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Prevalence of Bovine Trypanosomosis and Their Infection Rate in Vector in Sokoru Wereda, Upper Ghibe Valley, Western Ethiopia

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Abstract: The study was conducted to determine the prevalence of bovine trypanosomosis and infection rate in vector and total of 917 tsetse and other biting flies were caught from 72 deployed monoconical traps during the study period. Out of these, *Glossina* accounts n= 804(87.67%) and other biting flies includes n=113(12.3%). The apparent fly density was found to be 5.58 flies/trap/day for *Glossina* species and 0.78 flies/trap/day was accounted for other biting flies. From total of 150 tsetse flies dissected which includes 82 for *G. fuscipes* and the remaining 68 were *G pallidipes* the overall trypanosome infection rate was 6.6%. More trypanosome infections were observed in *G pallidipes* with an infection rate of (n=9) 6% and for *G. fuscipes* only (n=1) 0.6%. Generally, 2.66% (or 4/10) of the trypanosome infections carried by the female tsetse were identified as the Duttonella group; which were classified as *T. vivax* and the 2.2% (3/10) were Nanomonas; "T. congolense type. There was significant difference in the proportion of tsetse infected with trypanosomes between male and female flies. Moreover, there was a strongly significant difference (p = 0.00) between hunger stages which indicates that there was no infection of trypanosomes in teneral flies as more feeding and engorged flies were highly susceptible. The overall trypanosomosis prevalence was found to be 12.13 % including (n=1) 0.23%, (n=6) 1.38%, (n=26) 5.95% and (n=20) 4.58% Adama, Doyo kobota, Ghibe and Medalle peasant associations respectively was recorded. High prevalence was observed in Ghibe (n=26) 5.95% while oppositely, low trypanosomosis prevalence was observed in Adama (n=1) 0.23%. This study confirmed the presence of *T.vivax*, *T. congolonse* and Mixed infection (*T.vivax* and *T. congolonse*) with the prevalence of (n=16) 3.67%, (n=33) 7.56% and (n=1) 0.23% respectively. This finding indicates that the study sites (PAs) strongly affect the overall prevalence. PCV for all study animals was analyzed to estimate the degree of anemia. The mean PCV of the present finding of parasitemic n=39 (8.9%) was significantly lower than that of aparasitemic animals n=14 (3.2%). Moreover, most of parasitemic cattle are anemic so that trypanosomiasis strongly cause anemia. Therefore, vector controlling and treating infected cattle with prophylactic or chemotherapeutic measures should be given to mitigate the problem in the study area.

Key words: *Glossina pallidipes* · *Glossina fuscipes* · Infection rate · Prevalence · Trypanosomosis · Cattle · Sokoru Woreda

INTRODUCTION

Despite the large animal population; production and productivity is very low in Ethiopia and even below the average for most countries in eastern and sub-Saharan African countries. This is due to poor nutrition, reproduction insufficiency, management constraints and prevailing animal diseases [1]. 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.

Animal trypanosomosis is a disease of domestic animals resulting from infection with parasitaemic protozoa of the genus *Trypanosoma* transmitted primarily by tsetse fly and also by other haematophagous flies. Trypanosome parasitizes all classes of vertebrates including human beings and it is predominantly a parasite

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of blood. Animals affected with trypanosomosis become anaemic and weak, lose weight and body condition, reduced productivity and often mortality rates are high [2].

In Ethiopia about 180, 000 to 200, 000km^2 of agriculturally suitable land in the west, southwest and north western low lands and the associated river basins of the country are infested by tsetse fly and Trypanosomosis, making this land underutilized. In those areas, there are 14 millions heads of cattle, an equivalent number of small ruminants, nearly 7 million equines and 1.8million camels are at risk of contracting trypanosomosis at any time [3, 4].

To control the disease and its vector; tsetse fly, a base line data concerning tsetse density and trypanosomosis prevalence in the endemic areas like sokoru district is mandatory.

Therefore, statement problem of the study was based on lack of any organized finding on study area and study parasite, vector and associated risk factors at selected study areas including infection rate.

Therefore, the objectives of this study were:

- To determine the prevalence of bovine trypanosomosis in the study area,
- To determine the infection rate in vectors. *pallidipes*and *G. fuscipes* at the study area,
- To analyze risk factors

MATERIALS AND METHODS

Study Area: The present study was conducted in four peasant associations (PAs), namely: Medalle, Ghibe, Doyo kobota and Adama in Sokoru woreda of Jimma Zone in Oromia regional state South West Ethiopia. The woreda is located at about 180 km west of Addis Ababa with the altitude ranging from 1160 to 2940 m above sea level; the highest points include Ali Shashema, Ali Derar and Kumbi. Perennial rivers include the Gilgel Gibe a tributary of the Gibe and the Kawar; seasonal streams include the Melka Luku. A survey of the land in this woreda shows that 36.6% is arable or cultivable, 16.8% pasture, 17.2% forest and the remaining 29.4% is built-up or degraded. The Abelti-Gibe State Forest covers 159 square kilometers of the forested area. Teff is one important cash crop. Although coffee is another important cash crop of this woreda, less than 20 square kilometers are planted with this cropat longitude 8.27 N and 36’ 21 E and latitude 8.45° N and 36.35° E respectively [5].

Based on figures published by CSA [6] the woreda has an estimated total population of 157, 552, of whom 79, 305 were males and 78, 247 were females; 19, 676 or 12.49% of its population are urban dwellers, which is about the same as the Zone average of 12.3%. With an estimated area of 923.44 square kilometers, Sokoru has an estimated population density of 170.6 people per square kilometer, which is greater than the Zone average of 150.6.

Agriculture is the main stay of livelihood of people with a mixed farming system and livestock plays an integral role for agriculture. The major livestock kept in the study area are cattle, goats, sheep and equines. According to the information from Agricultural offices of the woreda 2000, there are about 68317 cattle, 24192 goats, 23607 sheep and 7134 equines poultry 48400. Animals are left in communal grazing area far away from farm area or residential areas under close supervision by the owners.

Study Design: A cross-sectional study was conducted to determine the trypanosome infection rate and population density of *G. pallidipes*and *G.fuscipes* and prevalence of trypanosomosis in cattle, at study area.

Study Animals: The target populations were local breeds of 437 cattle of all age group and sex found in each site and all G.*pallidipes*and *G.fuscipes*flies collected from deployed traps.

Sample Size Determination: The sample size was determined by usingThrusfield formula [7]. The simple random sampling technique was used for the study of trypanosomes infection rates in *G. pallidipes, G.fuscipes*and stratified sampling method in cattle based on the herd common characteristics of the population using simple random sampling method and sample sizes were allocated using proportional allocation under which the sizes of samples from different strata were kept. The sample size was determined based on the expected prevalence rate of 50% and absolute desired precision of 5% at confidence level of 95%. As a result, a total of 384 animals were needed to be sampled.

$$N = \frac{1.96^2(P(1-p)}{d^2}$$

where N= the sample size
d= the desired absolute precision
p= the expected prevalence

Study Animals Selection: Simple random sampling techniques were followed to select the animals to be used for the study of the prevalence of bovine trypanosomosis in the study area.

Sample Collection and Laboratory Processing

Sample Collection, Parasitological and Hematological Examinations: Paired blood samples were collected from the auricular vein (Marginal ear vein) of each animal using two haematocrit capillary tubes. The tubes were filled ¾ of its height and sealed with crystal sealant. The capillary tube was also used to measure the PCV values for the determination of anemia and comparison of infected animals with non-infected animals. The capillary tube was cut 1mm below the Buffy coat to include the top layer of RBCs. The content of the capillary tube was expressed on to a clean microscopic slid, mixed and covered with cover slip.

Then the slides were examined for trypanosomes based on the type of movement in the microscopic field. Confirmation of trypanosome species by morphological characteristics were done after staining with Giemsa and examination with oil immersion microscopy under×100 power of magnification according toMurray*et al.* [8].

During sample collection; age, sex, PAs, altitude and body condition of each animal were recorded. On subjective basis body condition of examined animals were evaluated during sample collection. They were classified as poor, medium and good relative to the average body condition of local animals (Zebu) [9].

Entomological Survey and Examination: The entomological data were collected only at one time during the dry period season in February 2017. A total of 72 monoconicalstandard traps were deployed in the study area for tsetse fly trapping. All the traps were baited uniformly with octanol (1-oct-3-nel), acetone and three weeks old cow urine [10]. All odours were placed on the ground about 30 cm upwind of the trap. The poles of traps were greased to prevent fly predators, mainly ants. Traps were allowed to stay at the site of deployment for a period of 48 h before collection. Trap deployment sites were selected to represent all vegetation type/habitat that could be related to fly multiplication, behavior, feeding and other related aspects. After 48 h of deployment, the catchments of each trap was sorted by fly species and then counted, identified and analyzed. The apparent density of the tsetse fly was calculated as the number of tsetse catch/trap/day [11]. The flies were collected from the trap and before dissecting them Sexing was done just by observing the posterior end of the ventral aspect of the abdomen by hand lens and stereomicroscope hence male flies were identified by enlarged hypopygium in the posterior ventral part of the abdomen. Tsetse fly apparent density mean catches in traps deployed was expressed as the number of tsetse catch /trap/ day.

Wings and the legs were removed from the flies. The dissection was carried out as described by the FAO Training manual for tsetse control personnel [12]. Then, freshly killed tsetse flies were dissected under a dissecting microscope by using 0.9% normal saline. Trypanosome infections in the tsetse flies were identified using a compound microscope at a magnification of ×400, using the methods ofLemecha[13]. Parasites found in the midgut, salivary glands and mouth parts were regarded as Trypanozoon; "T. brucei-type", those located in the mouth parts and midguts were Nanomonas; "T. congolense-type", while those found in the mouth parts only was put in the group of Duttonella; "*T. vivax*type infection", immature infections, when only the midgut was found infected. The Infection rate (IR) was calculated using the following formula:

Data Analyses: All statically analyses were performed using STATA-7 soft ware. The prevalence was calculated for all data as the number of infected individuals divided by the number of individuals sampled times 100. Categorical data were analyzed by using chi-square (χ^2) test of independence where as t-test was used to examine the difference in mean PCV between the study variables. In all cases, 95% of confidence intervals were used and p value less than 0.05 were considered as significant.

RESULTS

Entomological Survey: A total of 917 tsetse and other biting flies were caught from 72 deployed monoconical trapsduring the study period. Out of these, *Glossina* accounts n=804(87.67%) and other biting flies includes n=113(12.3%). The apparent fly density was found to be 5.58 flies/trap/day for *Glossina* species and 0.78 flies/trap/day was accounted for other biting flies. The composition of *Glossina* species identified in the present study were 19.65% (n=158) *G. morsitans sub morsitans*, 25.5% (n=205) G*fuscipes*and 54.85 %(n=441) were G *pallidipes*. Finally, from all collected *Glossina* species n=658 (81.84%) were females and other n=146(18.16%) were males.

Table 1: Proportion of *Glossina fly* species collected and their density in different peasant associations.

	Fly species								
PA's	*G. fuscipes*	*G. pallidipes*	*G. morsitans*	Total no of fly	FTD	No of trap	Alt	Long (N^0)	Lat (E^0)
Medalle	102	78	47	227	1.58	18	1088	08247.00	037539.44
Ghibe	87	92	48	227	1.58	18	1080	08137.49	037346.93
Doyo kobota	0	139	52	191	1.33	18	1501	08130.81	037288.40
Adama	16	132	11	159	1.10	18	1491	07400.22	037245.86
Total	205	441	158	804	5.58	72			

FTD= flay/ trap/ day

Table 2: The number of flies dissected and infection rate of *G. pallidipes* and *G.fuscipes* based on sex, age and hunger stage.

			Dissected fly species		Trypanosome species found				
Risk factors		No of fly dissected	*G.pallidipes*	*G.fuscipes*	*T.congolonse*	*T.vivax*	Total infected	Over All (IR %)	Significance (p value)
Sex	Male	27	11	16	1	2	3	2.0	
	Female	123	57	66	3	4	7	4.66	
	Total	150	68	82	4	6	10	6.6	0.05
Age	Young	58	23	35	1	1	2	1.33	
	Old	92	45	47	3	5	8	5.3	
	Total	150	68	82	4	6	10	6.6	0.05
Hunger stage	Teneral fly	34	11	23	-	-	0	0.0	
	Hunger	32	13	19	-	1	1	0.6	
	Feed	67	35	32	3	3	6	2.0	
	Engorged	17	9	8	1	2	3	1.3	
	Total		150	10	10	6.6	0.00		
Total number of		150	68	82	4	6	10	6.6	0.00

A total of 150 tsetse flies were dissected which includes 82 for G *fuscipes* and the remaining 68 were G *pallidipes* during the study period. The overall trypanosome infection rate was 6.6%. More trypanosome infections were observed in G *pallidipes* with an infection rate of (n=9)6% and for G *fuscipes only (n=1) 0.6%.* More trypanosome infections were observed in female tsetse with an infection rate of 4.66% (Table 2). Generally, 2.66% (or 4/10) of the trypanosome infections carried by the female tsetse were identified as the Duttonella group; which were classified as *T. vivax* and the 2.2% (3/10) were *Nanomonas;* "*T. congolense*type.There was significant difference in the proportion of tsetse infected with trypanosomes between male and female flies ($P = 0.05$) and also an age related effect in the number of trypanosome infections detected by microscopy with number of infected flies older than 31 days being significantly higher than those aged < 20 days *(P* <0.05). Moreover, there was a strongly significant difference (*p = 0.00*) between hunger stages which indicates that there was no infection of trypanosomes in teneral flies as more feeding and engorged flies were highly susceptible.

Parasitological Findings: The research was conducted in 4 selected peasant associations (PAs) of Sokoru wereda. Out of the total of 437 local breeds of cattle examined, 53 animals were found positive for trypanosomosis. The overall trypanosomosis prevalence was found to be 12.13 %including (n=1) 0.23%, (n=6) 1.38%, (n=26) 5.95% and (n=20) 4.58% Adama, Doyo kobota, Ghibe and Medallepeasant associations respectively was recorded. High prevalence was observed in Ghibe (n=26) 5.95% while oppositely, low trypanosomosis prevalence was observed in Adama(n=1) 0.23%.

This study confirmed the presence of *T. vivax*, *T. congolonse* and Mixed infection (*T. vivax* and *T. congolonse*) with the prevalence of (n=16) 3.67%, (n=33) 7.56% and (n=1) 0.23% respectively. Therefore, *T.congolonse* was the most prevalent parasite at the study area with 7.56% rate. This finding indicates thatthe study sites (PAs) strongly affect the overall prevalence.

To evaluate the debilitating effect of trypanosomosis in diseased cattle which were living under similar environment and management systems, observed the following prevalence in different body condition scores of animals: (n=7) 1.6%, (n=28) 6.4% and (n=18) 4.11% in Good, Medium and Poor body condition respectively. The prevalence of trypanosomiasis with in different age group was found to be (n=23) 5.3% in young's and (n=30) 6.9% in adults. Finally, at the study area male cattle's were more susceptible than female with the rate of (n=29) 6.64% and (n=24) 5.49%.

Table 3: Prevalence of trypanosomosis in four peasant association (PAS)

Sites (PAs)	Positive	Total	Prevalence	P-value, x^2
Adama	1	100	0.23	
Doyo kobota	6	100	1.38	
Ghibe	26	136	5.95	
Medalle	20	101	4.58	
Total	53	437	12.13	0.00, 29.96

P-value, $x^2$0.00 and *29.96*

Table 4: Summary of the prevalence of trypanosomosis in different peasant associations and by different species of Trypanosomes

Site (PAs)	Examined animals	Adama	Doyo kobota	Ghibe	Medalle	Total	Prevalence (%)	P-value, x^2
T. vivax	437	0	3	7	6	16	3.65	0.002, 30.7
T. congolense	437	1	3	17	12	33	7.55	
Mixed (*T. vivax* and *T. congolonse*	437	0	0	2	2	4	0.92	
Total	1	6	26	20	53	12.13		

P-value, x2, 0.002, 30.7

Table 5: The prevalence of trypanosomosis based on different risk factors

		Test result				
Risk factors		Positive	Negative	Total	Prevalence	P- value, x^2
Sex	Male	29	213	242	6.64	0.9, 0.15
	Female	24	171	195	5.49	
Age	Young	23	160	183	5.3	0.8, 0.06
	Adult	30	224	254	6.9	
Body condition	Good	7	58	65	1.6	0.7, 0.56
	Medium	28	215	243	6.4	
	Poor	18	111	129	4.11	

Table 6: Hematological finding result

				Buffy coat	
No	PCV	No	Prevalence (%)	Positive	Negative
1	Anemic	196	8.933	39	157
2	Non- anemic	241	3.2	14	227
Total	384	12.13	53	384	

P value, x^2 0.00 and 20.13

PCV for all study animals was analyzed to estimate the degree of anemia. From the total n=437 animals; (n=196) 44.85% are anemic and (n=241) 55.15%are non anemic. The mean PCV of the present finding of parasitemic n=39 (8.9%) was significantly lower than that of aparasitemic animals n=14 (3.2%). Moreover, most of parasitemic cattle are anemic so that trypanosomiasis strongly cause anemia (*p value, x^2 0.00 and 20.13)* respectively.

DISCUSSION

According to the present finding, total of 917 tsetse and other biting flies were caught from 72 deployed monoconical traps during the study period. Out of these, *Glossina* accounts n= 804(87.67%) and other biting flies includes n=113(12.3%). The apparent fly density was found to be 5.58 flies/trap/day for *Glossina* species and 0.78 flies/trap/day was accounted for other biting flies. This was in agreement withLeak *et al*. [14] who concluded the distribution and abundance of tsetse flies to be closely associated with the number and habitats of certain wild animals and also described that the highest densities of certain tsetse fly species are reported from areas with very high densities of wild animals and low human population areas.It's also in line with the finding of Bure wered, Iluababor zone of Western Ethiopia which was reported to be 7.23% flies/trap/day andreported 8.55 flies/trap/day for *Glossina* species in Diga and Sasiga districts, East Wollega Zone [15]. Highest prevalence 23.0% was also reported byAyele *et al.* [16] in Daremello district, Southwestern Ethiopia. These kinds of differences may be resulted due to previous control intervention with insecticides at study area.

The overall trypanosome infection rate was 6.6% in dissected *G. pallidipes* and *G. fuscipes* flies. More trypanosome infections were observed in *G pallidipes* with an infection rate of (n=9) 6% and for *G. fuscipes* only (n=1) 0.6% this finding was in agreement with Desta *et al.* [17] reported infection rate of G. *pallidipes* 6.6%. However, least infection rate 0.6% by G. *fuscipes* was due to the relatively low fly infection rate and trypanosome prevalence as compared to low tsetse challenge can be explained by the higher fly- animal contact and trypanosome-binding lectin proteins (D+ glucosamine and D+galactosamine) which makes *G. fuscipes* relatively resistant than *G. pallidipe* [18]. The reason for a higher infection rate in females might be due to their better life expectancy and lower infection rate found in male flies can be explained by the low average age of trapped male flies (20 days or less). There was significant difference (P <0.05) in tsetse flies density between surveyed peasant associations Medalle, Ghibe, Doyo kobota and Adama ranging from 1.58 to 5.58. This might be attributed to the altitude and vegetation type and coverage of the two sites. The trypanosome infection rate in a population of tsetse may vary with sex, age and the sampling methodLangridge[19]. Sex ratio and age composition of the flies were assessed in this study and higher numbers of female and adult flies were reported.

The predominance of *T. congolense* infection in cattle under sufficient number of cyclical and mechanical vectors of trypanosomosis may be due to the high member of *T. congolense* as compared to *T. vivax* and *T. brucei* regarding the development of better immune response to other species of trypanosomes by infected animals [20]. The lower infection rate in domestic animals by *T. brucei* than *T. congolense* and *T. vivax,* may be due to the seasonal absence of the parasite in circulation (Parasitaemia) as indiceted byLosos and Chovinard [21] and one might miss many latent infection which only become apparent after rat inoculation.

During the study period, cattle with PCV= 24% were considered anemic [22] which is said to be the principal sign of trypanosomosis in livestock [23]. Furthermore, PCV values can be affected by many factors other than trypanosomosis, but these factors are likely to affect both trypanosomosis negative and positive animals [24]. The resulting low PCV value may not solely be due to trypanosomosis; however, the difference in mean PCV between parasitaemic and aparasitaemic animals indicates that trypanosomosis significantly reduces the PCV values in infected animals (p=0.00).

CONCLUSION

This study presents findings on the trypanosome infection rate of *G. pallidipes* and*G.fuscipes* was 6.6%.Prevalence of cattle trypanosomes in sokoru wereda of Western Ethiopia was 12.13%. The trypanosome infections in vector and host animals wererelative. The study indicated that the presence of *G. pallidipes, G.fuscipes* and *G. m. submorsitans* with 5.58 fly/trap/day.The present study also disclosed that, the prevalence of different species of trypanosomes were found to be highest *T. congolense*; followed by *T. vivax* and mixed infection of both *T. congolense* and *T. vivax*. Therefore, vector controlling and treating infected cattle with prophylactic or chemotherapeutic measures should be given to mitigate the problem in the study area.

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