



## Epidemiology of Trypanosomosis in Domestic Ruminants and Donkeys in Asosa Zone, Northwestern Ethiopia: Prevalence and Vectors Involved

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**Key words:** Asosa, Bambasi, domestic ruminants, donkeys, trypanosomosis, vectors

**Abstract:** African animal trypanosomosis is among the most serious livestock diseases alleviating food sufficiency in Sub-Saharan Africa. Accurate information related to the prevalence, vectors distributions and densities have of great importance in designing the appropriate control and interventions strategies to combat trypanosomosis and its vectors. This study aimed to generate a base line data to implement the trypanosomosis and tsetse control operations. A cross-sectional involved dark phase ground buffy coat technique and deployments of baited traps was conducted from November 2014 to March 2015 in Asosa and Bambasi districts of Asosa Zone, Northwestern Ethiopia. Positive samples were stained by Giemsa's in thin blood smears, fixed with methanol for 5 min and examined under oil immersion using 100× objectives to identify the species of trypanosomes. *Glossina morsitans submorsitans* was the only caught tsetse fly species with an apparent density of 5.77 fly/trap/day. Among 42 *G. m. submorsitans* dissected, five (11.90%) were found to harbor *T. congolense* (9.52%) and *T. vivax* (2.38%). The prevalence of trypanosomosis was significantly higher in cattle (8.55%) than in donkeys (2.35%), goats (1.68%) and sheep (0.00%). In all infected study animals, *T. congolense* was the dominant trypanosome species (97.32%); *T. vivax* accounts 2.68% of the total infections. The mean PCV in trypanosome-infected animals was lower than in uninfected ones. African animal trypanosomosis is an important threat to animal health in the studied area and is not only a disease of concern in cattle but also in goats and donkeys. To this end, sustained interventions of trypanosomosis and its vectors, considering the epidemiological importance of cattle, small ruminants and equines is a prerequisite for the enhancement of livestock production in the areas where tsetse fly and trypanosomosis is prevalent.

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## INTRODUCTION

Ethiopia has an estimated 52 million heads of cattle, 46 million small ruminants, 9 million equines and 1 million camels. In spite of the large livestock population, the productivity is constrained by insufficient nutrition, poor husbandry systems and livestock ailments<sup>[1]</sup>. African animal trypanosomosis is among the most serious livestock diseases in sub-Saharan Africa<sup>[2, 3]</sup>. Trypanosomosis is a complex disease caused by unicellular parasites found in the blood and other tissues of vertebrates. It is transmitted cyclically by tsetse flies and/or mechanically by biting flies other than tsetse flies.

Trypanosomes can infect all domesticated animals but in parts of Africa, cattle are the main species affected due to the feeding preferences of tsetse flies. *T. congolense*, *T. vivax* and *T. brucei* *brucei* are the principal trypanosomes in domestic ruminants<sup>[4, 5]</sup>. In Ethiopia, the disease is considered as the most important in cattle but can also cause serious losses in pigs, camels, sheep and goats. However, the epidemiology and distribution of trypanosomes infection in domestic animals, cattle, small ruminants and donkeys is less investigated.

The degree of risk to which domestic animals are exposed to the trypanosomosis depends on the species and density of tsetse fly present, infection rate in the vectors, species and strain of trypanosomes, source of infections and feeding preference of the vectors. African animal trypanosomosis is mainly transmitted cyclically by tsetse flies which inhabit many parts of the continent that are restricted to latitude of about 15°N and 29°S of the equator. Of the three groups of *Glossina*, the savannah and riverine are the most important since they inhabit grazing lands.

In Ethiopia, *Glossina* species inhabit southwestern and northwestern regions which lies between longitude 33° and 38°E and latitude 5° and 12°N<sup>[6]</sup>. Five species of tsetse flies including *G. m. submorsitans*, *G. pallidipes*, *G. tachnoides*, *G. fuscipes fuscipes* and *G. longipennis* have been reported in Ethiopia. Currently, an estimated area of 220,000 km<sup>2</sup> is infested with one or more species of tsetse flies<sup>[7]</sup>.

In Benishangul Gumuz Regional State, tsetse fly infested area is estimated to be 31,000 km<sup>2</sup>. In the region, particularly in Assosa Zone the cyclical vectors of trypanosomosis, *G. morsitans submorsitans* were reported exist<sup>[8, 9]</sup>. In this region, some prevalence studies have been conducted on bovine trypanosomosis<sup>[10]</sup>. Nevertheless, information for other species of domestic ruminants and donkeys and the density of biological vectors is not well known. Therefore, the present survey was conducted before implementing the control strategies with the aim of determining the prevalence and distribution of trypanosomosis in domestic ruminants and donkeys, the species and density of tsetse flies in Asosa zone.

## MATERIALS AND METHODS

**Study area description:** The study was conducted from November to March 2014 in Assosa zone of Benishangul Gumuz region. The area is characterized by low land plane with altitude range of 580-1544 meters above sea level. Assosa is located between 8°30' and 40°27'N and 34°21' and 39°1'E. The average annual rainfall is 1316 mm with uni-modal type of rainfall that occurs between April and October. Its mean annual temperature ranges from 16.75 and 27.9°C.

**Study animals and sampling strategy:** The survey covered four species of domestic animals: bovine, Caprine, Ovine and Equine. Samples were collected from existing local zebu breeds managed under traditional communal grazing system. The sampling took place in November 2014 to March 2015. The number of animals required for the study was determined using the formula given by Thrusfield<sup>[11]</sup> for simple random sampling considering 95% level of confidence, 50% expected prevalence and 0.05-desired absolute precision. Although, sample from 384 animals is sufficient according to this formula, samples from 1146 cattle, 476 goats, 85 donkeys and 220 sheep were examined for trypanosomosis.

**Study design:** A cross-sectional study was used to estimate the prevalence trypanosomosis in domestic animals and to assess the distribution and density of tsetse fly involving in transmission of African animal trypanosomosis. The district's were purposively selected based farmers complaints of the problem and to avail base line data, prior to implementing trypanosomosis and its vectors control operations as one of NICETT project. Study sites were selected based on the accessibility of roads and sampling animals. All herds in selected peasant associations were included while individual study animals were sampled using simple random sampling techniques.

### Study methodology

**Entomological survey:** Mono-pyramidal shaped traps (N = 155) were deployed randomly in different parts of the district by baiting with acetone, octenol and cow urine to attract the flies. Traps were deployed at intervals of 100-200 m for 48 h in possible tsetse fly habitats like watering points and livestock grazing grasslands. Global Positioning System (GPS) supported each trap deployment. Fly catch per trap per day (F/T/D) was determined to estimate the fly density<sup>[12]</sup>. The species of the tsetse fly was determined based on morphological characteristics following the standard procedures<sup>[13]</sup>.

**Parasitological survey:** Blood samples were collected by pricking the marginal ear vein and blood was drawn into heparinized capillary tubes (Deltalab S.L, Barcelona, Spain). One end of the capillary tube was sealed with a crystal sealant (Hawksley Ltd., Lancing, United Kingdom (UK) and then centrifuged at 12,000 revolutions per minutes for 5 min to separate the blood cells and to concentrate trypanosomes using centrifugal forces as buffy coat using microhaematocrit centrifuge (Hawksley and Sons, UK). PCV was measured using haematocrit reader. The capillary tubes were broken just 1 mm below buffy coat and expressed on microscopic slide, mixed and covered with 22×22 mm cover slip. Then it was examined under 40× objective of a microscope using dark ground buffy coat technique to detect the presence of the parasite<sup>[14]</sup>. Positive samples were stained by Giemsa's, fixed with methanol for 5 min and examined under oil immersion using 100× objectives to identify the species of trypanosomes<sup>[15]</sup>.

**Statistical analysis:** All statistical analysis was performed using STATA soft ware version 12 (Stata Corp, Texas, USA). Descriptive statistics were used to summarize data. Chi-square test was used to analysis the differences in trypanosomosis prevalence between species of animals. The two groups mean comparison test was used to assess the differences in mean PCV value between trypanosome positive and negative animals. The prevalence was calculated for all data as the number of infected individuals divided by the number of individual sampled and multiplied by 100. The fly density of tsetse fly was calculated by dividing the number of flies caught by the number of traps deployed and the number of days of deployment.

## RESULTS AND DISCUSSION

**Entomological result:** By deploying 155 mono-pyramidal traps in two districts, 1791 tsetse flies were caught. *G. m. submorsitans* were the only tsetse fly species caught and its mean apparent density measured as fly/trap/day (FTD) was 5.78. The fly density of *G. m. submorsitans* was 6.93 and 0.35 in Assosa and Bambasi districts, respectively (Table 1). Furthermore, 42 *G. m. submorsitans* were dissected to estimate the infection rate of trypanosomosis in cyclical vectors. Proboscis, midgut and salivary gland of each tsetse fly were examined for trypanosomosis. Out of 42 flies dissected and examined, five (three in midgut, one each in proboscis and salivary glands) were positive for trypanosomes. *T. congolense* and *T. vivax* were the trypanosomes species detected in tsetse fly with the infection rate of 11.90%. The infection rate of *T. congolense* and *T. vivax* was in tsetse fly was 9.52 and

Table 1: Entomological survey result in different study sites of Assosa and Bambasi districts

Districts	Peasant associations	<i>Glossina m. submorsitans</i> caught			
		Female	Male	Total	FTD
Assosa	Kusmangel	130	114	244	8.71
	Albo	26	27	53	3.78
	Abenden	333	568	901	28.10
	Abermo	189	185	374	13.35
	Baro	26	27	53	2.20
	Bashabuda	46	57	103	3.22
	Tsentsuale	0	0	0	0.00
	Subtotal	750	978	1728	9.93
Bambasi	Dabous	0	0	0	0.00
	Kesmando	8	9	17	0.60
	Village16	16	14	30	0.94
	Village47	3	2	5	0.32
	Shobora	4	7	11	0.34
	Subtotal	31	32	63	0.46
Total		781	1010	1791	5.77

2.38%, respectively. The distribution and density of *G. m. submorsitans* in the studied area was indicated in Fig. 1.

**Parasitological findings:** The overall trypanosome prevalence in the studied area was 8.55, 1.68, 2.35 and 0.00% in cattle, goats, donkeys and sheep, respectively. In all species of infected animals, trypanosomes infections were mainly due to *T. congolense* (97.32%) and few infections with *T. vivax* in cattle (2.68%). Trypanosome infection rate was significantly ( $p < 0.05$ ) varied among domestic animals in both districts (Table 2).

**Hematological findings:** PCV significantly decreased among infected animal comparing with that of non-infected animal in cattle but no statistically significant association were observed in goats and donkeys (Table 3).

The entomological survey revealed that *G. m. submorsitans* were the only tsetse fly species caught in both districts. The current findings were in consistent with previous works by Mekuria and Gadissa<sup>[16]</sup> in Metekel and Awi zones of Northwest Ethiopia which indicated *G. m. submorsitans* is the common tsetse fly species in northwestern Ethiopia. The dominance of this species was probably due to an unspoiled savannah environment available in the area. The apparent density of Glossina species was 5.77 fly/trap/day. Aki and Godeso<sup>[17]</sup> reported a tsetse fly density of 3.92 FTD of *G. m. submorsitans* in Bambasi district.

The result was much higher than previous reports by Lelisa *et al.*<sup>[18]</sup> who reported 0.06 FTD of tsetse flies in Mandura district and Aki *et al.*<sup>[19]</sup> reported a tsetse fly density of 0.72 FTD in Bulen district, Benishangul Gumuz Regional State, Northwest Ethiopia. The lower apparent density of tsetse flies in Mandura and Bulen districts might be relate with tsetse fly and trypanosomosis interventions by NICETT-Ethiopia as these two districts are its project area.

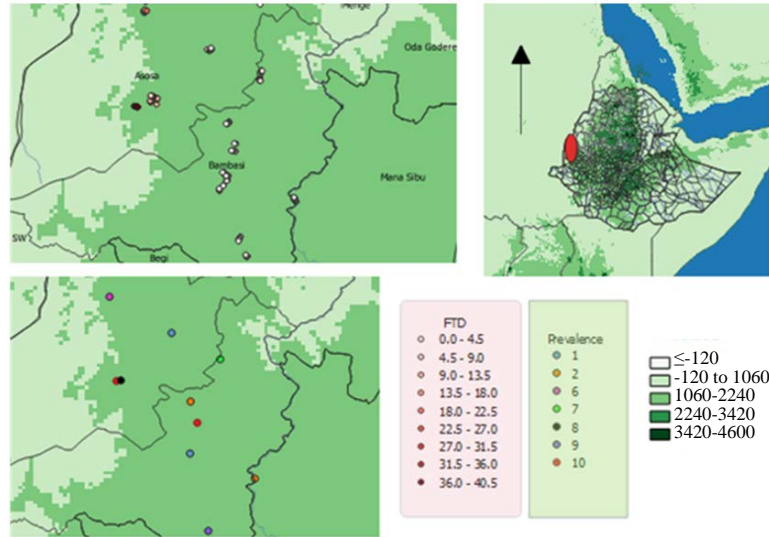


Fig. 1: Parameters of local area

Table 2: Relative proportion of trypanosome species in different animal species

Districts	Trypanosome species	Species of animals				$\chi^2$	p-values
		Cattle	Goats	Donkeys	Sheep		
Assosa	<i>T. congolense</i>	46(8.80)	8(2.19)	2(2.99)	0(0.00)	31.63	<0.001
	<i>T. vivax</i>	2(0.38)	0(0.00)	0(0.00)	0(0.00)		
	Sub total	48(9.11)	8(2.19)	2(2.99)	0(0.00)		
Bambasi	<i>T. congolense</i>	49(7.87)	0(0.00)	0(0.00)	0(0.00)		
	<i>T. vivax</i>	1(0.16)	0(0.00)	0(0.00)	0(0.00)		
	Sub total	50(8.03)	0(0.00)	0(0.00)	0(0.00)		
Overall	<i>T. congolense</i>	95(8.29)	8(1.68)	2(2.35)	0(0.00)		
	<i>T. vivax</i>	3(0.26)	0(0.00)	0(0.00)	0(0.00)		
	Total	98(8.55)	8(1.68)	2(2.35)	0(0.00)		

Table 3: Mean PCV of infected and non-infected animals in the study area

Species	Results	Mean PCV	SD	95% CI	t - values	p-values
Bovine	Negative	25.76	4.77	[25.47, 26.05]	10.4274	<0.001
	Positive	20.49	4.91	[19.50, 21.47]		
	Combined	25.31	5.00	[25.02, 25.60]		
Caprine	Negative	25.92	4.34	[25.52, 26.31]	0.3444	0.73
	Positive	25.38	7.09	[19.45, 31.30]		
	Combined	25.91	4.34	[25.51, 26.30]		
Donkeys	Negative	25.892	4.49	[24.91, 9.26]	0.9036	0.37
	Positive	23.00	2.83	[2.41, 48.41]		
	Combined	25.82	4.47	[24.86, 26.79]		

CI = Confidence Interval; PCV = Packed Cell Volume; SD = Standard Deviation

More female tsetse flies were caught than males. This finding was in comply with previous finding that indicated the mean catch of female tsetse fly is higher that of male<sup>[20]</sup>. Higher catches of female tsetse flies to could be attributable to their longer life span<sup>[21]</sup>.

The infection rate of trypanosomes in the biological vectors was 11.90%. Desta *et al.*<sup>[22]</sup> reported 6.93% trypanosome infection rate in tsetse fly in Amaro district, southern Ethiopia. *T. congolense* and *T. vivax* were the trypanosome species detected in tsetse fly with the infection rate of 9.52 and 2.38%, respectively. The

high ratio of *T. congolense* infections might suggest that *G. m. submorsitans* were more efficient transmitters of *T. congolense* than *T. vivax*<sup>[23]</sup>.

The prevalence of bovine trypanosomosis was 8.55%. Tafese *et al.*<sup>[24]</sup> who reported the prevalence of 8.55% in East Wollega and Mulatu *et al.*<sup>[25]</sup> who reported 8.50% in Dangur district, Northwestern Ethiopia, reported similar value. The prevalence of bovine trypanosomes in the studied area was lower than the previous reports from different districts of Northwestern Ethiopia: 28.1% in Asosa<sup>[26]</sup> and 24.7% in Mao-Komo special district<sup>[27]</sup>.

This finding was also lower than that of Yalew and Fantahun<sup>[28]</sup> who reported 21.50% infection rate of trypanosomosis in cattle in Bambasi district and Kenaw *et al.*<sup>[29]</sup> reported 22.8% prevalence of bovine trypanosomosis in Asosa district. The lower prevalence in this study might be associated with the differences in tsetse fly ecology of vectors of trypanosomosis and other epidemiological factors.

The prevalence of trypanosomosis in caprine (1.68%) agreed with the reports in Homesha and Asosa Kniepert<sup>[30]</sup> who reported 1.78% and Radostits *et al.*<sup>[31]</sup> who reported 1.96% prevalence of trypanosomosis in goats. The prevalence of trypanosomosis was 0.00%. This finding was far from that of Ayana *et al.*<sup>[32]</sup> and Lelisa *et al.*<sup>[33]</sup> who reported prevalence of trypanosomosis in sheep 8.70 and 4.50, respectively. This might be due to the low sensitivity of diagnostic technique used and the differences in the ecology of the areas.

The prevalence of trypanosomosis in donkeys was 2.99%. This result was higher than report from Awi zone, northwest Ethiopia by Kebede *et al.*<sup>[34]</sup> reported 1.60% trypanosomosis prevalence in donkeys. However, this finding was lower than reports on prevalence of trypanosomosis in donkeys: 6.3, 10.7 and 6.27% from different parts of Ethiopia<sup>[35-37]</sup>. These differences might be due to differences in sensitivity of diagnostic techniques<sup>[38]</sup> and the differences in the ecology of the areas.

The relative prevalence of *T. vivax* and *T. congolense* differed significantly between the animal species and between the study sites ( $p < 0.05$ ). In all studied animals except ovine and in all districts trypanosome infections were mainly due to *T. congolense* (97.32%) with some cases of *T. vivax* infections in cattle (2.63%). Nevertheless, *T. congolense* was detected in all animal species except ovine as well as in all the study sites *T. vivax* infection was only detected in bovine species in Asosa district. Indeed the result also agrees with other literature that indicated both *T. vivax* and *T. congolense* were generally economical important trypanosome species that mainly affect ruminants and equines<sup>[39,40]</sup>. The high proportion of *T. congolense* than *T. vivax* infection in all species and in all foci was similar with the findings of other researchers from different parts of Ethiopia.

The PCV value was performed having considered as normal 24-46 for bovine 22-45 PCV for sheep, 22-38 for goats and 32-50 for donkeys. The mean PCV were calculated to be 25.76 and 20.46 for bovine, 25.92 and 25.38 for caprine and 25.8 and 23 for donkeys' species for non-infected animals and infected animals, respectively. The difference in mean PCV value between parasitaemic and aparasitaemic animals indicates that trypanosomosis is one of the causes of anemia in domestic animals. Classically, infection with the trypanosome species that

are pathogenic in local breeds of cattle, *T. congolense* and *T. vivax* results in anaemia and an animal's nutritional status is key factor determining the outcome of any infection<sup>[41, 42]</sup>.

## CONCLUSION

*G. m. submorsitans* was the only tsetse fly species caught in the studied area with apparent density of 5.77 FTD. The prevalence of trypanosomosis in cattle, donkeys, goats and sheep was 8.50, 2.99, 1.68 and 0.00%, respectively.

Two trypanosome species were circulating in the studied area: *T. congolense* and *T. vivax* accounting for a proportion of 97.33 and 2.67%, respectively. Sustained interventions of trypanosomosis and its vectors considering the epidemiological importance of cattle, small ruminants and equines is a prerequisite for the enhancement of livestock production in the area to improve the livelihood of the society living in the studied area.

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