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A Study on Prevalence of Bovine Trypanosomosis and Associated Risks in Mao Komo Special District of the Benishagulgumuz Regional State, Western Ethiopia

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Abstract: A cross sectional study was carried out in Mao Komo special district of BenishangulGumuz Regional State, Western Ethiopia from December to November, 2016 to determine the prevalence of trypanosomosis, prevailing species of trypanosomes, associated risks and its vector density. Blood samples collected from 385 randomly sampled cattle (*Bos indicus*) was examined using parasitological (buffy coat technique) and haematological (Measurement of packed cell volume) procedures. An overall, 62 (16.10%) prevalence of trypanosomosis was recorded. The infection was caused mainly by *Trypanosoma congolense 34/62 (*54.84%), *Trypanosoma vivax* 19/62 (30.65%), *Trypanosoma brucei* 7/62 (11.30%) and to less extent mixed infection with *Trypanosoma congolense* and *Trypanosoma vivax* 2/62 (3.21%) and the infection rate was statistically significant among different trypanosome species (P<0.05). Mean packed cell volume (PCV) value of infected animals was lower (17.11% ± 5.42) than non-infected animals (25.42% ±3.66) and the variation was found statistically significant (P<0.05). Similarly, higher prevalence (26.35%) of trypanosomosis infection was registered in animals with poor body condition when compared to animals with medium (12.60%) and good (2.2%) body condition and the difference was statistically significant (P<0.05). In contrast, prevalence of trypanosomosis was not statistically significant among study sites, age categories and sex groups of study animals (P> 0.05). *Glossina morsitans, submorsitans, Glossina fuscipes* and *Glossina pallidipes* were the tsetse fly species caught and their mean apparent density measured as flies/trap/day was 1.41. In addition, other mechanical vectors such as tabanids and haematopota were captured with flies/trap/day 0.64 and 0.40, respectively. To wrap up, the result of the current finding reveals moderately high prevalence of trypanosomosis in the study district signaling the need for strategic and participatory approach to control the vector and to minimize the impact of the disease in the study district.

Key words: *Glossina* · PCV · Tabanids · *Trypanosoma* · Tsetse Fly

INTRODUCTION

African trypanosomes are flagellated protozoa that inhabit the extracellular compartment of host blood, in the face of the immune system which they float by switching among distinct antigenic types. The spindle-shaped parasites are about 20 µm long and 2 µm in diameter at their widest point, have a single flagellum and are motile. The African trypanosomes are transmitted to mammals in saliva deposited by biting tsetse flies (*Glossina* spp.) in which they undergo cyclic development. Tsetse flies infest the humid and semi-humid zones of sub-saharan Africa. There are several species of *Glossina* that are capable of transmitting trypanosomes, and also several species of African trypanosomes, some of which have a profound effect on the economy of sub-Saharan Africa. Among these, *Trypanosoma brucei brucei, Trypanosoma congolense* and *Trypanosoma vivax* are notable because they cause Nagana, a wasting and generally fatal disease in cattle [1].

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Nagana is endemic throughout the humid and semi-humid zones of Africa coincide with the distribution of tsetse flies, which infest an area of some 10 million Km^2 embracing 36 countries. About 30 % of African cattle graze on the fringe of the tsetse habitat, many sustained by chemotherapy and tsetse control programs. Regions of high tsetse and trypanosome challenge, which account for some 70 % of the humid and semi-humid zones of sub-Saharan Africa, are devoid of cattle and hence of integrated cattle and market garden systems. The absence of cattle and other domesticated animals for traction, forage conversion to fertilizer and livestock contribution to the small holder economy, makes the classic agrarian model of societal development inapplicable to this region [1].

Trypanosomosis is a major constraint contributing to direct and indirect economic losses to crop and livestock production [2] and has a significant negative impact on economic growth in many parts of the world [3-5].

The most important trypanosome species affecting livestock in Ethiopia are *T. congolense*, *T. vivax*, and *T. brucei* in cattle, sheep and goats, *T. evansi* in camels and *T. equiperdium* in horses [2]. The influence of tsetse flies on African agriculture through the transmission of trypanosomosis continues to be a major constraint to the development of national economies and their achievement of self-sufficiency in basic food production. The general distribution of tsetse flies is determined principally by climate and influenced by altitude, vegetation, and presence of suitable host animals [6]. Tsetse flies in Ethiopia are confined to southern and western regions between longitude of 33 ° and 38° East and latitude of 5° and 12° North which amounts to be about 200,000 Km^2. Tsetse infested areas lies in the low lands and also in the river valleys of Blue Nile, Baro Akobo, Didessa, Ghibe and Omo [7].

Benishangul Gumuz is one of the five regions of Ethiopia infested by more than one species of tsetse flies [8]. Five species of *Glossina (G.m submorsitans*, *G. pallidipes, G. tachinoides, G.f. fuscipes* and *G. longipennis)* have been registered in Ethiopia [8]. In the Benishangul Gumuz regional state, four glossina species namely, *G. tachinoides, G. m. submorsitans, pallidipes* and *G. fuscipes* were found [9]. Apart from the cyclical transmission of trypanosomosis by *glossina* species, it is highly considered that mechanical transmission is a potential threat to livestock production and productivity in some parts of Ethiopia [2].

As stated above, the negative impacts of tsetse flies and trypanosomosis in the Benishangul Gumuz reginal state are so vast and it should be underlined that no other development scheme or policy can generate the evidence of success in guaranteeing sustainable livestock development and alleviation of poverty in the rural areas of the region without addressing the problems posed by tsetse flies and trypanosomosis. Hence, intervening in the prevention and control of tsetse flies and trypanosomosis is very vital in the region.

Mao Komo special district is one of the twenty districts of the Benishangul Gumuz regional state with a serious problem of trypanosomosis. Controlling this economically important disease in this area could have a number of benefits to improve the livelihood of the poor people of the district by increasing milk, meat, surplus capital from the sale of livestock and livestock products and improving the availability of draft power (Oxen). Although the disease is one of the major obstacles of livestock production and productivity in the district, study on the prevalence of bovine trypanosomosis was carried out only by Daud and Molalegne [10] indicating an overall prevalence of 24.7% and no further study and control intervention has been carried out since then. Therefore, the present study is designed to determine the present situation and prevalence of bovine trypanosomosis in the district, to assess associated risk factors and to suggest possible control measures of the disease.

MATERIALS AND METHODS

Study Area: The study was conducted in Mao Komo special district of the Benishangul Gumuz regional state, western Ethiopia, from December 2015 to November 2016. It was carried out in five kebeles hereafter called sites namely: Tulu, Gure, Shoshore, Yamasera and Fafa. The district has 32 kebeles covering an area of 2,100 Km^2 with human population of 63,227 [11]. The district is characterized by low land plane (81%), mid altitude (19%), with altitude range of 567-1983 meter above sea level. Its annual average temperature is 33.5°c (27-40°C) and its rain fall range is 900-1400 mm [12]. The livelihood of the people in the district largely depends on mixed livestock and crop production having livestock population of 9,982 cattle, 8,692 sheep, 16,776goats, 2,352equines, 50,169poultry and 55,812beehives [13].

Study Design and Study Animals: Cross sectional study design was used. zebu cattle (*Bos indicus*), which are mainly kept under an extensive husbandry system grazing the communally owned pasture land throughout the year were randomly sampled. Study animals were herded together during the day time and returned to their

individual owner's farmstead every evening. The body condition of each of the study animal was scored as good, medium and poor [14]. Similarly, their age was determined based on De-Lahunta and Habel [15] principles as young (<2 years), matured (2-5 years) and adult (> 5 years).

Sampling Techniques and Sample Size Determination: The study sites were selected purposively as convenient. Study animals were sampled randomly involving both sexes, all age groups, and all types of body conditions. The desired sample size was calculated according to the formula given by Thrusfield [16]. The sample size was determined based on the expected prevalence of 24.7% by Daud and Molalegne [10] confidence level of 95% and 5% desired absolute precision. As a result a total of 286 cattle were calculated but increased to 385 to increase precision.

Study Methodology

Packed Cell Volume (PCV) determination: Blood samples were obtained by puncturing the marginal ear vein with sterile lancet and collected directly into a pair of heparinized capillary tubes (75 mm ×1.2 mm). The tubes were then sealed at one end with crystal seal. The capillary tubes were placed in microhaematocrit centrifuge with sealed end outermost. Then the tube was loaded symmetrically to ensure good balance. After screwing the rotary cover and closing the centrifuge lid, the samples were allowed to centrifuge at 12,000 rpm for 5 minutes. After centrifugation, the capillary tubes were placed in a haematocrit reader. The length of the packed red blood cells column is expressed as a percentage of the total volume of blood. Animals with PCV less than 24% were considered to be anemic [17].

Buffy Coat Technique: Heparinized microhaematocrit capillary tubes, containing blood samples were centrifuged for 5 minutes at 12,000 rpm. After the centrifugation, trypanosomes were usually found in or just above the buffy coat layer. The capillary tube was cut using a diamond tipped pen 1 mm below the buffy coat to include the upper most layers of the red blood cells and 3 mm above to include the plasma. The content of the capillary tube was expressed onto a glass slide, and covered with cover slip. The slide was examined under 40 × objectives and 10× eye piece for movement of parasite [18]. Trypanosome species were identified according to their morphological descriptions as well as movement in wet film preparations [17].

Data Analysis: During the study, data were collected using the data collection format and entered into Microsoft Excel. Hematological and parasitological data were managed very carefully. Then, the data from the Microsoft excel sheet were processed and analyzed by using a statistical software program. Chi square was used to compare the prevalence of trypanosomosis in different variables and to determine the relationship between variables and the result. Data collected on PCV values were analyzed by ANOVA to compare the mean PCV values of infected animals against that of non-infected animals. In all cases the difference between parameters were tested for significance at probability level of 0.05 or less. The prevalence of bovine trypanosomosis was calculated as the number of parasitologically positive animals examined by buffy coat method to the total animals examined by Thrusfield [16].

RESULTS

Prevalence of Trypanosomes Infection: Out of the total animals examined, 62(16.1%) were infected with trypanosomosis (Table 1). The trypanosome species responsible for the infection were *T. congolense, T. vivax* and *T. brucei*. The proportional prevalence of each species of trypanosome was 34 (54.84%) for *T. congolense*, 19 *(*30.6%*)* for *T. vivax*, 7(11.3%) for *T. brucei* and 2 (3.2%) for mixed infection with *T. congolense* and *T. vivax* was observed in the fresh blood examined during the study period and the proportional prevalence of trypanosome species was found to be statistically significant (P<0.05).

Trypanosomosis and Associated Risk Factors: The highest and the lowest prevalence of trypanosomosis were recorded in Yamasera (19.48%) and Shoshore (12.99%) study sites respectively. However, there was no statistically significant difference among the study sites and infection of animals with trypanosomes (P>0.05) (Table 3).

The Prevalence of trypanosomosis varies in both sexes; the infection in male is slightly higher (17.40 %) than in female 14.80%) and the association was not found statistically significant (P>0.05) (Table 3).

Out of the total sampled animals; 84, 176 and 125 were < 2 years, 2-5 years and > 5 years old, respectively and the prevalence was found to be 1.90 % for animals < 2 years, 18.75% for animals2-5 years and 15.20% for animals> 5

Table 1: Prevalence of single and mixed infection of trypanosomes in Mao Komo special district

Trypanosomes	No. positive	Prevalence (%)	χ^2	(P-value)
T. congolense	34	54.84	194.2868	<0.0010
T. vivax	19	30.65		
T. brucei	7	11.30		
Mixed (*T. congolense* & *T. vivax*)	2	3.21		
Total	62	100		

Table 2: Mean PCV comparison between infected and non infected animals in Mao Komo special district

Status	Frequency	Mean PCV (%)	SEs	Overall PCV	χ^2	P- value
Infected	62	17.11	5.42	1061	34.1967	<0.001
Non infected	323	25.42	3.66	8211		
Total	385	21.26	4.54	9272		

Table 3: Prevalence of bovine trypanosomosis and its association with various risk factors in Mao Komo special district

Risk factors	No. examined	No. positive	Prevalence (%)	x^2	P-value
Sites				1.6534	0.799
Tulu	77	14	18.18		
Gure	77	11	14.29		
Shoshore	77	10	12.99		
Yamasera	77	15	19.48		
Fafa	77	12	15.58		
Total	385	62	16.10		
Sex				0.4934	0.482
Male	183	32	17.40		
Female	202	30	14.80		
Total	385	62	16.10		
Age (years)				2.0840	0.353
<2	84	10	11.90		
2-5	176	33	18.75		
> 5	125	19	15.20		
Total	385	62	16.10		
Body conditions				27.1499	<0.001
Good	91	2	2.2		
Medium	127	16	12.60		
Poor	167	44	26.35		
Total	385	62	16.10		

years and the difference in the prevalence among the different age group was not statistically significant (P>0.05) (Table 3).

Similarly, during the study period, animals were categorized in to different body conditions as good, medium and poor. From a total of 385 animals examined; 91, 127 and 167 animals were registered as good, medium and poor body conditioned, respectively and out of which 2.2% prevalence was recorded in animals with good body condition. The rest 12.60% and 26.35%prevalence of trypanosomosis were obtained from animals with medium and poor body condition scores, respectively. Trypanosome infection and body condition scores of animals revealed statistically significant association (P< 0.05) (Table 3).

Packed Cell Volume: The mean PCV values for all examined animals were 21.26 ± 4.54. However, the mean PCV values for non-infected animals were 25.42 ± 3.66 and the mean PCV values of infected animals were 17.11 ± 5.42. There was statistically significant difference in the mean PCV values of non-infected and infected animals (P<0.05) (Table 2).

Entomological Survey Results: The present survey of tsetse flies depicted that *G.m.submorsitans, G. fuscipes* and *G. pallidipes* are species of tsetse fly responsible for cyclic transmission of trypanosomosis in the study area. Tsetse fly survey was carried out in five sites of the study district by deploying a total of 47 geo-referenced mono-conical traps in the river border, open wood land

Table 4: Flies caught in different areas of survey sites of Mao Komo special district

			Tsetse flies caught					Other biting flies	
Sites	Total flies caught	No. of traps	No.	Species	M	F	[a]F/T/D	*Tabanus*	*Haematopota*
Tulu	13	10	3	*G.m.submorsitans*	1	2	0.15	6	4
Gure	13	10	5	*G.fuscipes*	2	3	0.25	5	3
Shoshore	26	8	8	*G. pallidipes*	1	7	0.5	11	7
Yamasera	153	10	113	*G.fuscipes*	38	75	5.65	23	17
Fafa	26	9	4	*G.m.submorsitans*	1	3	0.22	15	7
Total	231	47	133		43	90	1.41	60	38

F/T/D=fly per trap per day, M=male, F=female

(savanna grass land) and on grazing fields of cattle; the number of tsetse flies captured in each study site is Tulu (3), Gure (5), Shoshore (8), Yamasera (133) and Fafa (4). The species of tsetse flies investigated in the current study were *G. m. submorsitans* in Tulu and Fafa, *G. fuscipes* in Gure and Yamasera and *G. pallidipes* in Shoshore study sites. The mean apparent density of tsetse flies in the survey sites was investigated as 1.41 f/t/d and mean apparent density of mechanical vectors such as Tabanus (0.64 f/t/d) and haematopota (0.40 f/t/d) were also recorded (Table 4).

DISCUSSION

The present study revealed an overall prevalence of 16.10% trypanosomosis infection in the study area. This finding was slightly higher than the findings of Getachew and Asmamaw [19] in Mandura district of the Benishangul Gumuz regional state, Western Ethiopia, who reported 13.30% prevalence in their study on epidemiology of cattle trypanosomosis and associated anaemia and Bayisa and Getachew [20] who reported 11.70% in their study on trypanosomosis and its associated risks in cattle population of Dangur district of the Benishangul Gumuz regional state, Western Ethiopia. However, the current finding was lower than the previous findings of Bayisa *et al.* [21] who reported 22.38% of prevalence in Assosa district of the Benishagul Gumuz regional state, Western Ethiopia and Mulaw *et al.* [22] whose result indicated 28.10% in their study on the prevalence of major trypanosomes affecting bovine in tsetse infested Assosa district of the Benishangul Gumuz regional state, western Ethiopia. The difference in the prevalence of trypanosomosis in the previous and the current findings might be due to the difference in agro ecology and climatic conditions of the areas and partly it might be the difference in seasons in the study period.

The current finding indicated that the infection was mainly caused by *T. congolense (*54.84%), *T. vivax* (30.65%), *T. brucei* (11.30%) and to less extent mixed infection with *T. congolense* and *T. vivax* (3.21%). This result was in concord with the reported proportions of *T. congolense* by Abraham and Zeryehun [23] who conducted their study on prevalence of bovine trypanosomosis in selected sites of Arba Minch district, Sothern Ethiopia, whose result showed proportional prevalence of *T. congolense* to be 61.4% [24] whose finding showed proportional prevalence of *T. congolense to be* 63.64% during their work on trypanosomosis and anemia in cattle population of Dale Wabera district of Kellem Wollega Zone, Western Ethiopia. This result was also in consistent with prior report of Mulaw *et al.* [22] who found proportional prevalence of *T. congolense* to be 66.7%.

The high proportional infection rate of *T. congolense* in cattle might be attributable to the high number of serodems of *T. congolense* relative to other species of trypanosomes. It could also be due to the possible development of better immune response to *T. vivax* by infected animals as demonstrated by Leak *et al*. [25]. Further, it might be attributed to the efficient transmission of *T. congolense* by cyclical vectors than *T. vivax* in tsetse-infested areas. Previous reports indicated that *T. congolense* and *T.vivax* are the most prevalent trypanosomes that infect cattle in tsetse infested and tsetse free areas of Ethiopia respectively [6] (Leak, 1999). Studies carried out by Leak *et al*. [25] and Rowland *et al*. [26] have indicated that *T. vivax* is highly susceptible to treatment while the problem of drug resistance is higher in *T. congolense.*

The effect of different risk factors such as sex, age categories, study sites and body conditions on prevalence of bovine trypanosomosis was studied and statistically significant associations were not observed in age categories, sex groups and study sites (P>0.05) while body condition was found to be statistically significant (@@ <0.05). This result was in agreement with previous reports of Mulaw *et al*. [22] whose result indicated that sex and study sites did not show statistically significant association with trypanosomosis infection; however,

unlike that of the present finding, their finding showed that body condition score of study animals did not show significant association with trypanosomosis infection. It was also in consistent with the previous work of Asmamaw and Mengistu [27] in the neighbouring district (Bambasi) whose result revealed that no statistically significant association was observed among study sites, sex groups, and age categories of study animals and trypanosomosis infection; however, in contrast to the present result their finding showed that no statistically significant association was seen between body condition score and trypanosomosis infection of study animals. Moreover, the current finding was in agreement with study conducted by Lelisa *et al.* [28] with regard to age category, sex group and study sites of study animals with trypanosomosis infection because their study revealed that no statistically significant association was observed among the above mentioned variables and trypanosomosis infection. The present finding was also in consistent with the previous work of Yehunie *et al*. [29] in that their finding indicated that there was statistically significant association between body condition score of study animals and trypanosomosis infection in the study sites.

The overall mean PCV values for all examined animals were 21.26±4.54. The mean PCV values of infected animals was lower (17.11±5.42) than that of non-infected animals (25.42±3.66) and there was statistically significant association between PCV values and trypanosomosis infection of the study animals. This result was in alignment with previous works of Mulaw *et al*. [22], Asmamaw and Mengistu [27], Lelisa *et al*. [28] and Gemeda [30] whose findings coincide with the present result in that they all reported statistically significant association between PCV values and trypanosome infection of study animals.

In the entomological survey, *G. m. submorsitans, G. fuscipes* and *G. pallidipes* were tsetse fly species captured and their mean apparent density measured as f/t/d was found to be 1.41. Tsetse flies account for 57.58% (133/231) out of the total flies caught. In addition, other mechanical transmitters of trypanosomosis such as tabanus and haematopota were registered and theyaccount for 25.97% (60/231) and 16.45% (38/231) of the total flies caught with f/t/d of 0.64 and 0.40 respectively. The present finding was in agreement with the previous reports of Asmamaw and Mengistu [27] in the neighbouring Bambasi district of the BenishangulGumuz regional state, Western Ethiopia, in that the previous and the current findings investigated tsetse flies and other mechanical transmitter of trypanosomosis in the study districts indicating that tsetse flies and other biting flies are responsible for the transmission of trypanosomosis from diseased to healthy animals.

CONCLUSION

The overall moderately high prevalence of trypanosmosis obtained in cattle of Mao Komo special district indicated the importance of the problem and its contribution to hampering the product, productivity, work performance and general health status of these animals. The most widely distributed and dominant species of trypanosome in the study sites are *T. congolense* (54.84%) followed by *T. vivax* (30.65%), *T. brucei* (11.30%) and to less extent mixed infection with *T. congolense* and *T. vivax* (3.21%) which was mainly transmitted by tsetse flies (*G. m. submorsitanas, G. fuscipes* and *G. pallidipes*) and other biting flies (*Tabanus* and *Haematopota*) with f/t/d/ of 1.41, 0.64, and 0.40 for the different species of tsetse flies, *Tabanus*, and *Haematopota,* respectively. Since the District lies within the tsetse belt area, the result of the present study (16.10%) showed the fact and expected prevalence. Significant association was not observed within study sites, sex and age groups of animals ($P > 0.05$) while there was statisitically significant association among body condition scores and PCV values of study animals and trypanosomosis infection ($P < 0.05$). These all revealed that Mao Komo special district is favorable for the successive breeding of tsetse and other biting flies that play a major role in the transmission of trypanosomes to susceptible hosts and hence, designing and implementing control strategies of trypanosomosis focusing on vectors and against the parasites will be undertaken in the study area and farmers of the district have to be educated about the impact of trypanosomosis on the health and productivity of animals so as to implement participatory approach in the control of the parasites and vectors.

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