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Assessing the impact of *Trypanosoma* spp. in cattle and its vector infestation in controlled and uncontrolled Kebeles in Kucha Wereda, Southern Ethiopia

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This study was undertaken in Kucha Woreda to assess the impact of *Trypanosoma* spp. on cattle and its vector infestation rate using cross-sectional study design. The study was done from November 2016 to June 2017 on 384 local cattle using buffy coat method and tsetse fly density was seen in intervention area with non-controlled area. The overall infection rate 4.17% (N=16) showed an insignificant (p>0.05) difference of prevalence rate in animal with different body condition score. Medium body condition animals were the most affected animal group. The study also revealed that adult (4.22%) and male (6.12%) animals are relatively susceptible to bovine trypanosomosis than young (4.00%) and female (2.95%) animals. Animals with PCV value of 21.56 and 26.32% were found to be parasitaemic and aparasitemic cattle, respectively. In both research site, 20 NGU trap was deployed to see the vector infestation and only *Glossina pallidipes* was found together with mechanical vectors. 2.86 and 26.27 FTD were found in both controlled and uncontrolled kebele, respectively, this big difference resulted in uncontrolled Kodo Wono kebele due to absence of intervention by Arba Minch tsetse fly suppression site. The total tsetse fly infestation rate in the area was found to be 14.67 FTD. Due to its adverse effect on cattle production, parasitic and vector control and prevention mechanism should be in place especially in those kebele neighbors to the controlled kebele.

Key words: Cattle, Kucha, NGU trap, trypanosoma, prevalence, vector.

INTRODUCTION

Trypanosomosis has long been recognized as a massive constraint to animal husbandry, livestock production and mixed farming in vast areas of rural sub-Saharan Africa (Oluwafemi, 2014). Ethiopia is known for its large and diverse livestock resource endowments. Livestock are primarily kept on small holdings where they provide drought power for crop production, manure for soil fertility and fuels, serve as sources of family diet and cash income (from livestock and livestock products).

Despite large livestock population, Ethiopia fails to optimally utilize this resource due to different constrains facing the livestock subsector (Bezabih et al., 2015).

Since more than 90% of crop production in Ethiopia are dependent on animal draught power mainly on ploughing oxen, many large fields lie fallow due to lack of these animals in trypanosomiasis infested area (Haile et al., 2016).

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This worsens the food supply and living conditions in affected areas. Trypanosomes are flagellated protozoan parasites that live in the blood and other body fluids of vertebrate hosts (International des Epizooties (OIE), 2013). Bovine trypanosomosis is one of the economically important diseases that are caused by this flagellated protozoal parasite belonging to the genus *Trypanosoma* which affects all domestic animals (Jember and Mitiku, 2013; Sharma et al., 2012; Singla et al., 2009; Sood et al., 2011).

The monitoring of impact assessment is a prime concern for the effectiveness of any control program which has to meet at least three criteria (Slingenbersh, 1999). First, they should be economically sound and sustainable, secondly the direct and indirect effect on the environment should be minimal and the third and most important is that they should fit into the rural development policy of a country. The impact of tsetse control can be best assessed by comparing changes before and after the implementation of a control intervention or alternatively by cross-sectional comparison of similar agricultural area under different level of trypanosomosis challenge or both (Van den Bossche and Rowlands, 2001).

According to annual report of Ethiopian Veterinary Association in 2008, most budget allocated to rural development bureau is to buy drugs and spray to treat and control animal trypanosomosis. In Southern region of Ethiopia, the problem of tsetse is common where a total of 75 districts are fully or partially infected. The infected area is about 48,000 km square. In the study area there was no information regarding the prevalence rate of the parasite and vector infestation level. Based on this, the study was conducted with the objectives of determining infection rate of *Trypanosoma* spp. in cattle, identifying the infestation level of vector (tsetse fly) and determining predisposing factors that contribute to the occurrence of the disease in controlled Masha Morka and uncontrolled Kodo Wono kebele, Kucha Woreda, South west Ethiopia.

MATERIALS AND METHODS

Study area

The research was done from November 2016 to June 2017 in controlled Masha Morka and uncontrolled Kodo Wono kebele, Kucha Woreda, South West Ethiopia, which is located in the southern rift valley of Ethiopia. The area is sub-humid climate with moderately hot temperature. It has a min and max annual rain fall of 900 and 1800, respectively. Belg starts from January to April, and long rainy season, Maher occurs from June to September. The vegetation is dominantly covered by wooded grass land (WGL) especially along the side of grazing area draining lines and there is a high gallery forest along the rivers (Van den Bossche and Rowlands, 2001).

Retrospective impact assessment

The retrospective data were collected from written documents, different booklets and annual report from Kucha Woreda rural development office and tsetse control project of Wolaita Sodo station.

Study animals and study type

This research was undertaken to assess the impact of *Trypanosoma* spp. in cattle and its vector infestation rate using cross-sectional study design in controlled Masha Morka and uncontrolled Kodo Wono Kebele, Kucha Woreda. Cattle with different sexes, age group and body condition score were included in the present study. Systematic random sampling technique was used to choose the study animals.

Estimation of sample size

Since there was no previous study conducted at controlled Masha Morka and uncontrolled Kodo Wono kebele to estimate the rate of *Trypanosoma* spp., therefore the sample size was allocated by taking an expected prevalence of 50%. Using the methods designed by Thrusfield (Thrusfield and Robert, 2018), the sample size was determined to be 384 cattle at 95% confidence interval and 5% absolute precision.

Study protocol

Collection of samples

The prevalence determination of *Trypanosoma* spp. in cattle was made by buffy coat technique. Blood sample was collected by piercing the marginal ear vein with a sterile lancet and blood was drawn by a hematocrit capillary tube. Then one end (the heparinized end) of capillary tubes was sealed with crystal sealant and centrifuged at 12,000 rpm for five minutes to separate the blood cells and to concentrate trypanosomes using centrifugal forces. Then the Packed Cell Volume (PCV) was determined by PCV reader and recorded. The PCV values ≥25 and <25 were considered as non-aneamic and aneamic, respectively. The buffy coat was dropped on microscopic slide and covered with a cover slip and then examined under 40X magnification power of microscope to identify and detect the presence of the parasites (Getachew, 1993). Those positive samples were further processed using geimsa staining for identification into species based on morphological characteristics under oil immersion using 100 x objectives (Parashar et al., 2018).

Entomological survey

A total of 20 NGU traps were deployed in both controlled and under controlled kebeles in the district (10 in Masha Morka and 10 in Kodo Wono) at approximate interval of 200-250m. All traps were baited with cow urine and left bottles open. Each trap poles near the ground were smeared with grease and the traps were deployed out of tree shade in order to prevent the ants from climbing up the pole. After 72 h of deployment, all cages were collected and tsetse flies in the cages were counted. Identification of tsetse fly was made based on their habitat and morphology to the genus and species level. Other biting flies were also identified according to their morphological structures such as size and proboscis at the genus level (Muturi et al., 2000). Male flies were identified by their enlarged hypopygium in the posterior ventral end of the abdomen using a hand lens. The apparent density of the tsetse fly was calculated as the number of tsetse catch/trap/day (Erkelens et al., 2000).

For the entomological study, tsetse flies were collected by 20 NGU trap in different positions of the study areas 10 in Masha

Table 1. Total No of *Glossina* and other biting flies caught in study areas.

Kebele	Altitude	Types of flies			Total	Fly/trap/day
		Fly	Sex			
			M	F		
Masha Morka	1360	G. *pallidipes*	27	59	86	2.86
		Tabanus	-	-	5	0.16
		Stomoxys	-	-	7	0.23
Kodo Wono	1355	G. *pallidipes*	297	491	788	26.27
		Tabanus	-	-	65	2.16
		Stomoxys	-	-	53	1.76

Buffy coat result.

Table 2. Level of *Trypanosoma* spp. in cattle on both kebele.

Kebele	Frequency		Rate (%)
	+ve	-ve	
Masha Morka	4	188	4
Kodo Wono	12	180	6.3
Total	16	368	4.17

χ^2=4.174, df=1, p=0.041.

Table 3. Level of *Trypanosoma* spp. on the basis of age of the animal

Age	No of animal	Frequency		Rate (%)
		Positive	Negative	
Young	100	4	96	4
Adult	284	12	272	4.22
Total	384	16	368	4.17

χ^2=88.1, df=1, p=0.00.

Morka and 10 (10 traps in Kodo Wono kebele. Bovine urine was used as a bait to attract the flies. Traps were positioned at approximate intervals of 100 to 200 m for 72 h in watering and grazing points in which the animals and the vector are believed to have frequent contacts (FAO, 1992). Fly catch per trap per day (f/t/d) was determined to calculate the fly density and distribution (Leak et al., 1987). The species of the dominant tsetse fly was determined following the standard procedures (Muturi et al., 2000) and biting flies according to their morphological characteristics such as size, color, wing venation structure, and proboscis at the genus level.

Data management

The data obtained from parasitology and entomological survey were entered into Microsoft Excel spreadsheet and decoded. The data were summarized and presented in tables and analyzed by using SPSS version 20. Tsetse fly density was calculated by dividing the number of flies caught by the number of traps deployed and number of days of deployment and expressed as Fly/Trap/Day (F/T/D). One way ANOVA was used to determine the association between risk factor and Trypanosoma infection. Descriptive statistic was used to determine the rate of cattle Trypanosoma in both infested and controlled areas.

RESULTS

Entomological survey

G. pallidipes was the only tsetse fly species found in the district and two genera of biting flies (*Tabanus* and *Stomoxys*) were identified during the entomological survey. The FTD of Glossina species and other biting flies were indicated in Table 1. The overall Glossina species caught per 72 h with sex proportion in controlled and uncontrolled kebeles are shown in Table 2.

Buffy coat result

16 (4.17%) animals out of 384 cattle in controlled Masha Morka and uncontrolled Kodo Wono kebeles harbored the parasite. There was statistically significant difference between the two sites (χ^2=4.174, p<0.05) with 2.1% in Masha Morka and 6.3% in Kodo Wono as indicated in Table 2.

As shown in Table 3 adult cattle are the most infected

Table 4. Level of *Trypanosoma* spp. on animal different body condition.

BCS	No, of animal	Frequency		Rate (%)
		+ve	-ve	
Poor	112	4	112	3.57
Medium	248	12	236	4.83
Good	24	-	24	-
Total	384	16	368	4.17

χ^2=2.252, df=2, p=0.324.

Table 5. Level of *Trypanosoma* spp. on the basis of sex of the animal.

Age	No. of animal	Frequency		Rate (%)
		+ve	-ve	
Male	147	9	138	6.12
Female	237	7	230	2.95
Total	384	16	368	4.17

χ^2=10.336, df=2, p=0.234.

Table 6. Level of *Trypanosoma* spp. on the basis of hematocrit result.

PCV reading (%)	Frequency		Rate (%)
	+ve	-Ve	
PCV≤24	10	144	15.4
PCV≥24	6	224	2.6
Total	16	368	4.17

χ^2=0.711, df=1, p=0.002.

and exposed age group for trypanosomosis as compared with young cattle and statistically significance difference exist ($p<0.05$). Depending on body conditions, animals with medium body condition score have high level of *Trypanosoma* spp. 4.22% (12) in their blood as compared with animal having poor body condition score 3.57% (4) and 0% (0) in good body condition score but this variation was not statistically significant ($p> 0.05$) (Table 4). As indicated in Table 5 male cattle are the susceptible group to trypanosomosis infection as compared with female and the variation was insignificant ($p>0.05$).

Hematocrit result

The mean PCV value for the parasitemic and aparasitemic cattle was 21.56 and 26.32%, respectively. Animals with PCV≤24% (anemic) were 15.4% while those with PCV>24% (non-anemic) were 2.6%. The proportion of anemic animals infected with the parasite was significant ($p<0.05$) as compared with non-anemic positive animals (Table 6).

DISCUSSION

Out of 384 blood sample examined from cattle 16 (4.17%) samples were harboring the parasite with higher prevalence in uncontrolled Kodo Wono compared to 12 (6.3%) in controlled Masha Morka which account for 4 (4%). This difference was due to the presence of different control mechanism of the disease vector in former kebele (Table 1). The results are in close association with the report by Getaneh and Tewodros (2017) and Adugna et al. (2017) in Amhara region having overall prevalence of 6.77 and 7.3%, respectively. Other reports in Oromia by Dano et al. (2014) (7.81%); Gamechu et al. (2015) (4.85%); Geremew et al. (2016) (3.9%) and Netsa et al. (2018) (5.76%) and in SNNPRS by Nigussu (2017) 5.83%) and Tamirat and Tsegaye (2018) (5.2%) were in line with the finding of this study.

The present study revealed that the vector infestation in the area was found to be 14.67 FTD. This finding was in strong association with the report by Achenef and Admas (2012) (16.0 FTD; Teferi and Biniam (2018) (11.77FTD); Kassaye (2015) (13.01FTD) in different parts of Ethiopia.

But the finding is not in line with the report by Sheferaw et al. (2015) 0.47FTD and Nigatuwa and Wondimagegnehu (2016) (0.067FTD). This difference may be due to difference in geographical area, season of the year and Muturi et al. (2000) which reported about 1.4 FTD in the southern rift valley of Ethiopia and the apparent tsetse fly density obtained in the present study was 2.86 and 26.27 flies/trap/day in Masha Morka and Kodo Wono, respectively; this high reduction of mean apparent density of the tsetse flies at Masha Morka kebele is because of the presence of different control measure and release of sterile male tsetse flies(the application of SIT) undertaken in the area.

Variations exist on infection rate of cattle with different age group by *Trypanosoma* species. Adults cattle were found to be the most affected age group 12 (6.3%) as compared with young animals 4 (4%) from the total positive animal in this study. This implies that adult animals are more exposed to vector (tsetse fly) bite than young animal. This could be due to the fact that adult animals are highly mobile for grazing and watering near forest (Maze national park), water point and other predisposing factors such as traveling long distance, work load on adult cattle. Young animals were more resistant than adults to the effects of trypanosomosis due to the fact that young animals were kept at home and not sent for grazing for this reason; they are not commonly exposed to the vector (Fines, 1970).

In the present findings; animals with different body conditions score harbour different level of *Trypanosoma* spp. in their blood which was 4.83% in medium, 3.57% in poor and none in good body condition score. Various reasons associated with this like high number of medium body condition scored animals during study period and the habit of local farmer to treat their animal by trypanocidal drug is very high. As far as the level of *Trypanosoma* infection in different sexes is concerned, the present study revealed higher level of infection occurred in male animal 9 (6.12%) as compared with female one 7 (2.95%). The variation was not statistically significant. This result is similar with the report of Biyazen et al. (2014) and Kassaye and Tsegaye (2016) in Dale Wabera District and Dale sedi and Wabera district. But the present finding is not in line with the report by Kitila et al. (2016) who revealed the prevalence of trypanosomosis was higher in the female cattle (8%) in Yayo District. The difference could be due to female cattle were kept for giving offspring and milking for a long period of time and low in numbers than male cattle used for ploughing purpose, leading to the continuous exposure of tsetse flies infestation.

The mean PCV of parasitemic animals was significantly lower than that of aparasitemic ones ($p<0.05$) (Table 6). The prevalence of parasitemic and anemic animals (62.5%) was significantly compared to the parasitemic and non-anemic animals 37.5% ($p<0.05$). Similar findings were reported by Zecharias and Zeryehun (2012), Haile et al. (2016, 2017) and Mekonnen and Negesse (2017). Packed cell volume has been demonstrated to be a good indicator of trypanosomal infection (Marcotty et al., 2008). The aparasitemic cattle with PCV≤24% in the current study could be either due to the precision of the techniques used and the experience of the laboratory technician or it might be due to other factors like poor nutrition and hemoparasitic disease which cause anemia (Picozzi et al., 2002).

Conclusion

The results of the present study on cattle *Trypanosoma* spp. proportion and its vector infestation survey in controlled and uncontrolled kebele of Kucha Woreda provides vital information of the status of the infection in infected and control area and the difference in fly density and disease condition in both infected and controlled areas. *G. pallidipes* together with biting flies is found to be the major flies' infestation and *G. pallidipes* is the only vector for bovine trypanosome infection of livestock in the study areas. The parasitological finding showed 4.17% infection rate of the woreda. The measurement of PCV gives a direct correlation of anemic animal with trypanosomosis infection and the detection of many cattle with PCV≤24% indicates the presence of more chronically infected cattle with low parasitemia, which is difficult to detect by the available diagnostic technique. Adult male animals with medium body condition had high level *Trypanosoma* spp. infection rate in this study. Above all tsetse fly suppression procedure should be implanted in all kebeles of the woreda to avoid re-infestation of tsetse flies from infected to controlled area. Farmers cooperative should be established to control the disease and its vector. This would support the effort of southern tsetse fly eradication project in the woreda and suppression of tsetse fly vector in all parts of the country to reduce the overall prevalence of bovine trypanosomosis. This can be done by applying advanced tsetse fly control system like the release of sterile male insect (SIT) across the tsetse infested areas.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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