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Prevalence of Bovine Trypanosomosis in Didessa District, Oromia Region, Ethiopia

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Abstract: A cross sectional study was conducted at Didessa District, Oromia region, south west Ethiopia. The study was carried out from November, 2011 to April 2012 on indigenous cattle breed managed under mixed crop-livestock production system, to determine the prevalence of bovine trypanosomosis. This study employs parasitological survey by the use of Buffy coat examination, thin smear and hematological study. The overall prevalence of bovine trypanosomosis in the present study was 5.47%. The predominant species recovered was *Trypanosme congolense* (61.9%) followed by *Trypanosome vivax* (23.8%) the *Trypanasome brucei* (9.5%). Mixed infection due to *trypanosome congolense* and *Trypanosome vivax* (4.7%) was also recorded in the study. Discrepancy in the prevalence of trypanosome infection was recorded in the different age groups, between sex and different body conditioned animals, but the difference was not statistically significant ($p>0.05$). The mean PCV of parasitemic animals was significantly lower (21.15 ± 4.675) than the aparasitemic animals (24.316 ± 4.93) ($p<0.05$). Although the present study came up with low prevalence of bovine trypanosomosis in the study area, the potential impact of this disease on production and productivity of cattle shall not be undermined. Therefore, sustainable community based tsetse and trypanosomosis control program should be implemented.
Key words: Didessa, Ethiopia, Prevalence, Trypanosomosis.

INTRODUCTION

Trypanosomosis is disease caused by unicellular parasites (trypanosome) found in blood and other tissue of vertebrates; including livestock, wild life and people [1]. It is a serious disease in domestic livestock causing a significant negative impact on food production and economic growth in many parts of the world, particularly in sub-saharan Africa [2, 1, 3]. Its epidemiology and impact on livestock production are largely determined by the prevalence, and distribution of the disease and its vectors in the affected area [4].

This disease is transmitted mainly by tsetse flies (cyclically), biting flies (mechanically) and by other means of transmission [5, 3]. The most important species that infected cattle include *Trypanosome congolense*. *T. brucei* and *T. vivax*. Mechanically transmission is particularly important in relation to *T. vivax* and T. evansi particularly on the fringe of tsetse areas. It can occur in the presence of biting files of genus *Tabanus. Haematopia, chrysopas and stomaxys* [6].

Tsetse flies ingest trypanosome in blood or lymph node while feeding on the host. The trypanosome undergoes a cycle of development and multiplication in digestive tract of the fly until the infective metacyclic trypanosomes (Meta trypanosome) are produced [3]. They undergo a transformation losing their typical trypanosome or trypomastogote and metacyclic traypanosomes which are infective form of the host [7]. African trypanosomes lose infectivity for mammals when they enter the tsetse fly gut and must complete their development cycle with differentiation to the metacyclic stage before infective parasites can be transmitted in the tsetse saliva [8]. Approximately 30% of the total cattle population in African continent and about 50 million people are exposed to animal trypanosomosis and human sleep sickness respectively [9].

Tsetse flies (Glosina) inhabit wide range of habitats covering over 10 million km^2 representing 37% of the African continent and affecting 37 countries including Ethiopia [39]. In Ethiopia tsetse flies are confined to the southern and western regions. Between

the longitude 33^0 and 38^0 E and latitude 5^0 and 12^0N. Tsetse fly infested areas lies in the low land and also in the river valley of Abay (Blue Nile), Baro, Akobo, Ghibe, Didessa and Omo [10]. Currently about 220,000km^2 area is ifested with tsetse flies normally *Glossina fuscipes, Glossina tachnoides, Glossina pallidipes, glossina morsitans, Glossina Longipennis* [11]. About 15-20 percent of the low land believed to be suitable for livestock production by one or two species of the tsetse flies [12].

Bovine trypanosomosis is a serious constraint to agricultural production in extensive tsetse infested areas of Ethiopia lowlands [13]. Trypanosomosis is prevalent in two main regions of Ethiopia i.e. the northwest and the southwest regions [14]. In Ethiopia, Trypanosomosis is one the most important disease limiting livestock productivity and agricultural development due to its high prevalence in the most arable and fertile land of south west part of the country following the grater basins of Abay, Omo, Ghibe, Didessa and Baroo with a high potential for agriculture [11].

The most important trypanosome species affecting live stock in Ethiopia are *T. congolense, T. vivax, T.brucei,* in cattle, sheep and goats, *T. evansi* in camels, and *T. equiperdum* in horses [6]. In Giemsa stained blood smears the species are distinguished by their size, shape, location, the size of kinetoplast, position of nucleus and the attachment and length of flagellum. Trypanosomes move actively and progress by movement of the undulating membrane and the free flagellum when present [3]. Trypanosomes are characteristically leaf like shape, they are a single flagellum and attached to the organism by undulating membrane [15].

Animal affected by trypanosomosis is manifested anemia, generalized enlargement of superficial lymph nodes, loss of body condition, fever, and loss of appetite [7]. In recent years, a number of drugs effective against cattle trypanosomosis have been introduced for both curative and prophylactic use [5, 7]. Curative treatment is the most effective in herds that are inspected at regular intervals [3]. Control and strategies in the trypanosomosis concentrate on vector control, parasites control with chemotherapy and chemoprophylaxis and use their inherent trypano tolerant in some breeds of animals [6].

The economic burden of trypanosomosis is not only due to the direct losses resulting from mortality, morbidity and infertility of the infected animals but also it is due to the indirect losses like exclusion of livestock and animal; power based crop production from the huge fertile tsetse infested areas [16]. In Ethiopia about 5.5 million heads of cattle are exposed to the risk of trypanosomosis [17]. This disease reduces meat and milk production of animals recovering from it. In addition to this some drugs are costly to treat animals that are diseased. Despite bovine trypanosomosis, have impacts on cattle production and prevalence of the disease in the study area. Therefore the objective of this research was: To estimate the prevalence of bovine trypanosomosis, determine the species of trypanosomes and Determining PCV of animals.

MATERILALS AND METHODS

A number of materials were required for both study of the disease and writing of this thesis. The list of the materials were sides, cover slips, PCV reader, pippet or stick, Bunsen burner, haematocrit clay, haematocrit centrifuge, microscope and equipment used for blood collection, (needle, needle holder, test tubes and haematocrit tube, the reagent used for laboratory test like iodine, methyl alcohol, giemsa stain, methyl blue stain and others.

Study area

The study area was located in Oromia Region, Illu Ababor Zone at Didessa district. Dembi is the administrative center of Didessa district. This district is bordered by Gechi Borecha distirict in north, Limmu Korsa in east, Sattama in west and Gumay in south and found at south west of Ethiopia at the distance of about 420km from Addis Ababa. The topography of this district is characterized by plateaus of central and western plains of Didessa valley. The main river for this district is Didessa River and the tributaries Mulade, Asha and Dibo. Didessa is the smallest district in Ilu Ababor Zone with area of 615km$^{2.}$ The elevation varies in this area from 1360-2340 meter above sea level. The annual mean temperature for most part of the district is 13^0c – 28^0c and annual rain fall is about 900-1000mm. the climatic condition of the area includes: Dega, woina Dega and kola cover 16%, 64%, and 20% of the district respectively. The lands used for cultivation are cultivated land 4719.57 hectares, grazing and 9848.45 hectare, forest 10682.7 hectare and the other 724 hectare [18].

Study Population

Bovine with any age and either sex in Dinge Beraha and Busi Settlement area were selected by simple random sampling for the collection of samples. The total cattle population of the two settlement were 21,867 which is greater in number than other species of animals in that area [18]. A total of 384 local cattle breed were selected with 192 from each peasant association. The cattle selected for sample collection were indigenous zebu cattle in Ethiopia.

Study design

A cross sectional type of study was conducted to determine the current prevalence of bovine trypanosomosis in the study area. The selected cattle were categorized according to their body condition (good, medium and poor), sex (female and male) and age (< year, 2-3 year and >3).

Sample size and sampling method

The simple random sampling technique was applied to collect from the ear vein. The sample size was determined based on the study type and sampling method for investigation, 95% confidence interval, 5% desired absolute precision and 50% average prevalence and 384 cattle were sampled by the following formula [19].

$$n = \frac{1.96^2 P_{exp}(1 - P_{exp})}{d^2},$$

where: n = required sample size;
P_{exp} = expected prevalence;
d = desired absolute precision.

Study methodology

Buffy coat technique

A small blood was collected from an ear vein using heparinized microhaematocrit capillary tube. A haematocrit tube with a whole blood sample and end was sealed with hematocrit clay. The tube was centrifuged at 12000 revolutions per minute for five minutes. After centrifugation trypanosome were usually found in or just above the buffy coat layer. The capillary tube was cut using a diamond tipped pen 1mm bellow the Buffy coat to include the upper most layers of the red blood cells and 3mm above to include the plasma. The content of capillary tube was expressed on to side, homogenized on to clean side and covered with cover slip. The slide was examined under x40 objective x10 eye piece for the movement of the parasites [20].

Thin blood smear

Blood drop from a micro haematocrit capillary tube the applied on the side of clean slide and spread by using another clean side at angle of 45^0, air dried and fixed for 2 minutes in methyl alcohol, then immersed in Giemsa stain (1:10 solution) for 30 minutes [21] drained and washed of excess stain using distilled water, allowed to dry by standing up right on the rack and was examined under microscope with oil immersion objective lens. In Giemsa stained smears the species were distinguished by their size, shape, location and size of the kinetoplast, position.

Measuring of packed cell volume

Blood samples were obtained by puncturing marginal ear vein with lancet and collected directly in to a capillary tube. The capillary tubes were placed in micro haematocrit centrifuge with sealed end outer most. The tube was loaded symmetrically to ensuring good balance after screwing the rotators cover and closing the centrifuge lid, the specimens were allowed to centrifuge at 12,000 revolutions per minute for 5 minutes. Tubes were then placed in a haematocrit and readings were expressed as a percentage of packed cells to the total volume of whole blood. Animals with PCV<24% were considered to be anemic

Body Condition Scoring

The body condition was characterized good, medium and poor according to Nicholson and Butterworth [22].

Data Management and Analysis

Raw data individual animals and parasitological examination were inserted in to Microsoft excel spreadsheets to create a data base and transferred to SPSS version 17.0 software program for data analysis. Chi-square was used to compare the prevalence association with risk factors.

RESULTS

Out of the total 384 cattle examined 21(5.47%) cattle were positive for trypanosomosis, the prevalence of trypanosomosis were not statistically significant (P>0.05) between different sex (P=0.611), age (p=0.676) or body condition score (p=0.4) of the animals as indicated in tables 1, 2 and 3 respectively. From the total infected animals 13 (61.9%), 5 (23.8%), 2(9.5%) and 1(4.76%) were infected with *T. congolense*, *T. vivax* and with mixed infection of *T. congolense* and *T. vivax* respectively (Table 4).

Table-1: Prevalence of bovine trypanosome with sex in Didessa district

Sex	Frequency	Percent	Trypanosome infected	Rate of infection (%)	P – value
Female	203	52.9	10	4.9	0.611
Male	181	47.1	11	6.07	
Total	384	100	21		

Table-2: Prevalence of bovine trypanosome with age in Didessa district

Age	Frequency	Percent	Trypanosome infected	Prevalence	P – value
(< 1)	8	2.1	0	0	
(2-3)	92	24.0	5	5.43	0.67
>3	284	74.0	16	5.63	
Total	384	100	21		

Table-3: Prevalence of bovine trypanosome with Body condition in Didessa district

Body Condition score	Frequency	Numbers of animal infected with trypanosome	Prevalence	P–value
Good	69	3	3(4.30	0.4
Medium	150	6	6(4)	
Poor	165	12	12(7.27)	

Table-4: Prevalence of bovine trypanosome species in Didessa district

Species	Total	Frequency	Percent
T. brucei	21	2	9.52
T. congolense	21	13	61.9
T. vivax	21	5	23.8
Mixed (*T. congolense* and T. vivax)	21	1	4.76

The mean PCV of the animals infected with trypanosomes (21.857 ± 4.67) was significantly lower ($p<0.032$) than the average PCV of the animals that were parasitological negative was (24.316 ±4.93).

Table-5: Mean packed cell volume and standard deviation of infected and non-infected animal cattle in Didessa district

Condition	No. Examined	(PCV<24%)	(PCV>24%)	Mean pcv%± sd	p-value
Parasitemic	21(5.47%)	17(80.95%)	4(19.05%)	21.857≠4.675	0.03
Aparasitemic	363(84.5%)	173(47.66%)	190(52.34%)	24.316≠4.93	
Total	384	190(49.48%)	194(50.52%)		

DISCUSSION

The result of the present study revealed an overall trypanosomosis of 5.45%. This finding was lower than the previously reported prevalence rate of 23% in western Ethiopia, 21% in Metekal district, 18.5% in Arba-minchi district, 17.5% in upper Dideesa valley areas, and 11.7% in Abay Basin north western Ethiopia of tsetse infested regions [23- 27]. The lower prevalence in the current study might due to the low sensitivity of the parasitological diagnostic method, the uncontrolled use of trypanocidal drugs, application of relatively well designed method of tsetse control and treatment, expansion of cultivation in the area which in directly affects flies distribution and awareness of the people towards the control and treatment of the disease were improved. Even though, the data was not collected during rainy season [28] revealed that there is significantly high infection rate following the months with high rain due the emergency of biting flies. This implies that the low prevalence of trypanosomosis in this study may be related with decrease in fly population during dry season.

The findings of this study revealed that the majority of the infection was due to *T. congolense* (61.9%) followed by T.vivax (23.8%) and T. bruecei(9.5%). Mixed infection of *T. congolense* and *T. vivax* was also prevalent (4.76%). The higher proportion of *T. Congolese* infection study area was in agreement with trypanosome species prevalence data from other tsetse infested regions of Ethiopia, where the *T. congolense* is the most prevalent species in cattle [14]. The ratio of *T. congolense*, *T. vivax* and *T. brucei* was 2.6: 1:0.4 indicating high trypanosome infection due to *T. Congolese*. The percentage species distribution in our finding was similar to Nigatu his co-workers finding in Abay Basin (*T. congolense* (66.1%) followed by *T. vivax* (20.8%) [25]. The predominant species of T.*congolense* compared to *T.vivax* and the development of better immune response to *T.vivax* infected animals [29, 30].

The prevalence of infection between sex categories was 6.07% in male and 4.9% in female animals. However, there was no significance difference between sex groups (p=0.611).

There no significance difference observed in age groups in the study groups in the study period but relatively higher rates observed in adult (>3 years) animals when compared to young animals. This may due to exposure of adult animals for the tsetse fly [40]. When they are freely grazing and also may be due to immune suppression as a result of stress factors such as lactation and when they are travelling a long distance through tsetse challenging are for drafting purpose in males. The low prevalence in young animals may also be due to the natural protection to some extent by maternal antibodies [31].

Although higher infection rate was observed in poor body conditioned animals as compared to good and medium, in the present study no statistically difference was observed between these groups (p=0.4). Similar results were reported from Tselemty district, Western Tigtay, Northern Ethiopia [32].

PCV is the most reliable indicator of anemia in trypanosomosis [33, 34]. In our study trypanosome infection results in a significant decline in PCV, this is in agreement with previous finding that are reported by different authors at different time [35, 36]. The mean

PCV values of studied animals was significantly ($p<0.05$) lower in parasitemic (21.857 ± 4.857%) than in aparasitemic (24.316 ± 4.93%) animals. This result was in agreement with the previous result by [28]. The appearance of parasitologically negative animals with PCV values of less than the threshold values (24%) may be due to the inadequacy of detection method used or delayed recovery of aneamic situation after a recent treatment with trypanocidal drugs; and the occurrence of positive animals with PCV of greater than 25% might it be thought of recent infection. Trypanosome infection and mean values obtained in this study in the parasitemic animals was found to be highly associated. Similar results were also reported by different authors in southern, north western and south western Ethiopia [37, 25, 30]. It was generally accepted that the mean PCV may be affected by many factors (helmenthiasis, tick born disease and nutritional imbalance) other than trypanosomosis. However, these factors are likely to affect both trypanosomosis positive and negative animals [38].

CONCLUSION

The overall prevalence of bovine trypanosome infection in study area was lower than the previous report but it is important disease that affects the health as well as productivity of cattle in Didessa district. There was no statistically significant difference between sex, ages, and body condition. The mean PCV of parasitemic animals due to *T. Congolese, T.vivax,* and *T. brucei* and mixed was significantly lower than aparasitemic animal.

Although, the present study low prevalence of bovine trypanosomosis in the study area potential impact of *T. congolense, T. vivax, T. burucei* infection on production and productivity of cattle shall not be undermined. Therefore Sustainable community based tsetse and trypanosomosis control program should be implemented and Use of the trypano-tolerant breeds of animals in the highly prevalent area should also be recommended.

Competing interest

The authors declare that they have no competing interest.

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