

Prevalence of Bovine Trypanosomosis in Selected Tsetse Infested Districts of West Gojjam Zone, Amhara Regional State, Ethiopia

Solomon Tesfaye

Department of Veterinary Epidemiology and Veterinary Public Health,
College of Veterinary Medicine and Animal Science,
University of Gondar, P.O. Box: 196, Gondar, Ethiopia

Abstract: A cross sectional study was conducted in selected tsetse infested districts of West Gojjam zone, North West Ethiopia. The purposes of the study were to determine the current prevalence bovine trypanosomosis. Simple random sampling was used to select 384 cattle from the purposively selected five kebeles for collection of blood sample. In the parasitological survey, blood samples of cattle were examined using a buffy coat technique and thin smear under Gimsa stain. The packed cell volume value of each animal was also measured using hematocrit reader. The overall prevalence of trypanosomosis in the study districts was found to be 10.15% and no statistical significant difference in the prevalence among Kebeles involved in the present study. The trypanosome species responsible for the infection were *Trypanosoma congolense* (7.29%) followed by *Trypanosoma vivax* (2.86%) and mixed infection of these two species during study period was no observed. The prevalence of trypanosomosis with assessed risk factors, body condition and coat color of animals showed statistically significant variation ($P < 0.05$). The age categories and gender groups were found statistically not significant ($p > 0.05$) risk factors for trypanosomosis prevalence. The mean packed cell volume value of the parasitemic animals (21.1%) was lower compared to the mean packed cell volume value of aparasitemic animals (26.7%). There was statistically significant difference in the mean packed cell volume value between parasitaemic and aparasitaemic animals in both districts. Generally, this study showed that trypanosomosis is still present and becomes a constraint for livestock production of the study area. So, sustainable tsetse fly control strategies should be continued to reduce prevalence of the disease and tsetse flies population.

Key words: Bovine • PCV • Prevalence • Trypanosomes • Trypanosomosis • Tsetse Fly

INTRODUCTION

Trypanosomosis is a fatal and debilitating disease of various domestic livestock and wild animals which is caused by a protozoan parasite of the trypanosomes species [1]. The Principal trypanosomes species affecting livestock in Ethiopia are *Trypanosoma congolense*, *Trypanosoma vivax*, *Trypanosoma brucei* in cattle, goat and sheep, *Trypanosoma evansi* in camels and *Trypanosoma equiperdum* in horses. Bovine trypanosomosis is one of the diseases in cattle caused by this protozoan parasite in the genus *Trypanosoma* [2, 3]. This disease is mainly transmitted biologically by several

species of the genus Glossina, commonly known as tsetse flies and mechanically by several biting flies (*Tabanids*, *Stomoxyes*, etc.) except *Trypanosoma equiperdum* which follows sexual means of transmission through coitus among equine species [4].

The impact of trypanosomosis is not only due to the direct losses resulting from mortality, morbidity, infertility of the infected animals and costs of treatment or controlling the disease but also due to indirect losses including exclusion of livestock and animal power from the huge fertile tsetse infested areas accounting for about 10 to 15% of the land believed to be suitable for livestock production [5, 6]. The impact of trypanosomosis is not

restricted to livestock production alone, but extends to changes in land use and exploitation of natural resources use, access to available and cultivable land and restriction of opportunities for diversification of agricultural production [7]. Furthermore, the disease reduces the efficiency of bulls and discourages the use of drought animals in crop production [8].

African trypanosomosis can be found wherever the tsetse fly vector exists. The distribution of tsetse flies is determined principally by climate and influenced by altitude, vegetation and, presence of suitable hosts [9]. The disease is distributed over approximately 10 million km² of sub Saharan Africa between latitudes 14°N and 29°S which directly coincide with distributions of tsetse flies [10].

More than 50 million cattle and more than 60 million people are affected by bovine trypanosomosis in 37 sub Saharan African countries [7].

In Ethiopia, Trypanosomosis is wide spread in domestic livestock in the Western, South and South-western lowland regions and the associated river systems (i.e. Abay, Omo, Ghibe and Baro/Akobo). Out of the nine administrative regions of Ethiopia, five (Amhara, Benishangul-Gumuz, Gambella, Oromia and Southern Nations and Nationalities and People Regional State (SNNPRS)) are infested with more than one species of tsetse fly [11]. Currently about 220, 000 km² areas of the above mentioned regions are infested with five species of tsetse flies namely *Glossina pallidipes*, *Glossina morsitans*, *Glossina fuscipes*, *Glossina. Tachinoides* and *Glossina longipenis* [12, 13].

The owners of cattle in Ethiopia locally know trypanosomosis as “Ghendi” and it is the most important and the first problem affecting livestock productivity and agricultural activities in many areas. Western part of Amhara Regional State bordering the Abbay river particularly west Gojjam zone is one of the north western tsetse belt areas of Ethiopia. Dembecha and Jabitehenan districts of west Gojjam zone are the most prominent districts where there are serious complaints of the disease. Although there have been few reports on bovine trypanosomosis in these two districts, there has been limited work on systematic investigation of the diseases. The current study at hand was planned to fill such gap in western Amhara region particularly in west Gojjam zone to estimate prevalence of bovine trypanosomosis. Therefore, the objective of this study was to determine the current prevalence of bovine trypanosomosis in selected tsetse infested districts of west Gojjam Zone in Amhara regional state, Ethiopia.

MATERIALS AND METHODS

Study Area: The study was conducted in Dembecha and Jabitehenan districts of west Gojjam administrative zone of Amhara regional state, North West Ethiopia. The zone is situated at 10°30' North latitude and 37°29' East longitude. The climatic condition of the study area alternates with long summer rainfall between June and September and winter dry season between December to March with mean annual rain fall of 1200-1600 mm. The mean temperature is between 10-20°C and the altitude ranges in these areas are from 1100-1500 m.a.s.l for Jabitehenan and 1400-2300 m.a.s.l for Dembecha. The river valleys altitude level ranges from 1700 m.a.s.l. from the main road of Addis Ababa to Bahirdar to 1300 m.a.s.l. to the lower valley of Abbay. The livestock populations found in the study districts include cattle, goat, sheep, horses, mules, donkeys and poultry. Among these animals, cattle are important in the agricultural activities where the farmers are dependent on oxen power for crop production [13].

Study Population: A study was conducted on randomly selected indigenous zebu cattle which are kept under traditional extensive husbandry management system. The body condition score was grouped in to poor, medium and good conditioned animals based on the appearance of ribs and dorsal spines applied for Zebu cattle [14]. The age of the animals was determined by dentition [15] and conventionally categorized as young (1-3 years) and adults (>3 years).

Study Design: A cross sectional study design was carried out to determine the current bovine trypanosomosis in five kebeles (Nebessa Kendamue, Enewend, Gedeb, Regeb Kebero Meda and Weynema Workema) of Dembecha and Jabitehenan districts of west Gojjam administrative zone of Amhara regional state, North West Ethiopia from February 2017 to January 2018.

Sampling and Sample Size Determination: Simple random and purposive sampling methods were followed to select the study animals and study districts respectively. Five kebeles were also being selected purposively with consultation of zonal livestock health experts to represent tsetse infested sites. Based on their accessibility, villages were selected from each kebele which were geographically representative to the rest of the villages in the study districts. A total of 384 cattle were selected by simple random technique for

parasitological survey from five selected kebeles (Nebersa Kendamue, Enewend, Gedeb, Regeb Kebero Meda and Weyenema Workema) during study period to determine the current status of bovine trypanosomosis in the study areas. To determine sample size, the following formula given by Thrusfield [16] and previous studies conducted in Dembecha and Jabitehenan by Dagnachew [17] were taken into consideration so the expected prevalence was 20% for absolute desired precision of 4% at confidence level of 95% were used for district.

$$N = 1.962 [P_{exp} (1-P_{exp})]/d^2$$

where N is the required sample size, P_{exp} was the expected prevalence and d is the desired absolute precision

Parasitological Study: A total of 384 blood samples were collected from aseptically prepared ear veins of cattle. Blood samples were collected by puncturing of the marginal ear vein of each animal with a lancet and drawn directly in to heparinized capillary tube. The tubes were filled with blood to $\frac{3}{4}$ of their height and sealed at one end with crystal seal. The capillary tubes were loaded on the microhaematocrit centrifuge symmetrically and centrifuge at 1200 revolutions per minute (rpm) for 5 min [18]. The capillary tubes were placed in a haematocrit reader after centrifugation. The length of the packed red blood cells column was expressed as a percentage of the total volume of blood. Animals with Packed Cell Volume = 24% were considered as anemic [19].

Hematological Examination: After packed cell volume was read, the Heparinised capillary tubes, containing blood samples, were cut using a diamond tipped pen 1 mm below and 3 mm above the buffy coat. The content of the capillary tube was expressed onto a glass slide, then covered with 22 x 22 mm cover slip by ground buffy coat technique and examined under $\times 40$ objective and $\times 10$ eye pieces for movement of parasite. Trypanosomes positive Buffy coat samples were analyzed and trypanosome species were identified based on their morphological structure from Geimsa-stained thin films [20].

Data Management and Analysis: The recorded data were entered and managed using Microsoft Excel spread sheets program to create a data base. Statistical analysis was done using IBM SPSS statistics version 20 and interpret the data. Descriptive statistics were used to analyze the qualitative data. An independent sample t-test was used

to employ to compare the mean packed cell volume of the parasitaemic animals with that of the aparasitaemic animals. Trypanosomosis infection rates with different variables like age, sex, body condition score and coat color were compared by using logistic regression. The test result was considered significant when the calculated P-value was less than 0.05.

RESULTS

Parasitological Findings: Out of the 384 local breeds of cattle examined during the study period, 39 animals were found to be infected with trypanosomes in the two districts. The result of the survey showed that the overall prevalence in cattle in the study area was 10.15% (39/384). The prevalence of bovine trypanosomosis in the two districts were found to be 10.95% (23/210) and 9.19% (16/174) in Dembecha and Jabitehenan district respectively. The trypanosome species responsible for the infection were *Trypanosome congolense* and *Trypanosome vivax*. The prevalence in terms of trypanosome species was 7.29% (28/384) *Trypanosome congolense* and 2.86% (11/384) *Trypanosome vivax*. The proportion of trypanosome species was 28/39 (71.79 %) for *Trypanosome congolense* and 11/39 (28.20%) for *Trypanosome vivax* with no mixed infection in study districts. The prevalence of trypanosomosis in cattle within five kebeles during study period were 12.85%, 10%, 10%, 9.18% and 9.21% at Nebersa kendamu, Enewend, Gedeb, Regeb Kebero Meda and Weyenema Workema respectively as shown in table 1. Despite the highest prevalence in Nebersa kendamu (12.85%) and the lowest in Weyenema Workema (9.21%) were occurred, the difference was not statistically significant ($P > 0.05$).

In the current study, the association of prevalence with the various risk factors including age, sex, body conditions and coat color were computed in Table 2. The highest prevalence was observed in the adult animals greater than or equal to 3 years old (11.3%) than young once below 3 years old age although variation in prevalence between the difference age groups was not statistically significant ($P = 0.550$). A comparison of trypanosome infection between males and females were made. Based on this factor, the overall prevalence of bovine trypanosomosis in males and females were 10.4% and 9.8% respectively. The prevalence of trypanosomosis was slightly higher in males as compared to female animals, but the difference was not statistically significant ($P = 0.912$). In animals with poor body condition, the prevalence of trypanosomosis was significantly higher

Table 1: The prevalence of bovine trypanosomosis species at different kebeles in the study districts

| District | Kebeles | No of animals examined | Total positive | Prevalence rate (%) | Trypanosome species | |
|-------------|------------------|------------------------|----------------|---------------------|----------------------|-----------------|
| | | | | | <i>T. congolense</i> | <i>T. vivax</i> |
| Dembecha | Nebersa Kendamue | 70 | 9 | 12.85 | 5 | 4 |
| | Enewend | 70 | 7 | 10 | 5 | 2 |
| | Gedeb | 70 | 7 | 10 | 6 | 1 |
| Jabitehenan | Regeb KeberoMeda | 98 | 9 | 9.18 | 7 | 2 |
| | WeyenemaWorkema | 76 | 7 | 9.21 | 5 | 2 |

Table 2: The prevalence of bovine trypanosomosis with host related different risk factors

| Risk factor | Total sampled | Number of positive | Prevalence (%) | OR | 95% CI | | P-value |
|-------------|---------------|--------------------|----------------|----------|----------|----------|---------|
| | | | | | Lower | Upper | |
| District | | | | | | | |
| Dembecha | 210 | 23 | 11 | | | | |
| Jabitehenan | 174 | 16 | 9.2 | .9694275 | .2925654 | 3.212238 | .959 |
| Sex | | | | | | | |
| Male | 221 | 23 | 10.4 | | | | |
| Female | 163 | 16 | 9.8 | 1.042549 | .4981703 | 2.181802 | .912 |
| Age | | | | | | | |
| Young | 100 | 7 | 7 | | | | |
| Adult | 284 | 32 | 11.3 | .7556356 | .3011994 | 1.895705 | .550 |
| BCS | | | | | | | |
| Good | 80 | 2 | 2.5 | | | | |
| Medium | 189 | 12 | 6.3 | .3859364 | .0823791 | 1.808066 | .227 |
| Poor | 115 | 25 | 21.7 | .0904601 | .0201242 | .406626 | .002 |
| Colour coat | | | | | | | |
| Red | 194 | 10 | 5.2 | | | | |
| White | 64 | 4 | 6.2 | .6312241 | .1786322 | 2.230526 | .475 |
| Black | 129 | 25 | 19.4 | .2104988 | .0925181 | .4789307 | .000 |

Table 3: Mean PCV comparison of parasitaemic and aparasitaemic bovine species in study area

| Infection status | Total sampled | No. of examined (PCV <24%) | No. of examined (PCV ≥ 24%) | Mean PCV±SD | T-value | P-value |
|------------------|---------------|----------------------------|-----------------------------|-------------|---------|---------|
| Parasitaemic | 39 | 32 | 7 | 21.1±1.16 | 5.46 | 0.000 |
| Aparasitaemic | 345 | 144 | 201 | 26.7±.67 | | |

($P=0.002$) than medium and good body conditioned cattle as indicated in table 2. The preset study revealed that prevalence of trypanosomosis was significantly different ($P=0.000$) among animals with different coat color where the prevalence was higher in animals with black coat color as shown Table 2.

Hematological Findings: To assess the relationship between trypanosome infection and packed cell volume (PCV) value, PCV determination was done by using hematocrit method and the mean PCV of parasitemic and aparasitemic animals were calculated. The PCV value in the sampled animals in the two districts ranged from 14-43%. The most frequently recorded PCV value was 23% and was recorded in 44 cattle from the overall studied animals in the district. The mean PCV values for all

examined animals were 26.13. From the total of 384 examined animals, 46.09% (177/384) of cattle were found to be anemic having PCV value less than 24%. Out of 46.09% anemic animals, only 8.33% (32/384) were found to be positive for trypanosomes whereas 37.76% (145/384) were trypanosomes free. Some animals 1.82% (7/384) were infected by trypanosome but their PCV value was found normal. The overall mean PCV value in parasitaemic animals was 21.1 ± 1.16 SD while in aparasitaemic animals was $26.7 \pm .67$ SD and the variation between the two was 5.6 % (26.7% - 21.1%). The mean PCV value of the parasitemic animals (21.1%) was lower compared to the mean PCV value of aparasitemic animals (26.7%). There was statistically significant difference ($P = 0.000$) in the mean PCV value between parasitaemic and aparasitaemic animals in both districts as shown in Table 3.

DISCUSSION

The result of the present study revealed that the overall prevalence of bovine trypanosomosis in the two districts during the study period was 10.15%. This finding was nearly similar value with the previous finding conducted by Kebede and Animut who reported 10.1 % from neighboring Awi zone [21]. The results was in close agreement with earlier works of Mekuria and Gadisa, Dagnachew and Shibeshi and Solomon and Fitta who reported a prevalence rate of 12.41%, 11.33% and 12.41% respectively from North West Ethiopia [22, 23, 24]. In this study, the prevalence of trypanosomosis in cattle in Dembecha and Jabitehenan districts were 10.95% and 9.19% respectively. The current finding was lower than the previous report of Wolde-Mariam in the same study area who showed prevalence rate of 23.36% and 24.5% in Dembecha and Jabitehenan respectively [25]. Dagnachew also reported with a total prevalence of 20% for Dembecha and Jabitehenan [26]. The lower prevalence in the current study might be due to expansion of veterinary clinic, deforestation for cultivation of land in the area which directly affects flies distribution, unsustainable of tsetse control intervention by National Tsetse and Trypanosomosis Investigation and Control Center and awareness improvement of the people towards the control and treatment of the disease.

In another way low sensitivity of direct parasitological buffy coat examination may contribute for low prevalence that chronic stage is characterized by low parasitemic which is difficult to confirm by parasitological diagnosis. In very low sensitivity of buffy coat method 50% of infected animals remained undetected using parasitological diagnostic tools as compared to the molecular analysis animals [27].

The present study showed that *Trypanosoma congolense* and *Trypanosoma vivax* were the pathogenic trypanosomes identified in the study area having a proportion of 8/39(36.4%) *T. congolense* and 14/39(63.6%) *Trypanosoma vivax*.

In the current study on bovine trypanosomosis, the association of prevalence with the various risk factors including district, age, sex, body conditions and coat color were computed. There was no statistical difference ($P>0.05$) in the prevalence of trypanosome infection in the two study sites. These might be because the areas are close to each other in almost a similar climatic and agro ecological condition.

The present study revealed that the prevalence of trypanosomosis in young animals ($1<3$ years) was lower than that of adult (≥ 3) animals but the difference was not significant ($P>0.05$). This finding was similar with the report of Muturi, Abraham and Tesfaheywet and Ayana [28, 29, 30]. Because, adult animals traveled long distance for grazing, watering and draft as well as harvesting of crops to tsetse challenge area. Calves were not allowed to move together with adult group and kept at homestead until weaned off [31]. The low prevalence in young animals might also be due to the natural protection to some extent by maternal antibodies [32]. Although the variation was not statistically significant association, the trypanosome infection in male animal was slightly higher than in the female animals. The result showed that both male and female cattle were equally susceptible to trypanosomosis infection. This finding was in close agreement with previous reports by Adane and Gezahegne; Ababayehu; Ayana; Bishawet) [33, 34, 30, 35]. This might be due to the management of animals where both male and female are allowed to graze freely in the field which consequently lead to similar exposure to tsetse flies.

The present finding indicates that higher prevalence of trypanosomosis was observed in animals with poor body condition when compared with animals with medium and good body condition and the association was found statistically significant ($P=0.000$). This finding was in agreement with study carried out by Solomon and Fitta in Awi and Metekel zones Northwest of Ethiopia [24]. The physiological status of the host, as well as nutritional and environmental factors, further play important roles in modulating the severity of the disease [36].

In this study, there was statistical significant difference ($P=0.000$) in prevalence among three hair coat colored animals at the present study area. Animals with black coat color were found to be highly infected than red and white coat color. This might be associated for *Glossina* species that are major transmitter of African animal trypanosomosis and the strongest landing responses were found to be on black surfaces. Consequently traps are made to be blue on the outside to attract the maximum number of tsetse flies and black on the inside to maximize the proportion of tsetse that land on the entrapment area [37]. The present finding also agreed with the above statement and the host preferences of tsetse flies; mostly tsetse flies favor and land on black objects so animals with black coat color were more susceptible.

The mean PCV value in parasitaemic animals was 21.1 ± 1.16 while in aparasitaemic a cattle was 26.7 ± 0.67 and the variation between the two was 5.6% (21.1% - 26.7%). There was statistically significant difference ($P < 0.05$) in the mean PCV value between parasitaemic and aparasitaemic animals. This result is in alignment with previous works [38, 39].

A similar finding was reported by Solomon and Fitta mean PCV value of parasitaemic and aparasitaemic animals were significantly different ($P < 0.05$) [24] where parasitaemic animals had low PCV value. Rowlands in Ghibe observed that with a decrease in the PCV value, the proportion of infected animals increased and hence mean PCV was a good indicator for the health status of herds in the trypanosomosis endemic areas [31]. Comparison of the mean PCV of infected animals within species of trypanosome out of 39, 28 were infected with *Trypanosoma congolense* and their mean PCV was 20.75% and 11 were infected with *Trypanosoma vivax* and their mean PCV was 22.09%.

CONCLUSION AND RECOMMENDATIONS

The study in Dembecha and Jabitehenan districts revealed that the overall prevalence of trypanosomosis in cattle was 10.15%. The trypanosome species responsible for the infection were *Trypanosome congolense* and *Trypanosome vivax*. The prevalence in terms of trypanosome species was 7.29% (28/384) *Trypanosome congolense* and 2.86% (11/384) *Trypanosome vivax* with no mixed infection during study period. The body condition and packed cell volume of cattle were negatively affected by trypanosomosis infection. This showed that trypanosome infection in cattle can cause the loss of body weight and production.

Therefore based on the above conclusion the following recommendations are forwarded:

- Sustainable tsetse fly control strategies should be strengthened to control bovine trypanosomosis effectively in the study area.
- Expand governmental and private veterinary services to provide chemotherapeutic service in the areas.
- Conduct further study on the occurrence of tsetse fly and trypanosomosis at different season of the year, at different altitude and different species of animals.

ACKNOWLEDGEMENTS

I would like to express my deepest appreciation to Dr. Shimelis Dagnachew from University of Gondar for his excellent guidance, follow up and regular supervision and supporting in all round during the study period.

REFERENCES

1. Zecharias, A. and T. Zeryehun, 2012. Prevalence of Bovine Trypanosomosis in Selected District of Arba Minch, Snnpr, Southern Ethiopia. Global Veterinaria, 8: 168-173.
2. Chau, N.V., M. Desquesnes, S. Herder, N.P. Lan, J.I. Campbell, N. Van Cuong, B. Yimming, P. Chalermwong, S. Jittapalapong, J.R. Franco and N.T. Tue, 2016. A clinical and epidemiological investigation of the first reported human infection with the zoonotic parasite *Trypanosoma evansi* in Southeast Asia. Clinical Infectious Diseases, 62: 1002-1008.
3. Abebe, G., 2005. Current situation of Trypanosomosis. In: review article on: Trypanosomosis in Ethiopia. Ethiop. J. Biol. Sci., 4: 75-121.
4. Broun, D.M., R.S. Reid, D.J. Rogers, W.F. Shnow and G.R.W. Wint, 2001. Environmental. Change and the automous control of tsetse and Trypanosomosis in Sub-Saharan Africa: Case histories from Ethiopia, Gambia, Kenya, Nigeria, and Zimbabwe, Environmental research Group Oxford Limited, Oxford, UK, pp: 9-24.
5. Awoke, K., 2000. Study of trypanosomosis and its Vectors in Humbo and Merabo Warheads. Ethiop Vet. J., 4: 1-61.
6. NTTICC (National Tsetse and Trypanosomosis Investigation and Control Center), 2013. Report on Tsetse and Trypanosomosis, Survey, Addis Ababa, Ethiopia.
7. Swallow, B.M., 2000. Impacts of trypanosomiasis on African Agriculture. In PAAT Technical and Scientific Series 2. FAO. Rome, pp: 52.
8. Omotainse, S.O., J.O. Kalejaiye, P.M. Dede and A.J. Dadah, 2004. The current status of tsetse and animal trypanosomiasis in Nigeria. Vom Journal of Veterinary Sciences, 1: 1-7.
9. Paris, J., M. Murray and F. Mcodimba, 1982. A comparative evaluation of the parasitological technique currently available for the diagnosis of African trypanomosis in cattle, Acta. Trop, 39: 1-11.

10. Radostitis, O.M., C. Gay and P.D. Constable, 2007. Veterinary Medicine: A text book of diseases of cattle, horses, sheep, pigs and goats. 10th edn. Elsevier, London, 1531-1540.
11. Bitew, M., Y. Amedie, A. Abebe and T. Tolesa, 2011. Prevalence of bovine trypanosomosis in selected area of Jabi Tehenan district, West Gojam of Amhara regional state, North Western Ethiopia, College of Agricultural and Veterinary Medicine, JU, Jimma. African Journal of Agricultural Research, 6: 1-5.
12. NTTICC, 2004. National tsetse and Trypanosomosis Investigation and Control Center report for the period 7 July 2001 – 6 July 2002 Badelle, Ethiopia, pp: 3.
13. CSA, 2013. Agricultural sample survey. Report on livestock and livestock characteristics (private peasant holdings), Vol II. Statistical Bulletin 570. Central Statistical Agency (CSA), Federal Democratic Republic of Ethiopia, Addis Ababa.
14. Nicholson, M.J. and M.H. Butterworth, 1986. A Guide to Condition Scoring of Zebu Cattle. ILCA, Addis Ababa, Ethiopia.
15. De Lahunta, A. and R.E. Habel, 1986. Teeth', in De Lahunta A. & Habel R.E. (ed.). Applied Veterinary Anatomy, W.B. Saunders Company, Philadelphia.
16. Thrusfield, M., 2005. Veterinary Epidemiology. 3rd ed., Black well science Ltd, pp: 233-250.
17. Dagnachew, S., 2004. Epidemiology of Bovine Trypanosomosis in the Abbay Basin Areas of Northwest Ethiopia. MSc thesis, AAU, FVM, Debre Zeit.
18. Murray, M., P.K. Murray and W.I. McIntyre, 1977. An improved parasitological technique for the diagnosis of African trypanosomiasis. Transactions of the Royal Society of Tropical Medicine and Hygiene, 71(4): 325-326.
19. OIE, 2008. Trypanosomosis (tsetse transmitted). Terrestrial Manual. Office International des Epizooties (OIE), Paris, France, 12
20. Murray, M., W.I. Morrison and D.D. Whitelaw, 1982. Host susceptibility to African trypanosomiasis: Trypanotolerance. Advances in Parasitology, 21: 1-68.
21. Kebede, N. and A. Animut, 2009. Trypanosomosis of cattle in selected districts of Awi zone, Northwestern Ethiopia. Tropical Animal Health and Production, 41(7): 1353-1356.
22. Mekuria, S. and F. Gadissa, 2010. Survey on bovine trypanosomosis and its vector in Metekel and Awi zones of Northwest Ethiopia. Acta Trop, 117: 146-151.
23. Dagnachew, S. and S. Shibeshi, 2011. Prevalence and vector distributions of bovine trypanosomosis in control (Sibu sire) and non-control (Guto Gida) districts bordering upper Anger valley of East Wollega Zone, Western Ethiopia. Ethiop Vet. J., 15: 77-86.
24. Solomon, M. and G. Fitta, 2010. Survey on Bovine Trypanosomosis and Its Vector in Metekel and Awi Zones of Northwest Ethiopia. Acta Tropica, 117: 146-151.
25. Wolde-Mariam, S., 1997. Trypanosome Survey in District of Abay Valley. In Some Woreda of Northwest Ethiopia, Amhara Region. Bureau of Agriculture, pp: 24.
26. Dagnachew, S., A.K. Sangwan and A. Getachew, 2005. The Epidemiology of Bovine Trypanosomosis in Abay (Blue Nile) Basin of Northwest Ethiopia. Global Veterinaria, 79: 151-157.
27. Simukoko, H., T. Marcotty, D. Berkvens, J. Vercruysse and P. Van den Bossche, 2011. Bovine trypanosomosis risk in an endemic area on the eastern plateau of Zambia. Research in Veterinary Science, 90(1): 51-54.
28. Muturi, K.S., 1999. Epidemiology of Bovine trypanosomosis in selected sites of the southern rift valley Ethiopia MSc Thesis. FVM, AAU. Debrezeit, Ethiopia.
29. Abraham, Z. and Z. Tesfaheywet, 2012. Prevalence of Bovine Trypanosomosis in Selected District of Arba Minch, Southern Ethiopia. Global Veterinarian, 8: 168-173.
30. Ayana, M., Z. Tesfaheywet and F. Getnet, 2012. A Cross-sectional Study on the Prevalence of Bovine Trypanosomosis in Amhara region, Northwest Ethiopia. Livestock Research for Rural Development, 24: 1-8.
31. Rowlands, G.J., M. Woudyalew, E.D. Authie, G.D.M. Ieteren, S.G.A. Leak and S.M. Nagda, 1995. A method for distinguishing new and recurrent trypanosome infections in a field survey of east Africa Zebu cattle in Ethiopia.
32. Fimmen, H.O., D. Mehlitz, F. Horchiners and E. Korb, 1999. Colostral antibodies and Trypanosome Congolese infection in calves. Trypanotolerance research and application GTZ, No, 116, Germany, 173-178.
33. Adane, M. and M. Gezahegn, 2007. Bovine Trypanosomosis in Three Districts of East Gojjam Zone Bordering the Blue Nile River in Ethiopia. Journal of Infection in Developing Countries, 1: 321-325.

34. Abebayehu, T., H. Eshete, M. Berhanu, A. Rahmeto and M. Solomon, 2011. Mechanically Transmitted Bovine Trypanosomosis in Tselemt Woreda, Western Tigray and Northern Ethiopia. *Journal of Agriculture*, 6: 10-13.
35. Bishaw, Y., T. Wudu, Y. Nuria and A. Sefinew, 2012. Prevalence of Bovine Trypanosomosis in Womberma Districts of West Gojjam Zone, Northwest Ethiopia. *Ethiopian Veterinary Journal*, 16: 41-49.
36. Ouma, E.M., 2010. Management of Trypanocidal Drug Resistance in Cattle in Identified Chemo resistance Hot Spots in the Administrative District of Sikasso, Southeast Mali. MSc, Thesis. Freie University at Berlin.
37. Leak, S.G.A., 1999. Tsetse Biology and Ecology: Their Role in the Epidemiology and Control of Trypanosomosis. Wallingford, Oxon, UK: CABI Publishing, p: 152-210.
38. Ali, D. and M. Bitew, 2011. Epidemiological of Bovine Trypanosomosis in Mao-Komo Special District, Benishangul Gumuz Regional State, Western Ethiopia. *Global Veterinaria*, 6: 402-408.
39. Bayisa, K., D. Getachew and T. Tadele, 2015. Bovine Trypanosomosis in Assosa District, Benishangul Gumuz Regional State, Western Ethiopia: Prevalence and Associated Risk Factors. *European Journal of Applied Sciences*, 7(4): 171-175.