected prevalence of 50% and absolute desired precision of 5% at confidence level of 95% according to the methods provided by Thrusfield [15].

$$N = \frac{(1.96)^2 \text{ Pexp-(1-Pexp)}}{d^2}$$

where: N=sample size; Pexp=expected prevalence; d=desired absolute precision. Then a total of 384 animals were sampled.

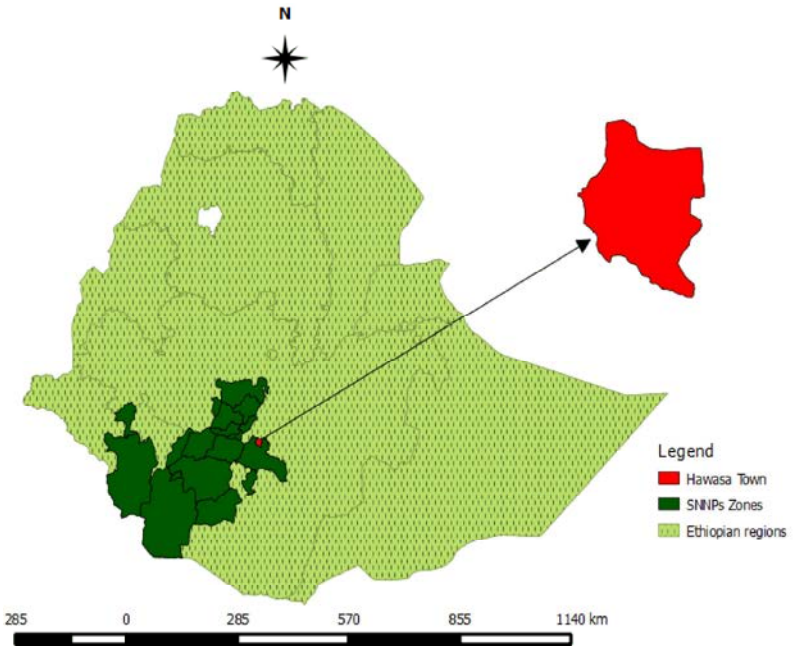

Fig. 1: Map of the study area

Study Methodology

Examination of Study Animals: The body condition of each of the study animal was scored using the guidelines established by Maurya *et al*. [16] (Appendix 2). Accordingly, on the basis of observation of anatomical parts such as vertebral column, ribs and spines, the study animals were classified as poor (score 1 to 3), medium (4 to 6), or good (greater than 6). The sex, age and breed of animal, address and management system were also recorded in a format (Appendix 1) during faecal sample collection.

Fecal Sample Collection: The fecal sample was collected directly from the rectum of randomly selected animals, by using new disposable plastic glove and placed in universal bottle. The bottles were labeled with a code that contains information about sex, breed and age, body condition score of individual animals, management, address and date of collection. After collection the samples were transported to the parasitological laboratory of Hawassa University. Samples were processed on the same day of collection and were reserved at refrigerator (4°C) to be processed within 48 hours.

Coproscopic Examination: For coproscopic examination of the fecal samples, a simple test tube flotation technique was employed and the procedure (Appendix 3) was followed as previously described by Hansen and Perry [17]. Floatation fluid, saturated sodium chloride (NaCl) solution was prepared and used to concentrate nematode eggs. Identification of eggs encountered was done by microscopic examination using compound microscope as described in Soulsby [18]. At the time of examination those fecal samples that were positive for *Strongyle* type eggs were subjected to faecal culture for third stage larvae (L3) recovery.

Faecal Cultures and Third Stage Larvae Identification: Faecal samples from animals which were positive for strongyle type eggs were cultured for harvesting third stage larvae and identification of the most important genera of non-distinguishable nematode eggs in cattle. The coproculture procedure (Appendix-4) was employed as previously used by Van Wyk and Mayhew [19]. The diarrheic faeces were finely mixed with sterilized bovine faeces. Then samples were transferred to petridish and were kept in room temperature at 27°C for two weeks. The cultures were moistened sufficiently every 1-2 days to ensure that they do not dry out whilst being managed, but without it becoming water-logged.

Third Stage Larvae Harvesting and Identification: Larvae from each culture were harvested by Modified Baermann technique (Appendix 4) as previously

described in MAFF [20]. Nematode species third stage larvae (L3) were prepared for examination by adding a drop of diluted Lugol's iodine solution to a drop of larval suspension from the harvested sample on a glass microscope slide. Iodine solution added to the larvae suspension was used for immobilizing as well as staining the larvae. Morphological identification of L3 of harvested nematode genera was conducted principally based on examination of the caudal and cranial extremities. However, other features such as the length and shape of oesophagus or cranial refractile spots were important identification keys (Appendix5) in some genera based on the description by Van Wyk and Mayhew [19].

Data Analysis: All the data collected about the age, breeds, body condition score and management system of the sampled animals were entered to Ms excel spread sheet. Descriptive statistics such as frequency tables mean and percentage were used to summarize the GI nematode occurrence. STATA 13 (Stata Corp LP, College Station, Texas USA) for statistical analysis and association between occurrence of GI nematode with hypothesized risk factors such as age, sex, breed, body condition, management and address of sampled animals) were tested by Pearson's Chi-square ($x2$) test statistics. In all the analysis, confidence level was held at 95% and the significance level was set at p =0.05.

RESULTS

Overall Prevalence of Gastrointestinal Nematodes: The present study revealed that 183 (47.7%) of the total 384 examined animals were found positive for at least one gastrointestinal nematode. The prevalence of gastrointestinal nematodes within different kebeles in the study area showed a relatively higher occurrence in Mehal kifile ketema (75.0%) followed by Tukurwuha (73.8%) and Addis kifile ketema (63.8%). However, the least prevalence of GI nematode was recorded in Chaffe (24.4%) and this difference was statistically significant (P = 0.000) as shown in Table (1) below.

The relative prevalence of gastrointestinal nematodes with different explanatory variables shows that age of animals have significant effect (P = 0.031) on the occurrence of gastro-intestinal nematodes. Relatively higher nematode positive animals were recorded in young age (57.4%) than adult (46.2%) and calf (36.7%). However, the difference in occurrence of GI nematode within other factors such as sex, breed, body condition and management system of the study cattle were not statistically different (Table 2).

Standard qualitative examination of faecal samples from study animals revealed three different types of nematode eggs. Strongyle type eggs were examined in 182 (47.4%) of the samples followed by *Strongyloides* spp. 64 (16.7%). Only three animals 3 (0.8%) were positive for *Nematodirus* spp.

Coproculture Result: Coproculture of 182 faecal samples which were positive for strongyle type eggs revealed four genera of GI nematodes, namely *Haemonchus*, *Trichostrongylus*, *Oesophagostomum* and *Bunostomum*. *Trichostrongylus* spp. (28.1%) followed by *Haemonchus* spp (23.2%) were recorded with relatively higher prevalence as compared to other gastrointestinal nematode parasites (Table 4). The composition of parasite genera in nematode positive animals showed a relatively higher proportion (68.5%) of mixed infection than single genera infection (31.5%). Half (50%) of animals positive for mixed infection were found with two parasite genera (Figure 2).

Table 1: Prevalence of GI nematodes per different kebeles in and around Hawassa

Kebeles	No.of examined	No. Positive (%)	95%CI
Addis kifileketema	47	30 (63.8)	48.5-77.3
Daka	30	13(43.3)	25.5-62.6
Dato	147	71(48.3)	40.0-56.7
Chaffe	82	20 (24.4%)	15.6-35.1
Mehalkifileketem	8	6(75.0)	34.9-96.8
Tesso.k.k	16	5(31.3)	11.0-58.7
Tula	12	7(58.3)	27.7-84.8
Tukurwuha	42	31(73.8)	58.0-86.1
Total	384	183 (47.7%)	42.6- 52.8

Pearson Chi^2 (7) = 39.1608; P = 0.000

Table 2: Prevalence and association of GIT nematodes of cattle with potential risk factors in and around Hawassa

Risk Factor	No. examined	No positive (%)	95%CI	x^2	*p-value*
Age				6.962	0.031*
Calf	60	22 (36.7)	24.6-50.1		
Young	101	58 (57.4)	47.2- 67.2		
Adult	223	103 (46.2)	39.5- 53.0		
Sex				0.001	0.975
Male	122	58 (47.5)	38.4-56.8		
Female	262	125(47.7)	41.5- 53.9		
Breed				0.657	0.72
Local	268	125 (46.6)	40.5- 52.8		
Cross	100	49 (49.0)	38.7- 59.2		
Exotic	16	9 (56.3)	29.9- 80.2		
Body condition				2.350	0.309
Poor	81	43 (53.8)	42.2- 65.0		
Medium	197	95 (48.2)	40.8- 55.2		
Good	106	45 (42.5)	33.0- 52.4		
Management				0.082	0.960
Intensive	28	14 (50.0)	30.6- 69.4		
Semi-intensive	201	96 (47.8)	40.7- 55.0		
Extensive	105	73 (47.1)	39.0- 55.3		
Over all	384	183 (47.7)	42.6- 52.8		

*Statistically significant

Table 3: Prevalence of GI nematode eggs identified in faecal examination.

GI nematode parasites	No. positive	Prevalence (%)	95% CI
Strongyle type	182	47.4	42.3- 52.5
Strongyloides	64	16.7	13.1- 20.8
Nematodirus	3	0.8	0.2- 2.3

Table 4: Prevalence of single and mixed GI nematode infection in coproscopy and coproculture.

Nematode genera	No. examined	No. of positive (%)	95% CI
Nematodirus	384	3 (0.8)	0.2 -2.3
Strongyloides	384	64 (16.7)	13.1 - 20.8
Haemonchus	384	89 (23.2)	19.1 - 27.8
Trichostrongylus	384	108 (28.1)	23.7 - 32.9
Oesophagostomum	384	44 (11.5)	8.5 - 15.1
Bunostomum	384	13 (3.4)	1.8 - 5.7

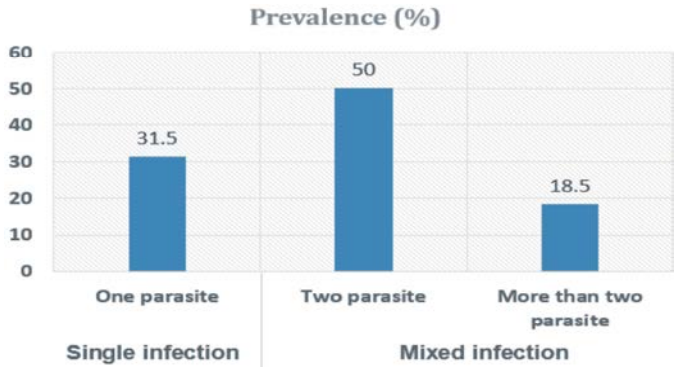

Fig. 2: Prevalence of single and mixed GI nematode infection as follows

DISCUSSION

Gastrointestinal parasitizes are prevalent in cattle and responsible for huge health problem to the animals as well as economic problems in Ethiopia [21-25]. They usually cause loss in body weight, digestive disturbance and emaciation [26, 27]. The findings of the present study, an overall of 47.7% positive for gastrointestinal nematode in cattle, suggest that GIT parasitism are still a major problem of cattle in and around Hawassa.

The findings of this study agree with the results of other researchers who have reported a prevalence of 41.2% [28] in Addis Ababa; 49% in West Arsi zone [23] and 47.0% in west Oromia regional state [8]. This finding is higher than 27.5% reported in and around Gonder [29] and 26.3% [30] in Western Amhara region, Ethiopia. But the present finding is lower than a very high prevalence rate of 82.8% reported by Etsehiwott [31] in Holleta Ethiopia. The difference in prevalence of gastrointestinal nematodes of cattle in different studies could be due to variation in de-worming practices, topography, season and climate that could favor the survival of parasitic stages.

The current study revealed that adult cattle have a relatively lower prevalence (46.2%, 95% CI = 39.5%-53.0%) of GI nematode than young cattle (57.4%, 95% CI = 47.2%-67.2%). It was also recorded that the prevalence in calves (36.7%, 95% CI = 24.6-50.1) is lower than both adult and young age groups of cattle. This agreed with study done by Tulu and Lelisa [32] in West Hararghe Zone and Addisu *et al*. [23] in West Arsi zone who reported a higher prevalence of gastro intestinal parasites in young and adult animals than calves. A statistically significant difference ($p = 0.031$) in infection rate of gastro-intestinal nematode parasites among age groups; observed in present study; is most probably due to variation in susceptibility and resistance to nematode infection among the different age groups. Adult animals may acquire immunity to the parasites through frequent challenge and expel the ingested parasites before they establish infection [33].

The study also presented that sex of the study animals did not show significant association with the prevalence of GI nematode parasites. This is in agreement with previous reports [8, 34, 35]. The insignificant association between occurrence of GI nematode and sex of animals is more probably due to an equal opportunity for infection when they are exposed to the parasites in the communal grazing pasture as well as the feed they are provided in intensive production system.

Higher infection was recorded on exotic breed cattle (56.3%) compared to cross breed (49.0%) and local cattle (46.6%). However, the difference on the occurrence of gastro-intestinal nematodes among breed of animals is statistically insignificant ($P = 0.07$). This is in contrary to the report of Bacha and Haftu [36] who reported a significant effect of breed on the occurrence of gastro-intestinal nematodes.

The difference in prevalence of gastrointestinal nematodiasis of cattle recorded in poor body condition (53.8%) than medium (48.2%) and good body condition (42.5%) is statistically insignificant ($P>0.05$). This agrees with previous report of Regassa *et al*. [8] and Hailu *et al*. [21] who described body conditions of the animal did not show significant association with the prevalence of the parasites. The reduction in body condition of animals might be due to malnutrition other concurrent diseases.

The study further revealed that kebeles in the study area show a significant association ($P = 0.000$) with prevalence of the parasites. Higher occurrence of nematode parasite was recorded in Mehal kifile ketema (75.0%) followed by Tukurwuha (73.8%) and Addis kifile ketema (63.8%). However, the least prevalence of GI nematode was recorded in Chaffe (24.4%). This is consistent with reports of Jelalu and Yitagele [22] who reported a significant difference in prevalence of gastrointestinal parasites of cattle among peasant association (PA's) in Gedebano Gutazer Wolene district, Ethiopia. The difference in prevalence of GI nematode among kebeles may be due to variation in management system, availability of communal grazing land and use of antihelmentics regularly.

In present study the predominance of mixed nematode infection (68.5%) than single nematode infection (31.5%) in positive cattle was not in line with the previous report of the predominance of single infection than mixed infections by Marskole *et al*. [37] and Olubukola *et al*. [38]. The possible explanation for this difference might be due to the variation in study methods and animals studied by researchers. The cited authors employed systematic random sampling method to identify helminths from slaughtered cattle but the present study was done by using purposively selected cattle grazing in a communal pasture land. Communal grazing pasture land has very great chance of becoming contaminated by different species of infective larval stages of helminths which can contribute to a higher mixed infection [39, 27, 40].

CONCLUSION AND RECOMMENDATIONS

The result of present study revealed high diversity of gastrointestinal nematodes in cattle with high overall prevalence of 47.7% suggesting that nematodes are still great enough to compromise cattle productivity. It was based solely on qualitative parasitological techniques (floatation and coproculture) for detection of gastrointestinal nematode eggs and larvae. Strongyle type eggs, *Nematodirus* and *Strongyloides* were the examined nematode eggs. Six different genera of gastrointestinal nematodes including *Haemonchus*, *Trichostrongylus*, *Oesophagostomum*, *Bunostomum*, *Strongyloides* and *Nematodirus* identified in present study, of which, *Trichostrongylus* spp. (28.1%) and *Haemonchus* spp. (23.2%) were identified as the predominant nematodes affecting cattle of the study districts. Most of infected animals were harboring more than one gastrointestinal nematode signifying that mixed infection is higher than single infection in cattle of the study area. The nematode parasites identified in the present study are already implicated as most pathogenic and cause huge economic losses in cattle around the world. Therefore, in line with the above finding, the following recommendations are forwarded. Strategic parasite prevention and control measures should be implemented. Awareness creation among the livestock owners on the proper management, feeding and use of anthlementics should be performed. Seasonal epidemiology of GIT nematode parasites should be studied.

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