



The impact of Ghibe-III hydroelectric dam on Bovine trypanosomosis situation: tsetse fly population dynamics, prevalence of disease and community perception in selected districts in up- stream and down-stream of the reservoir

A dissertation submitted to the College of Veterinary Medicine and Agriculture of Addis Ababa University in fulfillment of the requirements for the degree of Doctor of Philosophy in Veterinary Parasitology

By

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*To My wife, Selamawit Tamiru, my kids, Hasset Solomon, Nardos Solomon  
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## **ABBREVIATIONS**

AAT = African Animal Trypanosomosis

CSA = Central Statistics Agency

EEPA = Ethiopian Electric Power Authority

ETHB = Ethiopian Birr (currency)

FAO = Food and agriculture organization

FG D = Focus group discussion

FITCA = Farming in tsetse control area of Africa

FLDP = Fourth livestock development project

IAEA = International Atomic Energy Agency

MOA= Ministry of Agriculture

MOARD = Ministry of Agriculture and Rural development

NTTIC = National Tsetse and Trypanosomosis Investigation Center

UNDP = United nation development programme

SIT = Sterile Insect technique

SPP = Species

VSG = Variation Surface Glycoprotein

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## ABSTRACT

*African Animal trypanosomosis is one of the most economically devastating diseases of livestock in sub-Saharan Africa. The persistence of its tsetse vectors and consequently the severity of the disease in a certain population largely depend on the maintenance of favorable ecological conditions in the area. This study was primarily initiated to assess spatial and temporal changes in tsetse distribution and trypanosome prevalence upstream (Loma District) and downstream (Kindo Didaye District) the Ghibe - III hydroelectric dam area to appreciate if the dam has any impact. Questionnaire surveys with 189 respondents using semi-structured questionnaire and focus group discussions (FGD) with 15 groups each containing 6-20 members were conducted by matrix scoring with local community to assess their perceptions on the disease situation and its tsetse fly vectors. Triangulation was done to evaluate the relationship between farmers' perception and trypanosomosis prevalence and impact in cattle. Tsetse flies were trapped by deploying 160 NGU traps at an interval of 200m air distance starting from the fringe of water bodies and animals were bled in dry and wet seasons from selected kebeles' of both districts to assess the situation as we go away from the Ghibe III dam reservoir in the upstream and from the Omo River downstream. The findings from the interviews and FGD revealed that: 1) cattle herd size greater or equal to six have significantly increased after dam construction in Loma district, whereas in Kindo Didaye farmers with such cattle herd size have declined, 2) Bovine trypanosomosis was ranked as number one disease priority in both Loma (upstream) and Kindo Didaye (downstream) Districts, and 3) Strong concordance observed between diseases and their clinical symptoms as well as impacts on animal health and production. Trypanosomosis prevalence and tsetse distribution studies showed: A) no difference between the two study districts in disease prevalence and tsetse apparent density (TAD) before dam construction, B) both TAD and trypanosomosis prevalence were significantly lower upstream the dam (Loma) than downstream (Kindo Didaye) study sites five years after dam construction ( $P < 0.05$ ), C) prevalence of the disease was reduced from 17.9% in 2005 before dam construction to 6.4% in 2019/20 in upstream (Loma) and from 19.2% in 2005 to 11.7% in 2019/20 in downstream, D) the most prevalent trypanosome species were *T. congolense* and *T. vivax* while few cases of *T. brucei* were seen in Kindo Didaye, E) tsetse apparent density was reduced from 10 fly/trap/day (FTD) before dam construction to 1.09 FTD in Loma five years after dam completion and from 13.7 FTD to 5.3 FTD in Kindo Didaye in the same period, but significant reduction was noticed only for Loma ( $P < 0.05$ ), F) Tsetse density was more severe close to the fringe of Omo River in Kindo Didaye whereas it is more important as we go away from the fringe of the dam reservoir in Loma, G) the current study has clearly shown that despite the significant difference in tsetse fly apparent density, tsetse infection rates were almost similar between the two study sites; 12% in Loma and 17.8% in Kindo Didaye. In general, although bovine trypanosomosis is still a major problem in the study areas, Ghibe III dam construction appear to have produced a favorable outcome in areas close to the water reservoir probably by disrupting tsetse ecology and consequently reducing trypanosomosis prevalence. The finding was consistent with community perception. However, the problem is still rampant in areas downstream the dam and in places far away from the dam water reservoir. Based on the above findings, lists of practical recommendations are forwarded.*

*Key words: Bovine, Trypanosomosis, Tsetse fly, Ghibe-III dam, Downstream vs Upstream, Southern Ethiopia*

## 1.INTRODUCTION

Livestock production is a back bone of agriculture where it plays a role for crop production apart from its direct use as source of milk, meat and manure and determines socioeconomic status of individual farmers' (Perry, 2016). Despite significant contribution on agriculture subsector in sub-Saharan African countries, animal trypanosomosis remain a bottleneck in livestock productivity. African animal trypanosomosis is one of the major animal health constraints of animal production in sub-saharran Africa, nearly 37 countries are affected including Western, Northwestern and South-western parts of Ethiopia (NTTIC, 2004; Enwezer *et al.*, 2006). More than 50 million cattle are at risk and 10-30% of these cattle are infected. Major impact of this disease is associated with mortality, retarded growth rate, reduced reproductive performance and low milk production and poor draught power (Peter, 2013). Animal trypanosomosis is estimated to reduce cattle density by 37 – 70%, off take by 50% and reduce the calving rate and increase calf mortality by 20% (Swallow, 2000). Vector borne trypanosomosis is excluding some 180,000 – 200,000 km<sup>2</sup> of agriculturally suitable land in the Western, Northwestern and Southwestern parts of Ethiopia (Slingenbergh, J., 1992); more than 15, million livestock, which include cattle, goat and equines remain at risk of contracting trypanosomes every year in Ethiopia (CSA, 2018).

Livestock reproduction and productivity are severely hampered due to challenges of tsetse and trypanosomosis; as the result of advancement into new area and uncontrolled cattle movement from free area to infested area, as well as the role of non tsetse biting flies have contributed substantially for the severity and distribution of trypanosomosis outside tsetse belt (Slingenbergh, J., 1992).

The survival and breeding activity of tsetse flies is highly influenced by ecological zones; mean daily temperature, suitable shelter especially trees with sufficient canopy, source of food and suitable breeding ground are among the basic ecological factors. High altitude and cold temperature and lack of food source fasten their eradication (Jordan, 1986). It was reported that before 40 years distribution of trypanosomosis in Ethiopia was limited in the western and southern corner of the country in Abay, Baro Akobo River basin and Ghibe- Omo and Woito River basin, respectively (Langridge, 1976). However, recent evidence has indicated that tsetse flies are advancing to new ecological niches (NTTIC, 2004); for instance along Omo-Gibe River

course most of the tributaries are infested by tsetse- trypanosomosis. There is also report from North West Amhara regional state.

To mitigate the problem in Ethiopia various efforts have been made to control both vector and the disease in infested area. Mainly using trypanocidal drug to control the parasite in the host and secondly to control vector through odour- baited- insecticide- impregnated targets and traps as well as insecticide-treated cattle (Slingenbergh, 1992). Especially to knock down tsetse fly invasion to commiserates towards riverside and make the area suitable for agriculture, various operations have been implemented, mainly through specifically designed joint projects of MOA-FAO/UNDP/FLDP in the upper Didessa valley in 1986-1994, FITCA-Ethiopia (1997-2004) (MOARD, 2004). In recent years a joint project between Ethiopian science and technology commission and IAEA has tried to introduce sterile insect technique (SIT) in the southern rift valley of Ethiopia with the objective of eradicating *G.pallidipes*.

Although these efforts have produced some fruits in pocket areas, achievements were not sustainable and hence the vast majority of livestock in tsetse infested areas are still suffering from the disease. Prevalence of bovine trypanosomosis and tsetse fly catch per day per trap were reduced significantly during the project period because of continuous control activity in the area, but immediately after the end of the project reinvasion of tsetse fly and prevalence of trypanosomosis reappear or resume to the origin level (Hargrove, 2004).

In recent years, the construction of Ghibe-III hydroelectric dam and its water reservoir which has started in 2006 and inaugurated in 2016 is believed to have changed the ecological situation as it occupied vast area of tsetse suitable land. The Ghibe-Omo River has 17.9 billion m<sup>3</sup>/year runoff, covers 79,000 km<sup>2</sup> catchment areas and a variety of wildlife parks (Awulachew, *et al.*, 2011, EEPA, 2009). Ghibe III hydro-electric dam lied in the middle of Ghibe-Omo River, where the created Lake has covered 200 km<sup>2</sup> of tsetse infested area (EEPA, 2009). The dam height is 245 meter above the river table of Omo River and the water flooded back about 150kms from the site of dam foundation. Grazing land, wild life holes, bush and shrubs are flooded. Consequently, the impact due to dam construction include loss of common grazing land along the banks of the River; Loss of incense trees, gum and other important trees found along the banks of the River; loss of forest honey production as the result of flooding; Wildlife encroachment between water reservoir and agricultural activities have reduced substantially. The riverine forest and woodland

vegetation as well as wild life in main rivers and tributaries plays major role for tsetse infestation and survival (Leak, 1999; Duguma *et al.*, 2015).

Alteration of the ecosystem as a result of anthropogenic changes due to clearing of vegetation for such activities like dam construction and encroachment by settlements and agriculture could lead to habitat fragmentation and consequently affects tsetse population dynamics (Van den Bossche, *et al.*, 2010). Therefore, dam construction in the tsetse infested area could negatively or positively alter tsetse environment and consequently impact on livestock trypanosomosis prevalence and intensity. In this regards, the impact of Ghibe-III dam on tsetse and trypanosomosis has not been studied so far. Farmers have been moved from the area to the periphery with their livestock, vegetation has been cleared for the reservoir and construction camp establishment which may have altered the wildlife habitat in the area. Therefore, it is hypothesized that the ecological and settlement changes caused by the construction of the dam has a negative impact on tsetse distribution as well as composition and trypanosomosis prevalence in the upstream of the dam compared to down stream.

To address this issue systematically, this research project attempts to find answers to the following questions.

## **RESEARCH QUESTIONS**

1. Is Gibe-III dam making a difference in tsetse fly population density, composition and location/dispersal in comparison between upstream and downstream from the dam site?
2. Is there any difference in the prevalence and species composition of the parasites in animals as compared to upstream and downstream of selected district?
3. What is the perception of the local community on the situation of the disease before and after the construction of the hydroelectric dam in the area?

### **General objective**

To assess the impact of Ghibe-III hydroelectric dam on tsetse fly population dynamics and prevalence of trypanosomosis as well as community perception in downstream and upstream of Ghibe-III hydroelectric dam.

### **Specific objectives**

- To assess community perception on tsetse dynamics and prevalence of trypanosomosis in relation to dam construction
- To estimate prevalence and species composition of trypanosomes in cattle upstream and downstream of Ghibe-III dam during dry and rainy season
- To estimate tsetse apparent density, composition, dispersion in dry and rainy season
- To estimate infection rates in selected sites of upstream and downstream of the dam

## 2. LITERATURE REVIEW

### 2.1. African Animal Trypanosomosis

Tsetse transmitted animal trypanosomosis is a debilitating and wasting disease of livestock in sub-Saharan Africa which significantly hampers productivity of infected animals' thereby agricultural sectors. Infected animals chronically sick, emaciated, wasted and finally die if not treated. The disease caused by protozoan hemoparasite called *trypanosoma* which is unicellular, microscopic, elongated protozoan parasite that moves with the help of a single flagellum located anterior to the kinetoplast (Soulsby, 1986; Levine *et al.*, 1980). A typical characteristic of *trypanosoma* is having variation on surface glycoprotein antigen (VSG), which changes continuously against defense mechanism of the host. This surface glycoprotein antigen changing characteristics of the parasite has enabled to withstand immune defense of the host and will occur in the blood and tissue fluids, although a few may invade tissue cells (Soulsby, 1986). The disease mostly transmitted by blood sucking arthropods in which the development stage occurs and a few species are transmitted mechanically without development in the arthropods (Leak, 1999).

Historical background of trypanosomosis starts in South Africa in Zulu community as a disease called "Nagana". Nagana, a local word used for trypanosomosis in cattle, was referred to by David Bruce (1895) recognized an association between the disease and tsetse fly. Nagana was a Zulu word, meaning to be depressed or in low spirits. Bruce suspected that the tsetse fly was responsible but did not know how, but later identified one of the etiological agents which was named after his name *Trypanosoma brucei*. In 1909, Kleinfelder demonstrated the biological transmission of trypanosomes via tsetse bite, which led to the elucidation of the life cycle of the trypanosome in the fly by Robertson in 1913 (as cited by Williams, 2002).

#### 2.1.1. Classification and morphology

Protozoa in general are classified in the Kingdom Protista in the five kingdom classification of Robert Whittaker (Hagen, 2012) hence forming a subkingdom. The Subkingdom Protozoa is further classified into eighteen different phyla. *Trypanosoma* and *Leishmania* are classified under the same Phylum Kinetoplastida or Kinetoplasta because of their kinetoplast feature (<https://www.biologydiscussion.com/animals-2>). Whereas, Maudlin, et al. (2004) classified

trypanosomes under phylum Sarcomastigophora, subphylum Mastigophora, class zoomastigophora, Order Kinetoplastida, Family Trypanosomatidae, Genus Trypanosoma. Species of trypanosome infecting mammals comprises two distinct groups: Stercoraria and Salivaria.

A) **Stercoraria:** Includes subgenera Herpetosoma (Hoare, 1972), Megatrypanum (Hoare, 1964) and Schizotrypanum (Hoare, 1972) most of which are non-pathogenic to human and livestock. One exception in the subgenus Schizotrypanum is *T. cruzi* which causes Chagas' disease. Their life cycle in tsetse fly completes posterior unlike salivarian section parasite where they complete anterior. Trypanosomes of this group typically produced in the hindgut of tsetse fly and transmitted posterior direction via contaminative excreta. Morphologically they have large kinetoplast, not terminal, posterior extremity tapering, free flagellum present, undulating membrane not well developed (Urquart, 1996).

B) **Salivarian:** classified under subgenera *Duttonella*, *Nannomonas* and *Trypanozoon*. Transmission of trypanosomes of this group occurs in the anterior station through inoculation (Mauldin et al., 2004). Morphologically, they have small kinetoplast which is terminal or sub terminal, blunt posterior extremity, there may be no free flagellum, undulating membrane varying in development. They are pathogenic trypanosomes of human and animals (Soulsby, 1986).

**Subgenus *Duttonella*:** *Typanosoma vivax* Zeimann, 1905 is a recognized species in this subgenus. There is one species in this subgenus morphologically similar but smaller, *T. uniforme* which was last reported from a giraffe in Tanzania (Stevens and Brisse, 2004). The site of development in the tsetse fly is restricted to proboscis. Morphologically *T. vivax* is characterized by club shaped posterior end, which tapers towards the anterior; it has large kinetoplast placed terminally and free flagellum is present in all stages. Movement in wet film is rapid and distinctive, which cross the field rapidly.

**Subgenus: *Nannomonas*:** under this subgenus there are three recognized species; *T. congolense* Broden, 1904, *T. simiae* Bruce et al 1912 and *T. godfreyi* (McNamara et al., 1994). The most frequently encountered pathogen of African livestock is *T. congolense*, while *T. simiae* and *T. godfreyi* are primarily confined to Suids. Within *T. congolense* and *T. simiae* species a number of types are recognized. Site of development in tsetse fly is in proboscis and mid gut at different

developmental stage. Morphologically it is characterized by medium sized kinetoplast which is located marginally and absence of free flagellum in all stages. The posterior end of the body is either rounded or pointed; the polymorphism is especially pronounced in *T.simiae* (Stevens and Brisse, 2004).

**Subgenus *Trypanozoon*:** is the most homogenous group of salivarian trypanosomes. It contains three recognized species: *T.evansi*, *T.brucei* and *T.equiperdium* which are morphologically indistinguishable but exhibit distinct epidemiological, pathological and genetic characteristics. In Africa, the most important species is *T.brucei*, of which two subspecies, *T.brucei rhodesiense*, *T.brucei gambiense* are responsible for human sleeping sickness. A third subspecies *T.brucei brucei* infects a range of mammalian hosts in domestic livestock. It is tsetse borne parasite, where it develops in the mid gut and salivary glands of the vector. The other two species: *T.evansi* is transmitted by mechanical inoculators (e.g. Tabanid flies) and *T.equiperdium* is transmitted by coitus between equine hosts (Stevens and Brisse, 2004). Morphologically, they are polymorphic. It has long, intermediate and stumpy forms with long free flagellum, short free flagellum and no flagellum, respectively. Undulating membrane is well developed; kinetoplast is small and sub-terminal (Soulsby, 1986).

### 2.1.2. Mode of transmission

The nature of the parasite survival and its mode of transmission significantly affect for the occurrence of trypanosomosis. Cyclically transmitted trypanosomosis occurs in tsetse distributed sub-Saharan African countries; whereas mechanically transmitted trypanosomosis are more widely distributed than cyclically transmitted trypanosome species. Even it was reported that *T.evansi* occurs in Middle East southwest of Arabian Peninsula. On the other hand, *T.cruzi* is a causative agent of chagas disease can be transmitted in five different ways: by a vector (through the faeces of an insect), through vertical or congenital transmission (from mother to her child during pregnancy), by way of blood transfusions or organ transplants, by ingesting contaminated food or drinks, or because of laboratory work.

### 2.1.3. Lifecycle of trypanosome

Trypanosomes are typically digenetic protozoan parasites with life cycles alternating between a vertebrate host, where they exist in blood or tissues, and diverse haematophagous invertebrates. These hosts, mainly insects, act as intermediate hosts and vectors, transmitting the parasites to new vertebrate hosts. Tsetse flies are the only cyclical vectors of trypanosomes. Trypanosomes undergo a serious development stage inside tsetse fly. Trypanosomes ingested by a tsetse fly pass into the mid gut, where their life cycle continues. Relatively short trypanosomes that enter the fly transform into thin procyclic forms, which then multiply and become trypomastigotes in about 3-4 days. Trypomastigotes multiply and pass into different parts of digestive organs and move back to pharynx and hypo pharynx. Depending on trypanosome species some enter into salivary glands and others goes back to proboscis. In the lumen of the gland trypomastigotes transform into *epimastigotes* and multiply their then transform *metatrypanosomes* and this is the infective stage of trypanosome to vertebrates (William, 2002).

### 2.1.4. Pathogenesis

Pathological impact of the disease occurs in three successive stages: acute, stabilization and chronic stages. Fever and highest peak of parasitaemia followed by the development of anemia is the most prominent features of acute trypanosomosis. Virulence of the parasite population, age, nutritional status and breed of the host, etc. can influence the severity of anaemia (Katherine and Edith, 2004 as cited by Moudlin et al., 2004). Enlarged lymph node, weakness, lethargy and loss of condition, abortion and reduced milk production and high rate of neonatal mortality occur as the result of acute disease. In chronic stage, anemia is not strictly associated with the presence of parasites in the blood. Level of paracetamia is intermittently observed. Lymph node and spleen are become normal, even atrophy and sclerosis occur. Stunted growth, wasted and infertility are characteristics of cattle infected with chronic trypanosomosis. General lesions are congestive, inflammatory and degenerative and sometimes haemorrhagic. The disease affects various organs: heart, central nervous system (CNS), eyes, tests, ovaries and the pituitary gland. Congestive heart failure is an important cause of death in chronic cases which is related to the combined effect of anemia, myocardial damage and increased vascular permeability (Moudlin *et al.*, 2004).

Protective ability of the parasite from host immune defense mechanism is also one of the factors that allow the parasite to exist in the host for long time and become chronic stage. Meanwhile protective ability of the parasite creates an opportunity to spread wider in the area. African trypanosomes inhabit in the blood plasma and, in the case of *T. brucei*, also the interstitial fluids, the lymph, and cerebrospinal fluid. Their extracellular life-style presents two unique challenges. First, the parasites must avoid lysis by serum complement factors, a group of enzymes that can assemble into a donut-shaped protein complex on permissive lipid bilayer membranes, leading to loss of the membrane permeability barrier. Secondly, the parasites must evade immune elimination to establish chronic infections. Protection against complement activation and immune elimination is mediated by the variant surface glycoprotein (VSG coat) (Samuel and John, 2018). Trypanosomes contain over 1 000 different VSG genes within their genome. In addition, new variant antigen types (VAT) are created by VSG gene mutations and recombination. Immune evasion is achieved by antigenic variation of the VSG coat, a process that entails switching among any of a thousand VSG genes and their recombinants of which electron micrographs, the coat appears as an amorphous layer 12–15 nm thick on the outer lipid leaflet of the plasma membrane. On each trypanosome, it comprises about 10<sup>7</sup> VSG molecules encoded by a single VSG gene.

## **2.2. Epidemiology of animal trypanosomes and their vectors**

When the epidemiology of a disease is considered the triad determinant factors need to be explained. The characteristic of the host, the parasites and the environment including the cyclical and mechanical vectors may determine the distribution, transmission and intensity of infections by trypanosomes in a given area. Sub Saharan African countries are infested with the vector which plays role in the development and cyclical transmission of trypanosomes.

The disease trypanosomosis is transmitted from infected animals to healthier through cyclical and mechanical transmission, where cyclical transmission requires developmental stage within the vector before inoculated to final host; whereas mechanical transmission doesn't require development within the vector where transmission is immediate inoculation between infected and non infected animals. *Trypanosoma Congolense*, *T.simiae* and *T.brucei.brucei* are dependent on cyclical mode of transmission. Tsetse flies of different species are responsible for the

transmission. There are about 31 species and subspecies of *Glossina* play a role in the transmission of human and animal trypanosomosis. *T.vivax* transmitted in both cyclical and mechanical methods, where as *T. evansi* transmitted only mechanically. It could be tsetse or other biting flies like *Tabanid* spp, and *Stomoxysis* play a major role for mechanical transmission as mentioned by (Radostatis, *et al.*, 2010; Soulsby, 1986; Leak, 1999).

### 2.2.1. Trypanosome distribution and host range

Pathogenic trypanosomoses that are classified under salivarian section are found in at least 37 sub-Saharan African countries. Most of them, except *T.evansi* and *T equiperdium*, follow the distribution of tsetse flies; but *T. vivax* may extend little far away from tsetse belt area, because of the ability to be transmitted through mechanical transmission by biting flies (tabanids, stomoxys) in addition to cyclical transmission. Among African animal trypanosomosis causative agents, *T.congolense* and *T.brucei* are transmitted cyclically and circulating exclusively within tsetse fly belt in sub-Saharan Africa (Soulsby, 1986; Leak, 1999; Radostatis et al. 2010).

Cyclically transmitted trypanosomosis in sub-Saharan Africa depend on tsetse host preference, the common tsetse host are domestic and wild ruminants mainly bovids are preferred. Among these cattle, sheep, goat, buffalo, lesser kudu, greater kudu, warthog, bushbuck, gazelles including zebra and domestic equines are more affected (Fetene et al. 2021). In addition, some literatures also showed wide host range in *Trypanosoma vivax*; it was reported from cattle, dromedary camel, (Birhanu, *et al.*, 2015), goat, sheep, pig, dog (Nimpaye, *et al.*, 2011), horse, donkey (Pinchbeck, *et al.*, 2008) both domesticated and wild buffalo, warthog, hippopotamus, reedbuck, waterbuck (Anderson, *et al.*, 2011), antelope (Guedegbe, *et al.*, 1992), girafe (Auty, *et al.*, 2012), rhinoceros (Mihok, 1992), rodents, pangolins, primates, reptiles and different wild ungulates and carnivores (Njiokou, *et al.*, 2004).

### 2.2.2. Tsetse distribution and ecology

Members of the *morsitans* feed mostly on the mammals of savanna. One third to nearly a half of its food is from warthog, especially one of the forest-dwelling species in the savannah group (*G. austeni*) feeds exclusively on suids. Some other animals that are hardly ever used as host of *G.*

*morsitans* include: Zebra, impala, gazelles and wild beest. Members of the *palpalis* group feed manly on reptiles, bush buck, oxen and occasionally smaller mammals and humans visiting the watering spots. The *fusca* group feeds on a variety of host species, including bush buck, buffalo, other cattle etc. (Pollock, 1996; Clausen *et al.*, 1998). Human are not the preferred host of any of these fly species. Though biting flies (i.e. Tabanid and Stomoxysis) play important role as vector for mechanical transmission, tsetse flies contribution is an immense for the prevailing trypanosomosis related problems in sub Saharan Africa. Tsetse flies are blood sucking insects of the genus *Glossina*. They occur only in sub-Saharan Africa, and there are about thirty one species and subspecies reported as important vectors of African trypanosomosis in both humans and animals (Vreysen *et al.*, 2013). Tsetse flies are robust, 6-15 mm in length, and can be distinguished from other biting flies by their long mouthpart (proboscis) and characteristic wing venation and overlapping of wing like a scissors at rest (Pollock,1992). The identified 31 known species and subspecies of tsetse flies are belonging to the genus *Glossina* are divided into three distinct groups or subgenera based on morphological difference in the structure of the genitalia and further differentiated based on ecological characteristics (Leak, 1999). These are: *Austenia* (*G. fusca* group), *Nemorhina* (*G. palpalis* group) and *Morsitans* (*G. morsitans* group). Species of tsetse flies in each subgroup and their preferred habitat shown in Table 1.

Table 1. Species of *Glossina* and their habitat

Group	Species	Habitat
1. <i>Glossina</i>		Mainly found in savannah areas,
<i>Morsitans</i>	<i>G. morsitans</i>	including open areas and thickets
	<i>G. pallidipes</i>	
	<i>G. swyennertoni</i>	
	<i>G. austen</i>	
	<i>G. morsitans</i> sub <i>morsitans</i>	
2. <i>Glossina Palpalis</i>	<i>G. fuscipes</i>	mainly riverine and lakeshore habitats
	<i>G. palpalis</i>	
	<i>G. tachnoides</i>	
	<i>G. palpalis gambiensis</i>	
	<i>G. palpalis palpalis</i>	
3. <i>Glossina Fusca</i>	<i>G. fusca</i>	mainly forest in West and Central Africa
	<i>G. longipennis</i>	Primary forest in Lakes sides and riverine
	<i>G. brevipalpis</i>	
	<i>G. fusca congolense</i>	
	<i>G. schwetzi</i>	
	<i>G. fuscipleuris</i>	

Source: Kuzoe and Schofield (2005)

In general, tsetse flies are found over wide area of 37 sub-Saharan African countries; which covers more than 10 million km<sup>2</sup> between 15<sup>0</sup>N and 26<sup>0</sup>S latitude. The *morsitans* group inhabits in the vegetation of open wood land and in the wood land of savannah with plenty of game animal living in the area. It accounts for over 50% of total distribution with 4,052,000 km<sup>2</sup> total land cover. The environment of this group is found in the regions of tropical Africa having a mean annual temperature of 19-28 °C where it is neither very humid nor very dry. *G. morsitans* is also known to be adapted to areas along a busy cattle trekking route passing through a densely populated and heavily cultivated area well outside from its usual range (Pollock, 1996; Cecchi *et al.*, 2008).

The *palpalis* group is strongly associated with areas of high humidity and deep shades (gallery forests) along river banks in the middle of shrub or grass vegetation. These areas cover large portions of the transition zone to the north of the Congo basin and falls almost entirely (93%) within the predicted distribution of the *palpalis* group. The total area covered by this group is estimated to about 6,415,000 km<sup>2</sup> (Figure 1). The species of this group need the environment that has high humidity and some deep shade. Best conditions are about 25<sup>0</sup>C and 80-85% relative humidity with average rain fall 1000-2000 mm. *G. palpalis* lives in a wide range of climates and vegetation zones that closely associated with rivers and streams (Cecchi *et al.*, 2008).

The *fusca* group shows closed forest as a common habitat of which over 80% of the total area is suitable for them, that means they cover a total area of 4,132,000 km<sup>2</sup> in sub- Saharan Africa (Figure 1). The *fusca* group is found in and around the rain forest in Guinea to Ghana and the block of rain forest in Nigeria, Gabon, and Zaire etc. They inhabit well within the rain forest, riverine forest and isolated forest islands in the savanna. Because they are found deep in rain forest, little is known about their behavior, they rest on trunks of small trees, on vertically hanging creepers and on thin saplings with the head down wards (Cecchi *et al.*, 2008).

Recently, two species of tsetse flies reported in Southwestern region of Saudi Arabia. However, the distribution is not even throughout the region, due to variation in temperature, relative humidity, vegetation and bush availability (William, 2002). Human population, agricultural activities coupled with climate factors can limit the distribution of tsetse flies in each agro-ecological site. A suitable tsetse habitat commonly is called *tsetse belt*. Within these belts are

patches of forest and bush where environmental conditions, such as shade and high humidity, are suitable for tsetse survival and reproduction (Leak, 1999). Local communities living near the belt are well aware of the concentration of tsetse fly in the area. Extreme dry land like in case of Sahara desert and cold environment from south limits the distribution of tsetse flies. In figure 1 shown that the distribution and the ecological zone of each subgenus depicted.

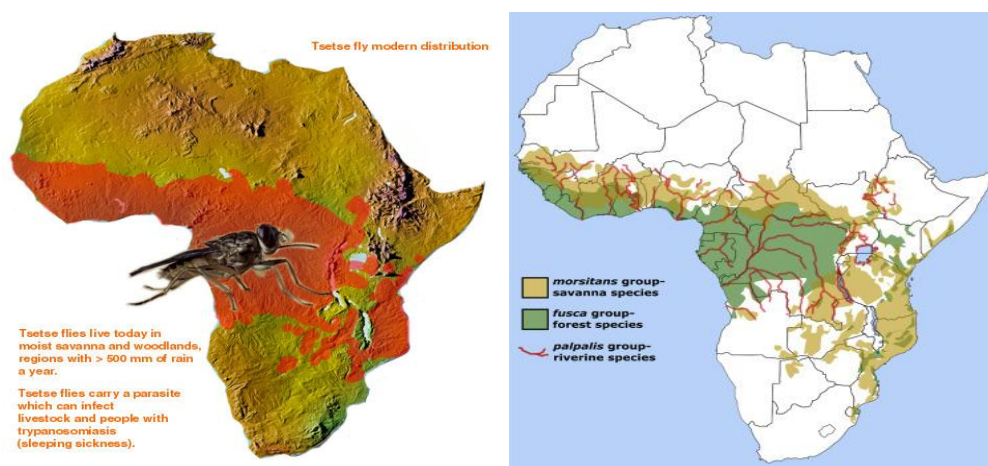


Figure 1. Distribution of morsitans group, fusca group and palpalis group tsetse flies in Africa.  
<https://analarcher.files.wordpress.com/2007/10/tsetsemap.jpg>

### 2.2.3. Climatic and ecological changes and their impact on tsetse and Trypanosomiasis

Tsetse populations mainly influenced by abiotic factors like temperature and humidity, which in turn depends on altitude and vegetation. The temperature limits for the survival of tsetse fly ranges from 17°C to 36°C and tsetse cannot survive in less than 300mm annual rainfall as well as without shelter site (Terblanche *et al.*, 2008). When average temperature is low in the morning tsetse fly inactive and become more active as the temperature rises before midday and late afternoon according to diurnal activity study conducted in Arjo escarpment, Ethiopia. Tsetse activity substantially reduced in the midday when the temperature is hot (Leak, 1999, Solomon, 1990). Feeding rate increases and inter-larval development period shortened when the temperature increases (Torr and Hargrove 1999). Seasonal variation during wet and hot dry season influences the spatial distribution of tsetse fly (Bett *et al.*, 2008). During hot season tsetse flies distribution shrunken towards moist environment whereas in wet season expands wider than dry season. Environmental changes considerably alter tsetse fly population dynamics and their

distribution, and thus influence disease transmission, pathogenicity and impact (Reid *et al.*, 2000; Van Den Bossche *et al.*, 2010). Tsetse fly density determined by suitable habitat, availability of host, expansion of human settlement, deforestation, agriculture, livestock movement (Roger, 1979, 1985). Another important factor limiting species distribution was the water balance stages within the puparium; the degree of resistance to desiccation is closely linked with habitat.

Climate change is an important determinant factor for the distribution of tsetse flies. The limit of distribution directly correlated with the tropical savanna climate. Climate depends on latitude that is modified by altitude and vegetation as well as by important animals host (Leak, 1999). Tropical rain forest climate limit the habitat of the *fusca* and *palpalis* group, whereas the surrounding wood lands are the habitat of the *morsitans* group. Altitude also influences the distribution of tsetse through temperature. In Ethiopia, 1600m.a.s.l. was considered the upper limit of tsetse distribution (Langridge, 1976). However, few studies indicated that it can extend from 1700m up to 2200m for *G. pallidipes* and *G.m.submorsitans*, respectively (Tikubet and Gemetchu, 1984). Apart from altitude and vegetation influence, tsetse abundance and distribution affected by human settlement and over population as it was observed in Nigeria and Uganda valley due to Tutsi migration (Leak, 1999), as the result of bush and tree clearance.

The effects of anthropogenic changes have influenced the epidemiology of AAT, as illustrated by Van Den Bossche *et al.* (2010). Accordingly, environmental change affects the distribution and occurrence of tsetse and trypanosomosis in three points these includes: the first factor occurs as the result of human population pressure when livestock are introduced into game reserve area. Where wildlife's are abundant and constitute the main source of feed of tsetse flies. Wildlife plays an important role as reservoir of trypanosomes in these areas. Therefore, the introduction of people and their livestock into such areas due to human encroachments leads to epidemics of animal trypanosomosis (Van den Bossche *et al.*, 2000).

The second factor involves livestock living at the fringe of tsetse-infested wildlife zones. These areas create a wildlife-livestock contact zone between tsetse infested and non-infested areas. This becomes a risk to livestock entering this zone. For instance, in Mago national park tsetse flies are more confined to this protected National Wildlife Parks, Game Reserves and Forest Reserves (personnel observation). During dry season pastoralists move their cattle into close proximity to the wildlife national parks in search of adequate pasture, which increases cattle-

wildlife contact and exposure to tsetse flies where animals will be more exposed and the incidence of trypanosomosis rampant (Gustafson et al., 2015). A similar situation occurs in West Africa (Reid et al., 2000; Bouyer et al., 2006). In such area, tsetse flies can move to the adjacent environment when the weather condition favors, especially during rainy season tsetse flies widely spread from their belt (Mamoudou et al., 2008).

The third factor, where domestic animals are the main source of feed, this happened because of human intervention and habitat change, in areas where the population density of wildlife is low and livestock share the main source of blood meal for tsetse flies (Reid et al., 2000). The progressive clearing of natural vegetation, expansion of human settlement and agricultural development, and disappearance of large wildlife species due to natural or artificial causes have had repercussions in the distribution, density and dispersal, and the life span of tsetse flies (Van den Bossche et al., 2000).

In addition to the three scenarios that affect distribution of the tsetse and trypanosomosis, species of tsetse prefer different habitat. The *fusca* group (subgenus *Austenia*), tend to occur in the low-land rain forests of West and Central Africa; *palpalis* group species (subgenus *Nemorhina*) occupy similar forest habitats throughout Africa and also extend into riverine and Lakeside forests or the moist areas between such forests; and finally the *morsitans* group of flies (subgenus *Glossina* s.s.) occurs in variety of savannah habitats lying between the forest edges and desert (Rogers and Robinson, 2004) as shown in Table 1 and figure 1.

Potential effect of climate change on tsetse and trypanosomosis distribution considered due to global warming. Increase in temperature has a strong influence on tsetse distribution and development rates of the parasite. Additionally, temperature causes shifting of the geographical range of tsetse flies or host populations, altering transmission dynamics or modifying host susceptibility to infection (Gubler et al., 2001; Patz et al., 2008). For example, increases in temperature will affect pupal development, larval production decreases above a certain threshold, and both pupal and adult mortality increase with temperature.

### **2.3. Socioeconomic impact of African animal Trypanosomosis**

Nagana is still continued to impose a major impact on livestock and crop production in tsetse infested and peri-tsetse infested area of sub-Saharan African countries. The infection of trypanosomosis on domestic animals resulted in low milk production, poor reproduction rate, insufficient draught power for use of crop production and absence of manure for natural fertilizer (Leak, 1999; William, 2002). Currently, more than 40 million cattle and millions of sheep, goats, horses and camels are at risk in 37 sub-Saharan African countries.

The direct impact (Figure 2) of the disease quantified using the parameters of mortality, fertility and milk production, animal traction output and weight loss and then to translate these into monetary terms (Alexandra as cited in Moudlin, et al., 2004). The impact of the disease also varies from situation to situation and it is difficult to generalize because of level of challenge. Annual calf mortality rates mostly ranges from 0-20 percent as suggested by Swallow (2000). Over fifty percent of the researchers quoted calf mortality ranges from 6-10percent due to the presence of trypanosomosis. In high challenge area calf mortality percentages lower because calves are not allowed to graze far outside home, which reduces fly contact and bitten less. Annual mortality rate in older animals are also very variable, with most in the ranges of 2-8%. This low percentage associated with high level of treatment cattle. Significant difference in calf and adult mortality was also observed before and after tsetse control activity, 69.9 to 64.9 and 8.7 to 7.2 percent before and after control activity, respectively; in Bukina Faso (Kamuanga et al., 2001a). In monitored herd in Ethiopia susceptible zebus showed high still birth and calf mortality (13.5 and 8.9%) than after tsetse control using pour-on was 4.1 and 5.3 %, accordingly. Whereas, calving rate was 62% before control and become 71% after control (Woudyalew, et al., 1999). Calving rates also reduced from 6 -19% lower in high challenge as compared to lower challenge areas. In terms of parasitaemia, individual animal comparison showed that calving rates were mostly around 7% lower in infected animals than non infected ones. On the other hand, oxen in high risk area are 38% less efficient than in a low risk area. In addition, in trypanosomosis village sheep and goats can lower annual lambing and kidding rates by around 20-30% even can also reduce twinning. These factors are important to extrapolate the impact of trypanosomosis by comparing infected with non-infected, before and after control or comparing between high challenge and low challenge.

Socio-economic impact of the disease is explained in terms of direct and indirect effects on livestock production and livelihood of the community. Long effect on livestock population growth, reduction on market off take of livestock, cost of trypanocidal drugs, reduction in traction efficiency and milk production considered to affect livelihood of households in high tsetse challenge area (Swallow, 2000). The program on African animal trypanosomiasis (PAAT) estimate that African animal trypanosomosis (AAT) causes approximately 3 million cattle deaths per year and farmers are required to administer approximately 35 million doses of costly trypanocidal drugs many of which fail because of drug resistance developed by the parasites (Geerts *et al*, 2001). Economic losses in cattle production are estimated at US\$ 1-1.2 billion annually and total agricultural losses are estimated at US\$ 4.75 billion per year

(FAO, [www.fao.org/aq/aqainfor/programmes/en/paat/home.html](http://www.fao.org/aq/aqainfor/programmes/en/paat/home.html)).

African trypanosomes cause nagana, a wasting and fatal disease in livestock. *Trypanosoma vivax*, *T. brucei* and *T. congolense* are regarded as major pathogens of cattle and other ruminants, while *T. simiae* causes high mortality in domestic pigs. Nagana has restricted agricultural development and has a profound effect on the economy of tropical Africa. Approximately 70% of the humid and sub-humid zones of sub-Saharan Africa are devoid of cattle. Only through continued chemotherapy and tsetse control programs, cattle can graze on fringe of the tsetse habitat. Sufficient financial and human resources are necessary to implement or sustain a comprehensive control program in all endemic countries (Johnson, 2007).

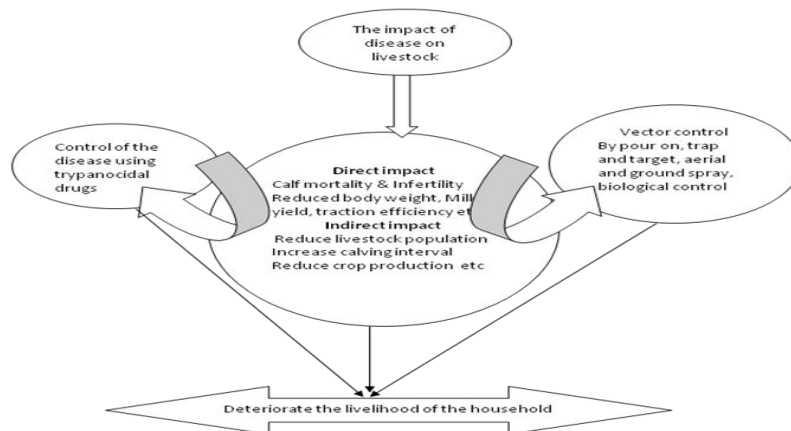


Figure 2. Schematic diagram indicating the socio-economic impact of trypanosomiasis

## 2.4. Control of trypanosomosis and its vectors

The economic and human health significance of trypanosomosis and their associated vectors imply that efficient and integrated control strategies be adopted to reduce the impact on livestock production and human health. The African animal trypanosomosis has been mitigated for centuries by means of direct intervention using different trypanocidal drugs and indirectly through vector control. Various strategies applied to control tsetse population, among these bush clearing, aerial spray, ground spray, sterile insect release, insecticide spray, baited traps and impregnated target are used in tsetse infected sub-Saharan African countries including Ethiopia (Leak, 1999).

### 2.4.1. Treatment against the parasite

Over most of sub-Saharan Africa and in other areas where different forms of trypanosomosis prevail, the use of trypanocides is a common practice. The most common trypanocidal drugs used are Ethidium/Homidium, Samorin, Isomethamidium, Diminazine aceturate. The drugs can be used as therapeutic and/or prophylactic. The main therapeutic drug for cattle is diminazene aceturate. Homidium chloride and homidium bromide are both therapeutic and prophylactic while isomethamidium is used as prophylactic drug. Melarsoprol, pentamidine, suramin and eflornithine are drugs of choice against sleeping sickness while Benznidazole and nifurtimox are the two drugs available for use against Chagas disease in the Americas (Barrett *et. al.*, 2007). It is cheaper and easier to attempt to kill the parasite than the fly vectors. However, besides their varying toxicity, the effectiveness of all trypanocidal drugs have dangerously declined with the emergence of more and more drug resistant trypanosome strains (Wilkinson and Kelly, 2009). For some of the above mentioned drugs, parasites have already developed resistant and drugs becoming less preferred in the market. Despite the known limitations and absence of vaccine development due to variant surface glycoprotein (VSG), the use of trypanocidal drugs remained the preferable choice by the farmers in most African countries (Leak, 1999).

#### 2.4.2. Vector control approach

##### Use of insecticides

Insecticides used to control tsetse flies listed below in Table 2. Aerial spray, ground spray, target impregnation and pour on techniques applied to control tsetse infestation. Among these spray techniques; aerial spray is less desirable due to its poor efficiency to reach the target, i.e. tsetse shelter site. Susceptibility to all insecticides can vary from one species to another, and between the different classes (age, sex, physiological state) of a species. Teneral male and female flies and older, fed males are generally similar in susceptibility to organochlorine insecticides, but fed pregnant females are much less susceptible by a factor of four to nine. Toxicity of insecticides can also be temperature dependent. Endosulfan has a positive temperature coefficient of toxicity whilst that of deltamethrin is negative. Deltamethrin is more toxic than endosulfan at any temperature, and can be up to 300 times more toxic at 10°C but the degree of greater toxicity varies widely with temperature(Leak,1999).

Table 2. Compounds used for the control of tsetse flies (*Glossina* spp.)(Leak, 1999)

<b>Organo chlorines</b>	<b>Pyrethroids</b>	<b>Avermectins</b>
DDT	Natural pyrethrum	Ivermectin
Dieldrin	Permethrin	
BHC (Gammexane)	Deltamethrin	
Propoxur (a methylcarbamate)	Alphamethrin	
Dimethylphthalate (indalone)	Flumethrin	
Diethyl toluamide (DEET repellent)	Lambda-cyhalothrin	
Endosulfan	Cyfluthrin-SOLFAC/Bayofly'	
Ethylhexane-diol (Formula 22)	Alpha-cypermethrin (Cypermethrin)	
	'Flectron' eartags	
	'ECTOPOR'	

##### Traps and target

Traps and targets are made of black and blue color cotton or lylon cloth. Different types of trap designs used for tsetse control namely: monopyrarnidal, NGU, ENZi, F3, Biconical are in use; among these biconical trap frequently used for experimental purposes. Flies are attracted by the blue segment, and they funnel upwards into a netting trap. To increase the efficiency of traps are baited with cow urine and acetone. These traps are mainly used for entomological surveys and

for tsetse control (Pollock et al., 1992). Targets are applied after insecticide impregnated for tsetse control activities. This control technique is efficient and ecologically friendly (Vreysen et al., 2013a). In addition, traps and targets can be used in combination with live baits to suppress tsetse population in accessible target sites before eradication programme started. Despite the effectiveness and environmentally friendly, use of traps and targets deployment very labourous and require frequent follow-up and replinshment to achieve the highest level. However, these control techniques are widely in use in most sub-Saharan African countries to control tsetse population.

### **Biological control**

Now a day due to environmental reason, there is an interest to use environmental friendly technology. Approaches that can be classified as biological control includes: genetic control, the use of natural enemies (parasites, predators, bacteria and fungi), bush clearing and elimination of wild hosts. However, bush clearing and elimination of wild hosts are not considered as environmentally friendly (leak, 1999). Sterile insect technique (SIT) has been used to eradicate tsetse fly in different African countries (Klassen and Curtis 2005). The technique involves release of sterile male in tsetse infestation area. This biological control activity usually applied after 95% tsetse population suppressed, which still depends on other control technology. The most commonly mentioned is the experience of Zanzibar-Tanzaania (Vreysen, *et al.* 2013a). The experience in Zanzibar has intiated Ethiopia to start biological control. In Ethiopia SIT project is almost in the final stage to release sterile male around Lake Abaya and Chamo, which covers about 25,000km<sup>2</sup>. Supression using ground spray, impregnated target, pour-on and aerial spray become regular activity by the project from the very beginning of the project until the desired percent of suppression achieved and the activity is ongoing.

#### *2.4.3. Use of trypanotolerant breeds*

There are cattle breeds know to have resistant to infection. West African cattle breed N'Dama and Muturu found tolerant. However, their productivity very limited similar with East African shorthorn breed and hence breeding these cattle breed not much attractive. Further genetic research regarding the gene responsible to resistance should be identified using genetic engineering. Experimental work on South African Buffalo has shown to develop broad- acting

trypanocidal activity in their blood plasma. This was mediated by production of a trypanocidal concentration of hydrogen peroxide as a result of catabolism of endogenous plasma purine by plasma xanthine oxidase which results in reduced paracitemia. The second mechanism is reducing parasite population growth. The mechanism is by producing trypanosome growth-inhibitory antibodies directed against conserved receptors for required macro- molecules, and the priming of a cell type that can accelerate the development of antibodies specific for trypanosome VSG thus limiting population growth in a VSG-specific manner (Samual and John, 2018). This green light finding should further extrapolated to qualify the finding with regards to trypanotolerant breed selection.

Vaccine production problem also associated with variant surface glycoprotein coat of trypanosomes. About 1000 antigenic shift and recombination of these could not be feasible to produce vaccine against parasite at least at this time. However, antibodies against tsetse flies are the future research direction. Vaccine development attempted against tsetse fly from mid gut showed a potential vaccine candidate (Kinyua, *et al.* 2005).

## **2.5. Trypanosomosis in Ethiopia**

### *2.5.1. Tsetse distribution and prevalence*

In Ethiopia tsetse and trypanosomosis has covered about 240,000km<sup>2</sup> (Slingenbergh, 1992), which is one fourth of arable land, in Western and South Western parts, of the country. It was reported that at least 6 million of the 45 million heads of cattle that are raised under trypanosomosis risk in Africa, are now found in the west and south west Ethiopia. The North West region of Ethiopia is also affected by tsetse and non- tsetse transmitted trypanosomosis (Afework *et al.*, 2000, Sinishaw *et al.*, 2005). Tsetse infested areas lie in the lowlands and also in the various river basins. The infested area extends from the southern part of the rift valley around the south-west corner of the country and along the western lowlands and escarpments to the Blue Nile. Restricting a further spread is the cold limits imposed by highlands that rise to the height above which tsetse can not survive, or the semi-desert condition along the southern border east of the rift valley.

Four species of tsetse flies reported in Ethiopia (Abebe, 2005), namely: *G.morsitans submorsitans*, *G.pallidipes*, *G.tachinoides* and *G.f.fuscipes* were recorded. Among the nine regions of Ethiopia, five are found to be infested with more than one species of tsetse flies. This includes Amhara, Beneshangul-Gums, Gambella, Oromiya and SNNPR (Afework *et al.*, 2000; Abebe, 2005).

Peer review article extracted from January 1990 – December 2014, has supported the aforementioned idea about tsetse flies distribution in Ethiopia, Uganda and Kenya. Extracted entomological data-sets have localized 1266 distinct sites using GIS. The review enabled mapping the occurrence of eight tsetse species in three study countries: *G. brevipalpalis* and *G. longipennis* under subgenus –*Austeninia*, *G. fuscipes fuscipes* and *G. tachinoides* under subgenus– *Namorhinia*; *G. pallidipes*, *G. morsitans submorsitans*, *G. austeni*, and *G. swynnertoni* under subgenus – *Glossina* s.s. Among eight species mentioned above, four species reported in Ethiopia: *G. pallidipes*, *G. morsitans submorsitans*, *G. tachinoides* and *G. fuscipes fuscipes* (Cecchi *et al.*, 2015).

Nineteen’s surveys were conducted in Omo-Ghibe River tsetse belt, to understand the distribution of tsetse and trypanosomosis in the area as shown in Table 3. Tsetse spp in the study area and prevalence of trypanosomosis was recorded in different districts.

Table 3: Tsetse and trypanosomosis survey result of Omo River tsetse belt

Zone	woreda	Month /year	Altitude range	Tsetse species	Flies/tr ap/ day	prevalence
N.omo	K/Koyssha	1996	1000-1400	<i>G.f</i>	2.1	20
N.omo	Kucha	5/1997	1160-1320	<i>G.P</i>	2.94	-
				<i>G.m.sub</i>	0.04	17
N.Omo	Daramalo	6/1996	980-1700	<i>G.p</i>	0.01	
				<i>G.f</i>	2.56	
N.Omo	Loma	10/1977	1000-1500	<i>G.m.sub</i>	0.08	
				<i>G.f</i>	0.61	5.2
Konso	Woyito	9/1995	400-500	<i>G.p</i>	15	10
N.Omo	Gofa zuria	4/1998	1400-1600	<i>G.f</i>	20	17.2
Gurage	Cheha	2/1999	1500-1650	<i>G.m.sub</i>	0.4	16
				<i>G.p</i>	1	
				<i>G.f</i>	3	
Kefa Sheka	Ginbo	1/1998	1350-1460	<i>G.m.sub</i>	0.16	

	yeki	2/1998	1150-1650	<i>G.p</i>	0.5	24
South Omo	Maki valley	11/1989	600-800	<i>G.p</i>	108.5	8.33
				<i>G.long</i>	1.05	
South Omo	Hammer	5/1996	450-800	<i>G.p</i>	2.5	8.6
				<i>G.f</i>	32	
Hadiya	Soro	3/1996	1400-1500	<i>G.p</i>	0.12	6.8
				<i>G.f</i>	0.7	
N.Omo	Loma	10/1994	1000-1500	<i>G.m.sub</i>	0.77	
				<i>G.f</i>	3	18.71
N.omo	K/ Koysha	12/1994	1000-1400	<i>G.p</i>	0.35	
				<i>G.f</i>	0.53	18.5
N.Omo	K/Koysha	11/1996	1000-1400	<i>G.m.sub</i>	0.28	20
KAT	Tembaro	3/1995	1320-1760	<i>G.p</i>	0.15	6.8
N.Omo	Boloso	2/1995	1120-1470	<i>G.p</i>	0.06	
				<i>G.f</i>	0.4	19.0

Source:SRVL, (1996; 1998), G.f. *G. fuscipes*, G.p.: *G.pallidipes*, *G.m.sub*:*G.m.submorsitans*, *G.long*: *G.longipennis*, KAT: *Kembata Alaba Tambaro*, N.Omo: *North Omo*

### 2.5.2. Trypanosomosis distribution and prevalence

In Ethiopia animal trypanosomosis widely prevail following Blue Nile River and its tributaries in the Western Ethiopia. Which includes major tributaries of Baro Akobo, Gilo in Gambella and Didessa and Tana Beless are among others in Oromia and Amhara region. In the South Western region Omo-Ghibe River and its tributaries are the main tsetse and trypanosome belt. In addition following Abaya and Chamo Lake and its surrounding which covers about 25, 000 km<sup>2</sup> is highly affected by trypanosomosis within Great Rift Valley around Arbaminch. Studies conducted in 11 districts along Omo-Gibe River in Dawro and Gamo Gofa zone escarpment revealed an average of 7.8% prevalence observed with the range of 1.0% to 27.2% (Abebe, *et al* 2017). Similar finding observed an average of 7.2% trypanosomosis prevalence from nine selected districts of Omo-Ghibe tsetse belt (Sheferaw *et al.*, 2016). Studies conducted for the last 20 years different region of the Ethiopia showed that the problem of trypanosomosis is huge and the prevalence more prevails in West and South West region (Table 4).

Table 4: Studies conducted in Ethiopia indicating apparent prevalence of trypanosomosis

Authors	Study year	Sample size	No. positive	Apparent prevalence	Region
Sheferaw et al. 2015	2014	1838	133	7.24	SNNP
Birhanu et al. 2015	2013	493	36	7.30	Tigray & Afar
Lelisa et al. 2015	2014	405	22	5.43	Benishangul
Terefe et al. 2015	2014	409	25	6.11	SNNP
Abera et al. 2014	2014	384	24	6.25	Oromiya
Biyazen et al. 2014	2014	384	11	2.86	Oromiya
Lelisa et al. 2014	2010	389	42	10.80	Oromiya
Tamiru et al. 2014	2013	436	6	1.38	Oromiya
Tafese et al. 2012	2011	386	33	8.55	Oromiya
Tesfaye et al. 2012	2009	1260	153	12.14	Benishangul
Fikru et al. 2012	2011	1524	81	5.31	Country wide
Bishaw et al. 2012	2011	384	30	7.81	Amhara
Bekele & Nasir, 2011	2011	384	33	8.60	Oromiya
Mekuria&Gadissa, 2011	2009	540	67	12.41	Benishangul
Dagnachew&Shibeshi, 2011	2009	368	33	8.97	Oromiya
Tadesse & Tsegaye, 2010	2009	250	11	4.40	SNNPRS
Kebede & Animut, 2009	2008	3200	322	10.06	Amhara region
Miruk et al. 2008	2007	341	40	11.73	SNNPRS
Mihret & Mamo, 2007	2005	3360	275	8.20	Amhara region
Sinshaw et al. 2006	2004	1509	92	6.10	Amhara
Cherenet et al. 2004	2001	7079	501	7.08	Amhara
Tewelde et al. 2004	2001	904	70	7.74	Oromiya
Kidan/mariam et al. 2002	2001	1008	151	14.98	SNNPRS
Afewerk et al. 2000	1997	484	83	17.15	Benishangul
Abebe et al., 2017	2017	1508	118	7.8	SNNPRS

Systematic review and meta-analysis conducted in twenty-four studies showed the apparent prevalence of bovine trypanosomosis varied from 1.38 to 17.15 %. The pooled estimate of bovine trypanosomosis prevalence across studies for the entire period was 8.12 % (95 % CI:

6.88; 9.35), ranging from 10.27 % (95 % CI: 7.34; 13.20) in the late 1990s and early 2000s, to 6.81 % (95 % CI: 5.00; 8.62) after 2010 (Leta *et al.*, 2016). The highest estimated regional prevalence was 13.30 % (95 % CI: 7.73; 18.88) in Benishangul Gumuz Regional state recorded.

## **2.6. Trypanosomosis and vector control practices in Ethiopia: successes and challenges**

### *2.6.1. Trypanocidal drug use*

In most tsetse and trypanosomosis infected sub-Saharan African countries, bovine trypanosomosis is primarily controlled by trypanocides (Peter, 2013). Historically, the three trypanocide compounds namely: Isometamidium chloride, homidium (bromide and chloride) and diminazene aceturate have been available in the market for the last 50 years. Recent evidences however show that currently Isometamidium and Diminazene appear to be widely used (Tekle *et al.*, 2018; Dagnachew *et al.*, 2017; Seyoum *et al.*, 2013). Isomethamidium is mostly used for prophylactic purpose and serve to protect for about six months. Whereas, diminazine aceturate is used for curative measures (Mouldin *et al.*, 2004). However, prolonged use, poor quality brands and miss utilization of the existing trypanocidal drugs have continuously threatened their efficacy (Tekle *et al.*, 2018). In this regards, several studies have ascertained the widespread occurrence of trypanocidal drug resistance in different parts of Ethiopia (Shiferaw, *et al.*, 2015; Dagnachew *et al.*, 2017, Mekonnen *et al.*, 2018; Degneh *et al.*, 2019).

### **Tsetse fly control**

In the face of trypanocidal drug resistance and in the absence of vaccines, trypanosome control heavily relies on control of vectors; more specifically tsetse flies. Success stories in the control of tsetse fly vectors have been documents in many parts of Ethiopia. The main control strategy is based on tsetse fly suppression and eradication, using pour-on, traps and insecticide impregnated targets and the release of sterile male tsetse flies. The use of monoconical and biconical traps as well as insecticide impregnated targets in areas around the Didesa river vally/Omo-Gibe valley in in the 80s and early 90s has been shown to have dramatically reduced tsetse fly population and trypanosomosis incidence from vast areas in Chelo, Limu Shay, Bedele and Dembi-Toba intervention sites (Slingenbergh, 1992; Meyar, *et al.* 2016). Since then, different methods of tsetse control including use of insecticides in the form of areal spray, animal spray or pour-on have been in use to suppress tsetse population (Zekarias *et al.*, 2014). Despite the tremendous

achievements however, tsetse and trypanosomosis remain a persistent challenge to Ethiopian Agriculture. Meyer, *et al.* (2016) in their systematic review indicated that tsetse and trypanosomosis control projects conducted in five sub-Saharan African countries namely: Burkina Faso, Cameroon, Ethiopia, Uganda and Tanzania were not fully sustainable.

More, recently, tsetse suppression using pour-on, targets and traps followed by the release of sterile male flies in the context of an area-wide integrated pest management strategy is being practiced in Southern tsetse infested pockets. This technique is area-wide in nature and has been successful in the control of several insect pests including screwworm fly and the Mediterranean fruit fly (Rogers and Randolph, 1990). The technique was also applied in large scale tsetse eradication programs in some parts of Africa including Burkina Faso (Clair *et al.*, 1990) and Northern Nigeria (Takken *et al.*, 1986). However, the success of this approach is yet to be evaluated as it has already been faced by various technical challenges. It also needs to be sustainable so as to produce lasting positive effect on the health and productivity of livestock and consequently on the livelihood of the community. The challenge of sustainability has already been experienced in countries where the technique has been implemented to eradicate tsetse flies (Klassen and Curtis, 2005). The major challenges in tsetse and trypanosomosis control include: Drug resistance of trypanocide, chemical residue of insecticide on environment, animals and human being are the main challenges to be considered. Apart from these challenges inaccessibility due to natural topography, political boundary, insecurity issues, natural conservation site for fauna and flora, lack of integration among countries, transhumance nature of pastoralist, uncontrolled cattle movement from intervention site to infected sites are the most challenging situation to control tsetse and trypanosomosis.

### **3. MATERIAL AND METHODS**

#### **3.1. Study area**

The study was carried out in selected districts of Dawro and Wolayta zone, namely: Loma and Kindo Didaye, around Ghibe-III hydro-electric dam. Ghibe-III hydroelectric dam is located within the Ghibe - Omo River Basin, in the middle reach of the Omo River around 450 km from Addis Ababa by road to 21<sup>0</sup> South west direction. The Lake created from the root of Ghibe-III hydroelectric dam to its tailrace outfall, extends back from the dam over a corridor of 155km long. The reservoir has surface area of 200km<sup>2</sup> and storage capacity of 2.24million m<sup>3</sup>. The dam height is 250m and crest length is 630m. The approximate geo-reference of the dam area lies at 06°50'10.35" N and 037°17'44.23" E. The construction of the Ghibe-III hydroelectric dam is concentrated in a small area of about 1 km<sup>2</sup> (EEPA, 2009). Administratively, the reservoir stretches over five zones and twelve districts. The downstream area which is below the dam that extends from the dam site up to Lake Turkana, which crosses over through Wolayta, Gamo Gofa, Konta and South Omo Zones. Here, Kindo Didaye district was selected from Wolayita zone representing the downstream study site. Zones located to upstream sites are Dawro, Jima, Kambata Tambaro and part of Wolyta, which are located bordering the water reservoir area. Loma district was selected from Dawro zone representing the upstream study site (Figure 3).

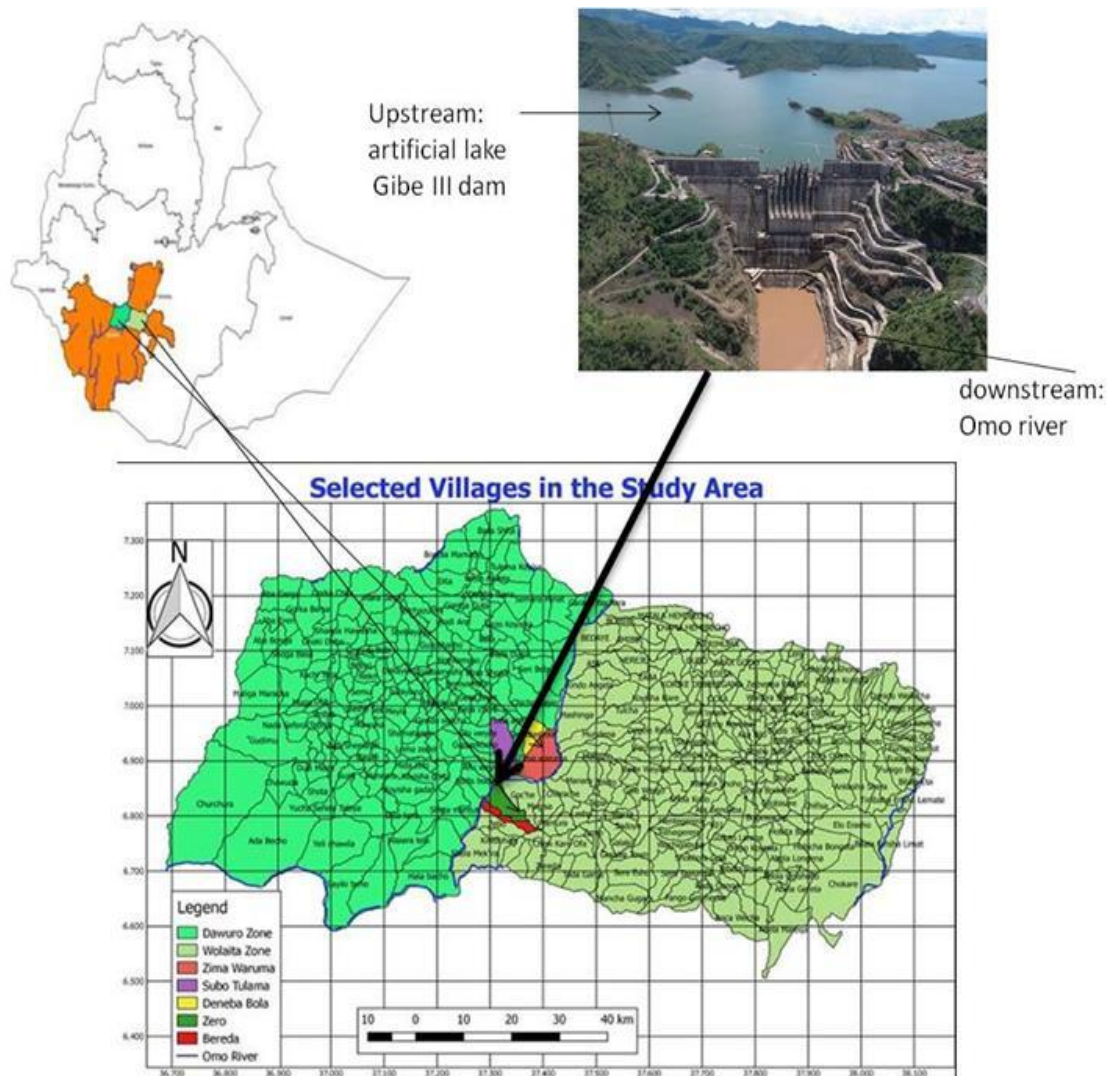


Figure 3. Map of Ethiopia showing study sites in Loma (left side) and Kindo Didaye (right side) district (Dam image: <https://www.salini-impregilo.com>)

### 3.2. Study site description

#### 3.2.1. Natural vegetation and forest

The riparian vegetation in the reservoir area was observed along the river side's and it occupied 1,839 ha of land (8.8 %), and deciduous woodland covers about 17,158 ha (82.2%) and it was characterized by approximately ; 2% tree cover and 98% grass at the time when the field survey was carried out. The exposed surface and silt/gravel land covers 4.7 % of the reservoir area and it mainly occurs along the lower parts of the river, on steep sides of rivers and degraded hillsides and rock outcrops. River/ water body covers 4.3 % of the reservoir area. Farming practices and

settlement are concentrated in areas outside the valley, which are not affected by the reservoir (personal observation; EEPA, 2009).

The vegetation on the hill slopes of the valley was characterized by deciduous phenology (seasonal) of the woodland species which shed their leaves during the dry season and regain them during the wet season as an adaptive mechanism for the prolonged dry season. The plant species of the Omo Valley have over time developed adaptive mechanisms and traits that allow them either to survive fire, germinating after the heat shock or to regenerate after a fire episode. There was a narrow band of riparian vegetation of almost similar species composition as the woodland on the hill slopes. Due to ample moisture, trees found at the edge of the riverbank were not affected by fire as the rest of trees in the upper parts of the study area (EEPA, 2009).

### *3.2.2. Climate*

The amount of rainfall decreases throughout the Omo-Ghibe catchments with a decrease in elevation and varies from a minimum of 1,200 mm to a maximum of about 1,900 mm. The average annual rainfall calculated over the whole Ghibe III basin, where the dam is located, is 1,426 mm. 75 to 80% of the annual rainfall occurs during a five months period from May to September. The mean annual temperature is 20.4°C.

### *3.2.3. Wild life Resources*

Based on the assessment, the population of wildlife species in the project area was low and did not rate equally with areas in downstream of Ghibe III dam like Mago and Omo National Parks and harbors only limited number of wildlife. However, the local residents and professionals from the offices of Agriculture interviewed during the field studies reported the presence of wildlife within the project area. Among mentioned wildlife; warthog, gazzles (greater kudu), buffalo, lion, deer, fox, apes, monkey, hyena etc were included. Vegetation was a good habitat for diverse wildlife species except that threatened by wildfires during dry season. On top of bushfire, intensive human activities at the time of dam construction affected the habitat especially in upstream area. At the time of deforestation before water reservation started, wildlife pushed from their original place towards the downstream where there was less human activities. The

maximum water level remains at around 850 m a.s.l. Therefore, most terrestrial animals took refuge in the area up to 1100 m.a.s.l. where human settlement limited.

### **3.3. Description of study districts**

Loma district was one of the study site, located in Eastern Dawro zone bordering Ghibe-III dam (upstream). The district has three agro ecological zones, which are lowland, midland and highland. The altitude ranges from 850 to 2000 m.a.s.l. Loma has 36 peasant associations and four towns. Out of these 9 PAs' bordering Omo River near Ghibe-III water reserve side. Three PAs bordering Omo River were used for the purpose of this study, namely: Zima-waruma, Deneba-Bolla and Subo-Tulema PAs'. Lomma district has four tributaries that supply Omo River. Namely: Kareta, Mala, Manta and Gindera rivers. Despite trypanosome and other diseases in the district, it was endow with livestock population according to the district veterinary officials. Accordingly, Cattle 183,832, Ovine 74,943, Caprine 92,768, Equine 18,205 and Poultry 207, 927 animal population was recorded(Loma district Annaul report of Livestock resource office, 2017).

Kindo Didaye district was the second study site and located in Wolyta zone bordering Omo River, downstream to the Ghibe- III dam. According to veterinary officials' livestock population in the district was recorded as follows: Cattle 113,122, Ovine 11,298, Caprine 24,452, Equine 6,186 and Poultry 96,504(District livestock resource office report ). The district has 20 Peasant associations and 3 towns. Thirteen PAs were affected by trypanosomosis problem in the district. Of these PA's seven of them were known to border Omo River. Three PA's were used for this study in order to compare downstream tsetse population dynamics and prevalence of trypanosomosis with that of upstream tsetse infestation in Loma district. There are four tributaries to Omo river; Deme, Walka, Site and Kile rivers. The altitude near to the river bank is 676 m.a.s.l whereas in upstream near to the water reservoir is 850m.a.s.l (Figure 4).

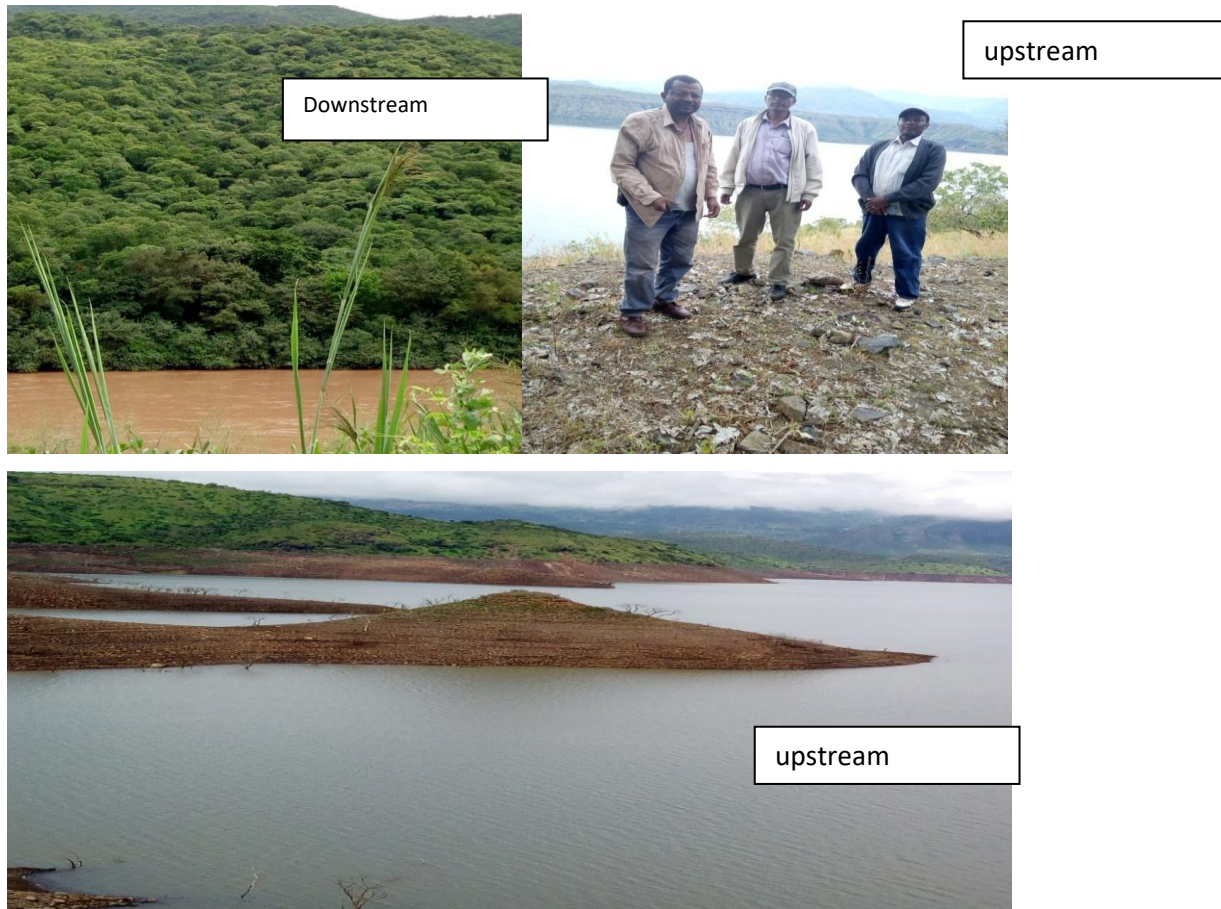


Figure 4. Birds' eyes view of Omo River downstream and upstream of Ghibe-III dam construction

### 3.4. Questionnaire survey

#### 3.4.1. Study population and design

Smallholder farmers located in selected PA's of both Loma and Kindo Didaye districts near the Ghibe-III dam, where people near to water reserve site (the upstream) and below the dam (downstream), were considered. Animal owners were selected randomly using lottery system after being listed in each selected tsetse and trypanosomosis affected peasant associations.

Cross-sectional study design was used for this particular questionnaire survey. Herd/flock size before/after dam construction, major diseases, perception on trypanosomosis and tsetse fly, cause and seasonality of trypanosomosis and other related questions included.

### 3.4.2. Sample size determination

The study area was stratified into two, Ghibe-III dam constructions being as a reference point: upstream which is Loma district and downstream which is Kindo Didaye district. The sample size for questionnaire survey was determined using the formula given by Arsham (2006) with the assumption of 5% standard error, and the study considers 95% confidence interval. For an item scored 0/1 for no/yes, the standard deviation of the item scores is given by  $SD = (p(1-p)/N)^{1/2}$  where p is the proportion obtaining a score of 1, and N is the sample size. The standard error of estimate SE (the standard deviation of the range of possible p values based on your sample estimate) is given by  $SE = SD / N$ . Thus, SE is at a maximum when  $p = 0.5$ . Thus the worst case scenario occurs when 50% agree, 50% disagree. The sample size, N, can then be expressed as largest integer less than or equal to  $0.25/SE^2$ . Therefore, SE of 5% given as follows.

$$n = \frac{0.25}{SE^2} = \frac{0.25}{(0.05)^2} = \frac{0.25}{0.0025} = 100$$

Accordingly, 100 animal owners were interviewed in each district. Most statisticians agree that the minimum sample size to get any kind of meaningful result is 100. If your population is less than 100 then you really need to survey all of them. However in this study we have two districts with more than 500 households in each Kebeles. Therefore we used a total of 200 questionnaires administered in two districts.

### 3.4.3. Questionnaire administration

Livestock owners' perceptions were assessed using semi-structured open and closed questionnaire survey. Face to face an interview method was used to generate information from individuals in selected peasant association of Loma and Kindo Didaye districts. Each questionnaire contained 21 questions. The first five questions were about interviewee biography; age, gender, primary occupation, spp and number of livestock owned before and after dam construction, major diseases that were encountered in the area included. The next eight questions were related to bovine trypanosomosis, clinical signs, means of transmission, seasonality, susceptibility among livestock owned. The last group of questions included the effect of dam on trypanosomosis, status of wildlife population, control practice, cost of drug and

frequency of treatment per month, drug availability and most commonly used trypanocidal drugs were recorded.

### **3.5. Focus group discussion using participatory epidemiology methods**

Participatory epidemiology (PE) tools were used as described by Mariner and Paskin (2000), Catley (2005) and Catley *et al.*( 2012). PE tools included: proportional piling, matrix scoring, socio-economic impact, control methods and seasonal calendar. Stones were used as counters for scoring and ranking purposes (Figure 5). Each group discussion took 2 to 3 hours depending on the level of understanding of the group. Only key informants who have shown willingness to participate and contribute in the discussion were allowed to participate in the investigation. Facilitators were attentively monitoring the discussion to ensure every group member had equal chance to reflect his/her opinion to avoid dominance of the discussion by few participants. At the end of the discussion, stones were used as means of reflecting opinion agreement of that group and this was counted and recorded. Three facilitators: lead facilitator (researcher), assistant facilitator (translator) and one recorder were involved during participatory investigation. Training was given to facilitators especially for recorders and assistant facilitators, prior to actual investigation and data recording methods practiced. Modeling all PE procedures was pre-tested in one non- selected village to familiarize the crew with the activities. The data collected in these pre-test activities were not included in the main data.



Figure 5: Participants actively involved in the allocation of stones for matrix scoring

### *3.5.1. Proportional piling*

Proportional piling was used to estimate the relative livestock population in the area and the most important cattle diseases encountered. A total of fifteen focus group discussions were made: eight groups from Kindo Didaye and seven of them were from Loma districts. At the beginning of the discussion, the researcher allowed the participants to mention types of livestock kept in their area and then listed by their own local language. A pile of 100 stones was given to group members in order to allocate them proportionally according to the size of animals existing in their village. They piled large proportion of stones to large size and small proportion to small size livestock population.

Following livestock population ranking, participants were asked to mention the most important cattle disease and allowed to pile proportionally (Catley *et al.*, 2012) according to severity and or occurrence of the diseases. The first eight diseases identified were used in all focus group discussion and for further analysis. High proportion of stone pile means the disease was more important in terms of severity, impact on productivity, mortality rate, drug cost, or year round occurrence. Every informant was asked to remove or add the already allotted stones to indicate the relative ranking proportion of cattle disease until all participants agreed upon allocated proportion. A semi-structured interview (SSI) and probing questions was also used to support better comparisons among the diseases. This activity was repeated in the fifteen focus groups of the two districts. Each group was composed of six to 20 (on average: 10) animal owners participated depending on their availability and interest to participate in the study.

### *3.5.2. Matrix scoring*

Matrix scoring method was used to understand the community perception about the diseases they have listed out including clinical signs, socio-economic impact, seasonality, age variability and control methods as described by Catley (2005) and Catley *et al* (2012). To verify how well they are responding to indicators of each disease, diseases that were commonly known by facilitators and participants were designated as control disease and used for cross checking (Mariner, 2003) as already described by Catley *et al.* (2012) within methods triangulation. Similar comparisons between diseases were done using matrix scoring to obtain socio-economic impact factors, seasonality, age variation and other indicators and the responses were compared with

descriptions in standard veterinary medical text books (Radostatis, *et al.*, 2007). Simple matrices were constructed to associate diseases against clinical signs and other epidemiological features (indicators). Disease names represented using known material pictures, which were placed along the top X-axis of the matrix while their indicators were placed along the left Y-axis. Group discussants' showed the relationship between each indicator against list of diseases using piles of 30 stones. During scoring process informants were allowed sufficient time to discuss and when necessary probing questions were also applied. The level of agreement across the groups was analyzed using Kendel's coefficient concordance (Seigel and Castellan, 1994).

### *3.5.3. Triangulation*

Triangulation was made using laboratory confirmation (Catley, 2002a; Catley, *et al.*, 2012) to demonstrate the parasite from suspected animal as indicated by clinical signs in the matrix score. Participants were allowed to select trypanosomosis suspected animals from the nearby herd. The selected animals were bled and the blood was examined first with wet film and then thin smear was made and fixed with methanol from wet film positive samples to stain with Giemsa at Soddo Regional Veterinary Laboratory for species identification on morphological basis.

## **3.6. Parasitological Studies**

### *3.6.1. Study population and study animals*

All zebu cattle regardless of their age and sex and which were grazing in selected sites used as the study population. Up on sampling, animal age, sex and body condition as well as reproductive status of cows registered based on information obtained from the owners and/or by physical examination. For body condition scoring the method of Nicholson and Butterworth (1986) was employed whereas to determine age of animals in case the owners did not provide correctly, the method described by Puja Mondal (html) was used.

### *3.6.2. Study design and sampling technique*

The study conducted using two-points cross sectional study design in both dry and rainy seasons from December 2018 to July 2020. Animals in the selected Kebele were brought together and hence sampling was conducted using systematic random sampling technique. The study Kebele's

were purposively selected, based on the fact that they were tsetse and trypanosomosis infected as well as bordering the created Lake (upstream) and Omo-Ghibe River (downstream). A total of six Kebele's namely: Subotulema, Deneba Bolla, Zima Waruma, Petere, Hamaya and Awasho were included.

### 3.6.3. Sample size and Sampling strategy

Three Kebele's from each district were selected purposively because of accessibility and tsetse infestation. For the purpose of sample collection and due to communal grazing system in the area, cattle population in selected Kebele considered as one herd and hence cattle from each Kebele proportionally sampled based on cattle population. Therefore, there were a proportion of 2353(45%), 1569(30%) and 1308(25%) cattle population recorded in Subotulema, Zima Waruma and Deneba Bolla Kebele's of Loma district, respectively. Whereas, a proportion of 456(34%), 473(36%) and 398(30%) cattle population was recorded in Petere, Hamaya and Awasho Kebele's of Kindo Didaye district, accordingly. To determine the sample size needed for the study, a prevalence report of 7.8% (Abebe *et al.*, 2017), 95% confidence interval and 5% desired absolute precision considered. Then a formula given by Thrusfield (2018) was used for simple random sampling technique. Accordingly, three hundred twenty animals from each district, a total of six hundred forty sample size in one season employed. The determined sample size was allocated proportionally based on cattle population in each selected Kebele. Therefore, 144, 96 and 80 bovines were sampled from Subotulema, Zimawaruma and Deneba bolla Kebele of Loma district; whereas, 110, 114 and 96 bovines were sampled from Petere, Hamaya and Awasho Kebele of Kindo Didaye district. In both dry and rainy season a total of 1280 animals were examined. The age of selected animals was estimated by dentition and the body condition status of the animals was assessed based on the criteria described by Nicholson and Butterworth, (1986). Despite nine classifications, in this study body condition score was categorized into two, where the animals showing emaciation, visible ribs, pointed dorsal spine, prominent transverse process, visible hip bone and tail head were considered as poor. Whereas animals with less visible ribs, smooth and well covered and heavy deposit fat clearly visible on tail, head, ribs, brisket and dorsal spine considered as good body condition score.

#### 3.6.4. Blood collection and Microscopic examination

Selected animals ear vein punctured using bleeding lancet and blood collected by heparinized capillary tube. Meanwhile information about the animals like coat color, sex, body condition score, age was recorded. Then blood was centrifuged in 12,000 rpm using heamatocrite centrifuge for 5minutes at field site and removed after centrifugation. Pack cell volume (PCV) measured by HCT reader and the results recorded to estimate anemia. The Buffy coat/ plasma junction with RBC/ was examined by cutting 1mm below buffy coat to include the upper most red blood cells; droplet expressed on a clean microscope slide then covered with cover slip and wet film examined under40x objective magnifications as shown in figure 6, using dark-ground or phase contrast illumination to detect motile trypanosome parasite (Murray *et al.*, 1977). Species of trypanosome confirmed by thin smear stained with Giemsa and examined under100x objective magnification (Luckins, 1992).



Figure 6: Blood collection and microscopic examination

### 3.7. Entomological survey

Entomological survey conducted in both upstream and downstream districts of Ghibe-III dam in selected Kebele's along water reserve and Omo River. Geographical position system (GPS) used to localize trap sites, coordinates were registered for further use. A total

of one hundred sixty NGU traps were deployed in dry and rainy seasons. Eighty traps in each season and 40 traps per district per season were deployed. The traps were deployed at an interval of 200 m air distance in four rows and remained at one site for 48 hrs (Ouma, *et al.*, 2016). Maximum air distance of 2000m in length and 800m in width was covered in each study site. The same trap deployment site was used in both dry and rainy seasons. Trap deployment started at 100m close proximity from the artificial Lake and Omo River, and then moving away from the riverside vegetation towards woodland vegetation in both upstream and downstream until the tenth trap deployed. Traps were baited with acetone and fermented cow urine. The coordinate of study site ranges from 06°81'18"N to 06°94'11"N and 37° 28' 15"E to 37°39'85"E using hand-held geo-graphical positioning system (GPS)(Figure 7). Tsetse fly and other biting flies from each trap were collected, counted and tsetse fly sexed (FAO, 1992; Leak, 1999) and apparent density per trap per day calculated in each district and season. Fly dispersion from the proximity to outside observed. Freshly died and live tsetse flies were dissected to estimate trypanosome infection in the proboscis, mid gut and salivary gland (Phelps and Lovemore, 2004).

### 3.7.1. Determination of tsetse Apparent Density (AD)

Apparent density (AD) is an estimation of flies' density and is given in terms of the number of flies caught per trap per day. It was calculated using the following formula: where fly caught is the number of total flies, trap is the number of functioning traps, and a day is the number of days for which traps were operational. The average AD for each trapping site was then calculated to obtain the data on fly distribution in the area (fly/trap/day) (Leak, 1999).

$$AD = \frac{NFC}{NTs * TDs}$$

Where: NFC = Number of flies captured

NTs = Number of traps

TDs = Number of trapping days



Figure 7: NGU trap deployment and tsetse fly caught

### 3.7.2. Determination of tsetse infection rate

Live flies caught were immobilized using ethyl acetate placed in a cotton wool at the bottom of a specimen tube. Dissection was carried out on freshly immobilized flies as trypanosomes are less likely to be found in dead dried flies (Figure 8). Non-teneral flies that were collected from different sites were dissected. Non-teneral flies were identified by the presence of dark or brown color on their abdomen which indicates the last blood meal. Freshly immobilized flies surface were sterilized by brief immersion in 70% ethanol, then dried by blotted on clean tissue paper, and then dissected in normal saline on a slide immersed with 0.9% saline to keep the trypanosomes alive (Leak, 1999). To avoid cross contamination, the dissecting instruments were sterilized by immersion in 3–5% (w/v) sodium hypochlorite for approximately 2 minutes, followed by extensive rinsing in distilled water and then finally were immersed in normal saline.

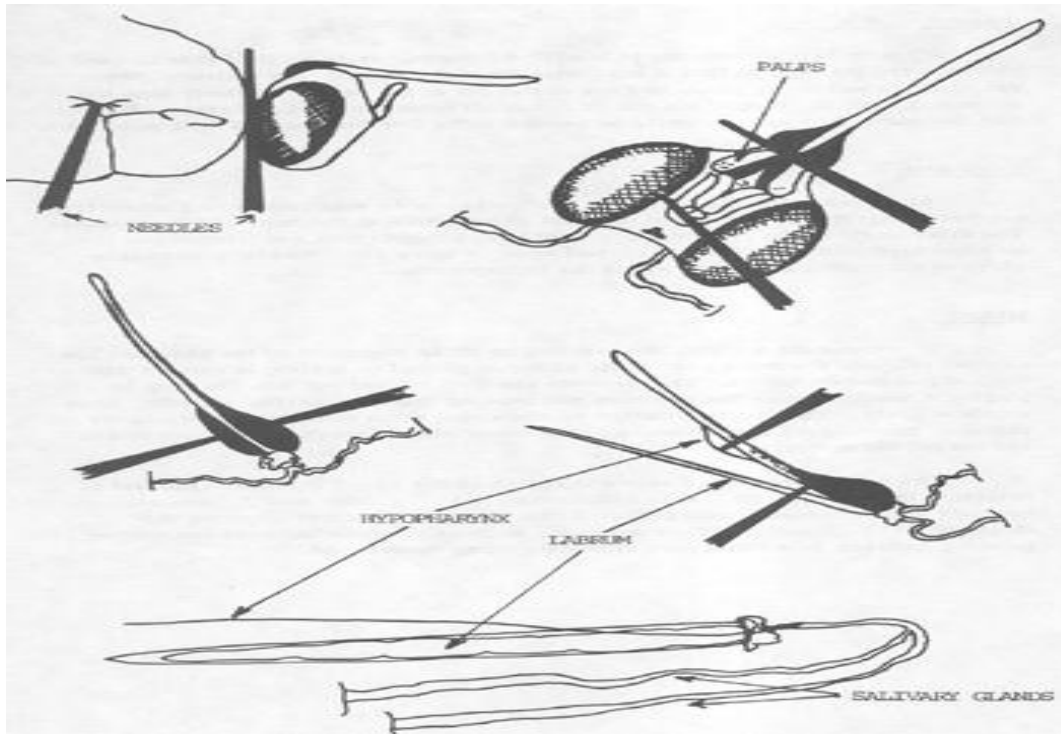


Figure 8. Tsetse fly dissection procedure (Phelps and Lovemore, 2004)

Using dissecting needles the proboscis was separated by applying pressure to the bulb (the cal bulb) at the base of the proboscis under dissecting microscope as shown in the fig 8. The three parts of the proboscis: the labium, hypopharynx and labrum were then separated using needles and spread apart special attention was given to examining the hypopharynx for trypanosomes (Phelps and Lovemore, 2004). To dissect the salivary gland it was necessary to remove the second segment of the abdomen. If the fly was lactating there was some fat in the abdomen and a large transparent structure; the salivary gland was removed using needles. After removing the salivary gland, it was easy to remove the midgut. After dissection, the proboscis, midgut and salivary gland were examined for the presence of trypanosomes using a Zeiss axiostar light microscope. Trypanosomes belonging to different subgenera develop in different parts of the fly; one can often recognize the subgenus involved by the location in the fly. *T. brucei* can be found in proboscis, mid gut and salivary gland, *T. congolense* can be found in proboscis and mid gut and *T. vivax* can only be found in the proboscis. However, in this study positive results recorded *T.vivax* when found in proboscis, *T.congolense* when mid gut is positive and *T.brucei* when salivary gland is positive.

## Secondary data

Retrospective data that was generated before Ghibe-III hydroelectric dam construction collected. Information mainly on trypanosomosis prevalence and tsetse fly apparent density in both study districts were extracted from annual report of Soddo Regional Veterinary Laboratory (SRVL) and Southern Tsetse Eradication Project (STEP).

### 3.8. Data Management

Information obtained from questionnaire survey entered into Microsoft Excel spread sheet and summarized using descriptive statistics. Farmers reported major cattle diseases using local terms; translated to the equivalent English terms were used in the analyses based on the clinical sign they have mentioned for each disease. For instance, to mention trypanosomosis disease, farmers described as gulfo or shulula, or gundia and the name “*Eduxinia or Chibebia*” for tsetse fly which was stated as cause of trypanosomosis. Farmers’ disease diagnosing capacity was either accepted when consistent with the clinical signs described by reference book, or rejected if the responses were inconsistent with the clinical signs. After farmers have mentioned diseases, they have to describe the clinical signs to fit with the standardized clinical sign e.g. descriptions such as ‘emaciation, staring coat hair and weakness’ were the most common signs for trypanosomosis described by farmers. Each clinical sign was then compared within the given two reference veterinary texts (Leak, 1999; Radostits *et al.*, 2010). A farmer’s diagnosis was accepted when at least half of the clinical signs described were consistent with those given in reference texts, otherwise it was rejected. In this respect, all clinical signs were given equal weight. Type of drug they used against trypanosomosis, the frequency of treatment per month and the most common drugs available in the area were summarized by pie chart and bar graph to compare the two districts.

Non parametric  $U$  test employed to compare response frequency between two districts, whether the response is different or not. The test statistic for the Mann Whitney U Test is denoted by  $U$  value where the smaller of  $U_1$  or  $U_2$  used as test statistic to compare with critical value from the table at 95% confidence interval ( $\alpha=0.05$ ). Where:  $U_1$  and  $U_2$  defined as  $U_1 = n_1n_2 + n_1(n_1+1)/2 - R_1$ ,  $U_2 = n_1n_2 + n_2(n_2+1)/2 - R_2$ : Where  $R_1$  = sum of the ranks for district 1 and  $R_2$  = sum of the ranks for district 2.  $n$ =total number of observation,  $n_1$ =number of observation in district one and

$n_2$ =number of observation in district two. The test considered to be significant when calculated U- value is less than the critical value (Siegel and Castellan, 1988).

### **Participatory Investigation**

Qualitative data collected from focus group discussion was entered into Microsoft excel spread sheet. The data were then exported to Statistical Package for Social Sciences (SPSS) base 20 (Inc. Chicago IL., USA) for statistical analysis. Kendall coefficient of concordance (W) was used to determine the level of agreement among groups. W (Kendall's coefficient of concordance) ranges from 0 to 1. The higher the value of 'W' is the higher the agreement amongst the informants. According to critical values for Kendall's coefficient concordance (W) provided by Seigel and Castellan (1994) agreement was termed as weak, moderate and good if W=values were less than 0.26, between 0.26 and 0.38 ( $p < 0.05$ ) and greater than 0.38 ( $p < 0.01$  to  $< 0.001$ ), respectively.

### **Prevalence and apparent tsetse fly density analysis**

Data collected were entered into Microsoft Excel spread sheet, edited and coded for the purpose of analysis. All statistical analyses were performed using statistical software STATA version 12 (Stata Corp, 4905, Lake way Drive, College station, Texas). The prevalence of trypanosomosis was computed according to the formula given by Thruisfield (2018). The association of trypanosomosis (dependent variable) with different independent variables was analyzed using univariable logistic regression analysis. Full model was developed using multivariable logistic regression by stepwise backward exclusion methods, log-likelihood and Wald test. Goodness of fitness model was evaluated by estat gof, estat class and Receiver operating curve (ROC) (Dohoo, *et al.*, 2009). Variables with significant Odds Ratio (OR) value and non significant variable that cause the smallest log-likelihood were included in the predictive final model. Mean PCV difference between trypanosome infected and non-infected animals, between season, and districts compared using student test statistics. Linear regression between trypanosome species infection and mean PCV were analyzed; level of significance was considered when p is less than 0.05. Apparent tsetse fly density expressed as tsetse fly per trap per day (F/T/D) and compared against two districts and season. Tsetse dispersion in relation between apparent tsetse density (AD) and distance from proximity of created Lake and Omo River was observed.

## **4. RESULTS**

### **4.1. Community perception on tsetse dynamics and prevalence of trypanosomosis**

#### *4.1.1. Farmers' characteristics*

Out of 200 questionnaires administered in Loma (upstream) and Kindo Didaye (downstream) districts, 99 and 90 questionnaires were completed, respectively. Few participants failed to complete the response due to suspicion or lack of adequate knowledge about the area. Respondents' age ranges between 25 and 70 years. In Kindo Didaye, 14 and 85 were females and males, respectively while in Loma District (upstream), 6 female and 84 male farmers participated. All of them were either livestock owner or livestock keepers. For the focus group discussion, participants in both districts were farmers who had experience in farming and willing to participate in the discussion.

#### *4.1.2. Perception on livestock herd size before and after dam construction*

##### **Questionnaire survey response**

Data generated from questionnaire survey indicates that livestock production systems in both districts were based on free grazing system. Only kids, calves and weak animals stay around homestead while all adults were allowed to move toward free grazing land near to Omo-Ghibe River. In these two districts, tributaries and the main river are used as water sources for their animals. All livestock species kept in both district were local breeds and breeding practice is using natural mating technique. Animals were kept to build economic status, serve as source of food through milk and meat. There was little draught power activity due to inaccessible and rugged land topography. The most common cereal crops produced were sorghum and maize, but production was very limited due to ragged and narrow arable land. Especially in Kindo Didaye landholding was very narrow, because of land shortage they use to plant more root crops. In Loma district, communities keep their animals on the grazing site for long period near to the artificial Lake created by Ghibe-III dam. At night animals kept in open air collecting pen. Animals allowed to comeback homestead either for mass vaccination or treatment and/or if cow was at trimester pregnancy and milking cow.

It was observed that livestock ownership before dam construction in both study districts had no significant difference ( $p>0.05$ ) as shown in Table 5. However, following construction of the dam, livestock herd size has decreased in Kindo Didaye District (downstream the dam) while cattle ownership appear to have improved in Loma District (upstream). Accordingly, in Loma district, out of 90 respondents 77(85%) of them had greater than 6 cattle herd size after dam construction compared to the percentage before dam construction 62(69%) which was significantly ( $p<0.05$ ) higher at present than before. Conversely, in Kindo Didaye district, out of 99 respondents within the district, 33(33.3%) of them had greater than 6 cattle herdsizes after dam construction compared to the proportion before dam construction 53(53.6%), which was significantly different ( $p<0.05$ ).

Proportion of questionnaire survey respondents having sheep and goat flock size of greater than or equal to six has significantly increased after dam construction in Loma District ( $p < 0.05$ ). On the otherhand, respondents in Kindo Didaye have seen a decline in goat ownership while there was no change for sheep herd size following construction of Ghibe - III dam.

Table 5: The current and the past (before dam construction) livestock herd size owned by respondents in Kindo Didaye and Loma districts

spp	Herd/flock Size/ HH	Frequency of respondents (percentage)			
		The current		The past	
		Kindo Didaye	Loma	Kindo Didaye	Loma
<b>Cattle</b>	$\leq 5$	66(66.8) <sup>a,c</sup>	13(14.2) <sup>b,c</sup>	46(46.4) <sup>a,d</sup>	33(36.7) <sup>b,d</sup>
	$\geq 6$	33(33.3) <sup>a,c</sup>	77(85) <sup>b,c</sup>	53(53.6) <sup>a,d</sup>	62(69) <sup>b,d</sup>
<b>Goat</b>	$\leq 5$	97(97.9) <sup>a,c</sup>	26(28.9) <sup>b,c</sup>	93(93.9) <sup>a,d</sup>	63(70) <sup>b,d</sup>
	$\geq 6$	2(2.1) <sup>a,c</sup>	64(71) <sup>b,c</sup>	7(7.07) <sup>a,d</sup>	27(30) <sup>b,d</sup>
<b>Sheep</b>	$\leq 5$	99(100) <sup>a,c</sup>	77(85.6) <sup>b,c</sup>	99(100) <sup>a,d</sup>	87(96.6) <sup>b,d</sup>
	$\geq 6$	0	13(14.4%) <sup>b,c</sup>	0	3(3.4%) <sup>b,d</sup>
<b>Poultry</b>	$\leq 5$	90(90.9) <sup>a,c</sup>	17(18.8) <sup>b,c</sup>	91(91.9) <sup>a,d</sup>	69(76.7) <sup>b,d</sup>
	$\geq 6$	9(9.1) <sup>a,c</sup>	73(81.2) <sup>b,c</sup>	8(8.1) <sup>a,d</sup>	21(23.3) <sup>b,d</sup>

$\leq 5$ = herd/flock size less or equal to 5 animals;  $\geq 6$ = herd/flock size greater or equal to 6 animals Superscript “a and b” compares between two districts at present and before dam construction; whereas superscript “c and d” compares the present and the past values within districts.

## Focus group discussion findings

The focus group discussions in Loma and Kindo Didaye districts gave a general picture on types of livestock kept and their relative proportion in the area. According to median score results in both districts, *Mizza* (cattle) ranked first followed by *Desha* (goat), *Quttuwa* (Poultry), *Dorsa* (sheep) and *Harria* (equine), as shown in Table 6. Livestock management system depends on natural grazing in two districts. Kindo Didaye group discussion participants unanimously agreed that farmers keep their animals overnight in the house and allow grazing in open land the whole day. On the contrary, in the selected village of Loma districts discussants keep milking cow in their homestead using tether whereas other cattle herds are kept far away from home near to Ghibe-III dam water reservoir. Except recently born lambs and kids, sheep and goat are also managed together with cattle. Grazing land, and land covered by forest, bushes and shrubs are much wider in Loma than in Kindo Didaye district.

Table 6. Proportional piling of major livestock population in Loma and Kindo Didaye districts

Kindo Didaye(n=8)				Loma(n=7)			
*Local name	Common name	Range	Median score	Local name	Common name	Range	Median score
<i>Mizza</i>	Cattle	34-59	41	<i>Mizza</i>	Cattle	32-46	36
<i>Desha</i>	Goats	20-31	24	<i>Desha</i>	Goats	17-25	23
<i>Dorsa</i>	Sheep	3-14	6	<i>Dorsa</i>	Sheep	4-16	8
<i>Quutuuwa</i>	Poultry	11-23	20	<i>Quutuuwa</i>	Poultry	13-18	14
<i>Harria</i>	Equine	3-12	8	<i>Harria</i>	Equine	3-8	5
<i>Matta</i>	Bee hive	0	0	<i>Matta</i>	Bee hive	6-14	9

\*local name is in *italics*

### 4.1.3. Knowledge on major livestock diseases

#### Questionnaire survey responses

During the semi-structured open and closed questionnaire survey, farmers have listed major livestock diseases chronologically based on significant impacts on their livestock productivity. In both district, all questionnaire respondents have listed main diseases which they perceived

prevalent any time in their experience. The first six diseases that were frequently mentioned by respondents were listed in Table 7 below while those less frequently mentioned were collectively categorized as “others”. Bovine Trypanosomosis ranked first for 97.8% and 93.4% followed by Blackleg with the proportion of 85.5% and 79.8%, in Loma and Kindo Didaye district, respectively. Lumpy skin disease (LSD) was reported with high frequency in Loma compared to Kindo Didaye district. Respondents believed that the presence of the artificial Lake coupled with wind contributed for the spread of the vector and the disease LSD towards west direction to Loma.

Table 7: List of major livestock diseases, frequency and percentage of the respondents in Loma and Kindo Didaye districts

Diseases	Kindo Didaye		Loma	
	N	% farmers	n	% farmers
Trypanosomosis	88	97.8	93	93.4
Blackleg	77	85.6	79	79.8
Anthrax	64	71.1	66	66.7
LSD	73	81.1	5	5.1
FMD	42	46.7	12	12.1
Pasteurolosis	47	52.2	0	0
Others*	5	5.6	42	42.4

\* GIT parasite, leech, babesia, bloat and etc. diseases mentioned by at least one farmer; n= frequency of respondents

### Focus group discussion findings

Participants’ in focus group discussion have listed a number of diseases in local names by describing clinical signs, severity and lesions. Probing questions were asked until agreement was reached on each disease under discussion. The seven most frequently mentioned and “other” disease designated to represent diseases with disagreement among groups were used for further matrices. The local name of each disease described by the group was translated by the facilitator: Accordingly, *Shulula* (Trypanosomosis), *Tilikia* (Blackleg), and *Duluwaa* (Anthrax) were ranked in the first three places, respectively (Table 8). In focus group discussion differences observed between two districts; *Aheeraa*/tick born or tick associated disease was mentioned only in Kindo Didaye whereas *Gerenda* (pasteurellosis) mentioned in Loma. Proportional piling score

was calculated for median score and minimum and maximum range. “*Shulula*” (trypanosomosis) became the highest among other diseases. Their main reason of high proportion piling was because of the disease impact throughout the year and its effect on productivity by reducing milk yield and bodyweight. In addition, cost of treatment for this particular disease was reported to be very high. The finding in group discussion was similar with questionnaire respondents; where in both FGD and questionnaire survey indicated that trypanosomosis was number one problem.

Table 8. Major Cattle disease listed by FGD participants in Kindo Didaye and Loma districts

Kindo Didaye(n=8)				Loma (n=7)			
Local name	Common name	Median Score	Range	Local name	Common name	Median score	Rang e
<i>Shululaa</i>	*Tryps	31	24-65	<i>Shululaa</i>	Tryp’s	22	8-28
<i>Karishuwa/ Tilikiaa</i>	Black leg	13	10-18	<i>Tilikia/ tinticho</i>	Blackleg	16	14-23
<i>Duluwaa</i>	Anthrax	8	0-9	<i>Duluwaa</i>	Anthrax	10	5-20
<i>Aheeraa</i>	TBD	16	9-19	<i>Garanda</i>	Pastuerela	12	4-12
<i>Galbbaa Harggiyaa</i>	Mange	6	4-12	<i>Quacha</i>	Mange	10	5-12
<i>Danquwanne cuuchaa</i>	Tick & lice	7	3-11	<i>Danquwa cuucha</i>	Tick & lice	11	8-13
<i>Ulloguxunniya</i>	GIT parasite/ diarrhea	6	5-14	<i>Gussuwa</i>	Diarrhea/ GIT parasite	12	6-14
<i>Dumma dummaa</i>	Others	7	3-12	<i>Duma</i>	others	12	6-22

\* Trypanosomosis, range = minimum and maximum score during focus group discussion

#### 4.1.4. Knowledge on clinical sign of animal trypanosomosis

##### Questionnaire survey response

Among the disease listed during questionnaire survey trypanosomosis received the highest frequency and number one priority disease. Knowledge of the community about clinical signs of

trypanosomosis was further assessed and frequently mentioned ones were summarized (Table 9). Out of seven clinical signs listed for bovine trypanosomosis, ‘emaciation’ and ‘rough hair coat’ were the most frequently mentioned signs by respondents. All seven clinical signs listed were consistent with conventional clinical signs written in veterinary text books (Radostatis, 2007, Soulsby, 1986). Response differences between two districts were compared statistically using Mann Whitney test. The smaller U test values were compared with tabulated critical value using the formula mentioned in the methodology. Therefore, the calculated results of  $U_1$  and  $U_2$  value were 17 and 32, respectively. The critical value based on degree of freedom in two district  $n_1=n_2=7$  was 8. Whereas the calculated smaller U value in this case was  $U=17$ . Since,  $17>8$ , there is no significant difference between the two districts on knowledge of clinical sign listed for bovine trypanosomosis (Table 9).

Table 9. Frequency of questionnaire respondents’ knowledge on clinical signs of animal trypanosomosis in Loma and Kindo Didaye districts

Loma (n=90)				Kindo Didaye (n=99)			
Clinical signs	freq	R <sub>1</sub>	Response %	Clinical signs	freq	R <sub>2</sub>	Response %
Emaciation	88	12	97.8	Emaciation	91	14	91.9
Swollen lymph node	64	7	71.1	Swollen lymph node	31	5	31.3
Fever and dry muzzle	72	9	80	Fever and dry muzzle	9	2	9.1
Rough coat hair	83	11	92.2	Rough coat hair	90	13	90
diarrhea	69	8	76.7	Diarrhea	8	1	8.1
Cough/salivation	77	10	85.6	Coughing/salivation	40	6	40.4
others <sup>a</sup>	10	3	11.1	others <sup>b</sup>	24	4	24.2
Sum =60				Sum =45			

a; alopecia, lacrimation, weakness, stop regurgitation, lethargy,

b; anorexia, shivering, lacrimation, constipation, tail lesion, bloat, death(each of these signs mentioned by at least one farmer)

R<sub>1</sub>=group one rank: R<sub>2</sub>= group two rank

### Focus group discussion findings

Perception of participants about clinical signs for each major disease listed above were assessed in both Loma and Kindo Didaye districts using matrix scoring technique among 15 focus group discussants’ as shown in Table 10. Particular emphasis was given to trypanosomosis as it is ranked the most important problem and other diseases are taken as controls in the description of

clinical signs. Diseases (Aheera and Gerenda) which were not common to the two districts were excluded. The finding shows clinical signs that were scored against trypanosomosis were generally in agreement with the descriptions in reference books (Radostatis, *et al.*, 2007, Maudlin, *et al.*, 2005; Leak, 1999). The scores allocated to the control diseases indicated that informants understood the scoring method and their reasonable ability to differentiate cattle diseases from one another based on clinical descriptions. For instance, Gelbahargia/quacha (mange mite) received high scores for itching and alopecia, Gussuwa/ulloguxinia (diarrheal syndrome/GIT parasite) received high scores for diarrhea. No itching in trypanosomosis or diarrheal syndrome. They have also clearly differentiated blackleg from anthrax, where they said lameness and skin swelling is a common sign in blackleg whereas sudden death is common in anthrax. Disease sign of “*Shululaa*” (trypanosomosis) was indicated mainly by signs of “emaciation, starring coat, diarrhea and inappetance among other signs, with significant ( $W=0.8^{***}$ ;  $p<0.001$ ) good level of agreement. As shown in figure 4 all focus group piled more proportion of stones to trypanosomosis clinical signs than other diseases. Individual questionnaire response and focus group discussion finding shows that communities have good experience in identifying and differentiation of most important disease.

Table 10. Summary of matrix scoring of disease signs based on FGD outputs

Disease signs	Disease name translated (n=15)							W
	<i>Trypanosomosis</i>	<i>Blackleg</i>	<i>Anthrax</i>	<i>Mange</i>	<i>Tick infestation</i>	<i>GIT parasite</i>	<i>others</i>	
<i>Chirises</i> (starring coat)	15 (7-18)	0	2 (0-6)	4 (0-10)	6.1 (0-9)	13 (7-18)	3 (0-6)	0.8***
<i>Matta mena</i> (inappetance)	5 (3-10)	6 (5-10)	7 (4-11)	(0)	(0)	2 (0-6)	4 (0-9)	0.8***
<i>Lapheses</i> (emaciation)	15 (10-18)	0	0	4 (0-9)	6.1 (0-9)	10 (6-18)	3 (0-6)	0.8***
<i>Kixes/kokorses</i> (skin swelling & shivering)	0	10 (8-24)	4 (0-7)	0	0	0	6 (0-10)	0.9***
Wobes (Lameness)	0	12 (7-20)	4 (0-8)	0	5 (0-9)	0	1 (0-5)	0.9***
<i>Woree/kerekeses</i> (sudden death)	0	11 (0-19)	25 (20-30)	0	0	2 (0-6)	7 (0-18)	0.9***
<i>Denquwaderes</i> (tick infestation & skinlesion)	0	0	0	5 (2-18)	15 (9-18)	0	4 (0-10)	0.8***
<i>Quachises</i> (Itching)	0	0	0	24 (16-30)	4.2 (0-6)	0	0	0.9***
<i>Xires</i> (alopecia)	4 (2-7)	0	0	18 (12-30)	4.2 (0-8)	0	0	0.9***
<i>Gussuses</i> (diarrhea)	10 (7-17)	1 (0-6)	1 (0-10)	0	0	16 (9-23)	2 (0-7)	0.8***

n=15 groups

(6–20 informant members participated in each group) W=Kendall’s coefficient of concordance (\*\*\*) $P<0.001$ ). The median scores in each box and the minimum and maximum scores are shown in parenthesis. Wolyta-Dawro words are shown in *italics*

## **Triangulation**

For the purpose of understanding how well the community has picked an animal with trypanosomosis, participants were allowed to indicate at least three animals they thought positive for trypanosomosis in a herd of cattle. Most of them selected animals with poor body condition, emaciated, staring hair coat and some of them were depressed. Accordingly, out of the fifty animals bled for wet film examination, six animals (6/50) were positive (12%) by buffy coat technique, a method that effectively detects trypanosomes only when parasitemia is above 60 trypanosomes/ml blood (Desquesnes and Tresse, 1996).

### *4.1.5. Knowledge and perception on transmission of bovine trypanosomosis*

#### **Questionnaire survey responses**

Majority of respondents in Loma district associated trypanosomosis transmission with biting fly and tsetse flies (58.9%) followed by animal contact (25.6%) during feeding and waterering whereas 7.8% farmers reported transmission is only through tsetse fly. Whereas 41.4% farmers in Kindo Didaye perceived transmission is mainly associated with watering site and tsetse fly bites (Table 11). In both cases, route of transmission associated with tsetse and biting flies scored high frequency than others. Knowledge of respondents on animal trypanosomosis transmission was compared between the two districts by using  $U$  test. Test statistics results of  $U_1$  and  $U_2$  were 49.5 and 31.5, respectively for Loma and Kindo Didaye. The smaller  $U$  was 31.5 and compared with tabulated critical value of  $n_1=n_2=9$  with two sided level of significance at ( $\alpha=0.05$ ). Tabulated critical value of  $U$  at 9 degree of freedom is 17. Therefore,  $31.5 > 17$ , hence there was no significant ( $p>0.05$ ) difference in the knowledge and perception of respondents on means of transmission of bovine trypanosomosis.

Table 11. Community perception comparison on transmission of trypanosomosis in Loma and Kindo Didaye districts

Route of transmission	Loma=90			Kindo Didaye=99		
	n	R <sub>1</sub>	%	n	R <sub>2</sub>	%
Transmission of disease						
Animal contact, feed, water, biting and tsetse fly	23	15	25.6	29	16	29.3
Biting and tsetse fly	53	18	58.9	10	14	10.1
Animal contact, feed & water	2	7.5	2.2	5	11	5.5
Tsetse fly	7	12	7.8	8	13	8.1
Feed and water	0	2.5	0	1	5.5	1.1
Water and tsetse fly	0	2.5	0	41	17	41.4
Water only	0	2.5	0	4	10	4
Others	3	9	3.3	0	2.5	0
I don't know	2	7.5	2.2	1	5.5	1
	Sum =76.5			Sum= 94.5		

### Focus group discussion findings

Trypanosomosis was being the center of interest and black leg and mange serving as cross check diseases to assess group response on disease transmission using matrix scoring against selected diseases as shown in Table 12. Understandings were created among participants about disease transmission. For instance, direct contact defined as when healthy animal got in close contact with sick animals. Contact with inanimate object or fly contact were not included in the concept of contact means of transmission. Water contamination, feeding, different fly bites were included in disease transmission. The discussion revealed that *shulula* (trypanosomosis) transmission is mainly through different species of biting flies where tsetse fly (*Chebebia*) was given the highest rank. *Tilikia* (blackleg) transmission was through direct contact, pasture feed and water; whereas participant agreed that *Quacha* (mange mite) transmission is mainly through direct contact (Table 12). The test agreement indicates that group respondents substantially agreed on means of transmission for each disease. Almost all group agreed that trypanosomosis transmission exists through tsetse fly and biting flies than other diseases with good level of agreement ( $W= 0.7^{***}$ ;  $p<0.001$ ).

Table 12. Summarized matrix scoring of focus group discussion findings on transmission of bovine trypanosomosis

Disease transmission	Disease				W
	<i>Shulula</i> Trypanosomosis	<i>Tilikia</i> Blackleg	<i>Danquwa</i> Tick&lice	<i>Quacha</i> Mange	
<i>Gaytyoga</i> (contact)	0	14(0-30)	21(0-30)	24(15-30)	0.7***
<i>Hatta</i> (water drinking)	0	3(0-7)	0	0	0.8***
<i>Quuma/Matta</i> (pasture feed)	0	7(0-30)	9(0-30)	0	0.6***
<i>Chibebia</i> (tsetse fly)	14(10-20)	1(0-10)	0	3(0-8)	0.7***
<i>Babanta</i> (Tabanus)	8(3-13)	2(0-11)	0	1(0-4)	0.7***
<i>Boyeboxia</i> (other biting flies)	8(5-11)	1(0-4)	0	2(0-5)	0.8***
<i>Quuphasa</i> (coughing)	0	2(0-6)	0	0	0.4**

*n=15 in each group ranges of 6-20 members participated. W=Kendall's coefficient of concordance (\*\*P<0.01; \*\*\*P<0.001). The median scores; the minimum and maximum scores are shown in parenthesis. Wolyta -Dawro words are shown in italics*

#### 4.1.6. Community perception on seasonality of bovine trypanosomosis

##### Questionnaire survey responses

Individual response of farmer's in both districts revealed about seasonality of trypanosomosis. All respondents agreed trypanosomosis was seasonal; but differed on which season the disease is more prevalent or severe. In Kindo Didaye district, 40% of respondents' perceived trypanosomosis is more prevalent in dry than rainy season whereas 42% responded the problem prevails in both dry and rainy season. On the other hand, in Loma district, 62.2% respondents believed trypanosomosis is more prevalent in rainy season followed by 25.6% respondents who thought the problem is common in dry season. Few Loma district respondents (7.7%) perceived the problem exists in both dry and rainy season; and the remaining 3.3% respondents couldn't decide in which season the disease exist. Their main reason for seasonality also differs among respondents within two districts. Substantial number of the respondents perceived that drought and feed shortage contributes more for the occurrence of trypanosomosis in dry season; whereas the other group of interviewee conceived trypanosomosis occurrence increases in rainy season due to increase tsetse population.

Perception on susceptibility of animal species to trypanosomosis indicates 98% respondent in Kindo Didaye district agreed cattle is the most susceptible followed by goats. Whereas, almost

all respondents' in Loma district has mentioned cattle, equine and goats followed by sheep are more susceptible in chronological order.

### **Focus group discussion**

Focus group discussants have also reflected on the seasonality of the occurrence of bovine trypanosomosis and tsetse/biting flies. Three seasons were identified according to participants: namely, "*Boniya* or dry season that occurs from October to March, *Bedessa* or short rainy season (April to June) and *Silla/Belgo* or long rainy season which extend from July to September. Trypanosomosis (*Shulula*) and tsetse/biting flies (*Udunxia*) were shown to have high occurrence in the rainy season as median score indicates in Table 13 whereas the lowest was indicated to be in dry season. Participants also mentioned that though tsetse flies are low during dry season, animals are unable to clear infection and signs reoccur every 3 weeks due to poor body condition of the animals arising from lack of adequate feed. Comparison between FGD and Individual interview indicates that there was minor difference; where FGD discussant agreed the problem occurs more during rainy season, whereas individual respondents didn't agree with one season because of their own justification.

### **Trypanosomosis occurrence in different age groups**

Focus group discussants categorize cattle ages into three categories: *Mara* (calf), *Mirgowa* (bull/heifer) and *Bora/ Mizza* (adult ox or cow) according to their own local names. Chances were given for group participants to allocate bean sized 30 stones in different age category, according to the occurrence of trypanosomosis in different age groups. The score allocated by fifteen groups were calculated for median and range score as shown in Table 13. Median score in both districts indicates the occurrence of trypanosomosis increases as animal age increases. Majority of participants from both districts have given higher score for old age group than calves and yearlings.

Table 13. Seasonality of trypanosomosis and susceptible age groups according to fifteen focus group discussants in two districts

Seasonality	Loma		Kindo Didaye	
	Trypanosomosis	Tsetse/ biting	Trypanosomosis	Tsetse/ biting flies
Dry ( <i>Bonnia</i> )	12(7-17)	5(0-8)	4(3-6)	3(0-5)
Short rainy ( <i>Bedessa</i> )	0	0	8(7-15)	5(5-8)
Long rainy ( <i>Belgo</i> )	18(13-23)	27(22-30)	18(16-23)	22(18-26)
Age	Loma		Kindo Didaye	
Calf ( <i>Marra</i> )	3(0-7)		3(2-4)	
Bull/heifer( <i>Mirgowa</i> )	8(8-11)		9(7-11)	
Ox/cow( <i>Bora/Miza</i> )	19(13-22)		18.5(15-21)	

\*values are median score; range score in parenthesis

#### 4.1.7. Community perception and practices on animal health management

##### Questionnaire survey response

According to the information from the respondents, the type of trypanocidal drugs commonly used in the area are diminazene aceturate (“*kishkish*”) and trypanidium group (“*duluwa*”) whereas Deltametrin (pour-on) was mentioned as the most common insecticide spray. All respondents use one or both trypanocides at least once in a month (Figure 9). Treatment cost also varied among individuals and between districts. The minimum cost recorded was 10 Ethiopian Birr per animal per treatment, and the maximum drug cost was 60 Ethiopian Birr as shown in pie chart (Figure 10) the frequency of response. In Loma district 97% of them expend within the range of 10 up to 30 Eth Birr, whereas in Kindo Didaye only 66% of them expend within the same range. 34% in Kindo Didaye expend more than 30 Eth Birr which is most costly than Loma.

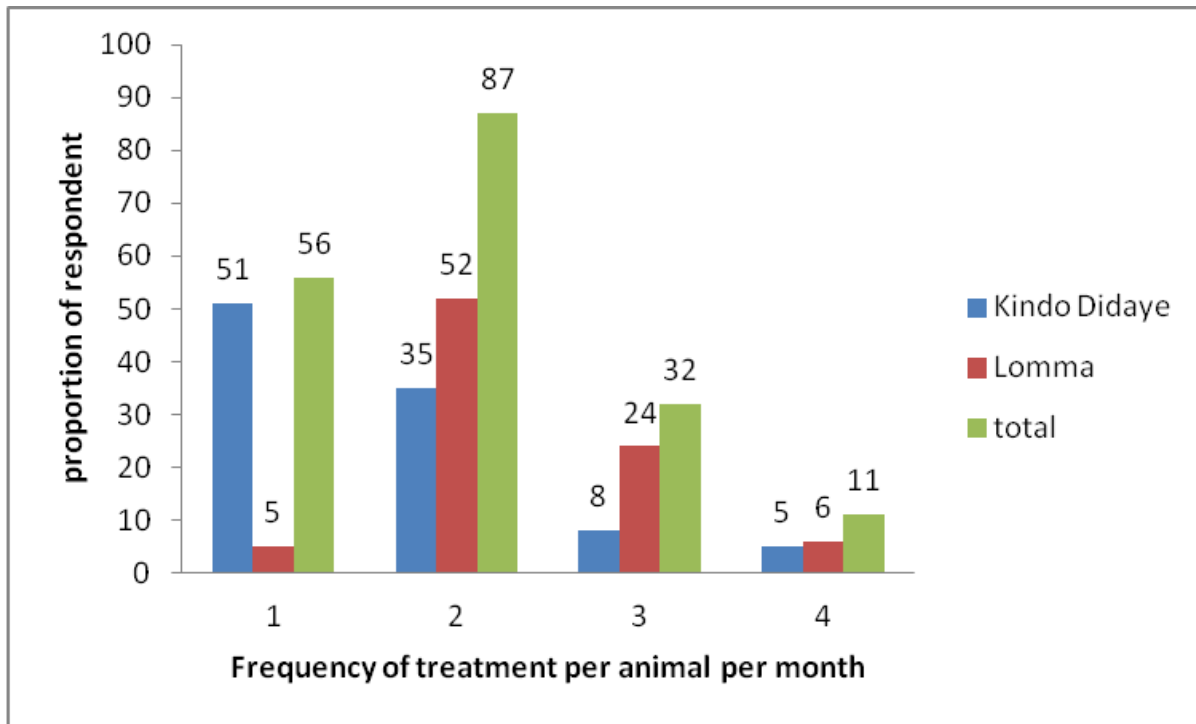


Figure 9. Frequency of treatment per animal per month according to animal owners' response

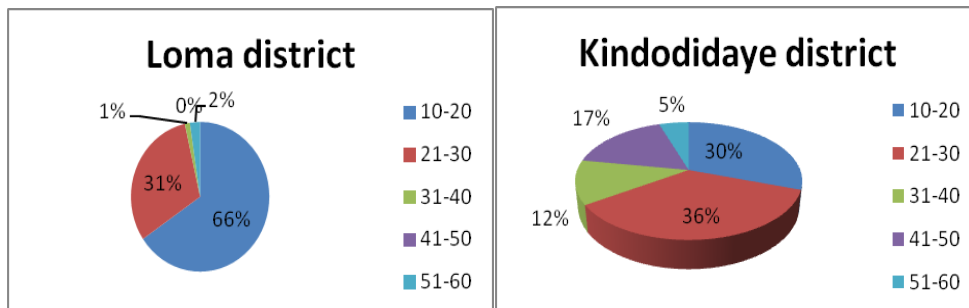


Figure 10: proportion of farmer's response for ranges of trypanocidal drug cost (ETHB) per animal per treatment in Loma and Kindo Didaye districts, respectively

### Focus group discussion findings

Knowledge, perception and practice of the participants were assessed about disease control methods. Trypanosomosis disease in association with other diseases (Blackleg, Anthrax, mange mite, tick and lice, GIT parasite) were compared with control methods, meanwhile response of participants monitored whether discussants responding properly or not. Therefore, treatment, spraying, vaccination and traditional medicine were used as an indicator for control method and analyzed against each disease. Good level of agreement ( $W= 0.75^{***}$ ;  $p<0.001$ ) was shown among the groups on vaccination, spray and treatment control method against diseases; whereas

moderate level of agreement ( $W=0.36$ ;  $p<0.01$ ) was observed between traditional methods against diseases as shown in Table 14. Participants were well aware that control methods for trypanosomosis is treatment and spray whereas vaccination and treatment were more allocated for blackleg as indicated by median score. According to participants anthrax can only be controlled through vaccination because it causes sudden death before it reaches to the treatment service.

Table 14. Summarized matrix scoring of disease control methods based on group discussion

Disease name	Control methods			
	Vaccine	spray	Treatment	Traditional medicine
<i>Shulula</i> (Trypanosomosis)	0	12(9-20)	18(10-21)	0
<i>Tilikia</i> (Blackleg)	16(5-30)	0	11(0-15)	4(0-12)
<i>Duluwa</i> (Anthrax)	29(23-30)	0	1(0-7)	0
<i>Quacha/ Gelbahargia</i> (Mange mite/skin disease)	8(0-24)	8(0-18)	14(6-20)	0
<i>Danquwacucha</i> (Tick and lice)	0	21(8-30)	8(0-18)	2(0-5)
<i>Gussuwa/Ulloguxinia</i> (Git parasite/ diarrhea)	0	0	28(21-30)	2(0-9)
W= Kendel's coefficient	0.89***	0.86***	0.75***	0.36**

n=15 informant groups in each group ranges of 6-20 members involved. W=Kendall's coefficient of concordance (\*\* $P<0.01$ ; \*\*\* $P<0.001$ ). The median scores; the minimum and maximum scores are shown in parenthesis. Wolyta -Dawro words are shown in *italics*

#### 4.1.8. Impact of Trypanosomosis in cattle

Community perception, through FGD, on the impact of trypanosomosis has revealed a number of indicators concordant with reduction in productivity of livestock caused by the disease. such indicators include: milk yield (*mataasisena*), body weight loss (*Ashuwasesena*), poor skin quality (*Gelbahergia*), preference for dowry (*woytisesina*), cause of abortion (*Awuchiyesis*), reduction in selling price (*Hirrayses*), cost of treatment (*Xilliya accoyes*) and cause of sudden death (*Wores*). To verify whether the responses were in line with the target disease (Trypanosomosis), mange and anthrax diseases were included as control items as shown in Table 15. Participants agreed that *shulula* (Trypanosomosis) reduces milk and meat production, causes abortion and is responsible for high cost of treatment compared to other diseases ( $W=0.61$ \*\*\*;  $p<0.001$ ). Whereas the impact of anthrax was unanimously agreed to be sudden death (*wores*), the impacts of external parasites such as tick infestation and mange were poor quality hide and skin followed

by reduction in body weight. The response of focus group discussion signifies how well experienced the community on livestock diseases in that area.

Table 15. Summarized matrix scoring of disease socio-economic impact based on group discussion

Indicators	Disease name (n=15)					w
	<i>trypanosomosis</i>	<i>anthrax</i>	<i>mange</i>	<i>Tick&amp;lice</i>	<i>GITparasite</i>	
Loss of milk <i>Mattasisena</i>	6(4-11)	0(0)	4(0-9)	6(4-8)	7(5-11)	0.61***
Loss of weight <i>Ashuwasisena</i>	4(2-5)	0(0)	3 (0-6)	5(4-7)	7(4-9)	0.68***
Poor hide quality <i>Gelbahargia</i>	0(0-2)	0 (0)	7 (3-11)	6 (4-8)	0 (0)	0.84***
Use for dowry <i>woytisesina</i>	3(2-4)	0(0)	3(0-8)	2(0-4)	2(0-4)	0.34**
Causes Abortion <i>Awuccayises</i>	3(2-5)	0(0)	0(0)	1(0-5)	1(0-7)	0.73***
Cheap price <i>Hirrayeses</i>	4(3-5)	0(0)	0(0)	5(3-11)	4(0-8)	0.72***
Drug cost <i>Xilliya accoyes</i>	7(5-12)	0(0-2)	6(4-10)	5(4-7)	9(5-16)	0.61***
Sudden death <i>Wores</i>	1(0-3)	29(28-30)	0(0)	0(0-2)	0(0)	0.84***

n=15 informant groups in each group. W=Kendall's coefficient of concordance (\*\*P<0.01; \*\*\*P<0.001). Numbers are median score and the minimum and maximum scores are shown in parenthesis. Wolyta -Dawro words are shown in *italics*

#### 4.1.9. Trend of trypanosomosis and tsetse fly population in relation to dam construction

In Loma district (upstream), 87.8% of respondents perceived trypanosomosis problem has been reduced significantly because of improved health care through trypanocidal drug treatment and insecticide sprays while only 12.2% of them said no change. They underlined that tsetse fly population was reduced since dam construction possibly because of deforestation and vast area previously occupied by vegetation is now covered by water reservoir. About 71% of respondents in Kindo Didaye district believed that there was no reduction in the prevalence of trypanosomosis and tsetse fly population despite treatment and insecticide intervention, whereas 29% respondents believed there is reduction in prevalence. A similar trend in trypanosomosis prevalence and tsetse infestation was described by the focus group discussants. Among those who believed there was reduction, 40% of them perceived the reduction of trypanosomosis was due to

treatment only whereas 45% of them believed the reduction were by treatment and insecticide spray. However, Kindo Didaye respondent emphasized there is still extensive tsetse challenge and trypanosomosis on their livestock and hence believed that dam construction had no effect in the situation of the disease.

#### *4.1.10. Wild life status before and after dam construction*

Questionnaire interviewee in both Kindo Didaye and Loma districts shared their experience on wildlife distribution in the area. Some 15 years back the Omo-Ghibe River gorge particularly where Ghibe-III dam built was naturally endowed with abundant fauna and flora. There were different species of wild life's, including lion, buffalo, woodland antelope, common warthog, bushbuck, Hyena's, monkey, apes and others. However, after ten years due to human activities and deforestation in the area for the purpose of Ghibe-III hydroelectric dam construction, wild life populations and diversity was believed to be significantly reduced. According to 87% of respondents in Loma districts, Lion, lesser Kudu, warthog, buffalo, bushbuck moved from upstream to downstream where there is less ecological disturbance due to less human activity. On contrary, monkey and apes significantly increased and moved into human settlement areas. In Kindo Didaye, which is located downstream from the dam, respondents confirmed there is significant change in wildlife population especially following seasonal trends due to natural phenomenon and dam construction. Findings from FGD also affirmed a similar trend in wildlife population in the two districts.

#### *4.1.11. Other changes following Ghibe III dam construction*

More FGD participants from Loma (upstream) district agreed that veterinary service was better, and animal disease outbreaks and cattle deaths were reduced after dam construction compared to those who perceived that it was good right from the beginning (Table16), on the contrary, findings of FGD from Kindo Didaye revealed no significant change in the perception on the above events despite better animal health services.

Table 16. Event comparison before and after Ghibe III dam construction in both Loma (upstream) and Kindo Didaye(downstream) using median score.

Events	Upstream		Downstream	
	Before	After	Before	After
Satisfaction on animal health services	8(6-12)	22(18-24)	10(7-13)	20(17-23)
Occurrence of animal diseases outbreaks	18(16-24)	12(6-14)	14(8-16)	16(14-22)
Number of cattle deaths	21(17-23)	9(7-13)	17(14-19)	13(11-16)

## 4.2. Prevalence of trypanosomosis in cattle in relation to Ghibe III dam

### 4.2.1. Secondary information

Kindo Didaye and Loma districts were assessed at least more than three times before Ghibe III hydroelectric dam construction has started. Due to the severity of trypanosomosis and tsetse fly challenge in the area, two NGO's namely SOS-Sahle and Action Aid supported veterinary health activities to reduce the impact of trypanosomosis and other diseases. Despite all the efforts made trypanosomosis prevalence and tsetse apparent density was high in both districts before dam construction (Table 17).

Table 17. Retrospective data extracted from 2002 to 2016 in Kindo Didaye and Loma districts

District	2002	2004	2005	Average	2006-2016
Loma					Damconstruction period bush clearing activities on water reservoir site upstream from dam
Tryps prevalence	18.71	20	15	17.9	
Tsetse AD	13	9	8	10.0	
Tsetse spp Identified	<i>G.fuscipes</i> <i>G.pallidipes</i> <i>G.m.submns</i>				
Kindo Didaye					Downstream intact with natural vegetation, minor human activities during dam construction
Tryps prevalence	18.5	20	19	19.2	
Tsetse AD	15	14	12	13.7	
Tsetse species identified	<i>G. pallidipes</i> <i>G. fuscipes</i>				

Source: unpublished report obtained in 2019 from Sodo Regional Veterinary Laboratory and Southern Tsetse Eradication Project

#### 4.2.2. Over all prevalence of trypanosomosis in district and season

Out of 1280 cattle examined 116(9.1%) were found infected by trypanosomes (Table 18). This was 6.4% in Loma (upstream) and 11.7% in Kindo Didaye (downstream) and the difference between the two sites was statistically significant ( $p < 0.05$ ). Prevalence of trypanosomosis in dry and rainy season was also significantly different ( $p < 0.05$ ) within and between districts as shown in Table 18. Prevalence of trypanosomosis in Zimawarum kebele near to the water reservoir in upstream showed low prevalence of 2.6% as compared to Petere kebele near to Omo River with prevalence of 15% in downstream. Though there were prevalence difference among Kebeles in downstream, trypanosomosis prevalence is not statistically significant ( $p > 0.05$ ). Whereas, prevalence among Kebeles studied in upstream were significantly different ( $p < 0.05$ ) as shown in Table 18.

Table 18. Over all prevalence of bovine trypanosomosis comparison between downstream and upstream in selected Kebele around Ghibe-III hydroelectric dam

Factor	Kindo Didaye				<i>p</i>	Loma			
	no.	+ve	%(95%CI)			no.	+ve	%(95%CI)	<i>p</i>
Kebele									
Petere	220	33	15[10.7, 20.6]	0.14	Zimawaruma	192	5	2.6[0.96,6.3]	0.05
Hamaya	228	21	9.2[5.9,13.9]		Denba/Bolla	160	13	8.1 [4.6,13.8]	
Awasho	192	21	10.9[7.1,16.4]		Subotulema	288	23	8.0[5.2,11.9]	
Season									
Dry	320	27	8.4[5.7,12.2]	0.01	Dry	320	15	4.7[2.7,7.8]	0.05
Rainy	320	48	15.0[11.4,19.5]		Rainy	320	26	8.1[5.5,11.8]	
Total	640	75	11.7[9.4,14.5]		Total	640	41	6.4[4.7,8.7]	
<i>Between district p-value</i>				<i>&lt;0.001</i>					

*No. = sample size, +ve= number of positive, 95%CI= confidence interval with 95%, p-value*

#### 4.2.3. Species of trypanosomes identified

Three species of trypanosomes were recovered during wet film examination. *Trypanosome congolense* (79.3%) being the dominant species observed followed by *T.vivax* (17.2%) and *T.brucei* (2.6%), only one mixed infection of *T.vivax* and *T.congolense* with the percentage of 0.08% was observed. *T.congolense* was dominant in both dry and rainy season as compared to *T.vivax*. Whereas, *T.vivax* observed relatively more during dry season than rainy season with proportion of 33.3% and 9.5%, accordingly. The number of *T.congolense* and *T.vivax* observed

in downstream district were high in both dry and rainy season as compared to upstream district shown in Table 19.

Table 19. Trypanosome species recovered in cattle during dry and rainy season in six peasant associations in Upstream and Downstream districts of Ghibe-III hydroelectric dam

$T_c = T.congol$   $T_v = T.vivax$ ,  $T_b = T.brucei$ , mixed  $T_c$  &  $T_v$ ,

Districts	Kebele	n	+ve	No. of positive		Spp of trypanosome				Total (%)
				Dry season	Rainy season	$T_c$	$T_v$	$T_b$	Mixed ( $T_c$ & $T_v$ )	
Loma	Zimawaruma	192	5	2	3	4	1	-	-	5(2.6)
	Deneb/ Bolla	160	13	5	8	10	3	-	-	13(8.1)
	Subotulema	288	23	8	15	19	4	-	-	23(7.9)
	Subtotal	640	41	15	26	33	8	-	-	41(6.4)
K/Didaye	Petere	220	33	14	19	24	7	2	-	33(15)
	Hamaya	228	21	5	16	17	3	1	-	21(9.2)
	Awasho	192	21	8	13	18	2	-	1	21(10.9)
	Subtotal	640	75	27	48	59	12	3	1	75(11.7)
	Total	1280	116	42	74	92	20	3	1	116(9.1)

#### 4.2.4. Risk factors

Assumed risk factors were analyzed against trypanosome prevalence using uni-variable logistic regression. Therefore, season, study district, body condition score, sex, age and skin coat colour of the animals were considered. Among the assumed risk factors season, districts, body condition score were significantly ( $p < 0.05$ ) different, where more trypanosomosis infection observed in rainy season, Kindo Didaye (downstream) and poor body condition score; whereas there was no significant ( $p > 0.05$ ) difference in age group, sex, coat color of skin. Significant risk factors using univariable logistic regression were further analyzed using multivariable logistic regression (Table 20). Those significant variables were checked by stepwise backward analysis. Hence season, and district were significant in multivariable logistic regression; but body condition score included in the fitted full model based on log likelihood and Wald test analysis, despite having non-significant value. Hosmer-Lemeshow goodness of fit model evaluation suggested that estat

gof  $\chi^2 = 14.9$  with  $p=0.979$  and estat class evaluation with correctly classified 90.94% as well as Receiver operating characteristics (ROC=0.6599) as shown in Table 21.

Table 20. Univariable logistic regression analysis of bovine trypanosomosis with all assumed risk factors in selected sites of Loma and Kindo Didaye Districts

Risk factors	n	n <sup>o</sup> .+ve	Prevalence[CI]	OR	95%CI	p-value
Season						
Dry	640	42	6.6 [4.8,8.8]	1		
Rainy	640	74	11.6[9.2,14.4]	1.9	1.3,2.8	0.002
District						
Loma	640	41	6.4[4.7,8.7]	1		
Kindo Didaye	640	75	11.7[9.4,14.5]	1.9	1.3,2.9	0.001
BCS						
Poor	1115	108	9.7[8.0,11.6]	1		
Good	165	8	4.9[2.3,9.7]	2.1	1.01,4.4	0.048
Age						
≤ 2 years *	137	17	12.4[7.6,19.4]	1		
2 ≤ 5 years	507	40	7.9[5.8,10.7]	1.7	0.9,3.0	0.10
5 ≤ 10 years**	544	47	8.6 [6.5, 11.4]	1.5	0.8,2.7	0.18
> 10 years	92	12	13.0 [7.2, 22.1]	1.1	0.5,2.3	0.88
Sex						
Female	822	77	9.4[7.5,11.6]	1		
Male	458	39	8.5[6.2,11.6]	1.11	0.7,1.7	0.61
Colour						
Black	28	2	7.1[4.0,14.3]	1		
Red/Brown	554	52	9.4[7.2,12.9]	1.3	0.6, 2.6	0.52
White-gray	698	62	8.9[6.9,11.3]	1.1	0.6, 2.4	0.74

\* Age group less or equal to two permanent tooth, \*\* age when all milk teeth replaced by permanent teeth, Kebele is for peasant association, +ve = number of positive

Table 21: multivariable logistic regression analysis for full model on trypanosomosis association with assumed risk factors

Risk factors	Category	OR	OR 95%CI	Z	P
			LL	UL	
Season	Dry	1			
	Rainy	1.83	1.23	2.73	2.97
District	Loma	1			
	Kindo Didaye	4.34	1.59	11.86	2.86
BCS	poor	1			
	Good	1.463	0.651	3.28	-0.92

LL=lower limit, UL= upper limit, log likelihood=-371.74 >  $H_0$  null model, estat gof  $\chi^2 = 14.9$  with  $p=0.979$ , estat class =90.94% and ROC=0.6599

#### 4.2.5. Packed cell volume association with trypanosomosis infection

The overall mean PCV of sampled animals was 24.40( $\pm$  4.78). According to (Latimer, 2011) the cut off PCV value is 24% to be considered. Animal considered as anemic when it is less than cut point and non anemic above the cut point. Sampled animals were compared between infected and non-infected, between seasons and districts. Mean PCV were significantly ( $p < 0.05$ ) (Table 22) different, in infected animals, dry season, and Kindo Didaye (downstream) district shown low PCV or anemic than the corresponding ones.

Table 22: Packed cell volume comparison between infected and non infected animals in dry and rainy season from selected Kebele of Loma and Kindo Didaye districts

Observation	n	Mean pcv	SE	SD	95%CI for the mean		<i>t</i>	<i>P</i>
					LL	UL		
Non infected	1164	24.54	0.140	4.77	24.27	24.82		
Infected	116	22.97	0.434	4.68	22.11	23.83	3.4	0.05
Dry season	640	22.9	0.187	4.74	22.54	23.28		
Rainy season	640	25.9	0.171	4.34	25.55	26.23	11.4	0.01
Loma	640	25.59	0.165	4.18	25.26	25.91		
Kindo Didaye	640	23.21	0.199	5.05	22.82	23.61	9.16	0.01
Total	1280	24.4	0.13	4.8	24.4	24.0		

Packed cell volume comparison with trypanosome species infection showed that all *T.congolense*, *T.brucei* and *T.vivax* infection were below 24% with mean PCV of 23.44 $\pm$ 4.6, 18.33 $\pm$ 3.0 and 21.52 $\pm$ 4.7, respectively; compared to the corresponding non infected animals mean PCV of 24.47 $\pm$ 4.7. In all trypanosome species, mean PCV significantly ( $p < 0.05$ ) lower than non-infected group. Regression analysis showed the effect of trypanosome species infection has statistically significant ( $p < 0.05$ ) negative impact on PCV value of infected animal. Animals infected either by *T.congolense*, *T.vivax* or *T.brucei* has reduced its PCV with odds of 1.07, 2.97 and 6.21 times more likely than non infected animals, accordingly. Statistical equation was developed for explanatory variables shown as follows:  $y = \beta_0 + \beta_1(T.congo) + \beta_2(T.vivax) +$

$\beta_3(T.brucei)$ ; Where  $y=PCV$   $\beta_0=constant$   $\beta_1 - \beta_3=$  coefficient of respective variables;  $x_1= T.congo$ ,  $x_2= T.vivax$ ,  $x_3= T.brucei$ .

Therefore,  $Y (PCV) = 24.54 - 1.07(T.congo) - 2.97(T.vivax) - 6.21(T.brucei)$

#### 4.2.6. Trypanosome Identification based on morphological characteristics

Among buffy coat examined positive animals, blood sample stained with Giemsa to identify trypanosome species under compound microscope morphologically. Three trypanosome species were identified namely: *T.congolense*, *T.vivax* and *T.brucei* during study period (fig.11). Free fallagella, the shape of posterior end, kinetoplast position were the most criteria used to appreciate morphological difference.

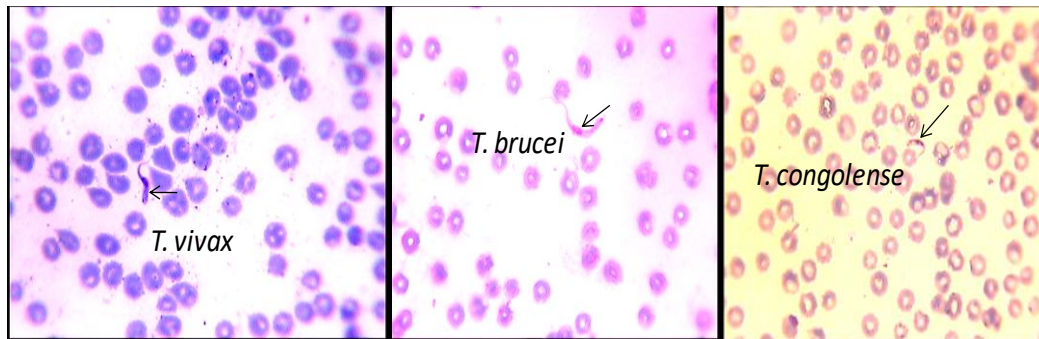


Figure 11. Trypanosome species observed in Giemsa stain from cattle blood (at x1000mg)

### 4.3. Entomological study

#### 4.3.1. Tsetse fly

During study period a total of 1030 tsetse flies and 2045 other biting flies were caught both in dry and rainy season from upstream (Loma district) and downstream (Kindo Didaye). Significant tsetse apparent density difference ( $p < 0.05$ ) was observed between the two sites. Fly per trap per day (FTD) was 5.34 for Kindo Didaye and 1.09 for Loma (Table 23). Seasonal distribution also shows significantly higher fly density with 7.1 FTD in wet season for Kindo Didaye compared to 3.6 for dry season ( $P < 0.05$ ) whereas no such pattern was detected for Loma District. In general *G.pallidipes* and *G.fuscipes* species caught, with different proportion in the two sites. There was high proportion of *G.pallidipes* in Kindo Didaye district, whereas *G.fuscipes* was in high proportion in Loma district. Unlike previous report in the area, there was no *G.m.submorsitans*

tsetse species observed both in dry and rainy seasons. In this study attention was given in species identification using hand lens and FAO manual book. Few tsetse flies were bitten inside the trap by insect predators; these were not counted and sexed. In downstream (kimndodidaye), out of eighty traps deployed in both seasons 78 (97.5%) of them had one or more tsetse flies captured. There was gradual decrease in tsetse apparent density from proximity of Omo River as far as 1900m away from the River. In upstream (Loma) District, out of eighty traps deployed only 16 (20%) of them had one or more tsetse flies captured. In upstream dispersion was irregular where it started with low number at proximity of dame lakeside and reaches high catch in the middle and then declined abruptly (Figure 12). This shows significant ( $p < 0.05$ ) difference in apparent tsetse population between the two districts. In rainy season, there was high apparent tsetse population catch as compared to dry season.

#### *4.3.2. Biting flies count and dispersion*

In this study during dry and rainy seasons a total of 2045 biting flies were caught, of these 736 *Stomoxys*, 329 *Tabanus* and 980 other unidentified species of biting flies were identified. In dry season the catch was low as compared to rainy season. A total of 306 and 1739 biting flies caught during dry and rainy season, respectively; fly catch was relatively high in rainy season. *Stomoxys* catch was only 107 in dry season, whereas during rainy season it was 620; *Tabanus* was also only 59 in dry season, whereas in rainy season it was 270. Likewise, other unidentified flies were also large during rainy season. Biting flies dispersed almost in all over the trap deployed sites (Figure 13). In both upstream and downstream had the same trend of fluctuation starting to the proximity of Lake or River bank as far as the last trap (Table 24).

Table 23. Tsetse species distribution, catch and apparent density based on season in Loma and Kindo Didaye

Study site	season	N <sup>o</sup> . trap deployed	trap With tsetse	<i>G.pallidipes</i>			<i>G.fuscipes</i>			Overall	F/T/D*	p-value
				Male	female	Total	male	female	Total			
Kindo Didaye (Downstream)	Dry	40	38	86	170	256	7	25	32	288	3.6	<0.05
	Rainy	40	40	145	397	542	8	17	25	567	7.1	
	Total	80	78	231	567	798	15	42	57	855	5.34	
Loma (Upstream)	Dry	40	8	1	2	3	27	54	81	84	1.05	>0.05
	Rainy	40	8	0	1	1	30	60	90	91	1.14	
	Total	80	16	1	3	4	57	114	171	175	1.09	
	G.total	160	94	232	570	802	72	156	228	1030		

\* F/T/D = fly catch per no.trap per no. day

Table 24. Biting flies distribution and catch based on season in Loma and Kindo Didaye

Study site	Season	N <sup>o</sup> of trap Deployed	N <sup>o</sup> of trap With biting flies	Range of			Total	AD	
				Fly catch	Tabanus	Stomoxys			Others*
Kindo Didaye (Downstream)	Dry	40	28	1-8	34	65	80	179	2.24
	Rainy	40	41	1-87	192	449	276	917	11.5
Loma (Upstream)	Dry	40	30	1-5	25	42	60	127	1.6
	Rainy	40	39	1-91	78	180	564	822	10.24
	Total	160	138		329	736	980	2045	

\* others=Majority of them were Musudae

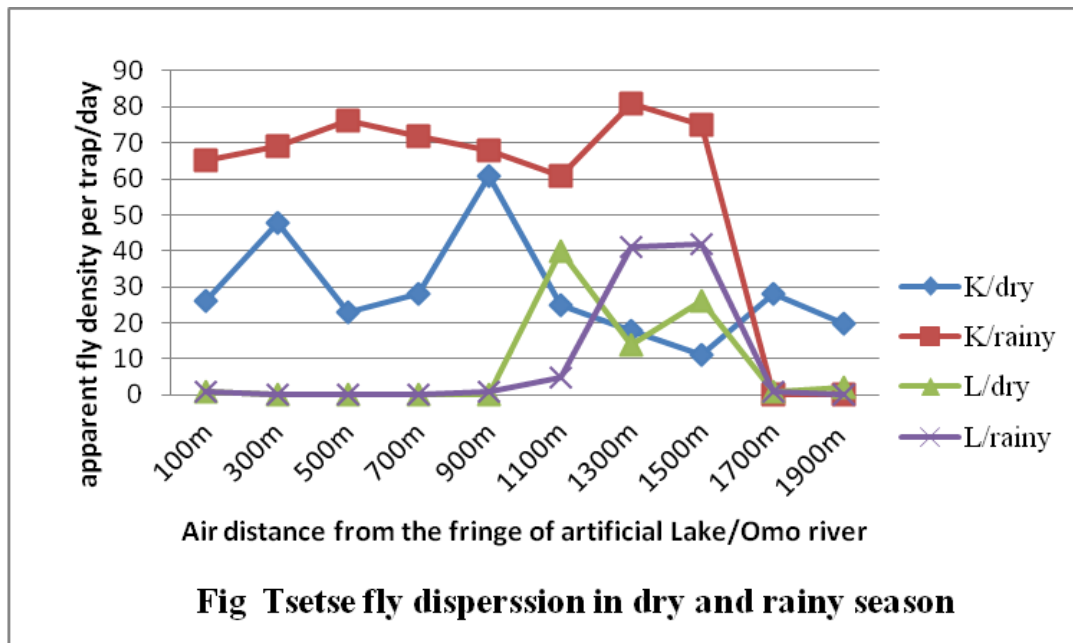


Figure 12. Tsetse flies’ distribution in dry and rainy season at 200m air distance interval from the periphery of downstream (K: Kindo Didaye) and upstream (L: Loma)

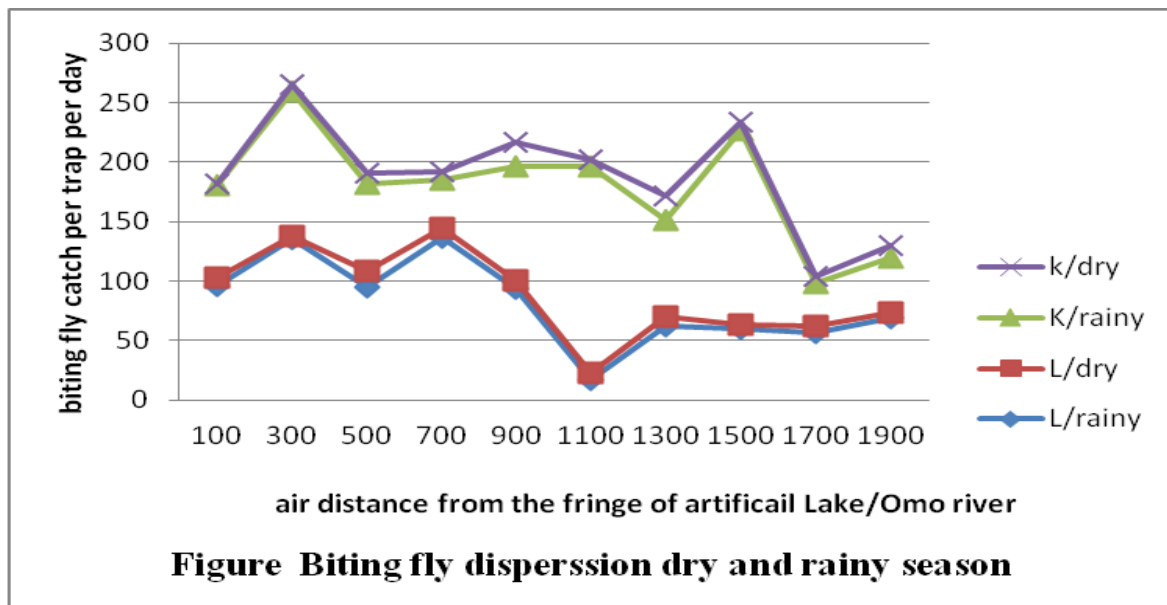


Figure 13. Other biting flies’ distribution in dry and rainy season at 200m air distance interval from the periphery of downstream (K: Kindo Didaye) and upstream (L: Loma)

#### 4.3.3. Trypanosome infection rate in tsetse flies

Out of 1030 tsetse flies caught from both Kindo Didaye and Loma districts in dry and rainy seasons, 182(17.7%) flies that were alive or freshly dead ones dissected to assess tsetse infection rate. Overall, 14.8% of them were infected with trypanosome. Tsetse infection rate was 12% in Loma and 17.8% in Kindo Didaye. There was no significant difference in fly infection rates between Loma and Kindo Didaye fly collection sites ( $P>0.05$ ). Similarly, when the two sites are combined, fly infection rate was 18.9% and 12%, respectively in dry and rainy seasons with no significant difference between the two ( $P> 0.05$ ). When infection rates are compared by season between the two study sites, again no difference was observed. *G. fuscipes* flies were dissected in Loma study site (Table 25).

Table 25: Trypanosome infection of tsetse fly species in dry and rainy season at downstream and upstream of Ghibe III hydroelectric dam

Season	Kindo Didaye (downstream)						Loma (upstream)			Over all	
	<i>G.pallidipes</i>			<i>G. fuscipes</i>			<i>G. fuscipes</i>				
	M	F	Σ	M	F	Σ	Σ Σ	M	F	Σ	ΣΣΣ
Dry season											
n°. dissected	8	26	34	-	1	1	35	8	31	39	74
n°. infection	4	4	8	-	-	-	8	-	6	6	14
Infection rate (%)	50	15.4	23.5	-	-	-	22.9	-	19.4	15.4	18.9
Rainy season											
n°. dissected	8	46	54	-	1	1	55	30	23	53	108
n°. infected	1	6	7	-	1	1	8	1	4	5	13
Infection rate (%)	12.5	13.0	12.9	-	100		14.5	3.3	17.4	9.4	12.0
Overall dissected	16	72	88	-	2	2	90	38	54	92	182
Overall infected	5	10	15	-	1	1	16	1	10	11	27
Overall infection rate (%)	31.3	13.9	17.0	-	50	50	17.8	2.6	18.5	12	14.8
Organ infected											
Proboscis	3	2	5	-	-	-	5	1	1	2	7
Salivary Gland	-	3	3	-	1	1	4	-	4	4	8
Mid gut	2	5	7	-	-	-	7	-	5	5	12

Σ = total at species level; ΣΣ= total of two species; ΣΣΣ= Grand total

## 5. DISCUSSION

### 5.1. Trypanosomosis and its vectors before and after Ghibe-III dam construction

#### 5.1.1. Community perception

Livestock production was more important component of agriculture in both districts. Community perception indicates that cattle population was more dominant followed by goats and poultry, whereas sheep were very small. This agrees with the data reported by the CSA (2018) for both Wolayta and Dawro Zones. According to questionnaire survey responses cattle herd size holding before dam construction was not significantly different between Loma (upstream) and Kindo Didaye (downstream). Whereas, after dam construction, most of the respondents agreed that farmers with cattle herd size greater or equal to six were significantly higher in Loma than Kindo Didaye district. In agreement with this observation within district comparison before and after dam construction was made, farmers with cattle herd size greater or equal to six have significantly increased after dam construction in Loma district, whereas in Kindo Didaye farmers with such cattle herd size have declined. Loma is a district located on the side of the dam reservoir (upstream) where the construction process has disrupted the fauna and flora of the area. It is not clear, if such changes have created favorable conditions such as improved feed availability, reduced diseases occurrence, etc in this area compared to the downstream district, Kindo Didaye. The finding is in agreement with Moudlin, et al. (2004), though there could be variation on trypanosomosis impact from place to place, and difficult to generalize because of variability. Swallow(2000) in his socio-economic impact assement study indicated trypanosomosis and tsetse challenge affect livestock population growth, reduction on market off take of livestock and others.

Community participants have listed more than six major livestock diseases in the study area. Among the diseases listed by participants, bovine trypanosomosis ranked as number one disease priority in both Loma (upstream) and Kindo Didaye (downstream). This was followed by blackleg, anthrax, LSD and FMD accordingly. Community response was in agreement with recent study conducted in Dawro zone by Shiferaw *et al.*(2016) and Abebe *et al.* (2017) have also indicated that trypanosomosis is a major problem in the study area. In Kaffa and Bench Maji zone, which is adjacent to the current study area, 94.1% of questionnaire survey

respondents considered bovine trypanosomosis as an economically important cattle disease accounting for 64.6% of the total annual deaths in the year 2011/2012 (Seyoum, *et al.*, 2013). Similar information reported in tsetse infested area elsewhere in sub-Saharan Africa. Study conducted in Kenya (Ohaga, *et al.*, 2007), Tanzania (Muangirwa *et al.*, 2001) and Nigeria (Njoku *et al.*, 2003) have shown the same report, indicating trypanosomosis is a major constraint in livestock production. Animal trypanosomosis is also prioritized the most important disease among others in West Africa (Gracea *et al.*, 2009).

Questionnaire survey respondents and focus group discussants were able to fairly describe the clinical manifestation of trypanosomosis in cattle in both study locations. Majority of the respondents agreed mainly on emaciation and rough coat hair. Knowledge on swollen lymph node, cough/salivation, fever, loss of appetite and diarrhea were also considered in focus group discussion. Study in Orma community in Kenya reflected similar clinical signs for chronic trypanosomosis infection but hemorrhage and sudden death for the acute form of the disease (Catley, 2006). Comparable knowledge was mentioned in agro-pastoralist area in Lamwat and Kwale, Kenya by (Ohaga *et al.*, 2007; Gracea *et al.* 2009). Majority of the responses of the communities were also in agreement with existing literature (Peter H., 2013; Radostatis, *et al.*, 2007; Soulsby, 1986). Many studies have substantiated that local communities have an accumulated knowledge about their animals and their wellbeing (Catley 2002b). The strong concordance observed between diseases and clinical symptoms listed above strengthens rich indigenous knowledge of the community.

Community indigenous knowledge on clinical sign was cross checked or triangulated by bleeding animals that were selected by study participants. While the finding was a little above the observed prevalence of the disease (9%) in the same study site under the same diagnostic technique (Solomon, *et al.*, 2021), the study expected a better agreement between the parasitological finding and participants' perception. The failure to detect more positive animals among cattle selected by the community for trypanosomosis could be ascribed to the weak sensitivity of the parasitological technique employed (buffy coat) or other animal health factors with similar debilitating conditions. The detection level (sensitivity) of the buffy coat technique, though depends on trypanosome species, is low sensitivity (37%) as compared to the PCR/RFLP (96%) (Marcotty *et al.*, 2008). The minimum parasite concentration per ml of blood below which

the technique can not detect parasitemia is  $2.5 \times 10^2$ ,  $5 \times 10^2$  and  $5 \times 10^3$  for *T.congolense*, *T.vivax* and *T.brucei*, respectively (Maudlin et al., 2005). Since trypanosomosis is mostly a chronic disease, parasites may not be detected in blood under such circumstances (Desquesnes and Tresse 1996; OIE, 2008). Other studies elsewhere in Africa have reported a better correlation between farmers' perceptions and laboratory findings (Oluwafemi, et al., 2007; Catley, et al., 2001; Kimaro, et al., 2017).

Most participants perceived that animal trypanosomosis is transmitted by tsetse and biting flies followed by tsetse and biting flies, animal contact during grazing and watering period. The response in both districts had no significant difference and it was in agreement with scientific descriptions (Leak, 1999; Radostatis et al., 2007, Soulsbey, 1986). Community response finding was also supported by many community perception studies including those in Southwestern Ethiopia, Serengeti community in Tanzania and in Southern Sudan community (Seyoum et al., 2013; Dismas and Kinyemi, 2017 and Catley, 2002b). It was reported that trypanosomosis causes reduction in productivity of livestock; including reduced milk yield, body weight loss, poor hide quality, abortion and costly treatment. Similar observations were recorded in various studies (Peter, 2013). Other studies have shown that more than half of the study participants incriminated draught power reduction and high drug cost to animal trypanosomosis in Metekel and Guji zones of Ethiopia (Tesfaye, et al., 2012; Mersha et al., 2013). Weight loss and milk reduction were also stated by Nigerian community (Oluwafemi, et al., 2007).

It was also evident from the discussions that trypanosomosis is more important in adult animals as compared to younger ones. This was supported by Catley et al (2002a) in Tana River district of Kenya and by Adama et al. (2012) in Ghana and by Von Wissmann et al., (2011) in West Kenya. This could be explained by the fact that tsetse feeding preferences for adult cattle due to size and olfactory cues (Torr et al., 2006) or an inherent resistance to trypanosome infections in young animals (Trail et al., 1994) and/or because calves usually remain around homestead where relatively exposure for tsetse fly is low (Rolands et al., 2001).

In general, the impact of dam on animal trypanosomosis and tsetse fly was explained differently by the upstream and downstream districts communities. Majority of respondent from Loma/upstream district reported that the impact of the disease and its vectors has significantly reduced after Ghibe-III dam construction while participants from Kindo Didaye district reported

no change in disease problem in relation to dam construction. Focus group discussion participants also have ascertained similar finding. On the contrary, Yewhalaw et al. (2010) in their studies around Ghibe-I hydroelectric dam reported that community members perceived an increase in mosquito abundance and malaria disease situation following construction of the dam. Such differences could be attributed to differences in the ecological requirement of the two flies, tsetse and mosquitoes. Stagnant water is the most favorable environment for mosquito multiplication. Water reserve in upstream of Ghibe-III hydroelectric dam could also contribute for mosquito multiplication, but tsetse breeding site might have been affected due to water flooding of breeding site; since most tsetse burrow moisture sandy area to deposit their puparium inside.

Altogether, the local community in both study sites has adequate experience with bovine trypanosomosis and the disease is considered a major threat to their livestock and their livelihood (Catley, 2002b). To combat this problem, the major control methods employed in the area were reported to be the use of trypanocidal drugs and pour-on insecticide spray. Mostly farmers buy trypanocidal drugs and treat their own animals in both districts, when there is drug shortage they look for private vender and open market similar activities indicated by Seyoum *et al* (2013). The dose and the frequency of treatment per month also depend upon their understanding; 53% of respondents practices inappropriately (Machila *et al.* 2003). Whereas in West Africa apply treatment following appropriate clinical signs (Gracea, *et al.*, 2009). However, farmers lack knowledge on appropriate dosage, poor hygienic injection (Ohaga *et al.*, 2007; Gracea *et al.* 2009). This knowledge gap might have contributed under dosage and repeated treatment, which could leads to drug resistance.

#### *5.1.2. Bovine Trypanosomosis situation before and after Ghibe III dam construction*

Trypanosomosis situation in the two study areas was assessed based on data generated before and after dam construction. It was also analysed to see the effect of season in both study districts encompassing upstream and downstream from the dam reservoir. As far as our literature search is concerned, this study is the first of its kind assessing the impact of dam construction on animal trypanosomosis prevalence and its vector. The overall trypanosomosis prevalence in the study area was 9.1% by using conventional buffy coat diagnostic technique. The finding was in agreement with reports of Duguma *et al.*, (2015,) from South western Ethiopia and comparable

with Leta *et al.*, (2016), Sheferaw, *et al.*, (2016) and Abebe *et al.*, (2017), who reported 8.1%, 7.2% and 7.8% prevalence, respectively. When data obtained from cross-sectional studies in Loma and Kindo Didaye were compared with a similar data obtained before construction of the Ghibe-III dam, it was clearly evident that trypanosome prevalence has dramatically declined in Loma, where dam reservoir is located and the ecology is significantly disrupted. This strongly supports the observations of the study participants who ascertained that there was an improvement in trypanosomosis and tsetse situation since dam construction in this particular study area. This is contrary to the malaria situation observed around Gilgel Ghibe I dam where there was an increase in malaria prevalence in the local community after construction of the later dam (Yewhalaw *et al.* 2010). Similarly, snail population and snail borne diseases such as Schistosomiasis has been reported to have increased in prevalence following construction of dams in several countries in Africa and the Middle East (Carter *et al.*, 1990). A study in Senegal has also shown that malaria situation at least remained the same and Schistosomiasis prevalence has increased after Diama dam construction (Sow *et al.*, 2002). It is possible that, the ecological changes that have occurred upstream of the Ghibe III dam and the resulting displacement of wildlife and low tsetse apparent density might have contributed to the reduction in the prevalence of trypanosomosis in cattle.

In a similar situation to the prevalence of trypanosomosis, tsetse apparent density was reduced from its level before dam construction to the current day. While there was also reduction in downstream areas, this reduction was much more pronounced in Loma/upstream sites suggesting that the closer the site to the dam reservoir, the lesser the tsetse challenge. Large scale changes in the environment, especially those involving increases in surface water in one area may modify host-vector-parasite contact that affect transmission patterns of vector born diseases. Lambrecht (1982) hypothesized that occupation of tsetse habitat by water bodies may create unfavorable condition for the flies but in the long run, if human settlement and livestock population increases in the area, reinvasion could be possible.

The 2005 retrospective data obtained from Sodo Regional Veterinary Laboratory and the Southern Tsetse and trypanosomosis Eradication Center revealed two species of tsetse flies from Savahana group (*Glossina pallidipes*, *G.m.submorsitans*) and one species from Forest group (*G.fuscipes*) in Loma District. Only *Glossina pallidipes* and *G.fuscipes* were recorded for Kindo Didaye at that

time. On the other hand, this study revealed that while the forest type species, *G. fuscipes* dominated the population of savahana type species were either absent or insignificant in number three years after completion of the dam in 2016 in the same site. On the other hand, in Kindo Didaye, where there was no major ecological disruption, the savahana type, *G. pallidipes* remain the dominant species followed by *G. fuscipes* suggesting that the downstream areas are still favorable for these groups of tsetse flies. Cecchi *et al* (2008) described that the Savahana/morsitans group of tsetse flies prefer savahana grassland with open woodland, plenty of game animals and a climate which is neither very humid nor very dry. Therefore, the change in vegetation from savahana to forest type and the high humidity near the water reservoir may have created unfavorable condition after dam construction. Altogether, the presence of the dam and its reservoir seem to have changed tsetse apparent density in the upstream area and consequently reduced trypanosome prevalence in cattle.

## **5.2. Effect of location on bovine trypanosomosis in reference to Ghibe III dam**

### *5.2.1. Perception of the local community*

Almost all respondents in this study agreed on the occurrence of trypanosomosis and its vector depends on season which is in agreement with (Seyoum *et al.*, 2013; Mekuria *et al.*, 2020). Questionnaire survey response in Loma district indicated that the problem is more prevalent during rainy season than dry season whereas in Kindo Didaye respondents agreed that the problem is more prevalent in dry season and/or in both seasons. However, focus group discussion revealed that trypanosomosis was more important in the rainy season. The abundance of flies in rainy season may predispose the animals to tsetse challenge and higher prevalence of trypanosome infections. In rainy season, despite the presence of adequate feed availability, trypanosomosis prevalence is assumed to be high due to the increased tsetse fly challenge (Seyoum, *et al.*, 2013; Tesfaye, *et al.*, 2012). Furthermore, 44% of the farmers perceived tsetse fly occurrence along river sides and grazing fields as well as wet season are the main cause of trypanosomosis (Machila *et al.* 2003). Rainy season might have contributed to favorable environment for vector growth and multiplication thereby increase tsetse population and increased tsetse challenge for the transmission of trypanosomosis. Similar perception of farmers was observed on signs of trypanosomosis and its transmission in high tsetse challenge area (Gracea, *et al.*, 2009), which was in agreement with current finding. In contrast, a study with

Tanzanian communities revealed that study participants perceived higher trypanosomosis challenge occur in dry season (Kimaro, et al., 2017) which can be explained by the fact that feed shortage and water scarcity may force animals to move down to river side where there is high tsetse fly challenge (Mamoudou et al.2016).

### 5.2.2. Tsetse fly density and infection rate

The main elements of the vectorial capacity are the relative density of vectors to hosts, the survival of vectors, their feeding preferences and their infectious rate (Tran et al., 2005). In this regards, apparent density of tsetse was very low and similar in both dry and rainy seasons in Loma while in Kindo Didaye, the fly catch was higher compared to Loma and this was much more important in the rainy season compared to dry season. This reduction in Loma could be associated to low humidity and high temperature in dry season that may increase tsetse mortality and reduces its dispersal significantly (Leak, 1999). The influences of rainfall and temperature on tsetse ecology have also been mentioned by other studies (Olwoch, et al., 2008). Such problem may be minimal in the upstream Loma District since the permanent water reservoir maintains constant humidity in both dry and rainy seasons. Ecological disturbance and changes in human settlement resulting in bush clearance and change in micro environment as a result of site preparation for dam construction and subsequent replacement by the artificial lake may have reduced domestic and wild ruminant populations as a source of tsetse blood meal and consequently affected tsetse population (Leak, 1999, Van Den Bossche et al. 2010). River with large size, width and slow speed water affect the temperature and humidity of river bank vegetation and hence favour the tsetse and biting flies' (Leak, 1999) as far as the breeding site is not affected unlike water reserve site. Therefore, if fly abundance is to determine trypanosome prevalence, obviously bovine trypanosomosis would be more prevalent in Kindo Didaye and mainly in the rainy season. The largest apparent density of tsetse flies in this study was higher than the findings of Simo et al (2015) in central region of Cameroon, Takele et al (2020) in Jimma Zone, Ethiopia and Adungo et al (2020) in Busia, Kenya. On the contrary, higher catches, 7.7 and 14.97 FTD, were reported by Amante et al (2020) and Teka et al (2012) respectively in different parts of Ethiopia. Such variations may suggest differences in ecology, host abundance, sampling season and tsetse species adapted to specific ecology. In this regards, the dominant species recorded in this study was *G. pallidipes* downstream in Kindo Didaye and *G. fuscipes*

upstream, in Loma district. Ngonyoka et al (2017) reported abundance of different species of *Glossina* among different habitats in Maasai steppe of northern Tanzania; *G. pallidipes* was common in riverine, *G. swynnertoni* in the ecotone habitat and *G. morsitans* in all habitats. Similarly, Salekwa et al (2014) has shown that *G. swynnertoni* was the most abundant species followed by *G. morsitans submorsitans* and *G. pallidipes* in Northern Tanzania. A study by Batu et al (2017) in West Wellega has also identified *G. m. submorsitans* and *G. tachinoides*; the latter being the most dominant species.

In this study, tsetse has also been captured from different locations away from the fringes of the Omo River and the dam's water reservoir. It was noticed that tsetse were almost absent in both dry and wet season from areas closer to the dam reservoir (up to 900m) and appear to have been displaced to relatively distant locations (1100-1700 metres) suggesting that the ecological conditions including reduced host availability near the dam reservoir has become uncomfortable for them to survive. This supports the idea that, whenever the need arises, tsetse can disperse 2-3kms from their original location (Vreysen et al., 2013b). On the other hand, tsetse density was much higher around the Omo river (where the ecology is relatively undisturbed) downstream dam for a distance of 300m and 1500m during dry and rainy seasons, respectively. This strongly supports the higher tsetse density in this part of the study location and hence, higher trypanosomosis prevalence may be expected. The seasonal abundance of tsetse downstream the dam is in line with the report of Happiness et al. (2017) from Tanzania.

The other factor in the tsetse vector competency is the fly infection rate which determines the probability of transmission to cattle of cyclically transmitted trypanosome species. The overall tsetse infection rate recorded in this study was in line with some of the reports in systematic review by Duguma (2016). Relatively lower infection rates were reported by two studies in Ethiopia by Meharenet and Alemu (2020) in *G. tachinoides* (1.76%) in Limu Kosa District of Jimma Zone and by Desta et al (2013) from *G. pallidipes* (6.93) in Ammaro Special Zone of the Southern Region compared to the current findings with an overall infection rates of 14.8% in Loma and 17.8% in Kindo Didaye Districts. Similarly, different infection rates have been reported from different countries in Africa (Adungo et al., 2020; Bouyer et al., 2013; Salekwa et al., 2014; Simo et al., 2015). Bouyer et al (2013) also reported that fly infection rate was higher during the rainy season in Burkina Faso. According to a study in Northern Tanzania by

Ngonyoka et al. (2017), infection rates by trypanosome species showed spatial variation among habitats and villages; ecotone (a transitional area of vegetation between two different plant communities) 21.1%, open woodland 9.0%, riverine 6.8% and swampy habitat 7.7%. Altogether, this suggests that, both tsetse apparent density and infection rate are detected by biotic and abiotic factors prevailing in each specific location.

### *5.2.3. Trypanosome prevalence and species composition*

The current study unequivocally revealed that the prevalence of bovine trypanosomosis was much lower in Loma/upstream when compared to Kindo Didaye/downstream. This perfectly goes in line with the trend in livestock population in the two areas. The decline in cattle number downstream the dam could be attributed to the higher impact of trypanosomosis and other diseases in the area whereas the increase in herd size in Loma is associated with the improvement in disease situation due to the reduction in tsetse population arising from biotic and abiotic changes that have occurred following dam construction. Adam et al (2012) concluded that low prevalence of trypanosomosis is associated with patchy tsetse density in Ghanaian river side. Similarly, Yewhalaw et al. (2010) and Lambrecht (1982) concluded that an increase in vector fly population in Gilgel Ghibe-I results in upsurge of malaria infection in humans near dam construction. Hence, high tsetse apparent density coupled with low cattle herd size in downstream might have contributed for repeated tsetse challenge and high trypanosomosis prevalence; whereas low tsetse apparent density and high cattle herd size might have contributed for reduction trypanosomosis prevalence.

Prevalence of bovine trypanosomosis was much higher in rainy season than in dry season in both study districts suggesting that season equally affects disease transmission in both study locations. Although tsetse apparent density was not changed with season in Loma district, the increase apparent density and small herd size in Kindo Didaye may partly explain for the higher prevalence of trypanosomosis in this area. In addition, biting flies increased significantly during rainy season might be additional reason for high prevalence in rainy season. Similar reports suggesting the role of rainy season in trypanosomosis prevalence have been documented (Cherenet et al., 2006; Dagnachew, et al., 2005; Majekodunmi et al., 2013, Kimaro et al., 2018). On the contrary, infection rate in dry season was reported higher than rainy season, mainly for

*T.vivax* (Woolhouse *et al.*, 1993) while such seasonality was not observed by Djohan *et al.* (2015).

Three species of trypanosome identified during study period, where *Trypanosome congolense* was the predominant species followed by *T.vivax* and *T.brucei*. This finding was comparable with the reports of Duguma *et al.*, (2015) and other studies in Ethiopia ( Cherenet *et al.*, 2006; Mekuria and Gadissa, 2011; Moti *et al.*, 2012; Duguma *et al.*, 2016; Sheferaw, *et al.*, 2016; Abebe *et al.*, 2017). However, *T. brucei* was absent in Loma study site for a reason yet to be investigated.

Biotic risk factor like poor body condition score (BCS) animals had more trypanosome infection than good BCS ones, though there are different factors contributing for poor body condition. Major clinical signs of trypanosome infected animals are loss of weight accompanied with lymph node enlargement, rough coat hair is in agreement with (Peter, 2013; Radostits, *et al.*, 2007). Further more, anemic condition is a typical pathological sign of bovine trypanosomosis, and considered when PCV of an animal is less than 24 (Latimer, 2011). Accordingly, infected group of animals had mean PCV of 22.97. Cattle in dry season and cattle from Kindo Didaye had mean PCV of 22.9 and 23.21, respectively. Despite low trypanosome prevalence in dry season mean PCV value was low as compared to rainy season. This might be due to feed scarcity during dry season, other parasitic infection and/or chronic trypanosome case with low parasitemia that usually considered as negative in BCT might have contributed. Kindo Didaye district with high trypanosome infection showed low mean PCV than Loma district. The finding was in agreement with other studies in Ethiopia and abroad (Marcotty, *et al.*, 2008; Mekuria and Gadissa, 2011; Abebe, *et al.*, 2017; Sheferaw, *et al.*, 2016, Mamoudou *et al.*, 2016) this might be associated with trypanosome infection, where significantly ( $p<0.05$ ) low mean PCV recorded in trypanosomosis infected group than non infected group. In this study, significant mean PCV reductions was observed in all species of trypanosome infection identified, and were equally important in causing anemia in cattle(Abebe, *et al.*, 2017).

Variation in the prevalence of trypanosomosis was observed among study sites which ranged from 2.6% prevalence at close proximity of created Lake to 15% at close proximity of Omo River. Similar village variation was observed in Tanzania (Kimaro *et al.*, 2018) and in Nigeria (Majekodunmi *et al.*, 2013). Other studies have indicated also the abundance of trypanosomosis

infection is influenced by spatial factors such as altitude, River drainage tributaries, presence of game and land use and human encroachment (Duguma, 2016). Furthermore, studies showed that vector borne diseases can be affected by anthropogenic and environmental changes that alter the interaction between the host, the vectors and the parasites in a given area (Van den Bossche et al., 2010, Imna, et al., 2011). Patz et al. (2000) and Norris Douglas (2004) have already shown that environmental changes and land use patterns can significantly affect vectors of parasites.

## 6. CONCLUSION AND RECOMMENDATION

This study was primarily initiated to assess the situation of tsetse and trypanosomosis in upstream (Loma District) and downstream (Kindo Didaye District) of Ghibe III hydroelectric dam area. When data are available, spatial and temporal factors are considered in the assessment of the perception of farmers on the disease situation, the prevalence of trypanosomosis, tsetse apparent density and infection rates.

Local communities in the two study areas have adequately described the livestock population structure and their diseases. Accordingly, cattle herd size greater or equal to six have significantly increased after dam construction in Loma district, whereas in Kindo Didaye farmers with such cattle herd size have declined. Bovine trypanosomosis was ranked as number one disease priority in both Loma (upstream) and Kindo Didaye (downstream) Districts. The strong concordance observed between diseases and clinical symptoms listed by the community ensures the presence of rich indigenous knowledge in the community. Majority of respondent from Loma/upstream district reported that the impact of the disease and its vectors has significantly reduced after Ghibe III dam construction while participants from Kindo Didaye district reported no change in disease problem in relation to dam construction.

Retrospective data for the period before dam construction revealed that there was no difference between the two study sites in the prevalence of bovine trypanosomosis and tsetse fly density. Now, it appears that bovine trypanosomosis and its tsetse vector prevalence and density have been significantly reduced upstream in Loma District following dam construction whereas these still remained important livestock health constraints in Kindo Didaye study site despite some reductions downstream the dam. Tsetse density was more severe close to the fringe of Omo River in Kindo Didaye whereas it is more important as we go away from the fringe of the dam reservoir in Loma. These all have in one way or another unequivocally corroborated the observations of the local community. The ecological changes arising from dam construction seem to have also changed Glossina species composition. Originally, *G. fuscipes*, *G. pallidipes* and *G. m.submorsitans* were reported for Loma and the first two for Kindo Didaye while the current study was not able to detect *G. m. submorsitans* in the upstream, Loma District. Data for tsetse infection rate was not available for the period before dam construction. However, the current study has clearly shown that despite the significant difference in tsetse fly apparent

density, tsetse infection rates were almost similar between the two study sites. Therefore, any difference in trypanosomosis prevalence in cattle may be attributed to fly availability and other factors. Seasonality of fly abundance was observed only at Kindo Didaye while fly infection rate was not affected by season in both study sites.

The current study also clearly demonstrated that the prevalence of bovine trypanosomosis was much lower in Loma/upstream when compared to Kindo Didaye/downstream, which perfectly aligns with the trend in cattle population in the two areas, the observations of the local community and the difference in tsetse density between the two study locations. It has also shown that cattle infection rate has significantly declined (three fold) after construction of the Ghibe III dam in the upstream Loma site and hence herd size was inversely proportional to trypanosomosis prevalence and tsetse apparent density. The most prevalent trypanosome species were *T. congolense* and *T. vivax* while few cases of *T. brucei* were seen in Kindo Didaye. In general, although bovine trypanosomosis is still a major problem in the study areas, Ghibe III dam construction has produced a favorable outcome in areas close to the water reservoir probably by disrupting tsetse ecology and consequently reducing trypanosomosis prevalence, However, the problem is still rampant in areas downstream the dam and in places far away from the dam water reservoir.

Based on the above conclusion, the following recommendations are forwarded:

- Rehabilitation of the ecosystem around Ghibe III dam should take into account in risk mitigation mechanisms as reinvasion of tsetse flies is a possibility in the upstream locations
- Strengthen Veterinary Services and tsetse and trypanosomosis control programs at Kindo Didaye by giving more emphasis than Loma, however Loma should not be overlooked.
- Further study is required to show year round situation of tsetse and trypanosomosis in the area
- Further study is needed to elucidate the socioeconomic gains from the reduction in tsetse and trypanosomosis challenge in Loma district compared to Kindo Didaye
- Integrated control measures need to be employed in pocket area of Loma to eliminate the remnant of tsetse flies.

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**ANNEXES**

**Annex I: KAP Questionnaire Survey on Animal Trypanosomosis**

Date: \_\_\_ / \_\_\_ / \_\_\_ G .C

Location: Region: \_\_\_\_\_ Wereda: \_\_\_\_\_ Kebele: \_\_\_\_\_

Site: \_\_\_\_\_ code: \_\_\_\_\_

1. How old are you? \_\_\_\_\_ Gender: \_\_\_\_\_

2. How long you have been here? \_\_\_\_\_

3. What is your primary occupation? \_\_\_\_\_

4. If it is agriculture/livestock, how many livestock do you have?

Currently: cattle \_\_\_\_\_ goats \_\_\_\_\_ sheep \_\_\_\_\_ poultry \_\_\_\_\_ equine \_\_\_\_\_

Before 2005:

cattle \_\_\_\_\_ goats \_\_\_\_\_ sheep \_\_\_\_\_ poultry \_\_\_\_\_ equine \_\_\_\_\_

5. What are the most five major livestock diseases in your area?

Currently: 1. \_\_\_\_\_ 2. \_\_\_\_\_ 3. \_\_\_\_\_ 4. \_\_\_\_\_ 5. \_\_\_\_\_

Before 2005

1. \_\_\_\_\_ 2. \_\_\_\_\_ 3. \_\_\_\_\_ 4. \_\_\_\_\_ 5. \_\_\_\_\_

6. If trypanosomosis is among the disease, tick from the lists of clinical signs

Emaciation  rough coat hair

Swollen lymphnode  diarrhea

Fever and dry muzzle  coughing and salivation

7. Do you know how trypanosomosis is transmitted? tick from the list

direct contact  biting fly

feed  tsetse fly

water  others (specify)

8. Is there seasonal difference in the occurrence of trypanosomosis? Yes/No

9. Which season of the year more prevalent? Dry/ rainy/ others

10. Why prevalent in the above mentioned season?

11. Which species of animals are more susceptible to trypanosomosis? Rank them accordingly.

1. \_\_\_\_\_ 2. \_\_\_\_\_ 3. \_\_\_\_\_ 4. \_\_\_\_\_ 5. \_\_\_\_\_

12. Is there change/reduction currently in the prevalence as compared to past years? Yes/No

13. If yes, what is the reason?

\_\_\_\_\_

\_\_\_\_\_

14. Do you think the Dam constriction contributed for reduction of typanosomosis & tsetse?

Yes/No

15. Is there any change in wildlife population? Yes/No

If yes, which wild animals increased list them?

Which wild animals decreased list them?

16. What is the common control practice to reduce trypanosomosis in your village? Tick

Trypanocidal drugs Rx  Other(specify)

Insecticide spray

Cattle movement

17. Where is the source of drugs/insecticide you are using?

Public \_\_\_\_\_ Private \_\_\_\_\_ other(specify) \_\_\_\_\_

18. Which source of drug provide regularly if not what is your alternatives?

19. Is it cheaper? Yes/ No

If no, what is the reason?

20. How much does it costs?

Drugs per animal per treatment \_\_\_\_\_, how often per month? \_\_\_\_\_

Insecticide per animal per treatment \_\_\_\_\_, how often per month? \_\_\_\_\_

21. What are the most commonly available trypanocidal drugs in your area?

1. \_\_\_\_\_ 2. \_\_\_\_\_ 3. \_\_\_\_\_ 4. \_\_\_\_\_ 5. \_\_\_\_\_

Thank you

**Annex II. Record format for trypanosomosis and tsetse survey**

		Gps							wet	tryps
s/n	Village	lat	long	sex	Age	colour	BSC	PCV	film	spp

Fly collection and identification record sheet

			Gps				total	sex	
s/n	Village	Distance from fringe	lat	long	Trap	Spp fly	count	M	F

Record sheet for infection rate

s/n	village	spp	Sex	Result	Probos.	midgut	Salivary gland

### Annex III: Age Determination Guide

TABLE 13.2 : AVERAGE AGE AT THE ERUPTION, WEAR AND TEAR OF TEMPORARY AND PERMANENT TEETH IN MURRAH TYPE BUFFALOES, CROSSBRED CATTLE, GOATS AND SHEEP (Saini, *et al.* 1993)

<i>Dentition</i>	<i>Buffaloes</i>	<i>Crossbred cattle</i>	<i>Goats</i>	<i>Sheep</i>
		AGE OF ERUPTION		
<b>INCISORS</b>				
(a) <i>Temporary</i>				
First pair	First week	At birth to first week	At birth to first week	First week
Second pair	Second week	At birth to first week	At birth to first week	First week
Third pair	1.5 months	First week	First week	Second week
Fourth pair	4 months		Third to fourth week	Fourth week
(b) <i>Permanent</i>				
First pair	34.0 months	27.0 months	17.0 months	18.0 months
Second pair	42.0 months	33.0 months	21.5 months	25.0 months
Third pair	52.0 months	40.0 months	26.0 months	31.5 months
Fourth pair	60.0 months	48.0 months	30.0 months	35.0 months
<b>A. PRE-MOLARS</b>				
(a) <i>Temporary</i>				
First pair	Before one month of age	Before one month	First to second week	Second week
Second pair	Before one month	Before one month	First to second week	Second week
Third pair	Before one month	Before one month	Second to third week	Third week
(b) <i>Permanent</i>				
First pair	30.0 months	23.0 months	14.0 months	15.0 months
Second pair	36.0 months	27.0 months	17.0 months	18.0 months
Third pair	42.0 months	33.0 months	21.0 months	25.0 months
<b>B. MOLARS</b>				
First pair	9.0 months	6.0 months	Fourth week	Fourth week
Second pair	24.0 months	18.0 months	2.0 months	2.5 months
Third pair	36.0 months	27.0 months	10.0 to 11.0 months	12.0 months
<b>WEAR AND TEAR</b>				
Just started	7.0-8.0 years	6.0-7.0 months	3.0-3 years	4-5 years
Table of tooth levelled	9.0-10.0 years	8.0-9.0 years	4.0-5 years	6-7 years
Reduced in size	12.0-14.0 years	More than 10.0 years	6.0 to 7.0 years	8-9 years
Reduced to stubs/ broken/absent	More than 15.0 years	More than 13.0 years	More than 8 years	More than 10 years

**Annex IV: Critical Values of the Mann-Whitney U test**  
(Two-Tailed Testing)

n <sub>2</sub>	α	n <sub>1</sub>																	
3	.05	--	0	0	1	1	2	2	3	3	4	4	5	5	6	6	7	7	
	.01	--	0	0	0	0	0	0	0	0	1	1	1	2	2	2	2	3	
4	.05	--	0	1	2	3	4	4	5	6	7	8	9	10	11	11	12	13	
	.01	--	--	0	0	0	1	1	2	2	3	3	4	5	5	6	6	7	
5	.05	0	1	2	3	5	6	7	8	9	11	12	13	14	15	17	18	19	
	.01	--	--	0	1	1	2	3	4	5	6	7	7	8	9	10	11	12	
6	.05	1	2	3	5	6	8	10	11	13	14	16	17	19	21	22	24	25	
	.01	--	0	1	2	3	4	5	6	7	9	10	11	12	13	15	16	17	
7	.05	1	3	5	6	8	10	12	14	16	18	20	22	24	26	28	30	32	
	.01	--	0	1	3	4	6	7	9	10	12	13	15	16	18	19	21	22	
8	.05	2	4	6	8	10	13	15	17	19	22	24	26	29	31	34	36	38	
	.01	--	1	2	4	6	7	9	11	13	15	17	18	20	22	24	26	28	
9	.05	2	4	7	10	12	15	17	20	23	26	28	31	34	37	39	42	45	
	.01	0	1	3	5	7	9	11	13	16	18	20	22	24	27	29	31	33	
10	.05	3	5	8	11	14	17	20	23	26	29	33	36	39	42	45	48	52	
	.01	0	2	4	6	9	11	13	16	18	21	24	26	29	31	34	37	39	

## **APPENDEXES**

**Appendix 1-2. Article published**

**Appendix 3: Curriculum vitae of the author of this manuscript**