International Journal of Advanced Multidisciplinary Research ISSN: 2393-8870

www.ijarm.com

DOI: 10.22192/ijamr

Volume 6, Issue 3-2019

Research Article

DOI: http://dx.doi.org/10.22192/ijamr.2019.06.03.001

Prevalence of Bovine Trypanosomosis and apparent fly density of tsetse and biting flies in step intervention area of Arbaminch Zuria Wereda

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Abstract

Cross sectional study was conducted from November 2016 to March 2017 to estimate prevalence of bovine trypanosomosis and apparent density of tsetse fly and biting flies in selected three tsetse controlled area of Arbaminch Zuria Wereda, Gamo Gofa zone. A total of 399 blood samples were collected and examined using a buffy coat technique for parasitological survey and the PCV value of each animal was also measured using hematocrit readers. A total of 240 NGU2 traps were deployed for consecutive 72 hours in selected area of three kebeles and flies in each trap were identified and recorded. The overall prevalence of trypanosomosis was found to be 1.75% where as 3% in kanchama, 0.7% in Elgo, and 1.57% in Sharakebles were recorded. The most positive cases were due to T. congolense (71.4%) followed by T.vivax (28.6%) and there was no mixed infection. The overall mean (±SD) PCV value of tested animals was 25.40±5.8. The study also demonstrated variations in prevalence among different age groups and between both sexes which were statistically insignificant (p>0.05). The prevalence recorded among different sex was 0.91% for males and 1.81% for females without statistically significant variation (P>0.05). In each study area, entomological surveys were conducted using 30 NGU traps 10 in each of the three PA's namely Elgo, Shara and Kanchama for eight round which was in total 240 times trapping. The overall mean apparent density of flies was 3.71flies/trap/day. This preliminary survey in controlled area shows that the use of spot-on and trypanocidal drugs at monthly basis had reduced substantially the prevalence of trypanosomosis. Furthermore, developing the effort in controlling bovine trypanosomosis at the area is recommended.

Keywords

Arbaminch, Biting flies, Prevalence, Trypanosomiasis, Tsetse fly.

1. Introduction

Trypanosomosis is the main haemoparasitic disease in domestic animals and is caused by the protozoan parasite Trypanosoma. The parasite is transmitted biologically by the tsetse fly (Glossina species) and infects animals over an area known as the 'tsetse belt', which extends approximately 10 million km² across 37 countries in Africa, from the Sahara desert in the north to South Africa in the south (Wagari et al., 2012). The disease is very economical in Ethiopia because of its highest prevalence in the most arable and fertile land with high potential for agricultural development in the South West and North West part of Ethiopia along the great river basins of Abay, Omo, Ghibe and Baro which are infested with vector tsetse fly. There are also studies which showed the disease to be equally important in non-tsetse infested highland part of the country as some species of Trypanosoma are transmitted non-cyclically (Cherenet et al., 2006).

The most important trypanosome species affecting livestock in Ethiopia are Trypanosoma congolense, Trypanosoma vivax and Trypanosoma brucei, in cattle, sheep and goats, *Trypanosoma evansi* in camels and Trypanosoma equiperdiumin horses (Wondwesen et al., 2012). The trypanosome caught the loss in huge livestock resource particularly cattle that are utilized as they provide traction power, provide milk, meat, cash income, manure and serve as a capital asset against risk and in general livestock are complementary to crop in highlands (MoA,1997). The relationship between the tsetse transmitted trypanosomes and the vector is highly specific, which effectively limits their distribution to that of the tsetsefly principally areas of Central and southern Africa between 15°N and 29°S. Tsetse-transmitted trypanosome species can usually infect a range of animal species, but cattle and horses are usually more severely affected than small ruminants such as sheep and goats. However, pigs can be severely affected by certain species of trypanosome. Exotic breeds of livestock are often more severely affected than indigenous breeds, which frequently show a degree of resistance to the effects of infection (trypan tolerance) (Service and Ashford, 2001).

In Ethiopia, there is no report of human trypanosomiasis which is the prevailing disease in most tsetse infested sub Saharan African countries. Animal trypanomosis is among the most important diseases which hinder livestock productivity and agricultural development in sub-Saharan Africa including Ethiopia (Ayana *et al.*, 2012).

Genus trypanosome dwells in bloodstream and tissues of final host. However, a few species are of overwhelming importance as a serious cause of morbidity and mortality in animals and man in tropical regions, with exception of *Trypanosoma eqiuperdum*, all others require an arthropod vector. Three elements influence the epidemiology of the disease, namely the distribution of the vectors, the virulence of the parasite (trypanosome) and response of the host (Urquhart *et al.*, 1996).

The life cycle of trypanosomes in tsetse involves cyclical development for a varying length of time, depending on the species and ambient temperatures. T. vivax completes its developmental cycle in the proboscis and pharynx and can be transmitted (as metacyclic trypanosomes) within a week of the initial infective feed. The cycle of *T. congolense* involves the midgut and proboscis and is completed in about 2 weeks. That of *T. brucei* is more complex: it takes 3 or more weeks and involves the midgut and salivary glands. Once infected, flies remain source of infection in life time which is about 1 to 2 months. After trypanosomes have been introduced into a herd, oncyclical transmission is possible even in the absence of Glossina species and presence of other biting files such as Tabanidae (Radostits et al., 2007).

The tsetse flies are widely distributed in western southern and southwestern lowlands and river valleys and 15% of the land believed to be suitable for livestock production is affected by one or more species of tsetse flies such as Glossina morsitans sub morsitans. G. paulidipes. *G*. tachinoides. G. fuscipesfuscipes and G. longipennis. Apart from cyclical transmission of Trypanosomosis by Glossina species, mechanichal transmission is a potential threat to livestock productivity in some parts of Ethiopia. Trypanosoma vivax infection can transmit mechanically by tabanide and large number of biting flies (Ayana et al., 2012). However, strict attention should be paid in preventing the occurrence of reinvasion of such freed sites and consolidate the sustainability of the achieved results. This necessitates a continued follow-up and evaluation of the current status of tsetse infestation and occurrence of trypanosomosis in such controlled sites and their surrounding villages. Currently, the study area is supported by Southern tsetse eradication program (STEP), whose main objectives were to suppress trypanosomosis and tsetse fly; and eventually eradicate tsetse flies from breeding site using sterile insect technique (SIT). Since the beginning of the

project suppression program have been undertaken for the last seventeen years using impregnated targets and traps, application of spoton and regular trypanocidal drug treatment implemented at monthly basis (STEP, 2013; IAEA, 1996). Therefore the objective of this study is to estimate the prevalence of bovine trypanosomosis and estimate tsetse fly and biting flies apparent density in the selected intervention areas of Arbaminchzuria district.

2. Materials and Methods

2.1. Description of the study area

The study was conducted in three selected intervention Kebeles or peasant associations namely Kanchama, Elgo and Shara in Arbaminch Zuria district, Gamo Gofa Zone, South Nation Nationalities and People Regional state (SNNPRs). Arbaminch is located at a distance of 505 Kms South West of Addis Abeba and 208 Kms from the regional town, Hawassa. Geographically, found in 37^{0} 5 east of longitude and 6^{0} North of latitude with altitude of 1200-3125 meters above sea level. The area has a sub-humid climate with a moderately hot temperature. The vegetation is dominantly occupied by wooden grass land. The annual rain fall ranges from 750-930millimeters with mean average temperature of 30° c. The town is situated in the well-known East African Rift valley and surrounded by Lake Chamo and Abaya as well as Nech-sar national park. The main occupation of the rural population is mixed farming system whereby crop and livestock production are managed. Main crop is maize and vegetation that includes coffee, banana, papaya, mango, avocado, inset, apple and others. The livestock populations in the Gamo Gofa Zone includes cattle 1,243,017, sheep 196,575, goats 543,385 and equine 58,664 (GZARDO, 2007).

2.2. Study design and study animals

A cross sectional study design was used to collect primary information to estimate the prevalence of trypanosome infection and the apparent density of its vector tsetse fly in the study areas. The study populations were local zebu cattle from three kebeles or peasant associations (PAs) such as Elgo, Shara and Kanchama. The age of animals was also determined by dentition in three different catagories as stated by Gatenby (1991).

2.3. Sample size determination

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 (2005) by using 50% of expected prevalence and 95% confidence interval as follows;

$$N=\frac{1.96^{2}(Pexp)(1-Pexp)}{d2}$$

Where N = the required sample size, Pexp= the expected prevalence and d = the desired absolute precision.

Accordingly, the calculated sample size was 384. However, the sample size was increased to 399 to increase the precision.

2.4. Study Methodology

2.4.1. Parasitological survey

То determine the prevalence of bovine trypanosomosis, blood that was taken from each animal into heparinized capillary test tubes was sealed and heparinized capillary tube containing blood was centrifuged for 5 minutes at 12,000 revolutions per minute. After the centrifugation, tubes were then placed in hematocrit reader and recorded for PCV. Then, the readings were expressed as a percentage of packed red cells to the total volume of whole blood. Animals with Packed Cell Volume (PCV < 25%) were considered to be anemic (Leak et al., 1987). Trypanosomes were usually found in or just above buffy coat layer. So, capillary tube was cut using a diamond tipped pen 1 millimeter below the buffy coat to include the upper most layers of the red blood cells and 3 millimeters above to include the plasma. The content of the capillary tube was poured on to slide, homogenized on to a clean glass slide and covered with cover slip. The slide was examined under 40x objective and 10x eye pieces for the movement of parasite. The Species were identified based on the movement characteristics of Trypanosomes under the microscopic field (Murray et al., 1977).

2.4.2. Entomological survey

For the entomological study, tsetse flies including biting flies were collected using 30 NGU traps in different positions of the study areas 10 traps in each kebeles. Acetone and cow urine were used as a bait to attract the flies. Traps were positioned at approximate intervals of 100 to 200 metters for 72 hours in cultivated lands in Elgo and Shara and wood and bush lands in Kanchama. Fly catches were employed for eight round after every fortnight. A total of 240 trapping were taken place during study period. The apparent densities of tsetse and biting flies were determined based on the mean fly caught. The flies caught per trap were identified, counted and apparent fly density per trap per day (f/t/d) was recorded. The tsetse flies were identified to species level (Marquardt *et al.*, 2000).

2.5. Statistical analysis

Raw data were entered into a Microsoft Excel spreadsheet and descriptive statistics were used to summarize the data. The point 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 logistic regression. All statistical analyses were conducted using STATA version 9.0 software. The test result was considered significant when the calculated p-value was less than 0.05.

3. Results

3.1. Parasitological survey

Out of total 399 examined animals, 7 were found to be positive for trypanosomosis. The overall prevalence of bovine trypanosomosis in the study area was 1.75%. The prevalence of bovine trypanosomosis in Kanchama, Elgo and Shara Kebeles were 3%, 0.7% and 1.57% respectively. Out of the three selected Kebeles (Peasant Associations), Kanchama had the highest prevalence (3%) and Elgo got the lowest prevalence (0.7%)(Table 1). The most prevalent trypanosome species in the study area was *Trypanosoma congolense*(71.4%) followed by *T. vivax* (28.6%). The highest prevalence of *Trypanosoma congolense* was recorded in Kanchama PA`s with a rate of 57.14%. During study period, mixed infection and *T. brucie have* not been recorded (Table 2).

3.2. Potential risk factors

Sex was one of the variables that were considered during the study period. Of 189 males and 210 females

examined, 4 (2.12%) and 3 (1.43%) were trypanosome positive respectively but there was no significant difference (p > 0.05) between sex categories. Age was categorized into three groups based on estimated age ranges and out of 399 animals sampled, 17 (4.3%), 181 (45.4%) and 201 (50.4%) were under age group less than 1 year, 1-5 years and >5 years respectively. In each group, there were 1 (5.88%), 4 (2.4%) and 2 (0.99%) trypanosome positive animals respectively. However, there was no significant difference (p > p)0.05) among the age groups as shown in Table 3 below. The color of the animals was also considered as a variable during data collection to detect whether the color difference has role on fly attraction or not. Accordingly, white and red colored animals were categorized as bright which are refractory to light; Black and brown colored animals were categorized as dark group and assumed as attractive for tsetse to perch. However, out of the total animals examined, 2 (1.49%) on white and related, 3(1.58%) on red and related colored and 2 (2.67%) on dark colored group were positive for trypanosomosis and statistically, there were no significance (p > 0.05) among the three groups (Table 3).

3.3. Hematological examination

PCV value of individual animals was measured using packed cell volume (PCV) reader. Statistical analysis was made to compare mean PCV value of infected and non-infected animals and other variables as shown in Table 4. As the findings revealed, infected animals had lower mean PCV than non-infected animals. The overall mean (±SD) PCV value of tested animals was 25.40 ± 5.7 . The mean PCV of infected bovines (24.6 \pm 5.6) was not significantly (p > 0.05) associated with that of non-infected bovines (26.88 ± 5.8) . In general, 36.8% of the total observed animals had PCV below the normal range that showed the anemic condition of animals. The current finding indicates that the animals affected with trypanosomosis were found in higher rate in kanchana and hence, the animals in the area with the higher prevalence got anemic (Table 4).

3.4. Entomological survey

A total of 2923 flies were caught in the current study area and of these, 2236 (76.5%) belong to Glossina species (tsetse flies), 687 (23.5%) were Tabanus and other remaining flies were belonging to biting flies. Furthermore, all Glossina species caught were *G. pallidipes* and both tsetse and biting flies were caught in all sites surveyed. The apparent fly density was 3.53 and 1.12 flies/trap/days (F/T/D) for both tsetse and biting flies respectively. The overall fly density was 3.71 flies/trap/days (F/T/D. The number of fly counted

was significantly associated (p < 0.05) with tsetse and other biting flie.

Table 1. The overall Prevalence of Trypanosomosis in three PA's of Arbaminchzuria district.

PA`s	N	Positive	Prevalence
Kanchama	131	4	3%
Elgo	141	1	0.7%
Shara	127	2	1.57%
Total	399	7	1.75%

Table 2; Prevalence of Trypanosoma species in each three PA's

Species	PA`s								
	Kanc	hama Elgo Shara		Total					
	n	%	n	%	n	%	Ν	Prevalence (%)	
T.vivax	-	-	-	-	2	28.6	2	28.6	
T. congolense	4	57.14	1	14.28	-	-	5	71.4	
T. brucie	-	-	-	-	-	-	-	-	
Mixed	-	-	-	-	-	-	-	-	
Total	4	57.14	1	14.28	2	28.6	7	100	

Table 3. Association between prevalence Trypanosomosis and the risk factors

	Risk factors	N	Positive	Percentage	95% CI	P-value	
Sex	Sex Female		3	1.43%	0.37 - 4.46	0.6	
	Male	189	4	2.12%	0.15 - 3.03		
Age	<1 year	17	1	5.88%	0.31 - 30.76	0.4	
	1≤5 years	181	4	2.21%	0.29 - 26.24		
	> 5 years	201	2	0.99%	0.53 - 72.35	_	
Coat	Black and related	75	2	2.67%	0.46 - 10.2	0.14	
color	Red and related	190	3	1.58%	0.28 - 10.43		
	White and related	134	2	1.49%	0.25 - 13.11		

Table 4. Mean PCV comparison

Factors		Ν	Mean PCV	<u>+</u> SD	p-value	
Health condition of	Non infected	392	26.88	5.8		
animal	Infected	7	24.6	5.6	0.30	
	Total	399	25.40	5.7		
Kebele (PA`s)	Kanchama	131	23.3	6.30		
	Elgo	141	26	5.61	0.80	
	Shara	127	26.7	4.80		

4. Discussion

The study showed that out of a total 399 randomly selected cattle in study area, 7 (1.75%) of them were positive for trypanosome, of which 2 (1.57%) in Shara, 1 (0.7%) in Elgo and 4(3%) in Kanchama. The trypanosomosis prevalence recorded in Kanchama was higher than the remaining two Peasant associations. Generally, high prevalence of the disease was found in place where with highest tsetse fly density area hence, the highest fly density among the three Peasant associations was recorded in Kanchama. This result agrees with previous result obtained by Soud (2008) who concluded that both the apparent density of flies and prevalence of trypanosomes were closely related. The present study revealed 1.75% prevalence of trypanosome infection which was lower than the finding reported by Wondwosen et al. (2012) at the same area (i.e. Elgo and Shara which was 4.43%). The possible suggestions could be frequent controlling of tsetse fly by Southern tsetse eradication project (STEP). The project applies variety of tsetse fly control methods, such as 1% deltamethrin pour-on and spot-on at monthly interval, yearly spraying of insecticides on the forest areas and bush lands where high numbers of flies are believed to exist, traps and insecticide impregnated targets and ground spraying which significantly reduced the prevalence of disease in the study area (STEP, 2013). Another possible reason for the low prevalence of the disease might be the prophylactic treatments with trypanocidal drugs, which obviously mask the epidemiological situation of the disease.

In the current study, the highest infection rate (3%) was recorded in Kanchama and the lowest was recorded in Elgo (0.7%). The prevalence among peasant associations in this study could be attributed to the tsetse fly and/or other biting flies' population in each peasant association which may be dependent on micro climate, animal herd density, distance between herds and breeding site. During the study period, the prevalence of bovine trypanosomosis was assessed between sexes of animals and from 7 trypanosome positive animals; 3 (1.43%) of them were female animals and 4 (2.12%) of them were male animals. This shows that both male and female cattle were at most equally susceptible to trypanosomosis infection. There was no significant difference between sex categories of study animals. This result agrees with previous results of Daya and Getachew (2008) who reported no significant difference in susceptibility between the two sexes.

Out of the two species of trypanosomes detected in study period, T. Congolese 5 (71.4%) was the most prevalent trypanosome species followed by T. viviax 2 (28.4%). The dominancy of T. congolense in the present study indicates that transmission was mainly by tsetse flies than biting flies. Entomological survey had also indicative that where tsetse flies catch was higher than biting flies. The finding was in agreement with previous results of Tewelde et al. (2004) at Kone (75%) settlement areas of Western, Ethiopia, Woldeyes and Aboset (1997) at Arbaminchzuria districts (85.2%) and Rowland et al. (1993) in Ghibe valley, south West Ethiopia (84%). However, Terzu (2004) in selected sites of southern region (63.4%), Bitewet al. (2011) in West Gojam (54.3%), Zecharias and Zeryehun (2012) in Arbaminch reported higher proportion of T. congolense than T. vivax. The increased proportion of infection with T. congolense in the study area might be due to the major cyclical vectors of the savannah tsetse flies, G. Palidipes, which are effective in transmitting *T. congolense* than T. vivax (Langrigde, 1976; Solomon, 1997) since the study area located in the tsetse belt of Ethiopia.

The overall mean (\pm SD) PCV value of cattle examined during the study period was 25.40 \pm 5.8. The mean PCV of non-infected cattle was slightly higher (26.88 \pm 5.8) than thatof infected animals (24.6 \pm 5.6). Though there was difference in mean PCV between parasitaemic animals and aparasitaemic animals which indicated that trypanosomosis may be involved in adversely lowering the PCV values of infected animals. Parasitaemic animals had generally lower mean PCV than the corresponding aparasitaemic ones. This might be due to inadequacy of detection method 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 (Murray *et al.*, 1977).

5. Conclusion and Recommendations

The finding of the current study in bovine trypanosomosis prevalence and apparent fly density survey in the three selected PA's of Arbaminch Zuria district indicated that an overall prevalence of 1.75% and overall apparent fly density of 3.71 flies/trap/day during the study period. Out of the three PA's,Kanchama was found to have higher prevalence of trypanosomosis and higher apparent fly density. *G. pallidipes* was the only species of tsetse fly identified in the study area. The age, coat color and sex were

taken as a risk factor in this study but none of the factors were found to be significant. From both the entomological and parasitological results, it concluded as low prevalence of the disease despite the presence of high number of both tsetse and biting flies was mainly due to the effectiveness of one of the control strategies that was spot on application of 1% deltamethrin together with regular trypanocidal drug treatment control strategies which implemented by Southern Tsetse Eradication Project. In light of the above conclusion, the following recommendations are forwarded:

• Designing and implementation of control measures for trypanosomosis focusing on participatory packages on public awareness creation should be undertaken in the Arbaminch ZuriaWereda.

✤ The current effort in controlling bovine trypanosomosis including its vector should be strengthened to the extent of tsetse fly eradication.

Studies on the resistance of trypanocidal drugs should be conducted in the study area.

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How to cite this article:

Ayele Anjulo, Biruk Alemu and Beredu Yohannes. (2019). Prevalence of Bovine Trypanosomosis and apparent fly density of tsetse and biting flies in step intervention area of Arbaminch Zuria Wereda. Int. J. Adv. Multidiscip. Res. 6(3): 1-8.

DOI: http://dx.doi.org/10.22192/ijamr.2019.06.03.001