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Epidemiological Distribution and Vectors of Bovine Trypanosomosis in Dangur District, Metekel Zone, Benishangul Gumuz region, North Western Ethiopia.

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Abstract: A cross-sectional study was carried out from February to June 2019 to determine the prevalence of bovine trypanosomosis, identification of circulating trypanosome species, vectors and associated risk factors in Dangur district of Benishangul Gumuz Regional State, Western Ethiopia. Blood samples were collected from a total of 390 cattle and examined using buffy coat technique. The present study revealed overall trypanosomosis prevalence was 18(4.62%). The major species of Trypanosoma identified were Trypanosoma Congolense (66.66%), Trypanosoma vivax (22.22%), Trypanosoma brucei (5.55%) and mixed infection (5.55%). Mean packed cell volume (PCV) value of the infected animals was lower (21.78% $\pm$ 2.44) than uninfected animals (26.69% $\pm$ 2.21) and the variation was statistically significant ($P< 0.05$). Overall, anemia prevalence of 26.43% (107/390) was recorded and it was significantly higher (66.67%) in infected cattle than in non-infected (33.33%). Statistical significant was not recorded between sex groups and age categories ($p>0.05$) but there was significant difference in the prevalence of trypanosomosis among study sites and body conditions ($P< 0.05$). Glossina tachinoide was the only tsetse fly caught and its mean apparent density measured as fly/trap/day was 0.25. In addition, mechanical vectors of trypanosomosis such as Stomoxys (0.11 f/t/d), Tabanus (0.08f/t/d) and Haematopota (0.02 f/t/d) were identified. Although the present study revealed low prevalence (4.62%) of Trypanosomosis in bovine in the study area, the impact of this desease on production, and the role of the bovine as the potential risk of transmissions to other livestock should not be underestimated. Therefore, appropriate intervention measures need to be taken.
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Key words: Dangur, Trypanosomosis, Tsetse fly, prevalence, Risk factors

1. Introduction

The livelihoods of more than 85% of the people of Ethiopia depend on the agricultural sector. This sector mainly possesses crop production, livestock production and mixed farming. Since people are dependent on this sector, the presence of livestock is one of the necessities to this sector. This fact has made Ethiopia to be one of the richest countries in livestock production in Africa (Azage and Alemu, 1997). Official figures gives a National Ethiopia animal population of 40.9 million cattle, 25.5 million sheep, 23.4 million goats, 2.7 million horses, 0.63 million mules, 5.2 million donkeys and 1.07 million camels (CSA, 2003).

Tsetse fly infest 10 million km^2 potentially productive land of Africa between 14 ^{0} N and 29 ^{0} S (Radiostits, 2006). There are 23 different species of tsetse fly and they exist in 37 countries of Africa. Five of them namely *G.m.submorsitans*, *G.pallidipes, G.tachinoides, G.fuscipes and G.longipennis* are reported in Ethiopia. Several reports made in Ethiopia revealed that tsetse fly occupy over 66,000 km^2 areas (Ford *et al*., 1976) based on 1500 masl, breeding limits in the south and southwestern valley of the country. Langridge (1976) has reported that some 98,000km^2 areas 1600 masl breeding limits in the southern and southern western of Ethiopia. The tsetse flies in Ethiopia are confined to the southern and western regions between longitude 33^0 and 38^0 E and latitude 5^0 and 12^0 N which amounts to about 200,000 km^2. Out of this 31,000 km^2 or (62%) Regional land area of Benishangul-Gumuz is infested with Tsetse fly (NTTICC, 1996).

Tsetse flies are hard to control and the tsetse fly infestation is becoming more and more serious in Africa. The clearing of large forest tracks some time cause the flies to spread to more populated areas and the deforest land covered with savannah grass

consequently newly invade by morsitans group (Jordan, 1986). Tsetse flies are enormous health risks in part of Africa. They can transmit a disease trypanosomosis. African trypanosomosis is heamoparasitic disease considered as the main obstacle to animal production development (Getachew A. and Yilma J., 1996). It is the wasting disease; affected animals are chronically unproductive in terms of milk, meat, manure, traction. The number of trypanocidal drugs available for treating and preventing infections in endemic areas is limited, and about 6 million doses are administered yearly in Africa. The drugs have been in the market for over 30 years, their range of therapeutic safety is small. The disease in Africa costs livestock producers and consumers an estimated US $ 1340 million each year (Radostits, 2006).

Mortality and the morbidity rate can be high. There is a direct association between increase prevalence and proximity herd pens watering points distance but no association of herd pens to grazing point distances which suggests that hydrological network played an important part in trypanosomosis (Enwezor *et al*., 2009). The disease distribution over 10 million km^2 of potentially productive land of Africa. The risk falls between 15^0 N and 29^0 S latitudes. As the result a total of 14.8 million cattle 6.12 million sheep and goats, 1 million camels and 1.23 million equines are at risk of contracting the disease (NTTICC, 2001) in Ethiopia. Therefore, the present study was designed to assess prevalence of bovine trypanosomosis; to identify vectors of trypanosomosis density and to identify the major associated risk factors and to forward possible control measures.

2. Materials and Methods

2.1 Study area

Dangur district is located in Metekel zone of Benishangul Gumz Regional State, and it is situated at 563 km away north west Addis Ababa. The district has 837,700 ha of land. The agro climate of the area alternates with long summer rain fall June to September and winter dry season December to March. The mean annual rain fall in the district ranges from 900 to 1400mm. The annual temperature in Dangur district ranges from 30 to 38°c. The district is located in Blue nile valley. The main rivers in Dangur district are Beles and Ayma with many others tributaries that enter these rivers, including Aypapo, Manbuk, Anja, Anzibuka, Kokel and Gublak which serve as sources of food for the tsetse fly. The area has got a number of wild animals which include; African buffaloes, Bush pigs, Warthog, Bush buck, Lion, Kudu, Hippopotamus, Crocodiles, Hyena, Velvet monkey and Antelopes. Many of these animals were serve as reservoir of infection for Trypanosomes.

2.2 Study design and sample size determination

A cross sectional epidemiological study design was employed for this study. The sample size was determined with 50% expected prevalence to increase the precision of the data using the formula: $n= (1.96)^2 \times Pexp (1-Pexp)/ d^2$ (Thrustfield, 2005).

Where

n = the required sample size for the district

Pexp = expected prevalence (50% in this case)

d^2 = desired absolute precision (5% in this case)

Therefore; $1.96^2 x 0.5 (1-0.5)/ (0.05)^2 = 384$

2.2.1. Questionnaire survey

To assess the socio-economic impact of trypanosomosis animals owners of the study group were interviewed about the husbandry practices, the farming practices, treatment cost (expenses against trypanosomosis control) and other trypanosomosis and its vector related questions.

2.2.2. Fly survey

During the study 60 monoconical traps were deployed and every trap was odor baited with acetone, octanol and cow urine. The underneath of each trap pole was smeared with grease in order to prevent the ants climbing up the pole towards the collecting cage that could damage the tsetse flies. The trap deployment time was 48 hours and after the flies captured in the collecting cage they were sorted by sex and species and recorded. The species of tsetse was identified based on the characteristic morphology. Other biting flies were also separated according to their morphological characteristics such as size, color, proboscis and wing venation structures at the genus level (Langridge, 1976; Fischer and Say, 1989). Sexing was done for tsetse fly just by observing the posterior end of the ventral aspect of abdomen by hand lens, as a result male flies easily identified by enlarged hypopygeum.

2.3. Study population

The study animals were bovine in Metekl zone Dangure district at three selected peasant associations which are managed under extensive management system. The study was carried out on 390 Cattle using random sampling method. The body condition can be classified as poor, medium and good according to (Asmare *et al*., 2012) and body condition Scoring is based on feeling the level of muscling and fat deposition over and around the vertebrae in the loin region (Thompson and Meyer, 1994).

2.4 Parasitological Examination

Blood samples were collected from cattle brought to three peasant association of Dangure district veterinary clinics and was examined for the presence of the parasites by using different parasitological examination techniques such as stained

thin blood smear and Wet blood smear techniques. The capillary tubes were loaded on micro hematocrit centrifuge systematically and centrifuged at 12000 rpm for five minutes to decrease the chance of false negative during diagnosis of parasites in case of mild infection where the parasites are very small in number (Murray,1997).

2.4.1. Wet Blood Films

A small drop of blood was placed on a clean glass slide, covered with cover slip to spread the blood as a monolayer of the cell. This was examined by light microscope (x40) objectives which was used detect any motile parasites, but this is not enough to identify the species of trypanosomes properly.

2.4.2. Thin Blood Smear

A drop of blood was placed on one end of a clean microscope slide and a thin film is drawn out in the usual way. The film was dried in air briefly, fixed in methyl alcohol for two minutes and allowed to dry. The smears were then stained by Giemsa and the stained slides must stand for 30 minutes. This techniques permits detailed morphological studies and identification of the Trypanosomes species. The same technique was used with the lymph nodes biopsies.

3. Data Management and Analysis

All data recorded in this study was entered in to Microsoft excel and subsequently analyzed using STATA version 7 soft ware. Chi-square test was used to determine the variation in Trypanosomes between sex, age, body condition, PCV and species.

4. Results

4.1. Parasitological Findings

In the current study a total of 390 head of cattle examined out of which 18/390 (4.62%) were infected with trypanosomes. The prevalence in terms of trypanosome species were 3.08 % T.congolense and 1.02 % T.vivax, 0.25% T. brucei and 0.25% mixed infection. The proportion of trypanosome species was 12/18(66.66 %) T. congolense, 4/18(22.22%) T. vivax, 1/18(5.55%) T.brucei and 1/18(5.55%) mixed (Table 1).

4.2. Packed cell volume Distribution and Anemia

The mean PCV value for whole examined animals was 26.47 ± 2.66 SE. However, the mean PCV value for uninfected animals was 26.69 ± 2.21 SE and mean PCV value of the infected animals were 21.78± 2.44 SE. The mean PCV values of cattle were significantly (= 0.000) influenced by trypanosome infection as 21.78% and 26.69 % PCV values in trypanosome positive and trypanosome negative animals were registered, respectively (Table 4). The overall anemia prevalence in the studied district was 27.43% (107/390). The anemia prevalence was significantly higher in trypanosome infected cattle (66.67%) than in non-infected cattle (25.53%) (<0.000). from the overall 27.43% anemia prevalence, 3.08% (12/390) was trypanosome infected animals. However, large number of animals 24.35% (95/390) had anemia (PCV < 24) without having trypanosome infection and Some of the animals 1.54% (6/390) were infected by trypanosome even though their PCV value was found to be normal (Table 5).

4.3. Prevalence of Trypanosomosis by Age, Sex, Sites and body Condition

The highest trypanosomosis prevalence (5.1%) was recorded in 2-7 years old animals whilst the lowest prevalence (2.85%) were >7years old. Slightly higher prevalence was registered in females 4.86% than in males 3.92 %, which was statistically non-significant. Trypanosomosis was recorded across the study sites with the highest (6.25 %) prevalence in Beles kutr hulet peasant association and the lowest 2.91 % in Azarti kitli peasant association. Trypanosomosis prevalence was statistically significant among study sites. There was a significant difference (P < 0.005) in the prevalence of trypanosomosis between good and poor body conditioned animals with highest prevalence in poor body condition category.

4.4. Entomological Findings

A total of 55 Tsetse and biting flies were caught during the study period from different sites. Out of the total, 30 (54.54 %) were belonging to tsetse of the genus Glossina, followed by 13 (23.63%) Stomoxys, and 10(18.18 %) Tabanid and 2 (3.63%) Haematopota. Only Glossina tachinoides were identified in the survey site with the overall apparent density of 0.25 F/T/D (fly/trap/day). The highest fly density were observed in beles kutr hulet peasant association 20 (0.5 F/T/D) and the lowest recorded in Azarti kitli 3 (0.075F/T/D) (Table 3).

Table 1: Prevalence of trypanosomosis by peasant associations

Peasant association (PA)	Location		Altitude	Sample size	No Negative	No positive	Prevalence %	X^2	p- value
	Latitude	Longitude							
Beles kutr hulet	11.28439◦	036.25727◦	1184m	128	120	8	6.25	11.101	0.049
Azarti kitli	11.29502◦	036.23600◦	1192m	137	133	4	2.91		
aypapo	11.31665◦	036.27746◦	1205m	125	119	6	4.8		

Table 2: The species of Trypanosoma identified from the study sites

Peasant association (PA)	Location		Altitude	Sample size	Parasites identified				Total	X^2	p- value
	Latitude	Longitude			T.congolense	T.vivax	T.brucei	mixed			
Beles kutr hulet	11.28439◦	036.25727◦	1184m	128	5	2	1	0	8	182.75	0.0001
Azarti kitli	11.29502◦	036.23600◦	1192m	137	3	1	0	0	4		
aypapo	11.31665◦	036.27746◦	1205m	125	4	1	0	1	6		

Table 3: Vectors of trypanosomosis identified from the study sites

Kebele	Latiteude	longitude	altitude	Site/river	Trap type	Tsetse fly			Biting fly					
						Glossinatachinoides			stomoxys		Tabanus		Hematopota	
						M	F	ftd	T	ftd	T	ftd	T	ftd
Beles kutr hulet	11.26271◦	036.23389◦	1108m	Manbuk river	Monoconical	5	15	0.5	7	0.175	5	0.125	1	0.025
Azarti kitli	11.29684◦	036.22934◦	1181m	Azibonka river	Monoconical	0	3	0.075	2	0.05	2	0.05	0	0
Aypapo	11.32116◦	036.26299◦	1185m	Aypapo river	Monoconical	2	5	0.175	4	0.1	3	0.075	1	0.025

Table 4: Mean PCV value between infected and uninfected Bovine of Dangur district

Status	**Frequency**	**Mean PCV (%)**	**SE**	**Overall PCV**	**X^2**	**p-value**
Infected	18	21.78	2.44	392	25.37	0.000
Uninfected	372	26.69	2.21	9932		
Total	390	26.47	2.66	10324		

Table 5: Proportion of anemia in infected and uninfected Bovine population of Dangur district

Status	**anemia**	**Frequency**	**Percent/%**	**Percent share per strata**
Infected	Anemic	12	3.08	66.66
	Non-anemic	6	1.54	33.33
Non-infected	Anemic	95	24.35	25.53
	Non-anemic	277	71.02	74.46

Table 6: prevalence of bovine trypanosomosis and associated risk factors in Dangur district

Risk factors	**No. examined**	**No. positive**	**Prevalence (%)**	**χ^2**	**p-value**
Sex					
Male	102	4	3.92	0.85	0.35
Female	288	14	4.86		
Total	**390**	**18**	**4.62**		
Age group (years)				0.110	0.946
< 2	120	5	4.16		
2 – 7	235	12	5.1		
> 7	35	1	2.85		
Total	**390**	**18**	**4.62**		
Body conditions				32.75	0.000
Good	140	2	1.42		
Medium	115	5	4.35		
Poor	135	11	8.15		
Total	**390**	**18**	**4.62**		

5. Discussion

The present study revealed an overall prevalence of 18/390 (4.62%) in the study area. This finding was lower than reported from earlier works of (Mekuria, S et al., 2011) which was reported 20.74% prevalence of bovine trypanosomosis and its vector in Metekel and Awi zones of North west Ethiopia and also reported 24.7% prevalence of bovine trypanosomosis by (Ali, D. et al., 2011) in Mao- Komo special district. The lower prevalence of trypanosomosis recorded in Bovine in this study may be due to the establishment of Assosa tsetse fly and trypanosomosis Control and surveillance center under national institute for control and eradication of tsetse fly and trypanosomosis,

which practice on application of control measures such as; pour on by Deltametrin 1% on the back of animals and deployment of traps and targets and treatment of sick animals.

The study showed that the infection was predominantly caused by *T. congolense 12/18 (*66.66%), *T. vivax* 4/18(22.22%), T. *brucei* 1/18(5.55%) and mixed infection 1/18(5.55%). This result was in agreement with prior reports of (Mekuria, S et al., 2011) who studied prevalence of major trypanosomes affecting cattle in the neighboring Asosa district of Benishangul Gumuz Regional State, Western Ethiopia and found *T. congolense* proportional prevalence of 66. 7%; (Abraham Z. et al.,2012) worked on the prevalence of bovine trypanosomosis in selected district of Arba Minch, Sothern Ethiopia and reported *T. congolense* proportional prevalence of 61.4%; (Biyazen, H. et al., 2014) reported *T. congolense* proportional prevalence 63.64% during his work on trypanosomosis and anemia in cattle population of Dale Wabera district of Kellem Wollega Zone, Western Ethiopia; Bayisa, K et al.,2015 demonstrated *T. congolense* proportional prevalence of 85% during his research on cattle trypanosomosis prevalence in Asossa district, Benishangul Gumuz Regional State, Western Ethiopia.

The high proportion infection rate of *T. congolense* in cattle might be attributable to the high number of serodems of *T. congolense* relative to *T. vivax*. It could also be due to the possible development of better immune response to *T. vivax* by the infected animals as demonstrated by (Leak, S ***et al***., 1993). 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 (Langridge WP.,1976; Leak, S *et al*.,1999). Different studies (Leak, S *et al.,* 1993; G. J. Rowlands *et al.,* 1995) have indicated that *T. vivax* is highly susceptible to treatment while the problems of drug resistance are higher in *T. congolense*, and mainly confirmed in the blood, while *T. vivax* and *T. brucei* also invade the tissues (L. E. Stephen.,1986).

There was a significant difference ($p<0.05$) in the prevalence of trypanosomosis among the study sites and body condition. This result is in agreement with previous reports (Mihreteab, B *et al*., 2011; 29-31, Ayele, T., *et al*., 2012; Lelisa, K *et al*., 2015). The overall anemia prevalence in the studied district was 27.43% (107/390). The anemia prevalence was significantly higher in trypanosome infected cattle (66.66%) than in non-infected cattle (25.53%) $p<0.05$. This is in concordance with previous results from different researchers (Mihret et al., 2007; M. Bekele et al.,2011, Biyazen, H et al., 2014). Out of 27.43% anemia prevalence, 3.07% (12/390) was trypanosome infected animals. Nonetheless, 24.36% (95/390) of non-infected animals were found to be anemic (PCV < 24). This indicates the fact that other factors such as gastrointestinal parasitism, nutritional deficiencies, fasciolosis and vector-borne diseases could affect the PCV value of cattle (P. van den Bossche et al., 2001).

This study revealed that 1.54% (6/390) of the cattle was infected by trypanosome; however, their PCV was laid in the normal range. This might be attributed to the capability of infected cattle to maintain their PCV within the normal range for a certain period of time. It could also be possibly due to inadequacy of the detection method used (Murray, M ***et al***., 1988), other anemia causing diseases (P. van den Bossche ***et al***., 2001), or delayed recovery of the anemic situation after current treatment with trypanocidal drugs. Furthermore, the occurrence of positive animals with PCV of greater than 24% might be thought of as recent infections of the animals (P. van den Bossche ***et al***., 2001).

The overall mean PCV value for examined animals was 26.47 ± 2.66 SE. The mean PCV value of the infected animals was significantly lower (21.78 ± 2.44 SE) than that of uninfected animals (26.78 ± 2.21 SE). This result is in alignment with previous works (Ali, D. Eta., 2011, Mulaw, S ***et al***., 2011, Bayisa, K ***et al***., 2015).

Glossina tachinoides was the only tsetse fly caught and its mean apparent density measured as f/t/d was 0.25. It accounts for 30 (54.55%) out of the total flies caught. In addition, other mechanical transmitters of trypanosomosis such as stomoxys 13 (23.64%), Tabanid 10 (18.18%) and Haematopota 2(3.63%) were recorded. The current findings were lower than previous works of (Solomon, M et al., 2010) at Metekel Awi zones of Northwest Ethiopia, who reported 6.49 f/t/d and 0.65 f/t/d for tsetse and biting flies, respectively. It was also lower than findings of (NTTICC, 2004) at Bure Iluababor zone of Western Ethiopia which was reported to be 7.23 f/t/d, 3.13 f/t/d and 0.06 f/t/d for tsetse fly, *Stomoxys* and *Tabanus*, respectively.

This result was also lower than the previous findings of (NTTICC, 2012-2014) at neighbouring Mandura district of western Ethiopia which was reported to be 3.59 and 1.16 f/t/d; 0.15, 0.20 and 4.5 f/t/d; 0.02, 0.05 and 0.33 f/t/d; 0.014, 1.38 and 4.5 f/t/d) for tsetse fly, *stomoxys, tabanus* and *haematopota*, respectively. Similarly, It was also lower than the previous findings of (NTTICC, 2012 & 2014) in Dangur districts of western Ethiopia which was reported to be 1.14 f/t/d; 4.04 & 0.09 f/t/d; 3.84 & 0.04 f/t/d and 0.4 & 0.6 f/t/d) for tsetse fly, *stomoxys,*

tabanus and *haematopota*, respectively. The current lower fly apparent density may be due to control measures such as deployment of targets and traps, and pour on application by Deltamethrin1% on the back of animals.

6. Conclusion

The most common trypanosomes species was *T.congolense* followed by *T.vivax*. The animal parameters such as sex and age were not found to be a risk factor; however, study site and body conditions were identified as risk factors. The mean PCV value of infected animals was significantly lower than that of uninfected animals indicating the adverse effect of trypanosomosis on the PCV profile of cattle. Trypanosomes were not detected in some anemic cattle indicating the occurrence of other causes of anemia in the area. *G.tachinoide* was the only tsetse fly species discovered in this study. Other mechanical transmitters of trypanosomosis such as stomoxys, tabanus and haematopota were recorded in the area. In wrapping up, trypanosomosis is an economically important disease threatening the health and productivity of cattle in Dangur district. Therefore, proper control strategies have to be designed and implemented to minimize its effect on livestock production in the studied district.

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