

Prevalence of Bovine Trypanosomosis and Apparent Density of Tsetse Flies in Sayonole District Western Oromia, Ethiopia

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Abstract

A cross-sectional study was carried out from January up to March 2013 to determine the prevalence of bovine Trypanosomosis and apparent density of the tsetse flies and other biting flies in Sayonole district. The methods employed during the study were deploying trap for the collection of tsetse flies and buffy coat technique for parasitological study. About 43 traps were deployed for 48 hr for collection of tsetse fly. Among five species of tsetse flies commonly found in Ethiopia four of them *G.m.submorsitans*, *G.pallidipes*, *G.f.fuscipes* and *G.tachinoides* were captured in the study area. The overall apparent density of tsetse flies trapped was 13.01 flies/trap/day and female tsetse flies were foremost in number. Other biting flies caught were *Stomoxys* and *Tabanus*. Blood samples collected randomly from 599 cattles were assessed for *trypanosoma* species by buffy coat techniques. *Trypanosoma* species faced in the study area were *Trypanosoma congolense* 80(79.2%), *Trypanosoma vivax* 11(10.9%), mixed infection (*Trypanosoma congolense* and *Trypanosoma vivax*) 10(9.9%) with the overall Trypanosomosis prevalence of 16.9%. Anemic cattles which Packed Cell Volume less than 25% were mainly endangered of Trypanosomosis and significantly different $P < 0.05$. Poor body condition, 4-5 years of cattle age, Male cattle were the most susceptible for Trypanosomosis and insignificantly associated $P > 0.05$. Mean of Overall, Aparasitic and Parasitic Packed Cell Volume were 22.38 ± 5.022 , 22.68 ± 4.966 and 20.86 ± 5.044 respectively and significantly different $P < 0.05$. Generally, the study concludes that tsetse flies were an important vector for the epidemiology of bovine Trypanosomosis in sayonole district. Therefore, Vector and disease control and prevention methods and further studies should be undertaken to improve livestock production and productivity in the study area.

Keywords: Prevalence; Trypanosomosis; Tsetse flies; Cattle; Sayonole district

Introduction

Trypanosomosis is the most serious veterinary and animal production problem in sub-Saharan Africa and prevents the keeping of ruminants and equines over 10 millions of square kilometers of potentially productive land. Hence, this study is the road map and contribution to the Pan African Tsetse and Trypanosomosis Eradication Campaign agenda [1]. It is a protozoal disease caused by different species of unicellular parasites found in the blood and other tissues of vertebrates including livestock, wild life and people [2]. Bovine trypanosomosis is one of the diseases that are caused by flagellated protozoan parasites belong to the genus *Trypanosoma*. Trypanosomosis limit the extension of natural herds particularly in Africa where the presence of tsetse fly density access to wood land and savannah areas with good grazing potential [3,4]. It is a serious constraint to agricultural production in extensive areas of the tsetse infested regions [5]. The traction power of African agriculture 80% is provided by animals which were reduced capacity of work by Trypanosomes infection [3]. Generally, there is a great threat of trypanosomosis which impedes the economic development of African continent and also reasonable for the incalculable toll of human health [6]. Currently six species of trypanosomes are recorded in Ethiopia and the most important trypanosomes in terms of economic loss in domestic livestock are the tsetse transmitted species: *Trypanosoma congolense*, *Trypanosoma vivax* and *Trypanosoma brucei* [3].

Tsetse flies in Ethiopia are confined to southwestern and northwestern regions between longitude 33° and 38° E and latitude 5° and 12°N covers an area of 220,000 km² [7]. Tsetse infested areas lie in the low lands and also in the river valleys of Abay (Blue Nile), Baro, Akobo, Didessa, Ghibe, and Omo [8]. Consequently, new areas are being invaded and settled communities are being continually evicted by the advancing tsetse. Five species of *Glossina* (*G. morsitans*, *G. pallidipes*, *G. tachinoides*, *G. fuscipes fuscipes* and *G. longipennis*) have been recorded in Ethiopia [3]. Apart from the cyclical transmission of trypanosomosis by the *Glossina* species, it is highly considered that mechanical transmission is a potential threat to livestock productivity in some parts of Ethiopia [3]. Annual report of National Tsetse and Trypanosomosis Investigation and Control Center indicated that tsetse transmitted animal trypanosomosis is still remain as one of the largest causes of livestock production losses in Ethiopia [7]. In Sayonole district trypanosomosis was found to be one of the factors that slowed down livestock rearing in most of its peasant associations. However, hard evidence on the occurrence of tsetse and trypanosomosis in the area is lacking [9,10].

Therefore, the objectives of the present study were to determine the prevalence bovine trypanosomosis and to assess the distribution and apparent density of its vectors in Sayonole district of Western Oromia, Ethiopia.

Materials and Methods

Study area, population and sample size

The study was conducted from January up to March 2013 in Sayonole district, Western Oromia, which is situated at 500 Km West of Addis Ababa. The mean annual rain fall in Sayonole district ranges from 1000-1500 mm. The annual temperature ranges from 15-31°C. The areas have got a number of wild animals, such as African buffaloes, Bush pigs, warthog, bush buck, kudu, hippopotamus, crocodiles, hyena, antelopes and snakes which are claimed to serve as sources of food for the vector of trypanosomes.

The cattle in the district are local breeds that are kept under traditional extensive husbandry systems with communal herding. Agriculture is the main livelihood of the society with mixed farming system and livestock play an integral role for agriculture. The district has 18 peasant associations and animal population estimated to be 122,985 cattle, 10,540 sheep, 16,933 goats, and 1,203 donkeys in 2012.

The sampling method applied was simple random sampling. The sample size was calculated at 95% confidence interval, 50% expected prevalence and 5% precision. The required sample size was 384 cattle; however, a total of 599 cattle were sampled to increase the precision [11].

Study design and protocol

Sayonole district was selected purposely based on the extent of the existing problems of Trypanosomosis, the complaints of farmers and the level of medium to high tsetse challenge in the area. A cross-sectional study design was employed and three peasant associations were selected based on the veterinarian and farmers reports of the Trypanosomosis and tsetse infestation in the district. The cattle were categorized into three age categories 1-3 years, 4-5 years and ≥ 6 years and three categories of body condition of Good, Medium and Poor. For the trypanosomosis prevalence study buffy coat techniques were used. Blood sample collection was performed by piercing the marginal ear vein with a sterile lancet and blood was drawn by a heparinized capillary tube. Then one end (the heparinized end) of capillary tubes were sealed with crystal sealant and centrifuged at 12,000 rpm for five minutes to separate the blood cells and to concentrate trypanosomes using centrifugal forces. Then the PCV was determined by PCV reader and recorded. The capillary tubes were then broken just below buffy coat using diamond pencil and expressed on microscopic slide and covered with a cover slip. It was examined under 40 × objective of microscope to identify and detect the presence of the parasites [12].

The entomological survey consists of a total of 43 mono pyramidal baited traps which were the most important to catch riverine species

and deployed in riparian and savannah that are suitable areas for tsetse habitats to assess the apparent densities, distributions and species of tsetse flies and other biting flies involving in transmission of trypanosomosis. All traps were baited with acetone, octenol (1-3-octane) and cow urine filled in separated bottles and labeled and deployed at an interval of 200-250 m. After 48 hr or 2 days of trap deployment the cages were collected and captured flies were identified and sexed according to morphological characteristics and counted. The tsetse flies were identified to species level and the other biting flies to the genus level. The apparent density elements were the amount of flies collected from the traps deployed, number of the traps deployed and period of deployment in days and it is calculated as the number of flies collected/functional trap/period of deployment in days [6].

Data management and analysis

Raw data were entered into a Microsoft Excel spreadsheet and descriptive statistics were used to summarize the data. The prevalence was calculated for all data as the number of infected individuals divided by the number of individuals examined and multiplied by 100. The association between the prevalence of trypanosome infection and risk factors were assessed by chi-square, whereas the student's t-test was used to assess the difference in mean Packed Cell Volume between trypanosome positive, negative and overall examined animals. All statistical analyses were conducted using SPSS version 20.0 software. The test result was considered significant when the calculated p-value was less than 0.05. The apparent density of fly population was calculated by dividing the number of flies caught by the number of traps deployed and the number of days of deployment and expressed as fly/trap/day (FTD).

Results

Entomological survey result

In the study area tsetse flies *G. morsitans submorsitans*, *G. pallidipes*, *G. fuscipes* and *G. tachnoides* and other biting flies were trapped. The overall apparent density 13.01 flies/trap/day of the tsetse flies in Sayonole district. Among Glossina species caught riverine species *G. fuscipes fuscipes* was the dominant 9.2 flies/trap/day followed by savannah species *G. pallidipes* 3.6flies/trap/day in the study area. Sex identification performed on 1,119 tsetse flies caught in the study area and counted. The male and female sexes were 503 and 616 respectively. Abichu peasant association was high in infestation of tsetse flies (30.31 F/T/D). However, Gutemikael peasant association has no infestation of tsetse flies. Biting flies *Stomoxys* and *Tabanus* were caught (Table 1).

Peasant Association	Altitude range of Trap deployed	Tsetse flies caught										Biting flies caught			
		<i>G.m.submorsitans</i>		<i>G. pallidipes</i>		<i>G. f. fuscipes</i>		<i>G. tachnoides</i>		Total	FTD	<i>Stomoxys</i>		<i>Tabanus</i>	
		M	F	M	F	M	F	M	F	No.	f/t/d	No.	f/t/d		
Abichu	1420-1445m	1	1	3	18	390	357	18	0	788	30.31	110	4.23	6	0.46
Korekokir	1391-1435m	0	0	77	210	14	30	0	0	331	11.03	189	6.3	3	0.2

Gutemikael	1545-1691m	0	0	0	0	0	0	0	0	0	0	103	3.43	2	0.13
Total		1	1	80	228	404	387	18	0	1119	13.01	402	4.67	11	0.26

Table 1: Distribution and apparent density of tsetse flies according to peasant associations. F/T/D=flies/trap/day, M=Male, F=Female, G.m.submorsitan=G. morsitans submorsitans, G. f. fuscipes = G. fuscipes fuscipes.

Trypanosomes survey results

Composition of cattle involved in the study were 1-3 years old male (73), 1-3 years old female (67), 4-5 years old male (152), 4-5 years old female (126), and ≥ 6 years old male (131), ≥ 6 years old female (50). Blood samples collected from those 599 cattle were centrifuged and examined under microscope. The examination revealed that *Trypanosoma congolense* 80(79.2%), *Trypanosoma vivax* 11(10.9%), mixed infection of *Trypanosoma congolense* and *Trypanosoma vivax* 10(9.9%) and null infection of *Trypanosoma brucei* and finally two trypanosomes species were the causes of Bovine Trypanosomosis in the study area (Table 2). The overall prevalence of Trypanosomosis in Sayonole district was 16.9%. Abichu peasant association was high in prevalence of Trypanosomosis 25.83% (Table 3). The prevalence of Trypanosomosis on anemic cattles (PCV<25%) was 70(18.5%) due to the infection of *Trypanosoma congolense* 61(16.1%), *Trypanosoma vivax* 2(0.5%) and mixed infection of *Trypanosoma congolense* and *Trypanosoma vivax* was 7(1.9%) and significantly different $P < 0.05$ with prevalence of trypanosomosis (Table 4). Prevalence of Trypanosomosis is frequent on poor body condition score 40(19.7%) followed by medium body condition score 21(18.5%) and

insignificantly associated $P=0.324$ with the body condition score (Table 5). Trypanosomosis prevalence of cattles with the age categories of 4-5, 1-3 and ≥ 6 years were 52(18.8%), 26(18.5%) and 23(12.7%) and insignificantly associated with age $P=0.282$ (Table 6). Trypanosomosis prevalent on male cattles 69(19.4%) than female cattles 32(13.1%) and insignificantly associated with sex $P=0.117$ (Table 7).

Trypanosoma species	Prevalence of Trypanosoma	
	Frequency	Percent
T.C.	80	79.2
T.V.	11	10.9
Mixed infection	10	9.9
Total	101	100

Table 2: Prevalence of Trypanosoma species. T.C.=*Trypanosoma congolense*, T.V.=*Trypanosoma vivax*, Mixed infection=T.C. &T.V.

Peasant Associations	Sample size	Non infected	Prevalence of Trypanosomosis			
			T.C.	T.V.	Mixed infection	Total
Abichu	240	178 (74.2%)	48 (20%)	9 (3.8%)	5 (2.1%)	62(25.83%)
Kore kokir	199	160(80.4%)	32(16.1%)	2(1.0%)	5(2.5%)	39(19.6%)
Gute mikael	160	160(100%)	0	0	0	0
Total	599	498(83.1%)	80(13.4%)	11(1.8%)	10(1.7%)	101(16.9%)

Table 3: Prevalence of Trypanosomosis in three peasant associations. T.C.=*Trypanosoma congolense*, T.V.=*Trypanosoma vivax*, Mixed infection=T.C. &T.V.

PCV value	Sample size	Non infected	Prevalence of Trypanosomosis				X ² -value	P-value
			T.C.	T.V.	Mixed infection	Total		
<25	378	308(81.5%)	61(16.1%)	2(0.5%)	7(1.9%)	70(18.5%)	16.014a	0.001
≥ 25	221	190(86%)	19(8.6%)	9(4.1%)	3(1.4%)	31(14%)		
Total	599	498(83.1%)	80(13.4%)	11(1.8%)	10(1.7%)	101(16.9%)		

Table 4: Comparison prevalence of Trypanosomosis in Anemic or non-Anemic cattles. T.C.=*Trypanosoma congolense*, T.V.=*Trypanosoma vivax*, Mixed infection=T.C. &T.V.

BCS	Sample size	Non infected	Prevalence of Trypanosomosis	X ² -value	P-value
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							6.968a	0.324
			T.C.	T.V.	Mixed infection	Total		
Good	282	242(85.8%)	28(9.9%)	7(2.5%)	5(1.8%)	40(14.2%)		
Medium	114	93(81.6%)	17(14.9%)	2(1.8%)	2(1.8%)	21(18.5%)		
Poor	203	163(80.3%)	35(17.2%)	2(1.0%)	3(1.5%)	40(19.7%)		
Total	599	498(83.1%)	80(13.4%)	11(1.8%)	10(1.7%)	101(16.9%)		

Table 5: Prevalence of Trypanosomosis by body condition score (BCS). T.C.=Trypanosoma congolense, T.V.=Trypanosoma vivax, Mixed infection=T.C. &T.V.

Age year	in	Sample size	Non infected	Prevalence of Trypanosomosis				X ² -value	P-value
				T.C.	T.V.	Mixed infection	Total		
1-3		140	114(81.4%)	24(17.1%)	1(0.7%)	1(0.7%)	26(18.5%)	7.445a	0.282
4-5		278	226(81.3%)	40(14.4%)	6(2.2%)	6(2.2%)	52(18.8%)		
≥ 6		181	158(87.3%)	16(8.8%)	4(2.2%)	3(1.7%)	23(12.7%)		
Total		599	498(83.1%)	80(13.4%)	11(1.8%)	10(1.7%)	101(16.9%)		

Table 6: Prevalence of Trypanosomosis by age.T.C.=Trypanosoma congolense, T.V.=Trypanosoma vivax, Mixed infection=T.C. &T.V.

Sex	Sample size	Non infected	Prevalence of Trypanosomosis				X ² -value	P-value
			T.C.	T.V.	Mixed infection	Total		
Female	244	212(86.9%)	28(11.5%)	2(0.8%)	2(0.8%)	32(13.1%)	5.883a	0.117
Male	355	286(80.6%)	52(14.6%)	9(2.5%)	8(2.3%)	69(19.4%)		
Total	599	498(83.1%)	80(13.4%)	11(1.8%)	10(1.7%)	101(16.9%)		

Table 7: Prevalence of Trypanosomosis by sex. T.C.=Trypanosoma congolense, T.V.=Trypanosoma vivax, Mixed infection= T.C. &T.V.

Haematological survey result

Blood samples collected from cattles were centrifuged by haematocrit centrifuge and its PCV read by PCV reader. Mean of

Overall, Aparasitic and Parasitic PCV were 22.38 ± 5.022 , 22.68 ± 4.966 and 20.86 ± 5.044 respectively and significantly associated $P < 0.05$ (Table 8).

Categories	Sample size	Mean	Std. Deviation	Std. Error Mean	T-test	df	Mean difference	P-value	95% interval difference		Confidence of the
									Lower bound	Upper bound	
Overall PCV	599	22.38	5.022	0.205	109.059	598	22.377	0.000	21.97	22.78	
Aparasitic PCV	498	22.68	4.966	0.223	101.936	497	22.685	0.000	22.25	23.12	

Parasitic PCV	101	20.86	5.044	0.502	41.566	100	20.861	0.000	19.87	21.86
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Table 8: Mean of Packed Cell Volume (PCV) of Overall, Aparasitic and Parasitic cattles. PCV=Packed Cell Volume, df=degree of freedom.

Discussion

In the present study four species of *Glossina* *G.m.submorsitans*, *G.pallidepes*, *G.tachinoides* and *G.f.fuscipes* were caught and 13.01 flies/trap/day of apparent density was recorded in Sayonole district. Other biting flies *Stomoxys* and *Tabanus* were caught. This study agrees with the apparent density of tsetse flies 14.97 flies/trap/day in selected villages of Arbaminch by Teka [13]. This study result is greater than study report from Mandura district, Northwest Ethiopia with 0.06 flies/trap/day apparent density of tsetse flies by Lelisa, et al. [14]. However, it is slightly lower than the finding reported by Ayele, et al. [15] of 19.14 flies/trap/day apparent density. The apparent density of tsetse flies in Sayonole district is huge because no control activities have taken yet. For this reason, it has gotten a favorable condition for the tsetse flies to multiply in the area. In this finding more female tsetse flies were caught than male tsetse flies which comply with finding of Efrem, et al. [16] in Lalo kile district, Kellem Wollega zone. This could be because of the female tsetse flies physiologically necessitated to feed more animal blood during pregnancy than males which exposes it to trapping than male tsetse flies.

The study revealed that the overall prevalence of Trypanosomosis from 599 cattles sampled in Sayonole district was 16.9%. It was slightly consistent with the finding of 13.44% Waktole [17] in Gawo Dale district Kellem Wollega zone and 20.74% of Mekuria, et al. [18] from Metekel zones. However, it was greater than the findings of 6.86% Efrem, et al. [14] in Lalo Kile district of Kellem Wollega zone; 6.9% Alamayehu et al. [15] in Chena district, South west Ethiopia and 5.43% Lelisa, et al. [14] in Mandura District, Northwest Ethiopia which were reported. The higher prevalence of trypanosomosis in this study may be endorsed to the lack of application of control measures such as target impregnated insecticides, insecticide treatment of cattle and regular treatment of sick animals [19].

The prevalent *Trypanosoma* species in the study area was *Trypanosoma congolense* 80(79.2%). It was marginally reliable with the finding in the Lalo kile district 75% reported by Efrem, et al. [16] and 84% in Ghibe by Muturi [20]. Even though, it was higher than other areas studies in southern Ethiopia 37% by Rowlands et al. [21]; 58.5% in tsetse infested areas of the country Abebe and Jobre [4]; 66.17% in Southern Rift Valley Rowlands et al. [22]; 71.8% in the Gawo Dale district Waktole [20] and 72.3% in Gawo Dale District [6]. Anaemia (PCV<25%) was dominant by infection of *Trypanosoma congolense* 61(16.1%) than other species of *Trypanosoma*. The predominance of *Trypanosoma congolense* infection in cattle may be due to the high number of serodems of *T. congolense* as compared to *T. vivax* and the development of better immune response to *T. vivax* by the infected animal [23].

Trypanosomosis was prevalent on the poor body condition scored 40(19.7%) cattles. This finding is similar to the report by Bacha et al. [24]; Daya, Abebe [24]; Teka, et al. [12] and Lelisa et al. [14]. So, poor body condition cattles are less immune to infection than medium and good body condition score cattles.

This study indicates that Trypanosomosis prevalent in male cattle 69(19.4%) than female cattle 32(13.1%). Different workers reported

that higher prevalence was observed in male cattle than in female ones Afewerk [25]; Tewelde [26] and Muturi [20].The possible suggestion to this finding could be that male cattles are more used for draught purposes, travel long distances to an area of tsetse challenge for grazing or plowing and stressed by draught power and as a result the risk of contracting trypanosomosis is higher.

In this study a higher infection rate was observed in age categories of 4-5 years 52(18.8%). This could be associated to the fact that majority cattles of age 4-5years are productive which travel long distance for grazing and draught as well as harvesting crops in areas of high to tsetse challenge. In the results of Rowlands et al. [22] in Ghibe valley indicated that suckling calves donot go out with their dams but graze at home steads until they are weaned off. Young animals are also naturally protected to some extent by maternal antibodies report of Fimmen et al. [27]. This could result in low prevalence in calves and also *Trypanosoma congolense* infection is a chronic disease increasing with age of animals. Infection by *Trypanosoma congolense* is usually higher in adult animals than young according to Mc Dermott et al. [28,29].

The overall mean of Packed Cell Volume (PCV) value of examined cattle was 22.38 ± 5.022 and significantly different $P < 0.05$. The mean PCV of Aparasitic cattle was higher 22.68 ± 4.966 than parasitic cattle 20.86 ± 5.044 . In fact the difference in mean PCV between parasitaemic cattles and Aparasitaemic cattles indicated that trypanosomosis may be involved in adversely lowering the PCV values of infected animals. Parasitaemic cattles had generally lower mean PCV than the corresponding Aparasitaemic cattles. Though, there is appearing of parasitological negative cattles within the PCV values of less than the threshold value. This may be due to inadequacy of detection method as Murray [12] or delayed recovery of anaemic situation after current treatment with trypanocidal drugs and may be other blood parasites infection, malnutrition associated with long draught in the areas. While the occurrences of positive animals with PCV greater than or equal to 25% might be thought of recent infection of animals.

Conclusion and Recommendations

The results of the present study revealed that *Glossina* species are the most responsible for the spread of bovine trypanosomosis in the study area which is the most important problem for agricultural activity and improvement of animal production and productivity. According to entomological survey result in the Sayonole district four species of tsetse flies *G.f.fuscipes*, *G.tachinoides*, *G.m.submorsitans* and *G.pallidepes* and other biting flies *Tabanus* and *Stomoxys* were captured. Parasitological survey result shows that *Trypanosoma congolense*, *Trypanosoma vivax* and mixed infection of both was identified in the study area and *Trypanosoma congolense* was the dominant trypanosomes species identified in the area. In case of heamatological survey result PCV<25% was principal on poor body condition score cattles. The situation of bovine trypanosomosis in the area is getting worse as no activities of the control and prevention of trypanosomosis was taken place in the Sayonole district.

Therefore, Designing and implementation of control strategies of trypanosomosis focusing on sustainable, community based, simple, cost effective, environmentally friendly and integrated approach should be undertaken and Proper and strict follow-up of trypanocidal drugs treatment should be conducted in the Sayonole district western Oromia, Ethiopia.

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