A study on Prevalence of Bovine Trypanosomosis and Associated Risk Factors in Bulen District of the Benishangul Gumuz Regional State, Western Ethiopia

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Abstract: A cross sectional study was carried out from December 2016 to March, 2017 in Bulen district of the Benishangul Gumuz Region, Western Ethiopia to determine prevalence of bovine trypanosomosi and associated risk factors. Blood samples collected from (n= 306) randomly sampled cattle (*Bos indicus*) was examined using parasitological (buffy coat technique) and hematological (measurement of packed cell volume) procedures. An overall,18/306(5.88%) prevalence was recorded. The infection was caused mainly by *Trypanosoma congolense 11/18(*66.11%), *Trypanosoma vivax* 6/18(13.5%) andto less extent by *Trypanosoma brucei* 1/18(5.56%). The infection rate was statistically significant among difference trypanosome species (P<0.05). Mean packed cell volume (PCV) value of parasitaemic animals was lower (18.21% ± 4.11) than aparasitaemic animals (28.12% ±2.67) and the variation wasstatistically significant (P>0.05). Higher prevalence 11/80 (13.75%) was registered in animals with poor body condition animals when compared with animals medium 5/147(3.40%) and good 2/77(2.60%) body condition and the difference was found statistically significant (p<0.05). Incontrast, prevalence oftrypanosomosis was not statistically significant across study sites, amongage categories and between sex groups (P> 0.05). To wrapup, the result of the present finding shows moderately high prevalence of trypanosomosis in the study sites indicating the need for strategic and integrated approach to control the vector and to minimize the impact of the disease in the study district.

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1. Introduction

Trypanosomosis is among the well-known constraints to livestock production in Africa as it causes a serious and often fatal disease of livestock mainly in the rural poor community and rightfully considered as a root cause of poverty in the continent (Vreysen, 2006). Most trypanosomes that are transmitted by tsetse flies inhabit many parts of the continent that extended about 15°N and 20°S of the equator, biting flies may also act as mechanical vectors of trypanosomosis (Urquhart *el al*., 1996).

Trypanosomiasis is a devastating disease of livestock caused by protozoal parasites of the genus trypanosoma that inhabits blood and other tissues of vertebrates including animals, wildlife and human (Adam *et al.,* 2003; Gupta *et al*., 2009; Bal *et al*., 2014). It is a vector borne disease that is transmitted biologically by tsetse flies and mechanically by other biting flies (FAO, 2002; OIE, 2009). It is a major constraint contributing to direct and indirect economic losses to crop and livestock production (Abebe, 2005) and has a significant negative impact on economic growth in many parts of the world (Taylor *et al*., 2007; Sharma *et al*., 2013), particularly in sub-Saharan Africa (Cecchi *et al*., 2008).

The disease is characterized by severe anemia, weight loss, reduced productivity, infertility and abortion, with death occurring in some animals during the acute phase of the disease. Animals which survive often remain infected for several months or years, exhibiting a low level of fluctuating parasitaemia which serves as a reservoir for the disease occasionally; however, the infected animals may undergo spontaneous recovery (Nantulyia, 1986).

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 equiperdium* in horses (Abebe, 2005). The influence of tsetse 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 (Leak,1999).

Ethiopia is situated at the East end of the African tsetse belt. In Ethiopia, tsetse flies are confined to south western and north western regions between longitude 33° and 38° E and latiude 5° and 12° N that covers an area of about 22,000 km^2 (NTTICC, 2004). Tsetse infested areas lies in the low lands and also in the river valleys of Blue Nile, Baro Akobo, Didessa, Ghibe and Omo. Benishangul Gumuz is one of the five regions of Ethiopia infested with more than one species of tsetse flies (Keno, 2005). Five species of Glossina (*Glossina morsitans submorsitans*, *G. Pallidipes, G. tachnoides, G. f. fuscipes and G. longipennis*) have been registered in Ethiopia (Keno, 2005). In the study region of Benishangul Gumuz regional state, four glossina species namely, *G. tachinoides, G.morsitant submorsitances, G. pallidipes* and *G.fuscipes* were found (ARVDSMSL, 2015). 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 (Abebe, 2005).

Bulen is one the seven districts of Metekel zone of the Benishangul Gumuz regional state, western Ethiopia 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 main obstacles of livestock production and productivity in the district, the prevalence and situation of the disease was assessed only once by (Asmamaw *et al*., 2016) and no further strategic and participatory control measures of have been made and the problem of the disease is still continuing in the district. Therefore, the present study is designed to determine the prevalence associated risk factors of bovine trypanosomosis and to forward possible prevention and control measures against the disease in the district.

2. Materials and Methods

2.1. Study Area period

The study was conducted from December 2016 to March, 2017in Bulen district of Metekel Zone, Benishangul Gumuz Regional State, Western Ethiopia to determine prevalence of bovine trypanosomosi and associated risk factors. Bulen district is located 550 km away from Addis Ababa. The area is located at 9° 00″ to 11° 07″ N latitude and 35°45″ to 3607″ E longitude. It was carried out in six kebeles hereafter called sites namely: Mata, Addis Alem, Chilanko, Bekuji, Dobi and Badore. Thedistrict has 19 kebeles covering an area of 2858 km^{2}with human population of 57567(CSA, 2014). It has an altitude range of 900-2300meter above sea level. Itsannual average temperature is 28.75^{0}c (23.5-34^{0}c) and its rainfallrange is 900-1500mm (NMSA, 2014). The livelihood of the people in the district largely depends on mixed livestock and crop production having livestock population of 102,904 cattle, 16,192 sheep, 48,034goats and 9,281equines (CSA, 2014).

2.2. Study Design and Study Animals

Cross sectional study design was used. A local zebu cattle (*Bos indicus*), that are mainly kept under an extensive husbandry system grazing the communally owned pastureland throughout the year were randomly sampled. They grazed together during the day time and returned to their individual owner's farmstead each evening. The body condition of each of the study cattle was scored as good, medium and poor (Nicholson and Butterworth, 1986). Similarly, their age was determined based on (De-Lahunta and Habel 1986) principles as young (<2 years old), matured (2-5 years old) and adult (> 5 years old).

2.3. Sampling Methods and Sampling Size Determination

The type of sampling methods was simple random sampling to establish the prevalence and associated risk factors of trypanosome infection in the study area. The desired sample size was determined using the formula given by (Thrusfeild, 2007).

$n = 1.96^2 p_{exp} (1-p_{exp})/ d^2$

where: n = require sample size

p_{exp} = expected prevalence

d = desire absolute precision

1.96^2 = z-value for the 95% confidence level

The prevalence of bovine trypanosomosis in Bulen district was reported to be 5.6% by (Asmamaw*et al*., 2016). Therefore, an expected prevalence of 5.6% was taken to estimate the sample size. Taking 95% confidence level, 5% precision and 5.6% expected prevalence 81 animals were needed to establish the prevalence. However, 306 cattle were sampled to increase the level of precision and randomness.

2.4 Study Methodology

3.4.1 Packed cell volume (PCV) determination

Blood samples were obtained by puncturing the marginal ear vein with lancet and collected directly into a pair of heparinised capillary tubes. The tubes were then sealed at one end with crystal seal. PCV was measured in a micro-haematocrit centrifuge (Hermmle Labortechnik, type Z, Germany). 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 (OIE, 2008).

2.4.2 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 x40 objective and x10 eye piece for movement of parasite (Murray and Dexter, 1988). Trypanosome species were identified according to their morphological descriptions as well as movement in wet film preparations (OIE, 2008).

2.5 Data Analysis

During the study period, data were collected using the sample 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 soft ware program (STATA 7). 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 parasitaemic animals against that of aparasitaemic animals. In all cases the difference between parameters were tested for significance at probability level of 0.05 or less. The prevalence of cattle trypanosomosis was calculated as the number of parasitologically positive animals examined by buffy coat method to the total animals examined (Thrusfield, 2007).

3. Result

3.1. Prevalence of Trypanosomes infection

Out of the total animals examined, 18/306(5.88%) were infected with trypanosomes. The trypanosome species responsible for the infection were *T. congolense, T. vivax* and *T. brucei*. The proportional prevalence of each species of trypanosome was 11/18(61.11%) for *T. congolense*, *6/18(*33.33%*)* for *T. vivax*, 1/18*(*5.56%*)* for *T. brucei and* the proportional prevalence of trypanosome species was found to be statistically significant (P<0.05) (table 1).

Table 1: Prevalence of trypanosomes infection in Bulen district

Trypanosomes	No. positive	Prevalence (%)	X^2	(P-value)
T. congolense	11	66.11	182.5627	0.000
T. vivax	6	33.33		
T. brucei	1	5.56		
Total	**18**	**100**		

3.2. Haematological Survey Results

The mean PCV values for all examined animals were 24.48 ± 3.34 SD. However, the mean PCV for non infected animals were 28.12 ± 2.67 SD and the mean PCV value of the infected animals was 18.21 ± 4.11 SD and the association was found significant between non infected and infected animals (P<0.05) (Table 2). The overall prevalence of animals with anemia in the study district was 165/306 (53.92 %). The prevalence of animals with anemia was statically significant in trypanosome infected cattle (88.89%) than in non-infected cattle (11.11%) (□<0.05) (Tables 2 & 3).

Table 2: Mean PCV comparison of parasitaemic and aparasitaemic animals in Bulen district

Status	***Frequency***	***Mean PCV (%)***	***SE***	***Overall PCV***	X^2	***p-value***
Parasitaemic	*18*	*18.21*	*4.11*	*327.78*	*9.4117*	*0.002*
Aparasitaemic	*288*	*28.12*	*2.67*	*8098.56*		
Total	***306***	***24.48***	***3.34***	***7490.88***		

Table 3: Proportion of anemia in parasitaemic and aparasitaemic cattle population in Bulen district

Status	Anemia	Frequency	Percent	Percent Share Per Strata
Infected	Anemic	16	5.23	88.89
	non anemic	2	0.65	11.11
Non infected	Anemic	149	48.69	51.74
	Non anemic	139	45.42	48.26

3.3. Trypanosomosis and Associated Risks

The highest and the lowest prevalence of trypanosomosis were recorded in Badore 7/61 (11.48%) and Mata 2/60 (3.33%) study sites respectively and no trypanosome positive animal was registered at Chilanko study site. There was no significant association among the study sites and trypanosome infection in the study district (p >0.05) (Table 4).

The Prevalence of trypanosomosis varies in both sexes; the infection in female is higher 13/164 (7.93 %) than male 5/142 (3.52%) however, the association was not statistically significant (P>0.05) (Table 4). In the present study animals examined were categorized in different age groups as < 2 years, 2-5 years and >5 years. Out of the total sampled animals, 67, 107 and 132, were < 2 years, 2-5 years and> 5 years old respectively and the prevalence was found to be 3/67(4.48%) for animals < 2 years, 5/107 (4.67%) for animals 2-5 years and 10/132(7.58%) for tested animals>5 years old and the difference in the prevalence was not statistically significant (p >0.05) (table 4).

Similarly, during the study, animals were categorized in to different body conditions as good, medium and poor. From the total 306 animals examined 79,147 and 80, were registered asgood, medium and poor body condition respectively and out of which 2/77 (2.60%), 5/147 (3.40%), and 11/80(13.75 %) prevalence of trypanosomosiswere recorded for animals with good, medium and poor body condition respectively. Trypanosome infection and body condition scores of study animals were found statistically significant (p < 0.05) (Table 4). The effect of age, sex, sites and body condition on prevalence of trypanosomosis is summarized in table 4 below.

Table 4: prevalence of bovine trypanosomosis and its association with various risk factors in Bulen district

Risk factors	No. examined	No. positive	Prevalence (%)	p-value	χ^2
Sites					
Mata	60	2	3.33		
Addis Alem	55	4	7.27		
Bekuji	47	3	6.38	6.9073	0.228
Chilanko	38	0	0		
Dobi	45	2	4.44		
Badore	61	7	11.48		
Total	**306**	**18**	**5.88**		
Sex					
Female	164	13	7.93	2.6682	0.102
Male	142	5	3.52		
Total	**306**	**18**	**5.88**		
Age (years)					
<2	67	3	4.48		
2-5	107	5	4.67	1.2052	0.547
>5	132	10	7.58		
Total	**306**	**18**	**5.88**		
Body conditions					
Good	77	2	2.60		
Medium	147	5	3.40	12.1809	0.002
Poor	80	11	13.75		
Total	**306**	**18**	**5.88**		

4. Discussion

The present study revealed an overall 18/306(5.88%) prevalence of trypanosomosis caused by different species of trypanosomes. This finding was in agreement with the previous studies conducted by (Belete, 2014) whose finding showed 6% prevalence in his study on prevalence of bovine trypanosomosis and host related risk factors in Jawi district of the Amhara region, south west of Ethiopia, (Asmamaw and Getachew, 2016) whose report showed 5.58 % prevalence in their study on trypanosomosis in Cattle Population of Pawi district of the Benishangul Gumuz Region, Western Ethiopia and (Lelisa *et al.,* 2015) who reported 5.43% prevalence in in their study on

prevalence of bovine trypanosomosis and a Apparent Density of Tsetse and Other Biting Flies in Dangur district of the Benishagul Gumuz region, western Ethiopia. Although it was slightly lower, it was found to be consistnent with the former results reported by (Dano *et al*., 2014) who reported an overall prevalence of 7.81% in Guto Gida district of Eastern Wollega Zone of Oromia region, (Tefese *et al*., 2012) whose finding was 8.55% in Sasiga and Diga districts of East Wollega Zone of Oromia Region and (Yehunie *et al*., 2012) who reported an overall prevalence of 7.81% in Wemberma district of West Gojjam Zone of the Amhara Region, Northwest Ethiopia. Incontrast, the present finding was much lower when compared with previous reports (Shimels *et al*., 2011) whose report indicated 26.3% prevalence in and around Assosa district of the Benishangul Gumuz region, Western Ethiopia, (Daud and Molalegne, 2011) whose finding showed an overall prevalence of 24.7% in Mao-Komo special districtof the Benishangul Gumuz region, Western Ethiopia, (Birhanu and Asmamaw, 2016) whose finding revealed an overall prevalence of 19.53% in their study on Prevalence of Cattle Trypanosomosis, Apparent vector density and Associated Risk Factors in Debate District, Western Ethiopia and (Zelalem *et al*., 2016) whose report showed an overall prevalence of 16.10 % in their study on Prevalence of Bovine Trypanosomosis and Associated Risks in Mao Komo Special District of the Benishagulgumuz Region, Western Ethiopia. The relatively low prevalence of trypanosomosis in the present study might be due to the differences in agro-ecology and climatic conditions of the localities.

Of the total cases registered, 11/18(66.11%), 6/18(33.33 %) and 1/18(5.56%) were found to be caused by *T. congolense, T. vivax, T. brucei* respectively. This indicates statistically significant difference among the distribution of trypanosome species ($p< 0.05$). This finding was in consistnent with the previous finding of (Biyazen *et al*., 2014) who reported 63.64%, 27.27%, and 9% for trypanosome species of *T. congolense, T. vivax, and T. brucei* respectively during their study in Dale Wabera district of Kellem Wollega Zone of Oromia Region, Western Ethiopia, similarly, it was in concordance with (Dano et al., 2014) whose finding showed proportional prevalence of T. congolense to be 53.33%, T. vivax 30% and T. brucei 16.66% in their study on Prevalence of Bovine Trypanasomosis in Guto Gida District of East Wollega Zone of Oromia Region, Western Ethiopia.

Among the study sites, the highest and the lowest prevalence of trypanosomosis were recorded in Badore PA 7/61(1148%) and Mata ePA 2/60(3.33%**)** respectively and no trypanosome infection was registered in Chilanko study site. However there was no significant difference ($p>0.05$) in the prevalence of trypanosomosis and the study sites. This finding was in agreement with the finding of (Asmamaw and Mengistu, 2016) in their study on Bovine Trypanosomosis and Apparent Vector density in Bambasi District of Benishangul Gumuz Region, Western Ethiopia, (Firaol *et al*., 2014) in their study on Post Control Survey on Prevalence of Bovine Trypanosomosis and Vector Distribution in Ameya District, South West Shewa, Ethiopia According to (Adale and Yasine, 2013), there is difference in prevalence of trypanosomosis in different study sites and the difference among kebeles/study sites/is due to difference in vegetation cover; reproduction and development of flies are highly influenced by climaticconditions.

The prevalence of trypanosome infection was higher in female animals 13/163(7.93%) than males 5/142(3.52%), although it was not statistically significant ($p>0.05$). This finding was in agreement with the previous findings of (Feyissa *et al*., 2011); (Tasew and Duguma, 2012). Similarly, (Bogale *et al*., 2012) found higher infection rate in females animals than males in some parts of Ethiopia. The possible reason for this difference might be due to physiological difference between female and male animals (Feyissa *et al*., 2011) because female animals are more exposed to physiological stresses than males.

Higher prevalence of trypanosomosis was observed 11/80(13.75%) in animals with poor body condition when compared with animals with medium 5/147 (3.40%) and good 2/77 (2.60%) body condition and the association was found statistically significant ($p<0.05$) and this finding was in agreement with study carried out by (Zewdu and Dessie, 2016), (Lelisa *et al*., 2015); (Teka *et al*., 2012) and (Ayana *et al*.,2012) who reported higher trypanosome infection rate in animals with poor body condition than in animals with good and medium body condition. Similarly, higher prevalence was registered in animals aged > years 10/132(7.58%) when compared with animals 2-5 years5/107(4.68%) and <2 years3/67(4.48%) and statistically significant associations were not observed ($p>0.05$) and this finding was in agreement with previous worker (Zewdu and Dessie 2016) who reported higher prevalence of trypanosome infection in adult animals than young in their study on Prevalence of bovine trypanosomosis in Chilga District, Northwest Ethiopia.

The overall mean PCV value of all examined animals was (24.48% ± 3.34 SD). The mean PCV of non infected cattle was higher (28.12% ± 2.67 SD) than that of infected animals (18.21% ± 4.11%) and the association was statistically significant. This finding was in agreement with the previous works (Bayisa and Getachew, 2015) and (Zelalem *et al*.,

2016) who reported lower mean PCV value in infected animals than noninfected ones. Similarly, (Daud and Molalegne, 2011) and (Molalegne *et al*., 2010) reported lower mean PCV value in infected than in the non-infected animal.

5. Conclusion and Recommendtions

The findings of the present study revealed the importance of trypanosmosis and its contribution to hampering the product, productivity, work performance and general health status of cattle in the district. The most widely distributed and dominant species of trypanosome in the study sites are *T. congolense (66.11%)* followed by *T. vivax (33.33%)* and *T. brucei (5.56%)* that were mainly transmitted by tsetse flies (*G.m.submorsitanas, G. fuscipes and G. pallidipes*) and other biting flies. Significant association was not observed within study sites, sex category and age groups of study animals ($p > 0.05$) while there was statistically significant association among body condition scores and PCV values of study animals and trypanosome infection ($P < 0.05$). These all revealed that Bulen 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. To wrap up the finding of the present study showed moderately high prevalence of bovine trypanosomosis indicating the need for strategic and participatory approach to prevent and control the disease in the study district.

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