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Prevalence of Bovine Trypanosomosis and its Vector Density in Selected Areas Bordering Omo River Basin, South Ethiopia

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Abstract: A cross-sectional study was conducted from November 2017 to April 2018 in selected sites bordering Omo river basin, south Ethiopia. The purpose of the study was to determine the prevalence of bovine trypanosomosis and apparent density of its vectors. A total of 384 cattle were randomly selected and examined for the parasitological survey. Buffy coat technique was used and a thin blood smear was prepared from positive samples for the determination of prevalence of trypanosomosis and biconical and NGU traps were used for the vector survey. The results revealed that overall prevalence of 6.51% with the highest rate of infection of *Trypanosoma congolense* 19 (76%) followed by *Trypanosoma vivax* 5 (20%) and *Trypanosoma brucei* 1 (4%). There was statistical significance ($p < 0.05$) between body condition, age, sex and packed cell volume of animals with prevalence but no significant difference was observed in peasant associations. The prevalence of bovine trypanosomosis was higher in poor body condition score 16(15.53%), old cattle 23(13.37%), female 22(8.9%) and lower value of packed cell volume 24(11.65%) animals and all the risk factors are significantly associated with trypanosomosis prevalence. The traps were deployed at grazing and watering points of animals in the six peasant associations. A total of 395 tsetse flies were trapped and among them 131 were *Glossina fuscipes*, 264 were *Glossina pallidipes* and other biting flies like tabanus and stomoxys were caught. Assessment of tsetse indicated the presence of *G. pallidipes* and *G. fuscipes* with the apparent density of 5.28 and 2.62 flies/trap/day, respectively. Due to its impact on cattle production, prompt control strategy has to be designed and implemented in the areas to minimize the distribution of tsetse as well as trypanosomosis prevalence.

Key words: Bovine · Omo River Basin · Prevalence · Trypanosomosis · Tsetse Flies

INTRODUCTION

Ethiopia has an enormous and diverse livestock population that plays an important role in the economy and livelihoods of farmers and pastoralists with a total contribution of 15% of Gross Domestic Product and 33% of the agricultural output. Current estimates of livestock population show that there are 57.83 million heads of cattle, 28.89 million sheep, 29.70 million goats, 2.08 million horses, 7.88 million donkeys, 0.41 million mules and about 1.23 million camels in Ethiopia [1]. Despite the large population of animals, productivity in Ethiopia is low due to poor nutrition, reproduction insufficiency, poor husbandry system and prevailing animal disease [2]. Trypanosomosis is one of the major disease impediments to livestock development and agricultural production, which negatively affect the overall development in agriculture in general and to the food self-reliance efforts of the nation in particular [3, 4].

Trypanosomosis is a disease caused by protozoan parasites of different species found in the blood and other tissues of vertebrates including livestock, wild life and man [5]. The disease is caused by the pathogenic species of trypanosomes transmitted cyclically by tsetse flies and non-cyclically by other biting flies except *Trypanosoma equiperdum*, which follows sexual means for transmission through coitus among equine species in its endemic area [6].

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African animal trypanosomosis and its vectors occur in vast areas of sub-Saharan Africa with devastating impact on livestock productivity. Its epidemiology and impact on livestock (especially cattle) production are determined largely by the prevalence and distribution of the disease and its vectors in affected areas. The limitations imposed by the tsetse and trypanosomes problem continue to frustrate efforts and hampers progress in crop and livestock production; there by contributing to hunger, poverty and suffering of entire communities in Africa [7].

Bovine trypanosomosis is a disease complex caused by several species of protozoan parasites of the genus *Trypanosoma*, mainly transmitted cyclically by the genus *Glossina* (tsetse flies), but also transmitted mechanically by a number of biting flies such as *Tabanus* and *Stomoxys* species [8]. Tsetse infests 10 million square kilometers and affects 37 countries, mostly in Africa, where the disease is known as 'nagana'. The disease can affect various species of mammals but from an economic point of view, tsetse-transmitted trypanosomosis is particularly important in cattle. It is mainly caused by *Trypanosoma congolense*, *T. vivax* and to a lesser extent, *T. brucei brucei*. Infection of cattle by one or more of the three trypanosomes results in subacute, acute, or chronic disease characterized by intermittent fever, anemia, rapid loss of condition, emaciation, collapse and death in untreated cases [9, 10].

The effects of trypanosomosis is not only the direct losses resulting from mortality, morbidity, infertility of the infected animals and costs of controlling the disease but also due to indirect losses which include exclusion of livestock and animal power based crop production from huge fertile tsetse infested areas [11].

Ethiopia, as part of the African continent shares a substantial loss from trypanosomosis [12]. Ethiopia is situated at the east end of the African tsetse belt. In Ethiopia, tsetse flies are confined to southwestern and northwestern regions between longitude 33°and 38°E and latitude 5°and 12°N an area covers 220, 000 km^2 [13]. Five species of *Glossina* including *G. morsitans submorsitans, G. pallidipes, G. tachinoides, G. fuscipes fuscipes* and *G. longipennis* have been recorded in Ethiopia [14]. Vector borne trypanosomosis is excluding 180, 000-200, 000 km^2 of agriculturally suitable land in the west and southwestern parts of the country; leaving 14 million of cattle, equivalent number of goat's and more than 7.5 million equines and 2.3 million of camels are at risk of contracting trypanosomosis [15].

The tsetse flies are widely distributed in the Western, Southern and South western low lands following the greater river basins of Omo-Ghibe and Baro, which has a high potential for agricultural development [16]. Ghibe-Omo river has 17.9 billion m^3/year runoff, 79, 000 km^2 catchment areas and a variety of wildlife parks [17]. In addition to this, the river has been hydraulically developed by the construction of ungauged cascade dams and reservoirs namely GIBE I and GIBE II and the huge GIBE III dam project [18]. Purpose of constructing Gibe III dam is to avoid severe drought periods and its reservoir operation also allows critical wet years [19]. Even though these dams should solve the power shortage problems of the country as well as increase the economic growth, construction of reservoirs and dams can lead to an increase in vector populations, provide breeding sites for them [20]. The primary forests in these areas together with the river networks constitute sanctuaries for tsetse flies and diverse wildlife [21]. However information on the prevalence and magnitude of the vector is important to plan appropriate control measures, previous studies on the prevalence of trypanosomosis and density of the vector are scanty. Hence, the study was aimed at determining the prevalence of bovine trypanosomasis and apparent density of tsetse flies in selected areas bordering Omo river basin (Gilgel Gibe III dam project), south Ethiopia.

MATERIALS AND METHODS

Description of Study Area and Animals: The study was conducted in six peasant associations (PAs) of 3 districts (Kindo-Koysha, Gena-Bosa and Loma) part of Omo-Ghibe tsetse belt located in Southern Nations, Nationalities and Peoples Regional State. The study area is characterized by wooded grasslands and riverine vegetation. The Omo Gibe river basin is almost 79, 000 km^2 in area and is situated in the south western part of Ethiopia, between 4°30' and 9°30' N and 35° and 38° E with an average altitude of 2800 m.a.s.l. It is Ethiopia's second largest river system after that of the Blue Nile, accounting for 14% of Ethiopian annual Runoff. There are two rainy seasons: the short one from the end of February to the end of April and the long one from June to September. All the study animals were indigenous cattle breeds that kept under an extensive husbandry system, free grazing and usually kept mixed with other livestock species in communal grazing areas [22].

Study Design: The study conducted during dry period from November 2017 to April 2018. A cross-sectional study type was used to estimate the prevalence of cattle trypanosomosis.

Sample Size: The sampling procedure was conducted using simple random sampling technique. The total number of animals required for the study was calculated based on the formula given by Thrus field [23]. The study considered 95% confidence interval and 5% desired absolute precision.

$$\frac{n = 1.96^2 * P\exp(1 - P\exp)}{d^2}$$

where,

n = required sample size
1.96 = the value at 95% confidence interval
P*exp* = expected prevalence of trypanosomosis
d = desired absolute precision level at 95% confidence interval (0.05).

An expected prevalence of 50% was used to increase the degree of precision and considering a 5% absolute precision and at 95% confidence level gave us 384 sample sizes.

Study Methodology

Parasitological Survey: For parasitological examination, blood sample were collected from ear vein of animal using microhaematocrit capillary tube and the packed cell volume, PCV was determined. The Buffy coat zone prepared in a microhaematocrit capillary was filled with 2/3 volume of blood and centrifuged for 5 min at 12, 000 rpm [24]. Some of the centrifuged blood filled capillary tubes were broken using diamond tipped pencil 1 mm below the Buffy coat to include the red blood cells layer and 3 mm above the Buffy coat to include the plasma. The content was expelled on the microscopic slides and then the slides were covered with 22x22 mm cover slip. It was examined under 40x objectives and ×10 eye piece using dark ground Buffy coat techniques, to see the movement of the parasite [25]. For species identification, a thin blood smear was prepared from positive samples and stained with Giemsa stain and examined under microscope using the oil immersion 100x [26].

Entomological Survey: For the entomological study, tsetse flies were collected by 15 NGU (named after Nguruman, where it was developed) traps and 10 biconical traps in different positions of the study areas. Octanol was used as a bait to attract the flies. Traps were positioned at approximate intervals of 100 to 200 m for 72 h in watering and grazing points in which the animals and the vector are believed to have frequent contacts [27]. Fly catch per trap per day (f/t/d) was determined to calculate the fly density and distribution [28]. The species of the dominant tsetse fly was determined following the standard procedures and biting flies according to their morphological characteristics such as size, color, wing venation structure and proboscis at the genus level [29].

Data Management and Analysis: The data collected from the study area were entered in to Microsoft 2010 excel spread sheet program to create data base and it was filtered before the analysis. Descriptive statistics was used to calculate the percentage of cattle in each peasant associations. For the analysis of data, statistical software program (SPSS version 20.0) was used. The risk factors like body condition, age, sex and PCV, of the animals were compared by using chi square test. In all cases $p < 0.05$ was used for the significance differences.

RESULTS

Parasitological Findings: The overall prevalence of bovine trypanosomosis in the study area was 6.51%. The prevalence of bovine trypanosomosis in the six PAs was determined to be in Loma (Afuki (8.69%) and Zima Waruma (6.25%)), Gena Bosa (Mela Galda (8.57%) and Bodi Ari (8.16%)) and Kindo Koisha (Moliticho (5.0%) and Soreto (2.94%)).

Among those six PAs, Afuki showed the highest prevalene rate 8.69% but the lowest being in the Soreto, 2.94%. The prevalence of bovine trypanosomosis and the corresponding infection rate in six selected PAs in the study area were summarized in table 1. Most of the infections were due to *Trypanosoma congolense* 19/25 (76%) followed by *Trypanosoma vivax* 5/25 (20%) and *Trypanosoma brucei* 1/25 (4%) as indicated in Table 1.

Out of infected cattle, 64% were from poor body condition, 32% with medium body condition and 4.0% were from good body condition. The higher prevalence rate 15.53% was seen in poor body condition followed by medium body condition 5.12% and that of good body condition 0.80%. Higher prevalence was observed in adult animals (>3 years) 13.37% and but lower in animals (≤ 3 years) of age 1.78%. The prevalence rate of infection in female and male were 8.9% and 2.18% respectively.

The prevalence rate of infection in female was higher than male. And also higher rate of infection in anemic (PCV < 25) was 11.65% but lower in non-anemic (PCV ≥ 25) about 0.56% were listed below in Table 2.

Table 1: The prevalence of bovine trypanosomosis in the six PAs of three selected districts

				Trypanosome species diagnosed					
Districts	PAs	Number of animals examined	Infected animals	Tc	Tv	Tb	P (%)	p-value	χ^2
Loma	Afuki	69	6	5	1	0	8.69	0.703	2.98
	Zima Waruma	48	3	2	1	0	6.25		
Gena Bosa	Mela Galda	70	6	4	1	1	8.57		
	Bodi Ari	49	4	4	0	0	8.16		
Kindo Koisha	Moliticho	80	4	3	1	0	5.0		
	Soreto	68	2	1	1	0	2.94		

Table 2: Prevalenceof trypanosomosis in relation to body condition, age, sex and PCV

Risk factors	Category	Number of animals examined	Infected animals	Prevalence (%)	p-value	χ^2
Body condition score	Good	125	1	0.80	0.00	20.96
	Medium	156	8	5.12		
	Poor	103	16	15.53		
Age	≤ year	112	2	1.78	0.016	5.79
	>3 year	272	23	13.37		
Sex	Female	247	22	8.90	0.011	6.53
	Male	137	3	2.18		
PCV	Lower (<25)	206	24	11.65	0.00	19.29
	Higher (≥25)	178	1	0.56		

Table 3: Apparent density of flies in six PAs of selected districts

Pas	Altitude Range(m)	*Stomoxys*	*Tabanus*	*Glossina*	No of traps	Mean catch F/T/D
Afuki	1048	8	5	96	5	10.9
Zima Waruma	1172	6	4	61	5	7.1
Mela Galda	1019	11	7	112	5	13
Bodi Ari	1051	7	5	76	5	8.8
Moliticho	1209	6	3	33	3	7
Soreto	1223	3	1	17	2	5.25

Table 4: Mean catch of *Glossina* species in six PAs at selected districts

		Glossina palidpes		*Glossina fuscipes*		
Pas	Altitude range(m)	M	F	M	F	Mean catch F/T/D
Afuki	1048	15	25	18	38	9.6
Zima waruma	1172	8	17	13	23	6.1
Mela galda	1019	27	69	5	11	11.2
Bodi ari	1051	16	47	3	10	7.6
Moliticho	1209	7	19	1	6	5.5
Soreto	1223	3	11	0	3	4.25
F/T/D	-	1.52	3.76	0.8	1.82	7.9

Entomological Findings: *Glossina pallidipes, Glossina fuscipes* and other biting flies like *Stomoxys* and *Tabanus* were caught. A total of 395 tsetse flies were trapped and among them 131 were *Glossina fuscipes*, 264 were *Gossina pallidipes* and a total of 41 *Stomoxys* and 25 *Tabanus* were caught during the study period. The apparent density of 2.62 for *Glossina fuscipes*, 5.28 for *Glossina pallidipes*, 0.82 for *Stomoxys* and 0.5 for *Tabanus* were recorded and summarized in Table 3 below.

In six PAs the already caught tsetse flies were determined and the higher fly per trap per day (FTD) catch for tsetse flies were obtained from Mela Galda (11.2) followed by Afuki (9.6), Bodi Ari (7.6), Zima Waruma (6.1), Moliticho (5.5) and Soreto (4.25) of PAs. A total of 395 tsetse flies caught during the study period were subjected for sexing. Accordingly 29.37% (116/395) males and 70.63% (279/395) were females. At all sites in each PAs more female tsetse flies were trapped than males are listed below in Table 4.

DISCUSSION

The overall prevalence of bovine trypanosomosis in the present study area was 6.51%, but it was low as compared to the previous studies done by different authors in different part of Ethiopia including 20.9% in selected sites of SNNPRS [30], 20.4% in Mareka, Dawuro zone [31] and 23% in Daramallo district [2]. But the current study result was higher than study in Arbaminch 17 (4.43%), south Ethiopia [32]. A slight decrease in prevalence from previous study attributed to the control measure of bovine trypanosomosis through a strategic tsetse control along with animal treatment.

Among the trypanosome species diagnosed, *T. conglense* was the predominant species (76%) followed by *T. vivax* (20%) and *T. brucei* (4%) is consistent with the previous work reports in different areas of Ethiopia of such as by Abebe and Jobre [33] in South west Ethiopia where they reported *T. congolence* (59%) and *T. vivax* (31%) infection and Muturi [34] who reported 66.86% *T. congolence* and 20.57% *T. vivax* infection in Southern rift valley. Such a high ratio of *T. congolense* may suggest that the major cyclical vectors of *Glossinia* species are more efficient transmitters of *T. congolense* than *T. vivax*. The transmission of *T. congolense* is cyclical; it requires the presence of tsetse flies whereas the transmission of *T. vivax* more readily transmitted mechanically by biting flies than tsetse flies [35].

The prevalence in six PAs also compared and high prevalence was observed in Afuki followed by Mela Galda but there was no significant difference ($P>0.05$). This might be due to similar agro-ecology and abundance vectors in all peasant associations [36].

The difference of infection rate between body conditions was statistically significant ($p < 0.05$) in which higher infectious rate was observed in poor than medium body condition cattle and was in agreement with the previous report by Tadesse and Tsegaye [37], Southwestern, Ethiopia. This might be due to weight loss caused by trypanosomosis. Since, loss of condition is a characteristics sign of trypanosomosis [38, 39].

The prevalence in sex groups also compared and it was higher in females than males and there was significance difference ($P<0.05$). The prevalence may be due to female animal are more likely exposed to different stress factors parity age, abortion, milk yield and lactation time and gestation. These factors may suppress the immune (defense) mechanism of animal which results in high infection rate. A similar finding also reported by Fetehanegest *et al*. [40] about 7 (3.46%) from a total of 202 female animals and 1 (1.02%) from 98 males were positive for bovine trypanosomosis and could also be due to female cattle were kept for purpose of rearing or giving offspring and milking for a long period of time that increases their exposure to the tsetse flies [41].

In this study, age was also taken as a risk factor. There was significance difference observed in age group in the study period and higher rates observed in older. These results agree with that of Dagnachew and Shibeshi [42]. As a higher prevalence was observed in adult animals (>3 years) and but lower in animals ≤ 3 years of age. This could be associated to long distance travel for grazing as well as for draught in areas of high tsetse challenge. Measuring PCV values for each animal samples in the study period in all six peasant associations were measured and marked difference was noticed. The overall average PCV of parasitaemic cattle is significantly lower than the aparasitaemic animals. Anemia is the common finding of chronic trypanosomosis [43]. The mean PCV value of studied animals was significantly ($P < 0.05$), varying between parasitaemic (<25) and aparasitaemic (≥ 25) animals. This result was consistent with the previous result reported by Terzu [44] in South Ethiopia and Tadesse & Tsegaye [37] in Southwestern Ethiopia.

The occurrence of trypanosomosis in the area was attributed to the existence of cyclical vectors, *G. pallidipes* and G. f. fuscipes and other biting flies. However, *G. pallidipes* was the predominant and most widely distributed. And an overall apparent tsetse fly density of 7.9 flies/trap/day caught in the study area include *G. pallidipes*, Glossina fuscipes and other biting flies like *Stomoxys* and *Tabanus*. A total of 395 tsetse flies were trapped and among them 131 were Glossina fuscipes, 264 were *Gossina pallidipes* and a total of 41 Stomoxys and 25 Tabanus were caught during the study period. That is higher than previous study by Rahmeto *et al*. [45] with an overall mean apparent tsetse density (AD) of about 3.5 F/T/D and the tsetse flies were identified to be *Glossina pallidipes* (85.1%) and *G. fuscipes* fuscipes (14.9%). Besides, other biting flies of the genus Stomoxys and Tabanus were trapped.

The result of present study is also higher than study by Abebayehu and Biniam [46] in Bench Maji zone, South Western Ethiopia with an overall apparent density of 2.83 flies/ trap/day, while the apparent density was 1.45 and 1.38 for *G. pallidipes* and *G. fuscipes,* respectively. The increase in the number of tsetse flies is due to favorable environment such as enough moisture, vegetation growth and suitable habitat. But the result is lower 14.97 flies/trap/day report in selected villages of

Arbaminch by Wondewosen *et al*. [47]. This finding also seems to be lower than the previous report 11.2 F/T/D. by SRVL [48] in Daremello district. The low level of tsetse population may be caused by the expansion of settlements and farmlands, which resulted in the migration of game animals from the study area.

Sex identification was performed on 395 tsetse flies caught in the study area and counted. The female tsetse flies 70.63 %(279) were dominantly caught than male ones 29.37% (116). Similar to study by Tola *et al*. [49] in Chora District of Illuababora Western Oromia, Ethiopia where a total of 237 tsetse flies caught among it higher number of female tsetse flies (124) than male tsetse flies (113). This indicates that female tsetse flies are playing important role in the cyclical transmission of Trypanosomosis than male tsetse due to the fact that female tsetse demands more blood when pregnant to feed their larva [50].

CONCLUSION AND RECOMMENDATIONS

Bovine trypanosomosis is an important disease and a potential threat affecting the health and productivity of cattle in the area. The results of the present study revealed that the overall prevalence of bovine trypanosomosis was 6.51%. *T. congolense* was the most commonly diagnosed parasite and followed by *T. vivax* and *T. brucei*. There was statistical significance between prevalence and risk factors like body condition, age, sex and PCV. The occurrence of trypanosomosis is associated to tsetse flies and other biting flies. Among tsetse flies *Glossina pallidipes*, *Glossina fuscipes* and other biting flies like *Stomoxys* and *Tabanus* were caught. Hence, negative impact on the production and health of cattle is determined largely by the prevalence and distribution of the disease and its vectors in study area. Thus, taking into account the above mentioned points, it is recommended that designing and implementation of control strategies for bovine trypanosomosis focusing on integrated disease management, vector control and awareness creation for the farmers for the control method is also essential.

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