academicJournals

Vol. 8(39), pp. 4922-4927, 10 October, 2013
DOI: 10.5897/AJAR2013.7350
ISSN 1991-637X ©2013 Academic Journals
http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Study on abomasal nematodes of sheep and goats: Comparison and characterization of vulvar morphology of *Haemonchus* in Hawassa, Ethiopia

Tilaye Demissie[1]*, Dawit Tesfaye[2], Amene Fekadu[2] and Isayas Asefa[3]

[1]College of Veterinary Medicine and Agriculture of Addis Ababa University, P. O. Box, 34, Debre-Zeit Ethiopia.
[2]School of Veterinary Medicine of Hawassa University, P. O. Box.05 Ethiopia.
[3]Ministry of Agriculture Department of animal health, Ethiopia.

Accepted 24 September, 2013

The study was conducted from November, 2011 to April, 2012. Two hundred and twenty two sheep and 174 goats' abomasum were examined according to the standard procedures. The overall prevalence of abomasal nematode was 82% for sheep and 76.4% for goats. The parasitic species specific prevalence was 80.6, 25.7 and 14.8% for Haemonchus species, *Trichostrongylus axei* and *Teladorsagia circumcincta*, respectively in sheep and 75.2, 9.8 and 14.2% in goats. Sex related prevalence for sheep was 79.6, 25.7 and 17.1% in male and 81.9, 25.5, and 11.7% in female for Haemonchus species, *T. axei* and *T. circumcinecta* respectively. The sex related prevalence in goats was 71.9, 5.7, 6.1% and 81.6, 21.6, 16.6% for *Haemonchus* species, *T. axei* and *T. circumcinecta*, respectively for male and female. The overall mean worm count was 7459.4 for sheep and 6244.9 for goats. The sex related mean worm burden was significantly higher (P<0.05) in female than male for both sheep and goats. Female *Haemonchus* species vulvar morphology was characterized and linguiform vulvar morphology was the most and knobbed type vulva morphology was the least frequently identified vulvar type both from sheep and goats' worms with higher proportions of linguiform vulva from goats than sheep. However, this difference in vulvar morphology was not statistically significant (P>0.05). It was concluded that the variation in prevalence and vulvar morphotype was almost similar with little deviations between sheep and goats. So importance of role of sheep to goats or goats to sheep as reservoir should be assessed. Investigations using advanced molecular techniques should be carried on genetic diversity and pathogenecity of *Haemonchus* in special as drug resistance is another emerging challenge in addition to the economic loss.

Key words: Abomasal nematodes, Haemonchus, Hawassa, prevalence, vulvar morphology.

INTRODUCTION

Ethiopia, with over all 42 million heads has the third largest numbers of sheep and goats among Africa nation and ranks eighth in the world (CSA, 2008). Small ruminants are particular resources for their owners; because they require small investment, have shorter production cycle, faster growth rate and greater environmental adaptability than cattle (Anon, 2005). Although, small ruminants represent a great resource for the nation, the productivity per animal is low. Small ruminant disease particularly the gastrointestinal helminthes (*Haemonchus*

*Corresponding author. E-mail: tilayedemissie2000@gmail.com Tel. 251-913-299098.

and liver fluke), are among the major causes of reduced productivity in Ethiopia (Sissay et al., 2007).

Fragmented studies indicated the importance of nematode as cause of decreased production in different parts of Ethiopia (Abebe and Esayas, 2001; Bersissa and Abebe, 2006; Sissay et al., 2007) but different studies should be done at different parts of the country to get national representative figures and most of the previous studies in Ethiopia were based on coprological examination which is less sensitive for identification of nematode species. Traits of vulvar morphology are considered as markers of ecological adaptation. The variation of vulvar morphology indicates manifestation of genetic factors during worm establishment and development (Gharamah et al., 2011). Le Jambre and Whitlock (1968) and Eysker and Ploeger (2000) indicated vulvar morphology helps to understand and know more about the biology of *Haemonchus* species and determine the type of population that occurs in area. The present work was conducted to study prevalence of abomasal nematode species in sheep and goats, determine their burden, and to compare the vulval morphology of *Haemonchus* species with the aim of complementing the study gap from the Southern part of the country.

MATERIALS AND METHODS

Study area

The study was conducted in Hawassa town. Hawassa town is the capital city of Sidama zone and Southern Nation Nationality and peoples (SNNP) region which is located in the northern part of SNNPR, 275 km South of Addis Ababa the capital city of Ethiopia. It geographically lies between 4°27' and 8°30' latitude N and 34°21' and 39°11' longitude E. The area receives 800 to 1000 mm annual average rain fall. During the study period the mean minimum and maximum temperature of the area was 20.1 and 30°C respectively and mean relative humidity was 71.8%. Hawassa lies at an altitude of 1790 m above sea level. The area mainly covered by dry savanna and bush type of vegetation (SNNPR, 2011).

Study animals

The study was conducted on sheep and goats slaughtered in certified restaurants in Hawassa town. The ten restaurants were randomly selected. A total of 222 sheep (20 to 24 sheep per-restaurant) and 174 goats (15 to 20 goats per-restaurants) were examined. Most of these animals were purchased from local markets surrounding the town. The age and body condition of these animals was determined and ranged from 1year to greater than 4 years according to Gatenby (1994) and Steel (1996).

Study design and sample size

A cross-sectional study type was used from November 2011 to April 2012. Of the animals ready to be slaughtered some were randomly selected and marked for sampling. The sample size was calculated according to Thrusfeild et al. (2005). Using 91.1 and 87.1% abomasal nematode prevalence for sheep and goats respectively from previous study (Thomas et al., 2007) and 95% confidence level, 5% desired absolute precision. The calculated sample size was 124 for sheep and 172 for goats but the sample size was raised to 222 for sheep and to 174 for goats in this study.

Sampling methods: Worm recovery, identification and count

The two end of the abomasum was ligated, separately collected in plastic bag and immediately transported to laboratory for examination. Worm collection, identification and counting were made in accordance with procedures and techniques described by Hansen and Perry (1994). In the laboratory the abomasum was placed on a clean tray and opened along the greater curvature and then thoroughly washed in the tray several times, paying particular attention to the folds of the mucous membrane. Finally the total volume of contents and washings in the bucket was made two liters' by pouring additional water. The total content was stirred vigorously until all food material, mucous and water were mixed. Finally 200 ml of the contents were transferred to another container in 5 steps of 40 ml per stir by stirring the mixture continuously. Then 20 ml of the content were poured in to Petri dishes and a few drops of iodine solution was added and mixed with the sample (Hansen and Perry, 1994). Species identification and worm count was made according to MAFF (1986) and Hansen and Perry (1994). Finally, the number of parasites found in 20 ml multiplied by 100 and then by 20 gave the total number of parasite found in the abomasums (MAFF, 1986).

Vulvar morphology of *Haemonchus* species

The vulvar morphology of representative female *Haemonchus* worms (400 worms from sheep and 400 from goats) were thoroughly observed under the microscope and characterized according to Jacquiet et al. (1995) and Eysker and Ploeger (2000).

Data management and analysis

The data were coded and fed to Ms Excel (2007). STATA computer software (Stata IC, version 11) was used for analysis. Descriptive statistics were used to calculate prevalence of abomasal nematodes and P-value was used to look the significant differences between the factors.

RESULTS

Prevalence

The overall prevalence of abomasal nematodes was 82% (182/222) for sheep and 76.4% (133/174) for goats. Abomasal nematode species specific prevalence in sheep was 80.6, 25.7 and 14.8% respectively for *Haemonchus* species, *Trichostrongylus axei and Teladorsagia circumcincta*. There is statistically significant prevalence difference (P<0.05) between *Haemonchus* species and the rest two parasitic nematodes but there was no statistically significant difference between *T. axei* and *T. circumcincta*. Out of the 182 positive sheep 67% were found positive for a single abomasal nematode species (most of which was *Haemonchus*), 21% were positive for two abomasal nematode species and 12.3% sheep were found infected with all three abomasal nematode species.

Table 1. Evaluation of the effect of the observed risk factors on the prevalence of nematode.

Animal species	Putative factors		Parasitic nematodes identified, and their prevalence with 95% CI*					
			Haemonchus species		*T. axei*		*T. circumcinecta*	
			Prevalence (%)	CI*	Prevalence(%)	CI	Prevalence (%)	CI
Sheep	Sex	Male	79.6	73-87	25.7	17-29	17	7-20
		Female	81.9	74-90	25.5	19-30	11.7	9-18
	Body condition	Good	80.3	74-86	24.4	18-31	14.3	9-20
		Medium	81.4	71-91	29.6	17-42	16.8	7-27
	Age group	≤2years	78.2	71-85	24.8	17-32	15.7	9-22
		2-4years	81.3	76-93	27.4	17-37	12.3	7-20
		>4years	71.3	61-100	25	29-47	18.7	11-39
Goat	Sex	Male	71.9	64-80	15.7	9-23	6.1	4-14
		Female	81.6	72-92	21.6	11-32	16.6	4-19
	Body condition	Good	64.7	48-79	27.4	12-35	14.3	4-18
		Medium	56.2	52-81	20.7	15-26	16.8	2-20
	Age group	≤2 years	77.8	70-86	16.3	9-24	10.5	5-17
		2-4 years	74.5	63-86	22.3	11-33	8.4	2-15
		>4 years	54.5	23-86	9	5-27	9	5-27

CI*=95% confidence interval.

The prevalence of *Haemonchus* species was 78.2% for young sheep (<2 years), 84.9% for adults (2-4 years) and 81.3% for old sheep (>4 years) while that of *Trichostrongylus axei* was 24.8, 27.4, and 25% for the three age groups, respectively. *Teladorsagia circumcincta* was isolated at the prevalence rate of 15.7, 12.3 and 18.7%, respectively for the age group <2years, 2 to 4years and >4years (Table 1).

The prevalence of *Haemonchus* species was 79.6 and 81.9% for male and female sheep, respectively and that of *T. axei* was 25.7% for male, and 25.5% for female. *T. circumcincta* was detected at prevalence rate of 17.1 and 11.7% for male and female sheep, respectively (Table1). There was no statistically significant prevalence difference (P>0.05) between male and female animals for single parasitic nematode comparison but statistically significant difference was observed in prevalence when *T. axei* and *T. circumcincta* were compared in reference to *Haemonchus species* between male and female sheep (P<0.05).

In goat abomasal nematode species specific prevalence was 75.2, 29.8 and 14.1% for *Haemonchus* species, *T. axei* and *T. circumcincta*, respectively (Table 1). Out of total positive goats 8.6% were positive for all the three parasitic species, 11.5% for two species and 79.9% for single parasitic species.

Age specific prevalence of *Haemonchus* species, *T. axei* and *T. circumcincta*, respectively was 77.8, 74.5, and 54.5% for young goats (< 2 years); 16.3, 22.3, and 9% for adults (2-4 years) and 10.5, 8.4, and 9% for older goats (>4 years). Abomasal nematodes prevalence was statistically significant among the three age groups (P<0.05) with all the three nematode species more prevalent in young goats.

The prevalence rate of *Haemonchus* species was 71.9% for male and 81.6 for female goats. The prevalence of *T. axei* was 5.7 for male and 21.6% for female goats while that of *T.circumcincta* was 6.1 and 16.6% for male and female goats respectively (Table 1). The prevalence difference was statistically significant (P<0.05) for *T. axei* and *T. circumcinecta* with female goats more being positive than males. Female goats were also more positive to *Haemonchus* species than males. However, this difference was not statistically significant (P>0.05).

Worm burden determination

The overall mean total worm count during the study period was 7459.4 worms per animal for sheep and 6244.9 worms per animals for goats. This overall mean total worm count was significantly difference between sheep and goats (P<0.05). The highest and lowest monthly overall mean worm count was shown in Table 2. The highest mean *Haemonchus* count both for sheep and

Table 2. Prevalence of the three abomasal nematodes of sheep and their mean worm counts per months of study.

Month	Monthly *prevalence* and mean *worm count*						
	***Haemonchus* species**		***T. axei***		***T. circumcinecta***		**Total**
November	42.5%	450.7	5 %	266.6	1.5%	288.6	1005.9
December	82.5%	559.8	9.4 %	364	6.6%	233.3	1157.1
January	72.5%	552.6	7.5 %	344.3	12.5%	296	1192.9
February	67.5%	541.6	6.6 %	372.7	14.8%	308	1222.3
March	86%	769.5	14.4 %	414	19.5%	347.7	1531.2
April	83.5%	689	17.5 %	373.3	26.5%	287.7	1350
Total	80.6%	3021.7	25.7%	2134.9	14.8%	1761.3	7459.4

Table 3. Prevalence of the three abomasal nematodes of goat and their mean worm counts per months of study.

Month	Monthly prevalence and mean worm count						
	***Haemonchus* species**		***T. axei***		***T. circumcinecta***		**Total**
November	74.6%	446.7	22.3%	223.3	18.5%	255	925
December	67.7%	400	36.5%	320	5.6%	133.5	853.5
January	50%	439.2	42.5%	329.3	13.5%	333.5	1102
February	61.7%	534.3	33.6%	306.4	11.5%	279.5	1120.2
March	80.5%	586	25.8%	219.6	15%	287.5	1093.1
April	63%	554.5	34.6%	316.4	11.5%	279.5	1151.1
Total	75.2%	2825.8	29.8%	1400	14.1%	1442.8	6244.9

goats was observed in March which was 769.5 and 586, respectively. However, the lowest mean *Haemonchus* count was observed in November (450.7) in sheep and in December (400) in goats. In sheep the highest mean count both for *T. axei* and *T. circumcinecta* was observed in March which was 414 and 347.7, respectively but the lowest *T. axei* count (266.6) was observed in November and *T. circumcinecta* (233.3) in December. In goats the average worm count for *T. axei* was highest in January (329.3) and lowest in March (219.6), for *T. circumcinecta* the highest mean count was also in January (333.5) but the lowest mean count was observed in December (133.5) (Table 3). Generally sheep with 7459.4 overall mean worm counts were found to be more heavily infested than goats with 6244.9 overall means count during the study period and this difference was statistically significant.

Vulvar morphology of female *Haemonchus*

Of a total female *Haemonchus species* collected from the two species representative worms (400 from sheep and 400 from goats) were randomly selected and thoroughly looked for the morphology of their vulva under stereomicroscope. Out of the total 400 female *Haemonchus* species of sheep that were studied for vulvar morphology, 176 (44%) showed linguiform vulva, 120 (30%) showed smooth vulva and the rest 104 (26%) worms manifested knobbed vulvar morphology (Table 4). The detail of goat's *Haemonchus* vulvar morphology was shown in Table 4. Linguiform vulvar morphology was the most and knobbed type vulva morphology was the least frequently identified vulvar type both from sheep and goats worms with higher proportions of linguiform vulva from goats than sheep. However, this difference in vulvar morphology was not statistically significant (P>0.05).

DISCUSSION

The overall prevalence of abomasal nematode in sheep in the present study (82%) was high and this was in agreement with the reported of Abebe and Esayas (2001), Bersisa and Abebe (2006), and Sissay et al. (2007), all of which reported prevalence higher than 80%. The overall prevalence of abomasal nematode was 76.4% for goats and this was lower than result of Sissay et al. (2007), who reported 88.2% prevalence. This difference in prevalence among the different study might be due to agro-ecological variation, time and season of sampling (temperature and moisture), it may be due to the nutritional status of sampled animals as nutrition affects the immune status of the host. El-Azazy (1995) from Saudi Arabia reported lower prevalence of abomasal nematodes than any previous report from Ethiopia. Although not confirmed by excluding other parameters this difference most probably be due to agro-

Table 4. Type of vulvar morphology of female *Haemonchus* and frequency of observation.

Animal species	Vulvar morphology	No. positive and prevalence		χ2	P-value
Sheep	Linguiform	176	44%	13.7	0.48
	Smooth	120	30%		
	Knobbed	104	26%		
Goats	Linguiform	261	65.2%		
	Smooth	85	21.3%		
	Knobbed	54	13.5%		

climatic factor as Saudi Arabia's climate is hot and dry comparing to the most reports from Ethiopia that was carried out in relatively wet and humid area.

There was statistically significant difference of prevalence among the three nematodes with the prevalence of haemonchosis much higher than the two species in both sheep and goats. Similar high prevalence rate of *Haemonchus* was reported by Bersissa and Abebe (2006) from Ogaden, and from abroad Githigia et al. (2005) from Kenya and Wang et al. (2006) from china all reported higher prevalence of *Haemonchus* over other abomasal nematodes and this might be due to various factors like its ability to produce large number of eggs (Getachew et al., 2007) and this allows *Haemonchus* an advantage over other parasites in that it can easily contaminate grazing areas or its ability to survive adverse climatic conditions through hypobiosis (Waller et al., 2004). *Haemonchus* uses various extrinsic and intrinsic factors for survival and hence development in host (Getachew et al., 2007).

The prevalence of *T. axei* (25.7 and 9.8%) and *T. circumcincta* (14.8 and 14.1%) in sheep and goats were lower than Sisay et al. (2007) who reported 47.2 and 39.4% of *T. axei* and 19.4 and 20.5% of *T. circumcincta* for sheep and goats, respectively. The result of this study was in support with that of Haileleul (2002). Many epidemiological factors may be attributed to this difference in prevalence in different study sites in Ethiopia. Conder and Jonson (1996) stated seasonal difference for *Haemonchus* and *Trichostrongylus* with *Trichostrongylus* preferring time of the year when temperatures are normally moderate, and significant rainfalls occur. These conditions are favourable for the free living stages of *Trichostrongylus* spp. which are inactive at the temperatures optimum for *Haemonchus contortus*. In addition although *Trichostrongylus* eggs may have been passed throughout the grazing season and a large proportion killed by desiccation, those which manage to become embryonated remain latent, but viable, and do not turn into larvae until suitable climatic conditions, in the form of mild weather and rain, occur. At that time there is massive hatching, overwhelming pasture infestation and clinical trichostrongylosis which may persist until well into the wintering period. Since at this time most lambs are approaching market weight, trichostrongylosis may cause losses by delaying marketing for several months.

The overall prevalence of abomasal nematodes was higher in females than males in both sheep and goats and this might be due to prolactin which suppress the immunity of the ewe and brings an apparent increase of the numbers of worms by the resumption of development of previously inhibited larval stages, increased rate of establishment of newly acquired larvae, failure in the elimination of existing infections, uninhibited development to maturity of newly acquired larvae, increase in the fecundity of egg laying adult female nematodes, which were repressed in ordinary healthy non-lactating ewes (Bowman, 1999). The condition in general brings increased worm egg which output which is epidemiologically significant not only by contaminating the pasture but also its adjustment with the period of existence of new susceptible population of lambs (Bowman, 1999). The higher prevalence in female animals may also be related to increased susceptibility to new infestations and enhanced prolificacy of female parasites arising from stress associated to pregnancy (Getachew et al., 2007).

The overall total mean worm count showed statistically significant difference between sheep and goats but the monthly lowest and highest mean count showed similar trend for both sheep and goats with little variation among few months of study. The highest monthly mean count in April in goats and in March in sheep might be due to favorable humidity and temperature that support larval development and survival of nematode as these months are time of short rainy season in Ethiopia and at the same time the lowest mean count for both animal species was at the pick of dry season in the study area. Muhammad et al. (2009) showed high humidity, at least in microclimate of the faeces and the herbage is also essential for parasite larval development and their survival.

Vulvar morphology

The result showed that the linguiform vulvar type was the most predominant morphotype followed by smooth and knobbed vulvar type in both sheep and goats. The vulvar morphology also showed consistency in the months of study and between animal species with very little variation and this was in consistence with previous works of Abebe and Esayas (2001), Bersissa and Abebe (2006), but Thomas et al. (2007) reported linguiform vulvar type followed by knobbed morphotype in sheep which was in contrast to the present finding. These variations may be due to ecological difference as Bersissa and Abebe (2006) study on Ogaden sheep that lives in hot arid areas comparing hot and humid area of the present work. Jacquiet et al. (1995) indicated vular morphology vary with ecology and taken as an adaptation to specific ecology. The variation of vulavar morphology among the different authors (mentioned above) may also be due to a manifestation of some genetic factors during worm establishment and development in the host (Gharamah et al., 2011). Le Jambre and Whitlock (1968) and Eysker and Ploeger (2000) indicated vulvar morphology helps to understand and know more about the biology of *Haemonchus* species and determine the type of population that occurs in sheep and goats of the area.

Cross infections between sheep and goats might be possible as trends of worm recovery were almost similar between these species of animals. The variation in vulvar morphotype is also similar with little deviations between sheep and goats. So importance of role of sheep to goats or goats to sheep as reservoir should be assessed. Investigations using advanced molecular techniques should be carried on genetic diversity and pathogenecity of *Haemonchus* in special and other nematodes in general as drug resistance is another emerging challenge besides economic loss they created.

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