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Abattoir and Coprological Prevalence of Fasciolosis and its Vectors: Infection Intensity and Species Diversity

[1,2]Abebayehu Tadesse, [1]Lencho Terefe, [1]Hagos Ashenafi,
[3]Getachew Tilahun, [1]Dinka Ayana and [3]Tadesse Eguale

[1]Addis Ababa University, College of Veterinary Medicine, Debre-Zeit Ethiopia
[2]Hawassa University, Faculty of Veterinary Medicine, Hawassa, Ethiopia
[3]Aklilu Lemma Institute of Pathobiology, Addis Ababa University, P.O. Box, 1176, Addis Ababa, Ethiopia

Abstract: A cross-sectional study was carried out to estimate the coprologic and abattoir prevalence as well as the burden of bovine and ovine fasciolosis. The study also identified the density of its vectors in endemic areas of central highland district of Ethiopia. The standard sedimentation and the McMaster egg counting techniques were used to determine the egg counts, estimation of herd egg output and subsequently the egg shedding index. Overall fasciola infections were only diagnosed in 605 (53.8%) animals coprologically. The highest prevalence was for sheep (60.1%) and followed by cattle (49.2%). The overall herd level infection prevalence, as estimated from the egg-shedding index, was 50.9±29.3. In both sheep and cattle, the highest and the lowest prevalence were observed during the months of November and April, respectively. In abattoir, about 1061 (81%) livers examined were positive for fasciolosis. *F. hepatica* was a dominant (87.9%) species identified followed by *F. gigantica* (6.3 %). Among the genera collected, *Lymnaea* species were the most abundant (1470) snail encountered. The present study plainly disclosed the high prevalence of fasciolosis both at herd and individual animal level and at abattoir survey. The eggs shedding index seemed to be useful approach in current epidemiological survey than the individual animal coprologic examination. The high pathogenecity of fasciolosis substantiate the need of at least one or two strategic treatment during the dry season. Further studies concerning the effects of altitudinal and climatic factors associated with the epidemiology of fasciolosis should be undertaken.

Key words: Fasciola · Prevalence · Coprology · Abattoir · Cattle · Sheep · Central Ethiopia

INTRODUCTION

In Ethiopian highlands, sheep and cattle production has remained as an important sector of the country's agricultural economy where three fourth of the national sheep flock and a significant number of cattle are located in the highlands [1-4]. However, their potential has been exploited far less than expected due to several constraints including shortage of forage, poor livestock management and diseases. Yilma and Malone [5] suggested that fasciolosis is widespread in Ethiopia encompassing the major productive highland plateaus except very limited areas in arid escarpments. In affected areas lacking strategic and systematic parasite control and prevention measures, fasciolosis still continued as one of the most devastating animal diseases in sheep and cattle resulting in morbidity and associated mortality [4 - 6]. However, these have been exacerbated in highland regions particularly in sheep [7, 8].

Apart from the altitudinal variations on the prevalence of fasciolosis, a study has demonstrated the differences in survival rate during fasciolosis among breeds of sheep. According to this study, the Ethiopian Menz sheep breed had shown superior survival rates, where other breeds have succumbed [9- 11]. It is also recently recognized as important re-emerging zoonotic diseases of public health important [1, 3,12]. Despite the inconsistent reports of fasciolosis in central highland

Corresponding Author: Abebayehu Tadesse, Addis Ababa University, College of Veterinary Medicine, P.O.B. 34, Debre-Zeit Ethiopia and Hawassa University, Faculty of Veterinary Medicine, P.O.B. 05, Hawassa, Ethiopia.

districts, the disease is considered as one of the most significant constraints on livestock production in Ethiopia [2, 3, 14-16].

The study of liver flukes in live animals depends on the use of faecal egg counts (FECs). However, these detect only patent infections and their interpretation is constrained by the paucity of information about how they relate to parasite burdens and pathology. Information collected at abattoirs can be used for cross-sectional studies to compare animals' management and performance with enumeration of parasites and other indices of infection [17].

The present research attempts to assess the epidemiology of bovine and ovine fasciolosis and its vectors with the appraisal of the associated risk factors in endemic central highland region of Ethiopia. The abattoir based survey and coproscopic examination in long dry season were determined. The comparison of the intensity of the liver infections and liver lesions, the occurrence of the vector species and its abundance, fluke prevalence in the abattoir and morphometric measurement of the flukes has been established.

MATERIALS AND METHODS

The Study Area: The study was conducted from November 2012 to April 2013 in and around Debere Berhan, Basona Worana, North shoa zone, Central Ethiopia. Debre Berhan lies at 09^{0}31N latitude and 39^{0}28E longitude with an altitude of 2780 m.a.s.l. located some 130 km away in northeastern part of Addis Ababa. The area is mountainous with large plain grazing land, dissected by rivers and streams. Five peasant Associations (PA's) namely Angolela, Birbirsa, Kormargefya, Weiniye and Goshbado located in close proximity of Debre Berhan were part of study. This area comprises large plain grazing land with extensive type of management system. The faecal samples and laboratory works for abattoir study were conducted at Debere Berhan Veterinary Clinic and Research Center. The snail identifications were carried out at Aklilu Lemma Pathobiology Institute and College of Veterinary Medicine and Agriculture of Addis Ababa University. The dominant livestock population is sheep followed by cattle where they are raised under traditional management system [18].

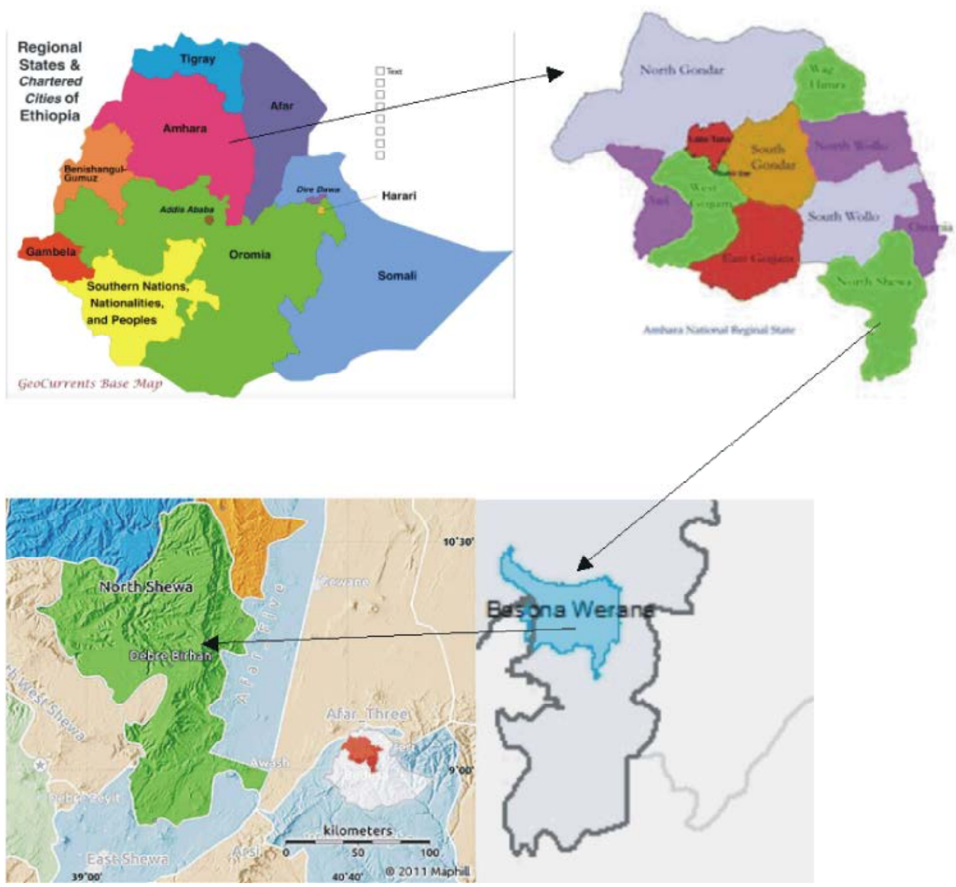

Fig. 1: Map of the study area showing Debre Berhan in Bosana district of North Shoa zone, Amhara region

Study Animals: Local zebu cattle and their crosses with the Holestien-Freiesian cattle as well as Menz sheep and their crosses with Hawassi breed sheep were used for both the abattoir and coprological study. However, the significant number animals used in this study were the local breed cattle and sheep managed extensively.

The Study Design and Sample Size: The cross sectional study was taken place following the subsequent selection of district, PAs and herds. The sampling strategy involves purposive (accessibility) selection of a district and random selection of PAs within the district from the available list of PAs as sampling frame. This was followed by random selection of herds that will bring their animals for the examination and then selection of study animal for faecal sample collection using simple random sampling technique. Therefore, cattle and sheep from herds in different PA's were taken part in the investigation. The sample size was determined according to the formula given by Thrusfield [19] with expected prevalence of 50% and precision of 5% with confidence level of 95%. However, to increase the precision, the sample size was made to 448 sheep and 667 cattle and this was allocated proportionally based on population number of animals in each PA. Accordingly, the PAs used in this study were; Debre Berhan (199), Angolela (222), Birbirsa (211), Kormargefya (161), Weiniye (179), Goshabado (153).

On each herd animals were selected in different age groups as young (calves and heifers or lambs and weaned lambs) and adults. Age estimation was done by inspection of the incisor teeth according to a method by Yeates and Schmidt [20] which is based on incisor teeth temporary teeth replacement and the degree of wear of permanent teeth. In addition sex of animals and their body condition status (poor and good) were recorded. The body conditions were estimated based on descriptions of Nicolson and Butterworth [21] for zebu cattle and Thompson and Meyer [22] for sheep.

Hence, attempt was made to determine the prevalence of fasciola species during the long dry season in slaughtered sheep and cattle with the help of both coprological examinations and abattoir survey at Debre Berhan Municipal Abattoir.

The Coprologic Study: A total of 1125 randomly selected animals comprising cattle (677) and sheep (448) managed under extensive traditional system were sampled. For the estimation of herd level prevalence fresh faeces were randomly collected, on a regular basis, directly from the rectum of all animals and transported to the laboratory in an airtight condition. The specimens were then subjected to qualitative coproscopic examination for the presence of characteristic *Fasciola* eggs by direct sedimentation technique employing a standard procedure [23]. The mean prevalence was then compiled on monthly basis and analyzed with respect to the different risk factors considered such as age, sex, breed and body condition categories.

Egg Output at Herd Level and the Egg Shedding Index: The McMaster egg counting technique was used to determine the faecal egg count, herd egg output and subsequently the egg shedding index [23, 24]. Accordingly, pooled faecal specimens from a herd of 6-10 animals belonging to different owners and from a total of 170 herds (80 sheep and 90 cattle herds) were examined. The pooling of samples was done regularly during the daily sampling of each animal. Prevalence and EP2G were used to calculate the herd shedding index, a parameter which estimates the herd-infection prevalence, using the following formula: HERD EGG-SHEDDING INDEX = PREVALENCE*EP2G [24].

Postmortem Examination: Adult fasciola parasites specimens were collected from condemned livers and associated gallbladder of cattle and sheep in Debre Berhan Municipal abattoir. Each liver was placed in a large basin and all the flukes in the gall bladder and the major bile ducts were collected into a small plastic container for subsequent counting. After visual observation and palpation of the liver, sharp incisions were made on the surface, through the major bile ducts into the parenchyma. The liver was then sliced into strips of about 1 cm in thickness and soaked in normal saline for about 5 h and washed extensively (incubated in physiological saline, 0.9% NaCl) in order to regurgitate the intestinal contents. Flukes emerging from the cut bile ducts were put into the same jar and each sliced strip was thoroughly squeezed from end to end, washed in saline and discarded. The contents of the basin were sieved, put into a petridish and the adult, immature and cut pieces of flukes were added to the container.

Fluke Identification: Individual flatworms were identified to species level according to existing keys and descriptions using their morphologic, morphoanatomic characters and morphometeric measurements [25]. Accordingly, they were classified as adult *F. hepatica*, *F. gigantica,* mixed and immature flukes. Adult *F. hepatica* are smaller than *F. gigantica* and have well developed 'shoulders' distal to the oral sucker whereas, the shape of *F. gigantica* is more streamlined without 'shoulders' [25]. Counts of the heads of cut flukes was made and added to the appropriate count of adult flukes.

Snail Survey: The snail survey was conducted in areas suspected of harboring the vector that include the water bodies (low-lying swamps, water lodged areas and slow flowing streams) and possible transmission sites (grazing and watering sites). Across the study district "six permanentand temporary" water bodies were selected based on their accessibility and animal-water contact frequency. Sample collections from these sites were made at monthly intervals from November 2012 to April 2012. A five meter by five meter quadrant was thrown on each sampling site. Snail abundance was estimated from the number of snails collected per unit of time (30 min) using kitchen sieves and the counts extrapolate to give the number of snails per man per hour. The number of snails collected and the water pH were recorded. They were moved at frequent intervals to fresh vessels and were observed for cercaria shedding. The identification was carried out on the basis of the snail's shell morphology and classified into major categories as per the criteria described by Hansen and Perry [26] Malek [27] and Frandsen and McCullough [28].

Statistical Analysis: The descriptive statistics was used to describe the overall prevalence of fasciolosis and the associated risk factors (age, sex, body condition, species, season and PA). A univariable logistic regression followed by multivariable logistic regression model was used to investigate the relation and statistical significance between positivity for coprology and liver examination and the factors considered. Data analysis was undertaken for the prevalence with species, breed, season, sex, body condition and age as independent variables. Fluke infection and egg detection with positive samples was considered as dependent variable. The results were analyzed for statistical significance by using STATA Version 11 (STATA, Stata corp. LP, 4905, Lakeway drive, College Station, Texas, USA).

RESULTS

Coprology: Among the 170 herds (80 sheep and 90 cattle) at the animal level, the overall prevalence of fasciolosis on the basis of coprologyy was 53.8% (605/1125). The highest prevalence was for sheep (60.1%) followed by cattle (49.2%). Among others, Birbrisa (77.7%) PA was shown the highest overall prevalence and Angolela (31.5%) the lowest. Among the risk factors considered the overall prevalence was highest for cross breed (62%) and female animals (56.6%) as well as those in young groups (59.8%) and animals with poor body condition (65%) (Table1).

The prevalence of eggs of fasciola from bovine species was highest in Birbirsa PA (76%) and the least goes to Angolela (31.7%). This similar trend was also recorded for sheep. In addition the other risk factors were noted to exhibit similar pattern as the case for the overall prevalence of fasciola eggs discharged from both sheep and cattle species (Table 2 and Table 3).

Table 1: Overall coprological prevalence (OR and 95% CI) of fasciolosis in ovine and bovine species with regards to the different risk factors assessed (species, PA, Age, breed, sex body condition).

Risk factors		Number examined	Number positive	Prevalence (%) 95%	OR (95% CI)	P value
Species	Overall	1125	605	53.8	.095 [0.4, 0.7]	0.49
PA	*Debre Berhan	199	130	65.2	-	-
	Angolela	222	70	31.5	0.31 [0.19, 0.49]	<0.001
	Birbirsa	211	164	77.7	1.72 [1.0 3.0]	0.04
	Kormergefya	161	76	47.2	0.83 [0.48 1.40]	.479
	Weiniye	179	95	53.3	0.67 [0.42 1.1]	0.108
	Goshbado	153	70	45.8	0.51 [0.38 0.84]	0.08
Age	Young	584	347	59.8	1.6 [1.3 20]	<0.001
	Adult	541	258	47.6		
Breed	Cross	610	378	62	0.41 [.27 .59]	<0.001
	Local	515	227	44.1		
Sex	Male	466	231	52	1.1 [.78 .15]	0.65
	Female	659	374	56.6		
Body C	Good	545	228	51.8	0.44 [0.35 0.56]	<0.001
	Poor	558	377	65		

*Debre Berhan is a reference PA (Peasant Association), OR, odds ratio; CI, confidence interval; P value, Probability value; P value <0.05 considered significant

Table 2: Over all prevalence (OR and 95% CI) Bovine fasciolosis with regards to the different risk factors assessed (species, PA, Age, breed, sex body condition). P value <0.05 considered significant

Risk factors		Number examined	Number positive	Prevalence (%) 95%	OR (95% CI)	P value
Bovine	677	333	49.2	-	-	
PA	Debre Berhan	118	76	64.4	-	-
	Angolela	126	40	31.7	0.32 [.19 .36]	<0.001
	Birbirsa	121	92	76	1.7 [.99 3.0]	0.05
	Kormergefya	99	58	58.6	0.39 [.23 .68]	0.001
	Weiniye	98	67	68.6	0.39 [.22 .67]	<0.001
	Goshbado	153	70	45.7	0.25 [.15 .45]	<0.001
Age	Young	324	175	54.0	1.4 [1.1 1.9]	0.016
	Adult	353	158	44.8		
Breed	Cross	346	183	52.8	0.34 [.11 1.0]	0.05
	Local	331	150	45.3		
Sex	Male	290	134	46.2	0.46 [0.15 1.6]	0.181
	Female	387	199	51.2		
Body Condition	Good	129	129	38.4	0.21 [.12 .38]	<0.001
	Poor	205	205	60.6		

*Debre Berhan is a reference PA (Peasant Association), OR, Odds ratio; CI, confidence interval; P value, Probability value

Table 3: The prevalence of Ovine fasciolosis (OR and 95% CI) with regards to the different risk factors assessed (species, PA, Age, breed, sex body condition). P value <0.05 considered significant

Risk factors		Number examined	Number positive	Prevalence (%) 95%	OR (95% CI)	p
Ovine	448	272	60.1	-	-	
PA	Debre Berhan	84	57	67.9	0.16 [.08 .36]	<0.001
	Angolela	96	24	25	0.16 [.08 0.36]	<0.001
	Birbirsa	90	72	80	3.2 [1.4 7.4]	0.01
	Kormergefya	62	35	56.5	0.77 [.33 1.8]	0.55
	Weiniye	64	48	75	2.1 [.94 .84]	0.68
	Goshbado	55	39	70.9	1.6 [.68 3.7]	0.29
Age	Young	180	100	55.6	1.93 [1.1 3.3]	0.014
	Adult	260	188	72.3		
Breed	Cross	184	77	41.8	0.30 [.19 0.4]	<0.001
	Local	264	195	73.6		
Sex	Male	176	97	55.1	2.2 [.14 .35]	0.001
	Female	272	195	71.9		
Body Condition	Good	205	100	48.8	0.38 [.22 .6]	0.001
	Poor	243	172	71		

*Debre Berhan is a reference PA (Peasant Association), OR, Odds ratio; CI, confidence interval; P value, Probability value

In cattle the egg output for fasciola species ranged from 14 to 890 with mean EPG of 57.1±34.3 (mean±SD) while in sheep the EPG ranged from 18 to 1300 with mean EPG of 63.5±88.3 and the difference was not significant (P<0.05). The highest overall mean EPG (74±87) was reported in Birbirsa PA and the least was in Angolela (42.4±33.8) (Table 4).

The Herd Egg Output: Across herds, the proportion with at least one infected animal, varied between 63 and 75 %. This proportion varied for sheep (63-70) and cattle (65-75) herds. However, there was a significant sampling effect at the herd-level; all herds where at least 605 animals sampled over the study period exhibited evidence of fluke infection (53.8%). There was significant variation in terms of within-herd infection prevalence (<0.05) (Table 4).

The egg shedding index estimated for the whole dry period (November to April) was 50.9±29.3. According to this indices, the value of herd egg output estimated for bovine and ovine species was 42.41±30.75 and 59.4±27.9, respectively and the variation was statistically significant (P<0.05). The overall mean egg shedding indices for young (58.9±39.8) and adult (37.8±29.6) as well as cross (60.4±60.6) and local (32.3±27.7) breeds were statistically significant (P<0.05). On the other hand, the herd egg output values obtained during this long dry seasons for male (44.4±23.4) and female (50.5±25.8) animals were not statistically significant (P> 0.05) as had been the case reported for good (51.7±41.5) and poor (48.1±39) body condition (P>0.05). The highest egg shedding index was recorded at Birbirsa PA (77.7±68.1) and the least was at Angolela (22.4±11.7).

Table 4: The prevalence (%) for faecal positive animals and the mean faecal egg counts of fasciola species with respect to the different risk factors assessed (species, PA, Age, breed, sex body condition). P value <0.05 considered significant

Risk factors		Number examined	Prevalence (%)	Mean EPG±SD	P value	Mean Herd EPG±SD	Mean Egg Shedding Index±SD	P-value (EP2G*P)
Species	Bovine	667	49.2	57.1±34.8		86.2±89 .4		<0.001
	Ovine	448	60.1	63.4±88.3		98.8±46.5	59.4±27.9	
PA	Debre Berhan	199	65.2	59.7±37.2		88.8±53.4	57.9 34.8	Reference
	Angolela	222	31.5	42.4±33.8	0.061	71.2±37	22.4 11.7	<0.001
	Birbirsa	211	77.7	73.7±87.9	0.310	100±87.7	77.7±68.1	<0.001
	Kormargefya	161	47.2	59.7±57.8	1.000	89.2±70.4	42.1±33.5	<0.001
	Weiniye	179	53.3	65.6±75	1.000	85.6±74.9	45.5±39.6	0.023
	Goshbado	153	45.8	58.2±58.5	1.000	84.8±62.3	38.8±84.1	0.025
Age	Young	584	59.8	64.5±66.6	0.049	98.5 80.8	58.9±39.8	<0.001
	Adult	541	47.6	56.2±58.3		79.4±60.6	37.8±29.6	
Breed	Cross	610	62	61.8±71	0.024	97.8±51	60.4±60.6	<0.001
	Local	515	44.1	57.1±49.2		73.3±62.3	32.3±27.7	
Sex	Male	466	52	56.2±58.3	0.027	85.3±57.8	44.4±23.4	0.043
	Female	659	56.8	64.5±66.6		88.9±44.9	50.5±25.8	
Body condition	Good	545	51.8	59.2±59.7	100	78.6±80	51.7±41.5	0.830
	Poor	580	65	60.2±64.1		74±60	48.1±39	

*Debre Berhan is a reference PA (Peasant Association), OR, Odds ratio; CI, confidence interval; P value, Probability value

Table 5: The overall abattoir prevalence of fasciolosis (OR and 95% CI) with regards to the different risk factors assessed (species, PA, Age, breed, sex body condition) and the associated risk factors.P value <0.05 considered significant.

Risk factors		Number examined	Number positive	Prevalence (%)	OR(95%CI)	SE	P value
Species	Bovine	803	623	77.7	.25[.15 .39]	0.6	<0.001
	Ovine	509	428	84			
Age	Young	874	428	81.6	0.99[.67 .48]	0.99	0.04
	Adult	438	623	77.7			
Breed	Cross	247	714	85	0.49 [0.62 1.3]	0.49	0.63
	Local	1312	338	79.2			
Sex	Male	762	210	81.1	0.91 [0.00 1.2]	0.18	<0.001
	Female	550	1040	78.7			
Body condition	Good	360	618	95	13.7[6.7 0.1]	7.12	<0.001
	Poor	952	433	74.2			
F.species	*F. hepatica*	1050	923	87.8	11.3[8.5 14.9]	1.6	<0.001
	F. gigantica	1051	67	6.3			
	Mixed	1051	61	5.8			

Abattoir Survey: Out of a total of 1312 sheep (509) and cattle (803) slaughtered, 1061 (81%) were found positive for fasciola species. However, the prevalence in sheep (84%) was significantly (P>0.05) higher than that of cattle (77.8%). The disease is 0. 25 times more likely occurs in sheep than in cattle. *F.hepatica* was a dominant (87.9%) species identified followed by *F. gigantica* (6.3 %) with some (5.8%) mixed infections. Age and sex appears to have no effect on the overall prevalence of fasciolosis. However, the prevalence was relatively higher in young (81.6%, 714/814) and male (81.1%, 618/762) groups than the adult (77.7%, 338/438) and female (78.7%, 433/550) animals. The effect of breed and body condition differences on the overall occurrence of fasciolosis was statistically significant (P<0.05) (Table 5).

The proportion of cattle and sheep slaughtered at Debre Berhan abattoir indicated that 79.2% (247/1312) were local breed. In sheep, the probability of the occurrence of the disease in Awassi-Menz cross bred animals was 49% compared to the local Menz breed. Similarly, 20% of the Holstein-Friesian crosses are more prone to acquire the disease than the local Zebu cattle. In general, poor conditioned animals have high risk of acquiring fasciolosis with the respect to animals in good body condition.

In sheep, most of the livers (248/509) examined were lightly affected (47.8%) and about a third of those (170/509) were affected severely (33.3%). However, the severity of the infection was more pronounced in cattle (53%, 426/803) while the remaining 31% (249/803) and 18%

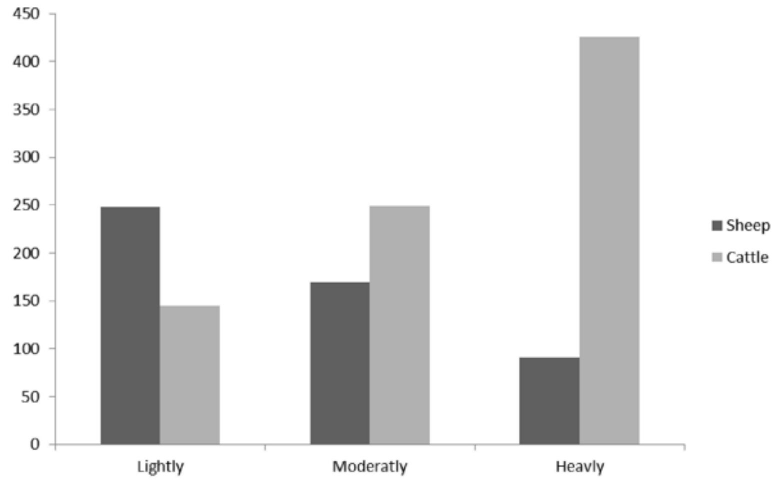

Fig. 2: The level of severity in fasciola infected liver of sheep and cattle

Table 6: The mean live weight measurement of the fluke infected liver and the corresponding mean fluke burden

Risk factors		Live weight (g) (M±SE)	Range (M)	Fluke burden (M±SE)	Range (M)
Species	Bovine (N=438)	539±41	483-610±30	77.5±0.6	24 -174±20
	Ovine (N=438)	344.4±38.5	254- 392±28	59.6±0.6	16-154±15
Breed	Awassi-Menz cross	386±48	296-423±34	63.2±0.3	36-154±12
	Menz sheep	302.7±29	245-338±21	56±0.3	16-145±13
	Holeistin-Fresien	583±29	522-647±22	81±0.7	32-175±20
	Zebu (Bos indicus)	504±34	470-563±31	74±0.6	24-162±16

Prevalence = Number positive/Total number; OR, Odds ratio; CI, confidence interval; P value, Probability value

(145/803) of the infected livers were affected moderately and lightly, respectively (Figure 2, vertical axis represent the number fluke infected liver while the horizontal axis the severity of the fluke infection).

The live weight of 1061 (sheep, 438; cattle, 623) fluke infected livers were measured. The mean (mean±SE) live weight of fluke infected liver of local Menz breed sheep was 302.7±29gm and this was lower than Aawassi Menz cross bred sheep (386±48 gm). On the other hand, the corresponding mean live weight of a liver of local zebu cattle and Holiestine-Frezian crosses were 504±34 and 583±29gms, respectively (Table 6).

The overall mean number of flukes recovered from sheep and cattle were 67.5±5.6 and this was ranged from 16 to 175. However, the mean number (77.5±0.7) recovered from the cattle was relatively high in comparison to the lower count (59.6±0.6) obtained from sheep. The live weight of fluke infected liver and fluke count of both sheep (r= 0.65) and cattle (r=0.73) have been positively correlated.

Both adult *F. hepatica* and *F. gigantica* were found to be leaf-shaped dorsoventrally flattened flukes with tapered anterior and posterior ends. From a total 17800 intact flukes used for morphometric analysis in cattle, 13172 (91.5%) were matured and 4628 (8.5%) immature. Similarly in sheep, from a total of 8560 intact flukes 6755 (78.9%) were matured and 1805 (19.1%) were immature. Accordingly, the mean length (mm) and width (mm) of matured *F. hepatica* in cattle was 27.8±0.23 (21-30±4.5) and 5.92±0.05, respectively. The corresponding length and width *F. hepatica* in sheep was 23.4±0.6 (20-25±2.4) and 4.12±0.54. In sheep, the mean length and width of matured *F. gigantica was* 25.1±2.38 (22-28±3.4) and 8.3±3.2, respectively. In cattle both the length and width of *F. gigantica* are relatively larger than sheep. Hence, the mean length and width were 25.1±2.3 and 8.3±3.2 in sheep and 31±3.2 (22-28±3.4) and 9.5±2.7 in cattle, respectively (Table 7).

Snail Survey: A total of 4350 snails belonging to different genera (*Lymnaea*, *Biompholaria*, *Bulinus*, *Physa, Bivalvia* and *Ancylus* species) were collected during the long dry season of the snail survey in those villages around Deber Berhan town. The snail species identified were clearly disclosed the existence of *Lymnaea truncatula, L. natalensis, Biomphalaria pfeifferi, Bulunus truncates, Bulinus forscale,* Physa, Bivalvia and Ancylus species. Among the genera collected, *Lymnaea* species

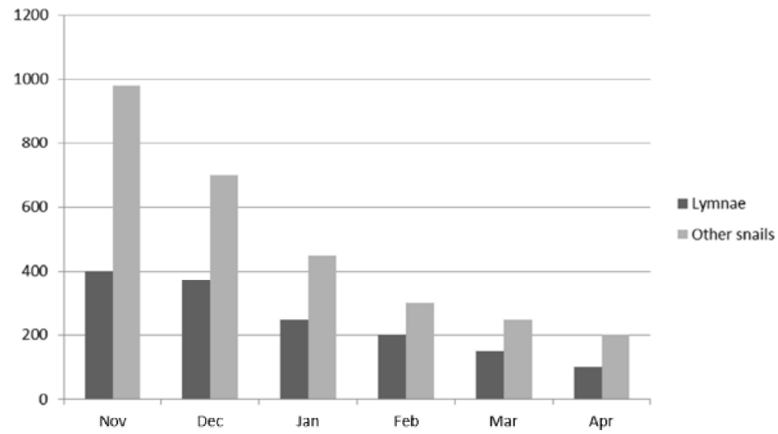

Fig. 3: Monthly prevalence of *Lymnaea* and other snail vectors in the study area (the vertical line represent the number of snail collected while the horizontal line is months within a year)

Table 7: The morphometric analysis of the length and width of *F. hepatica* and *F. gigantica* from sheep and cattle

	Fluke status		*Fasciola hepatica*			*Fasciola gigantica*		
Species	Mature	Immature	Length	Width	Range	Length	Width	Range
Bovine (N=17800)	13172	4628	27.8±0.23	5.92±0.05	21-30±0.45	31±3.2	9.5±2.7	25-35±2.5
Ovine (N=8560)	6755	1805	23.4±0.6	4.1±0.54	20-25	25.1±2.3	8.3±3.2	22-28±3.4

N= number of flukes recovered

was the dominant and most abundant (1470) snail encountered. The majority (905) were actually *L. truncatula*. However, *L. natalensis* (465) was encountered in those areas at the dearth of 1800masl and around Angolela river with higher elevation above 2600 masl. The study clearly demonstrated the presence of the two recognized snails serving as intermediate hosts of fasciola species (*L. truncatula* and *L. natalensis)*.

The pH of the different water bodies ranged from 6.9 to 8.35. The abundance of Lymnaea and other snails was highest during the cooler months with the highest record during the month of October and progressively decrease across the dry period with the lowest record during the month of April (Fig. 3). The cercaria shedding pattern of the Lymnaied snails had shown that, out of those 1470 snails, 82 (5.6%) were positive for *Fasciola* cercariae and 45 (3.0%) were releasing cercariae of other trematodes.

DISCUSSIONS

In Ethiopia, the prevalence of fasciolosis in sheep and cattle had been reported in different altitudinal ranges where the levels of climatic (temperature, rain fall) and ecological factors affect the prevalence, epidemiology and species of the parasite [6, 14, 29] Accordingly, the dearth of the *Fasciola hepatica* and *Fasciola gigantica,* determined by different authors [6, 24], was below 1200 masl and above 1700 masl respectively.

The present study had clearly disclosed the occurrence of higher prevalence of fasciolosis in sheep and cattle from highland areas. The overall coprologic, herd and abattoir prevalence of fasciolosis in cattle and sheep were, 53.8%, 50.9±60.8%, 81%, in that order. The subsequent findings in sheep were, 65.2%, 59.4±27.9% and 84%, respectively. Unlike in sheep, significantly (P>0.05) lower prevalence were reported in cattle for coprology, herd and abattoir survey with a prevalence of 49.2%, 42.41±30.8%, 77.8%, respectively. This result is higher than the coprologic findings of both individual animal (33.4 %) and herd (41.2%) level prevalence reports from North Gondar and lower than the abattoir reports (90.7%) of the same authors in cattle during the long dry season [6]. Contrary to the present finding (49.2%), higher prevalence (64.6%) was reported from the different PA's of the same districts around Debre Berhan town; however, the prevalence in sheep (54.2%) was relatively lower than the present (65.2%) finding [30]. Birbrisa (77.7%) PA was showed the highest overall fluke egg prevalence and Angolela (31.5%) the lowest. The difference in climatic and ecological factors (altitude) may have positively contributed to the observed differences in prevalence. Treatment provision (fasciolicidal) and vector control (endod) are also other factors that can affect the prevalence of fasciolosis.

The abattoir prevalence of fasciolosis in cattle (77.8%) was nearly in agreement with the 83% report of other workers in the same site [30, 31]. Comparable reports were observed in some parts of the country with similar ecological and climatic conditions in cattle and sheep. Accordingly, the report from the predominantly (75%) highland neighbor district of Menz Gera Midir of North Shoa zone indicated a prevalence of 40.5% in sheep [32]. However, other authors [33, 34] revealed, significantly lower prevalence of 15.5% and 14.6% of fasciolosis in sheep from highland district of Dabat in North Gondar and mid land areas of Jimma, respectively.

The predisposing factors like the PA's, species, age, breed and body condition observed were statistically significant ($P< 0.05$) variables on both individual animals and herd level by coprological survey. However, the abattoir study indicated that age and sex appears to have no effect on the overall prevalence of fasciolosis. On the other hand the effect of breed and body condition differences on the overall occurrence of fasciolosis in abattoir was significant ($P<0.05$). In sheep, the probability of the occurrence of the disease in Awassi-Menz cross bred animals was 15% compared to the local Menz breed. Similarly, 20% of the Holstein Friesian crosses are more prone to acquire the disease than the local Zebu cattle. In general, poor conditioned animals have 35% risk of acquiring fasciolosis with the respect to animals in good body condition. Furthermore the abattoir survey indicated that the disease is 0. 25 times more likely occurs in sheep than in cattle.

The present study in abattoir indicated that the dominant fluke species of the study area is *Fasciola hepatica* (87.8%). The proportions of cattle and sheep infected with *F. gigantica* alone (6.3%) or mixed infection with both species (5.8%) was relatively small.

The limitations of the gold standard method (post mortem examination) for fasciolosis in terms of labour, time and cost relatively makes it difficult approach compared to the egg-shedding index, as an estimate of herd-infection prevalence and individual coproscopic examination. This is mainly depends on the selection pressure for animals to be slaughtered at post mortem examinations [24]. Since, Selectionpressures are external agents which affect an organism's ability to survive in a given environment. Selectionpressures can be negative (decreases the prevalence fasciolosis) or positive (increases the prevalence). However, from the findings of the present study the approach with egg-shedding index was relatively stronger than the results obtained by individual coproscopic examination. Similar findings were reported by other authors for the superiority of the egg shedding index in terms of labour and cost [6, 22].

The number of flukes recovered and the fibrosis of the liver were used to classify the severity of fluke infections as light, medium and heavy. Accordingly, the worm burden in lightly affected sheep liver had relatively higher fluke counts compared to the heavily affected liver. However, in cattle, fluke counts in moderately affected livers exceeded that obtained from heavily affected ones. The less worm burden in heavily affected livers of sheep and cattle was associated with the severe fibrosis that impedes the passage of immature flukes and acquired resistance that resulted in the expulsion of flukes from the bile ducts [40].

The present result on the mean fluke burden in sheep and cattle was 59.6±0.6 and 77.5±0.7, respectively. This finding relatively exceeds the mean fluke count of 66.23 reported in Gondar [6]. Similar high mean fluke burden of 73.3 and 67 was reported in the Gondar and Bahrdar by other authors [35, 36, 47, 48]. The reports from other parts of the country in Nekemte area had also shown a mean fluke count of 85.5 in cattle [42]. Adem [43] and Abduljebar [43] were reported similar high mean fluke burden of 74 and 68 in Zeway and Sinana districts from cattle, respectively.

According to Soulsby [44] the presence of more than 50 flukes per liver indicates high pathogenecity. Other authors [45] were also concluded that as the number of *F. hepatica* adult forms increases, the likelihood of developing liver fibrosis will also increase in cattle. Therefore, the present finding clearly disclosed the high pathogenecity of fluke infections in both sheep and cattle of the study area and thereby in agreement with aforementioned authors. The mean (mean±SE) live weight of fluke infected liver of local Menz breed sheep was 302.7±29g and this was lower than Aawassi Menz cross bred sheep (386±48 g). On the other hand, the corresponding mean live weight of a liver of local zebu cattle and Holiestine Frezian crosses were 504±34 and 583±29g, respectively. The live weight of infected liver and fluke count of both sheep ($r= 0.65$) and cattle ($r=0.75$) have been positively correlated. That means, the larger the weight of the infected liver, the higher the count of the liver fluke.

CONCLUSIONS

The malacological survey clearly disclosed the existence of different genera of snails in the study areas. The vectors of *Fasciola* species (Lymneaa species) were the dominant snails collected during the survey. The presence of both Lymnaea species (*L. truncatula* and *L. natalensis*) demonstrated the potential of the intermediate hosts to transmit both *F. hepatica* and *F. gigantica* in the study areas (in all agro climatic zones despite the altitudinal difference). The existence and the report of *L. natalensis* at higher elevations were not uncommon. The finding of this snail above 2600 masl in river angolela was also reported by other authors. Abegaz [30] indicated the presence of the intermediate host in the same river and Rahmeto (1992) reported the existence of both species at 2070 elevations in West Shoa. From other snails collected, Bulinus species were dominant. The effect of the soil, temperature and rainfall on the occurrence, abundance and distribution of the snail intermediate hosts has been demonstrated by Yilma and Malone [14].

The presence study plainly disclosed the high prevalence of fasciolosis both at herd and individual animal level coprologically and at abattoir survey. *F. hepatica* was the dominant fluke species prevailing in the study area with the presence of some *F. gigantica* and mixed infections. The morphometric analysis clearly distinguished *F. hepatica* from *F. gigantica* where both adult matured *F. hepatica* and *F. gigantica* are leaf-shaped with tapered anterior and posterior ends with differences in their measure of mean body length and width.

The high prevalence of the disease in both sheep and cattle with the occurrence of the intermediate snail hosts (*L. truncatula* and *L. natalensis*) along with favorable climatic and agro-ecological conditions necessitate and urge the application of control measures focusing on curative and preventive measures that are integrated with management practices. Furthermore, the findings on the vector survey and the prevalence of the disease on host animals are dispensable for strategic application of control measures in study areas. The strategic treatment of animals at least once or twice during the long dry season will help to control the disease in addition to treatments which are commonly restricted to the rainy season by farmers of the study areas. Given the significant effect of the risk factors assessed on the prevalence of the disease, due attention has to be given in managing the risk of mortality and morbidity of animals associated with the difference in breed and other factors like age and body condition. Furthermore the emerging nature of the disease in human endemic parts of the world signifies the need to study the public health aspect of fasciolosis in endemic areas of animal fasciolosis.

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