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ASSESSMENT ON THE EFFECT OF DELTHAMETHRINE 1% POURS ON AT DIFFERENT RIGIME IN CONTROL OF TSETSE FLY AND TRYPANOSOMOSIS IN TWO DISTRECT OF GAMO ZONE, SOUTHERN ETHIOPIA

MSc THESIS

BY:

MULUKEN TEMESGEN SHIFERAW

ARBAMINCH UNVIRESTY
JUNE, 2020

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A THESIS SUBMITTED TO THE DEPARTMENT OF ANIMAL SCIENCE, COLLEGE OF AGRICULTURE, SCHOOL OF GRADUATE STUDIES, ARBAMINCH UNIVERSITY IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN VETERINARY EPIDEMIOLOGY

ARBMINCH UNVIRESTY
JUNE, 2020

DECLARATION

I hereby declare that this MSc thesis is my original work and has not been presented for a degree in any other university, and all sources of material used for this thesis have been dully acknowledged.

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I

LIST OF ABERBATION

AAT	Africa Animal Trypanosomosis
BCS	Body Condition Score
CSA	Central Statics Agency
DG	Dark Ground
DNA	Deoxyribose Nucleic Acid
F/T/D	Fly per Trap per Day
FEAV	Facultative Ectoparasites and Arthropods Vectors
OAU	Organization for African Unity
OIE	Office International des, Epizooties
PCV	packed Cell Volume
SAT	Sequential Aerial Technique
SIT	Sterile Insect Technique
SSA	Sub-Saharan Africa
WHO	World Health Organization
WOAH	World Organization of Animal Health

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Assessment on the effect of Delthamethrine 1% pours on at different rigime in control of tsetse fly and Trypanosomosis in two distrect of Gamo Zone, Southern Ethiopia

By: Muluken Temesgen Shiferaw *Dr Minale Gethachew * Aschenaki Kalsa

ABSTRACT

A study aimed at assessing the temporal effect of Deltamethrin 1% in controlling tsetse was carried out in two Kebel of Gamo zone south rift valley from October 2019 to April 2020. The objective of the study was to assess the temporal effect of Deltamethrin 1% in controlling tsetse. The strategy followed to accomplish the study was by implementing pre-intervention phase (entomology and parasitology) and intervention phase by applying insecticide (Deltamethrin 1%) pour-on' on cattle at a rate of 1 ml/10 kg body weight on quarterly in Fura Kebel and monthly 'in Kanchama Kebel. During The intervention phase hematological and entomological results in each Kebele was monitored on a monthly basis. The results indicated that in Fura Kebel the relative abundance of tsetse fly (Glossina pallidipes) there was no significant effect of Deltamethrin were seen p (0.870) in pre-intervention and last monitoring time (2.1 flies per trap per day to 1.73 flies per trap per day) and there was no significant effect of delthametrin were seen p (0.385) in the incidence of Trypanosomosis during first and last monitoring time (5.5% first monitoring) to 4.7% last monitoring. In Kanchama Keble there was significant effect of Deltamethrin were seen p (0.000) in the relative abundance of tsetse flies revealed during pre-intervention and last monitoring time (17.73 flies per trap per day to 8.867)).There was also significant effect of Deltamethrin were seen p (0.006) in the incidence of Trypanosomosis in cattle's also declined from first monitoring (9.6%) to last monitoring (3.2%). This work finally disclosed that monthly application of Deltamethrin 1% has effect in controlling tsetse and Trypanosomiosis. Wet and dry season has no significant effect on controlling tsetse flies, however further research should be required to give concrete recommendation.

***Key words*:** *Delthamethrinv1%; Fura, Kanchama, pour-on, Trypanosomosis; Tsetse fly,*

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1. INTRODUCTION

1.1. Background

According to different research work Ethiopia is known for its large and diverse livestock resource endowments, livestock is primarily kept on small holdings where it provide drought power for crop production, manure for soil fertility and fuels, serves as a sources family diet and sources of cash income (from livestock and livestock products, despite large livestock population, Ethiopia fails to optimally utilize this resource due to different constrains facing the livestock subsector (Abebe *et al*., 2004). Since more than 90% of crop production in Ethiopia are dependent on animal draught power mainly on ploughing oxen, many large fields lie fallow due to lack of these animals in trypanosomiasis infested area (Kenaw *et al,* 2015), Bovine *Trypanosoma* is one of the diseases that are caused by this flagellated protozoal parasite belonging to the genus trypanosome (Jember *et al.,* 2013). Which has long been recognized as a massive constraint on animal husbandry, livestock production and mixed farming in vast areas of rural sub-Saharan and worsen the food supply and living conditions in affected areas Africa (Oluwafemi, 2014).This group of diseases caused by protozoa of the genus trypanosoma affects all domestic animals. (Kenaw *et al,* 2015).

The major veterinary important species are *Trypanosoma congolense, Trypanosoma vivax, Trypanosoma brucei brucei, Trypanosoma equperdium, Trypanosoma evansi and Trypanosoma simiae. The only zoonotic important Trypanosoma are Trypanosoma brucei rhodesiense and Trypanosoma brucei gambiense*, with people as the predominant host (OIE, 2013). The transmission of infection is during blood feeding by trypanosome infected tsetse fly (*Glossina* spp), initial replication of trypanosomes happens at the site of inoculation in the skin; this causes a swelling and a sore (chancer), then spread to lymph nodes and blood continue to replicate. *T. congolense* localizes in small blood vessels and capillaries. *T. brucei brucei* and *T. vivax* localize also in tissue. Wild animals can be infected with pathogenic trypanosomes and many of these remain carriers of the organisms, Cattle, sheep, goats, pigs, horses, camels, dogs, cats and monkeys are susceptible to Africa Animal Trypanosomosis(AAT) (WOAH, 2012).

Tsetse transmitted African Trypanosomosis is found between latitude 15°N and 29°S covering across over 37 countries in Africa from the southern edge of the Sahara desert to

Zimbabwe, Angola and Mozambique (OIE, 2009). African Trypanosomosis can be found wherever the tsetse fly vector exists.*T. vivax* can spread beyond the "tsetse fly belt" by transmission through mechanical vectors (WOAH, 2012). Tsetse flies *(Glossina* spp.) are found only in Africa; they are the biological vector of trypanosomes and constitute a potent and constant threat to humans and livestock over much of sub-Saharan Africa (Bouyer, 2006). Tsetse flies are robust, 6–15 mm in length, and can be distinguished from other biting flies by their forward-pointing mouthparts (proboscis) and characteristic wing venation (Bouyer, 2006).

Tsetse and Trypanosomosis control are a series of efforts employed to reduce or eliminate the incidence of African Trypanosomosis, transmitted by tsetse flies, two complementary strategies have been used in the attempts to reduce or eliminate the disease, the strategies are chemotherapeutic, targeted at reducing the incidence of Trypanosomosis in livestock and entomological, aimed at disrupting the cycle of transmission of the disease by reducing or eliminating the vector of Trypanosomosis (Bouyer *et al*., 2010). The problem has always been a relapse after any control effort due to the complex mitigating factors such as rural poverty, civil strives and the lack of a concerted effort to sustain the individual national control efforts (Kgori *et al*, 2006). Despite this situation in Africa, examples of successful tsetse elimination efforts can be sighted in Botswana (Kgori *et al.*, 2006), Namibia and the Islands of Zanzibar (Vreysen *et al*., 2000).

Tsetse control techniques have gone through evolutions, with earlier crude methods recently being replaced by methods that are economically cheaper, more directed, and ecologically sustainable, these techniques include: environmental management (bush clearing and game destruction), pesticide campaigns, traps/targets, animal baits and the use of Sterile Insect Technology (SIT) (Sow, 2013). To embark on a national tsetse intervention programme, a number of pre-control operations are undertaken to ensure a well monitored and successful control programme.

1.2. Statement of the Problem

In the study area Different tsetse flies control measures and *trpanosoma* treatments have been implemented for more than two decades to control this disease chemically by prophylactic treatment using isometamidium bromide (veredium). Clinically sick animals are treated using diminazene aceturate and, to control tsetse flies the available used

technology is mainly pour on Deltamethrin 1% application on cattle's within three months intervals and deploying target. Despite implementing the above control measures, the tsetse flies density and disease problem was still reported higher and which results reduction of production and productivity of the livestock sector.

The most striking direct effects of the disease are observed in the field of milk production, calving rate and draught animals. Apart from these direct economic effects of the disease, there were also considerable indirect losses due to unbalanced land use and settlement patterns, difficulties to integrate livestock breeding and agriculture, reduced use of draught power and massive expenditures on trypanocidal drugs in tsetse infested areas. Therefore, the aforementioned crucial problems in the study area initiated to conduct this study using alternative Deltamethrin 1% application method to control tsetse flies which were indispensable vectors of trypanosomes.

1.3. Objectives

1.3.1. General objective of this study was to investigate the effect of different application regime of Deltamethrin 1% treated cattle on control of tsetse fly and Trypanosomosis.

1.3.2. Specific objectives:

- To assess the effect of monthly Deltamethrin 1% application on the control tsetse flies density.
- To assess the effect of quarterly Deltamethrin 1% application on the control tsetse flies density
- To assess the seasonal effects on Deltamethrin 1% application to control of tsetse and of Trypanosomosis.

1.4 .Research Questions

What is the effect of monthly Deltamethrin 1% application in control of Tsetse fly?

. What is the effect of quarterly Deltamethrin 1% application in control of Tsetse fly?

What is the effect of season on controlling of Tsetse Fly by pour on Deltamethrin 1%?

1.6. Significance of the Study

The study helps in increasing productivity of all farmers' cattle in study area by decreasing flies effect. It also helps national institute of tsetse and Trypanosomosis

control and eradication institutes to select the effective and cost benefit Deltamethrin application regime. Based on the result of the study concerned bodies (government, Nongovernmental organization) used for decision making, strategy development. This study helps as source of information for the researcher works.

1.7. **Delimitation**

It was good to conducted the present study in wide study area, but because of time and financial constraint the study was conducted in Fura and Kanchama kebeles from October 2019- April2020 for seven month.

1.8. Operational Definition

- The packed cell volume (PCV) is measurement of proportion of blood that is made of cell. The value is expressed a percentage or fraction of red cells in the blood.
- Pour-on is application of insecticide along the line starting from in front of the shoulder running back to behind the hip
- .Tsetse fly they are the biological and/or mechanical vector of trypanosomes and can be distinguished from other biting flies by their forward-pointing (proboscis) mouthparts and characteristic wing venation.

2. LITERATURE REVIEW

2.1. Bovine Trypanosomosis

According to different researchers indicates bovine *Trypanosomosis* is a disease that affects cattle resulting from infection with protozoa of the genus *Trypanosoma* transmitted primarily by tsetse fly and also by other haematophagous fly (Urquart *et al.,* 1995). *T. vivax, T. congoiense, T. brucei, T.equperdium* and *T. simiae* are the four main species responsible for African trypanosomoses affecting virtually all domestic mammals. *T. vivax and T. congoiense* are the main pathogens of cattle (Radostitis *et al*., 2007). African animal Trypanosomosis is a disease complex caused by tsetsefly transmitted*T. congolense, T. vivax, or T. brucei brucei,* or simultaneous infection with one or more of these trypanosomes (Radostitis *et al*., 2007). Trypanosomosis depends on the distribution of the vectors, the virulence of parasite and the response of the host (Langridge, 1976). In southern Africa the disease is widely known as Nagana, which is derived from a Zulu term meaning “powerless/useless" (Vreysen, 2001). Also in Ethiopia widely as known Ginde(Jember *et al.,* 2013).

2.1.1. Etiology

Animal Trypanosomosis is caused by protozoa in the family Trypanosomatidae genus *Trypanosoma* (FAO, 2006). *T.congolense* resides in the subgenus *Nannomonas,*a group of small trypanosomes with medium-sized marginal kinetoplasts, no free flagella, and poorly developed undulating membranes, *T. vivax* a member of the subgenus *Duttonella*, a group of trypanosomes with large terminal kinetoplasts, distinct free flagella, and inconspicuous undulating membranes, *T. vivax* is a large (18-26 μm) monomorphic organism that is very active in wet-mount blood smears, *T. brucei brucei* resides in the subgenus *Trypanozoon*. *T. b. brucei* is an extremely polymorphic trypanosome occurring as short stumpy organisms without flagella, long slender organisms with distinct flagella, and intermediate forms that are usually flagellated (FAO, 2006).

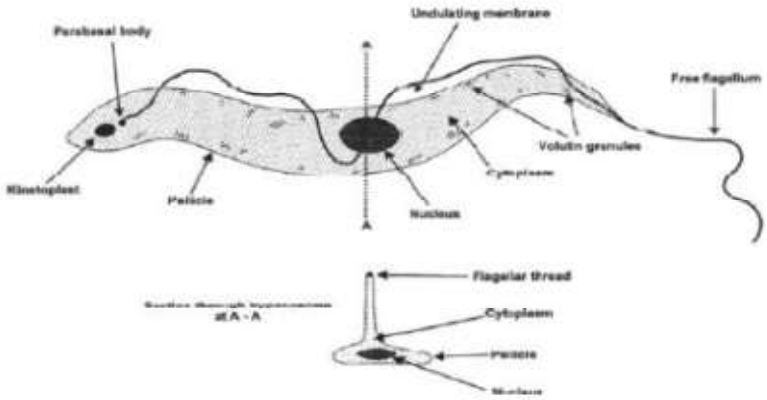

Figure 1 Diagram of trypanosomis showing fundamental morphology features

Source (FAO, 2006)

2.1.2. Epidemiology of Trypanosomosis

The epidemiology of African Trypanosomosis is determined mainly by the ecology of the tsetse fly which is found only in tropical Africa (Radostitis *et al.*, 2007). Most tsetse-fly transmission is cyclic and begins when blood from a trypanosome-infected animal is ingested by the fly, the trypanosome alters its surface coat, multiplies in the fly, then alters its surface coat again, and becomes infective (Merck, 2005). Tsetse flies (genus *Glossina*) are restricted to Africa from about latitude 15° N to 29° S, the three main species that inhabit relatively distinct environments are: *G. m. submorsitans* usually found in savanna country, *G. p. palpalis* prefers areas around rivers and lakes, and *G. fusca* lives in high forest areas, all three species transmit trypanosomes, and all feed on various mammals (Merck, 2005).

2.1.3. Pathogenesis and clinical signs

Initial replication of trypanosomes happens at the site of inoculation in the skin; this causes a swelling and a sore (chancer), then spread to lymph nodes and blood continue to replicate. *T. congolense* localizes in small blood vessels and capillaries. *T. brucei brucei* and *T. vivax* localize also in tissue. Antibodies developed to the glycoprotein coat of the trypanosome lyse the trypanosome and result in the development of immune complexes, however, does not clear the infection, for the trypanosome has genes that can code for many different surface-coat glycoprotein's and changes its surface glycoprotein to evade

the antibody, as result there is a persistent infection that results in continuing cycle of trypanosome replication (Luckins, 1992).

Marked immune suppression lowers the host's resistance to other infections and thus results in secondary disease, which greatly complicates both the clinical and pathological features of Trypanosomosis (Aderem and Underhill, 1999). The breed of affected animals and the dose and virulence of the infecting trypanosome, Stress, such as poor nutrition or concurrent disease, plays a prominent role in the disease process (Taylor and Authie, 2004). The cardial clinical sign observed in AAT is anaemia, with a week of infection with the haematic *trypanosomes* (*T. congolence* and *T. vivax*) there is usually a pronounced decrease in packed cell volume (PCV), haemoglobin and red blood cells and within 2-3 months the PCV may drop to below 24% of their preinfecion values (Taylor and Authie, 2004).Also intermittent fever, oedema, loss of condition and abortion in cow and infertility of males (Taylor and Authie, 2004).

2.1.4. Host range

Cattle, sheep, goats, pigs, horses, camels, dogs, cats and monkeys are susceptible to AAT. More than 30 species of wild animals can be infected with pathogenic trypanosomes and many of these remain carriers of the organisms this include buffalo, deer, wild Equidae, lions, leopards and wild pigs (Leak, 1999).

2.1.5. Geographical distribution

African Trypanosomosis can be found wherever the tsetse fly vector exists, *T. vivax* can spread beyond the "tsetse fly belt" by transmission through mechanical vectors, Tsetse transmitted African Trypanosomosis is found between latitude 15°N and 29°S covering across over 37 countries in Africa from the southern edge of the Sahara desert to Zimbabwe, Angola and Mozambique (OIE, 2009).It is the most economically important livestock disease of Africa, especially of cattle (WOAH, 2012).

2.1.6. Transmission

In Africa, the primary vector for *T. congolense, T. vivax and T. b. brucei is* the tsetse fly, these trypanosomes replicate in the tsetse fly and are transmitted through tsetse fly saliva when the fly feeds on an animal, Trypanosomosis (*T. vivax)* is also mechanically transmitted by other biting flies the most important mechanical vectors are flies of the

genus *Tabanus,, Liperosia, Stomoxys* and *Chrysops* flies have also been implicated (OIE, 2009).

2.1.7. Vector

Tsetse flies *(Glossina* spp.) are found only in Africa. They are the biological and/or mechanical vector of trypanosomes and constitute a potent and constant threat to humans and livestock over much of sub-Saharan Africa, (Bouyer, 2006,). Tsetse-fly, in general adult tsetse, are narrow, yellow to dark brown flies (Veterinary Entomology, 2015) 6 to 15 mm in length and have along, rigid, forward projection proboscis, characteristic wing venation the thorax is a dull greenish brown color and is marked with inconspicuous stripes and spots (Bouyer, 2006, FEAV, 2015),

There are about 23 known species of tsetse flies can be divided into three groups, each with different habits and requirements. *G. palpalis* group are riverine species which feed primarily reptiles and ungulates, flies of the *G. morsitans* group are savannah and dry thorn-bush species which is mainly on large animals, members of *G. fusca* group occur in rainforest, preferring dense shade and riverine thickets (FEAV, 2015).

Life cycle of both male and female flies suck blood and although the various species of tsetse may have some host preferences, generally they will freedom a wide variety of animals (Urqhart *et al*., 1995). The puparial period can range from 20 days (at 30°C) to 47 days (at 20°C) (on average 30 days at 24°C). Development in the puparium is generally unsuccessful below about 17°C and above about 32°C. The entire life cycle from egg to adult usually takes about 30 days , the historical classification of tsetse, based on habits and requirements, divides the species into three groups, the genus is further subdivided into well marked species groups (subgenera) identified based on differences from ecological characteristics (Leak, 1999).

1. The *moristans* group (savanna tsetse): this class occupies the savanna land of tsetse belt, and includes *G.m.moristans, G.pallidipes, G.austeni, and G.swynertoni.*

2. The *palpalis* group (riverine tsetse): this group invades river and lakeshores, and encompasses *G. fuscipes, G.palpalis, G.tachinoides* and their subspecies.

3. The *fusca* group (forest tsetse): this forest parts of tsetse belt, and includes *G.fusca, G.brevipalpalis, G.longipennis*.

2.2. Diagnosis of Trypanosomosis

2.2.1. Clinical findings and lesions

The general clinical picture is as follows but there are many variation determined by the level of tsetse fly challenge, the species and strain of the trypanosome and the breed and management of the host (Radosititis *et al.,* 2007). Severity of disease varies with species; age of animals infected and the species of trypanosome involved, the incubation period is usually 1 to 4 week (Merck, 2005).

The primary clinical signs are intermittent fever, anemia and weight loss, cattle usually have achronic course with high mortality, especially if there is poor nutrition or other stress factors(Merck, 2005). The anemia results in a progressive drop in packed cell volume it is useful indicator in endemic areas (Radostitis *et al*., 2007). Necropsy findings vary and nonspecific in acute and fatal cases, extensive petechication of the serosal membranes specially the peritoneal cavity, also the lymph nodes and spleen are usally swollen (Merck, 2005).definitive diagnosis of the disease is ultimately dependent on the detection of the trypanosome in blood samples from infected animals (Abebe, 2005, Mercky, 2005).

2.2.2. Diagnostic methods

The diagnosis of trypanosome infection is based on clinical signs and on the demonstration of the parasites by direct or indirect methods, the clinical signs of the AAT are indicative but are not sufficiently pathognomonic and diagnosis must be confirmed by laboratory methods (Abebe *et al*., 2004).

The classical direct parasitological methods for the diagnosis of Trypanosomosis, namely microscopic examination of blood or lymph nodes, by wet mount of blood films and haematocri centrifugation technique are not highly sensitive, but anumber of techniques including enrichment of the sample, rodent inoculation and molecular methods may increase the sensitivity (Murray *et al*., 1982). Blood sample was collected by puncturing of the marginal ear of each animal with a lancet and drawn directly into heparanized capillary tube and centrifuged with capillary haematocri centrifuge and positive samples were further processed for thin blood smear for confirmation of trypanosome species using their morphological characteristics with Giemsa staining techniques (Tekele, 2012).

Indirect methods rely on serological test by detecting specific antibodies developed by the host against the infection or inversely to demonstrate the occurrence of circulating parasitic antigens in the blood by the use of characterized specific antibodies, the detection of antibodies indicates that there has been infection, but as antibodies persist for some time after all trypanosomes have disappeared from the organism either by drug treatment or self cure appositive result is no proof of active infection, on the other hand circulating trypanosomal antigens are eliminated quickly after the disappearance of the trypanosomis and the presence shows almost always the live trypanosome present in animal (FAO,2004). The three test used most often are the indirect immune fluorescent antibody test (IFAT), the capillary agglutination test(CAT),and enzyme linked immune sorbent assay (ELISA) (Radostitis *et al*., 2007).

2.3. Associated Factors That Affect Distribution of the Diseases

The study of different researchers indicates that the prevalence of trypanosomisis associated various risk factors including,age, body condition and colour coat (Adane and Gezahegne 2007; Abebayehu *et al*., 2011; Ayana *et al*., 2012; Bishaw *et al*., 2012).The finding Solomon and Fitta (2010) in Awi and Metekel zones Northwest of Ethiopia indicates that, the physiological status of the host, as well as nutritional and environmental factors, further play important roles in modulating the severity of the disease, the prevalence in poor body condition animal was higher than the prevalence in medium body condition and good condition animals and the infection in poor body condition animals were significantly higher than medium and good body condition animals. similarly the finding of (Ouma, 2010)indicates the occurrence of disease in three different body condition (poor, good and medium) animals shows the highest prevalence in poor body condition (12.22%) followed by in medium (2.32%) and good body condition (2%). Due to poor body condition; animals are highly susceptible to diseases.

Based on the age groups of studied animals, adult groups of animals was highly infected than young group and significance difference was observed ($P<0.05$).A relatively higher prevalence of Trypanosomosis in adults than young cattle has been previously reported in the country (Abraham and Tesfahiwet (2012); Ayana *et al*., (2012).Since adult animals traveled long distance for grazing, watering and draft as well as harvesting of crops to tsetse challenge area. Calves were not allowed to move together with adult group, kept at

homestead until weaned off and protective material immunity in high tsetse challenge areas. The effect of the maternal antibodies which could afford protection young animals might have contributed to the lower prevalence of Trypanosoma in these animals (Fimmen *et al*., 1992).

Prevalence was also observed between animals with different coat color. Animals with black coat color were found to be highly infected than red and white coat color the strongest landing responses were found to be on black surfaces. (Leak, 1999)Comparison conducted between the different skin color of cattle indicated that slightly higher prevalence was observed in cattle's having mixed skin color (7.25%) followed by 4.88% in red, 3.57% in black, 1.56% in white and 0% in gray skin color. Tsetse flies by nature are attracted toward a black color, so in animals having black skin color there is high prevalence of Trypanosomosis recorded. The possible suggestion for the low prevalence in black skin color animals in the current study may be the low number of samples taken from black skin color animals.

2.3.1. Sex factor

The prevalence of bovine Trypanosomosis was assessed between sexes of animals and among 17 trypanosome positive animals; 9 (4.35%) of them were female animals and 8 (4.52%) of them were male animals, this shows that both male and female cattle were equally susceptible to Trypanosomosis infection (Daya and Abebe, 2008). Similarly the study results of Getachew (1993), Adane (1995) and Wellde *et al.* (1979) who obtained no significant difference in susceptibility between the two sexes.

2.3.2 Seasonal factors in tsetse abundance and trypanosois infection

According to WHO (2013).The ecology, development, behavior, and survival of insects and the transmission dynamics of the diseases they transmit are strongly influenced by climatic factors such as temperature, rainfall, and humidity. Similarly the research results of Simwango *et al*., (2017) and Ngonyoka.*et al*., (2017) reveled that habitat factors , host present and as well as time of the year and also the repot of Ngonyoka.*et al*.,(2017).in Maasai Steppe indicts Tsetse fly abundance is influenced by vegetation type, climate, distance from protected areas and the availability of wildlife species due to reduction available land for cattle grazing by conversion to crop farming . Since the available land for cattle grazing has been reduced by conversion to crop farming.

Seasonal livestock movements often lead to encroachment of cattle into protected areas that increases exposure of cattle to tsetse fly bites and trypanosome infections (Msoffe. *et al.*,2011,Nnko.*et al*.,2017) Climate plays an important role in the geographical distribution and abundance of vector species that are responsible for the transmission of a number of human diseases(Mashingo, 2010)

The findings from interviewees and key informants revealed that tsetse fly challenges have wide seasonal variations; on the one hand, the dry season (June –October) was reported to be of high risk of tsetse fly bites in the area. On the other hand, majority of the respondents admitted that the challenges of tsetse fly bite become mild in the rainy season, particularly in the months of November, December, January and February. Climate change, gender roles and perceived biting risk from tsetse flies: a case of communities neighboring ikorongo and grumeti game reserves in serengetin district, Tanzania (Ngongolo *et al.,* 2019)

Based on farmers' opinions, the major driver for movement of their livestock is search for pasture and water, and this is exacerbated during the dry season. Time of the year and grazing in areas close to the wildlife interface has been shown to influence prevalence of trypanosome infections in cattle. Kinyemi jabir (2016) Similar findings were reported in this study that cattle grazed along river banks and in swampy areas during dry seasons are exposed to higher risks of AAT and HAT trypanosome infection due to close contacts with humans, livestock, and tsetse flies (Rutto *et al*., 2013).

According to Tsegaye *et al*. (2015) and Emmanuel (2015)) July as well as January are dry months of the year; accordingly, shortage of pastures on grazing lands and drought are expected to lead to nutritional stress and decreased immunity of the cattle, making them vulnerable to Trypanosomosis as was noted by other researchers Concurrently, lower trypanosome prevalence was indicated in the month of October. Climate change is a reality which may not only explain the increase of density of arthropod vector, but also of their hosts, changes in periods of activity and variations in geographical distribution (Beugnet and Chalvet-Monfray, 2013).

2.4. PCV Value and Trypanosomosis

According to Stephen (1986) and (Van den Bossche *et al*. (2000) cattle with PCV≤24% were considered anemic is one of the most important and principal sign of

Trypanosomosis in cattle, The level of anaemia or the PCV usually gives a reliable indication of the disease status (Van den Bossche *et al*. 2000)) or delayed recovery of the anaemic situation after current treatment with trypanocidal drugs. The appearance of parasitologically negative animals with PCV values of less than the threshold value set (25%) may be due to the inadequacy of the detection method used (Abebe *et al*., 2004) Furthermore, the occurrence of positive animals with PCV of greater than 25% might be thought of recent infections of the animals. The report of Rowlands *et al.* (1993) in Ghibe valley at South Western Ethiopia, in which was stated that the average PCV of parastiologically negative animals was significantly higher than the average PCV of parasitological positive animals.

The previous result of Garoma (2009) indicates that almost 94.30% cattle's having PCV≤24% but they react negatively for Trypanosomosis infection and this may have occurred due to the inadequacy of detection method used (Murray et al., 1977) or delayed recovery of anemic situations after recent treatment with trypanocidal drugs or may be due to the compound effect of poor nutrition and hematophagus helminth infection such as haemonchosis and bunostomiasis (Afework, 1998). However, PCV values can be affected by many factors other than Trypanosomosis, but these factors are likely to affect both Trypanosomosis negative and positive animals (Van den Bossche and Rowlands, 2000)

2.5. Trypanosomosis and Tsetse control

According to OAU (2001).There are three main strategies for the controlling of the disease the first is to use drugs to combat the parasite itself, the second strategy is to control of vector of disease, tsetse fly. The third strategy is to use animals that are inherently tolerant to the effect of the use of the disease and are able to remain relatively productive even when infected

2.5.1. Chemotherapy and chemoprophylaxis

Trypanosomosis disease control is based on the use of trypanocidal drugs. Depending on the control strategy, drugs are used for curative or preventive purposes. Curative or preventive trypanocidal drugs are used to maintain susceptible livestock in trypanosomosis enzootic area. So far, the protection of livestock by immunisation is not possible. The use of preventive drugs can help to prevent susceptible animals from

contracting the disease for a period of two to four months (Sow 2013).The control of trypanosomosis in enzootic countries involves control of tsetse fly population, prophylactic treatment, good husbandry of animals at risk, and use of trypano-tolerant animals (Radostitis *et al*., 2007). Most have a narrow therapeutic index, which makes administration of the correct dose essential (Merck, 2005). Therapeutic drugs for the treatment of trypanosomosis includes diminazenum aceturate, homidium bromide and homidium chloride. Prophylactic drugs for cattle include homidiumbromide, homidium chloride and isometamidium (Ayisheshim *et al*., 2015).

There are three anti trypanosomal compounds upon which treatment and prophylaxis of trypanosomosis currently depend. These are isometamidium chloride, homidium chloride or bromide and diminazine aceturate, whereas, quinapyramine, suramin and melarsomine are primary used as therapeutic druges for infection caused by T.evansi in equidae, camels and buffaloes, although quinapyramine is also used for prophylactic purpose (Venturia *et al*., 2008). In West Africa, only two molecules are used to tackle the disease, namely DA and ISM. However, there has been widespread incorrect use of these drugs which has leads to the development of drug resistance by the parasite (Geerts *et al*., 2001).

These compounds disrupt or block one or more of vital processes, which are essential to the invading trypanosome. Certain drugs have specific effects on the enzyme systems or block metabolic processes/pathways, and this is true of most of the trypanociodal drug (FAO, 1998).

In the case of sleeping sickness, the most widely used drugs to treat the patients are Suramin, Melarsoprol and Eflornithine (difluoromethylornithine) depending on the trypanosome species and the clinical stage of the disease. A new treatment using a combination of NifurtimoxEflornithine is available for sleeping sickness (Priotto and Sevcsik, 2008). There is no vaccine available to control both human and animal trypanosomosis.

2.5.2 Vector Control Strategy

Vector control intends to disrupt the cycle of disease transmission by reducing or eliminating the fly population in a given geographic area with the objective to eliminating trypanosomosis related problems in both human and livestock. Tsetse control techniques

have gone through evolutions, with earlier crude methods recently being replaced by methods that are economically cheaper, more directed, and ecologically sustainable. These techniques include: environmental management (bush clearing and game destruction), pesticide campaigns, traps/targets, animal baits and the use of Sterile Insect Technology (SIT) (Sow, 2013).

The choice of the control method to be applied will depend on the target zone, the impact on the environment, the tsetse species, and the possibility of isolation of treated areas, the economic impact, the possibilities of post control land use and the available funding of the operations now and for the future (Sow, 2013). So far, insecticides remain the most frequently used method of control due to their efficacy and they constitute the first step of tsetse control before any other method can be applied (Allsopp, 1984). Vector control must be combined to cattle treatment to avoid the spread of resistance (Bouyer *et al*., 2013).

Bush and game clearance early attempts to control tsetse included extensive bush clearance designed to eliminate the shaded places where tsetse rest and lay their larvae and extensive shooting of wild game animals designed to eliminate the wild blood sources used by the tsetse(WHO, 2004). Bush clearing and game destruction is targeted at disrupting the ecology of tsetse flies, thereby bringing down tsetse populations to sustainable levels for human habitation and livestock development, although widely effective such methods can no longer be recommended because of their adverse effect on biodiversity (Bouyer *et al*., 2010).

Pesticide campaign is one of Current vector control interventions involve the use of insecticides either through aerial spraying; ground spraying; insecticide-treated targets or insecticide-treated animals (live baits) and the use of other-baited traps or screens (Hargrove *et al*., 2000, Vale and Grant, 2002).According to WHO (2013) trials with insecticides against tsetse started in 1945, when DDT and BHC (HCH) were the only synthetic compounds available, the application of residual deposits of persistent insecticides to tsetse resting sites was very widely used, but is now discouraged due to concerns about effects on non-target organisms.

The area-wide application of the Sequential Aerosol Technique (SAT) involves the use of Deltamethrin insecticidal concentrates at minimal doses with little or no residual effects on flora and fauna an extensive area of tsetse control or elimination is achieved by the

application of Deltamethrin insecticidal aerosols from air-crafts over large tracts of land (Kgori, 2006).

SAT, which can effectively clear large areas of tsetse in a relatively short time, requires substantial economic and infrastructure support, .recent advances in aircraft guidance systems have considerably increased the accuracy and the efficiency of insecticide delivery as shown in recent control operations in the Okavango delta in Botswana, Namibia, Ghana, Burkina Faso (Kgori *et al*., 2006). The main limitation of aerial spraying as with other methods is the reinvasion pressure if the area is not isolated, and the occurrence of dense vegetation cover lowering the penetration of insecticide droplets (Kgori, 2006).

Sterile Insect Technique (SIT) exploits the particular mating biology of tsetse, whereby female flies rarely mate more than once, Male flies are therefore mass reared in the laboratory, sterilized by irradiation, and released to mate with wild females (Kuzoe and Schofield, 2004). Females mated with sterile males are unable to produce offspring, unlike all other tsetse control techniques, SIT has no effect on non-target organisms also becomes more efficient at lower fly densities, and is ideally suited to the final phase of local tsetse eradication Kuzoe and Schofield, 2004). The proportion of infertile males to fertile wild males must be at least 10:1, if the sterile males vastly outnumber the fertile wild males, the wild fly population quickly dies out, the most notable success of SIT against tsetse was in eradicating *G. austeni* Newstead on Unguja Island and Zanzibar with 50:1 infertile to fertile male proportion (Hargrove, 2003).

Insecticide treated cattle has been used to control tsetse and trypanosomiasis in various sub-Saharan African countries including Burkina Faso (Bauer *et al*., 1999).Tanzania (Hargrove *et al*. 2000) Zimbabwe (Torr *et al*., 2007). Ethiopia (Bekele *et al*., 2010)., Degrees of success differ between control programme, being affected by the size and shape of the control areas, and the number and density of treated cattle (Hargrove *et al.,* 2003). If the area treated is small and is surrounded by a tsetse-infested area, invasion from the untreated area can re-infest much or the entire controlled region (Torr and Vale 2011).

Two techniques of application of insecticides on cattle, that is, whole-body (WB) and restricted application (RAP) of insecticides, are considered (Hargrove *et al*., 2012). These

are now easy to apply and remain on animal skin for at least two weeks. The effectiveness of this strategy depends on cattle density, grazing patterns and fly distributions. The strategy works best where livestock are the main host of tsetse. The insecticide pyrethroid (Deltamethrin 1%) applied on the middle back of the animals (cattle) in the experimental sites (belly, leg, middle back line and neck which are marked and insecticide applied at the dose of 1ml per 10kg body weight by using T-bar applicator as recommended by manufacturers (STEP,2012).

Different theoretical and field experimental studies suggesting that the use of the ITC approach can provide a highly cost-effective approach to tsetse and trypanosomiasis control (Hargrove *et al*., 2000, 2012; Torr and Vale 2011; Torr *et al*., 2007; Bourn *et al.,* 2005) and is the method of choice whenever there are appropriate densities of cattle in a tsetse area (Shaw *et al*., 2013).

the study of Vale and Torr (2004) they reported that monthly application of pyrethroid was the most effective method to kill tsetse flies .Also Alemu *et al.*(2013)which revealed that the insecticide effectively works through week 5 but start declining at week 6 and totally fails on week 7. Hargrove *et al*. (2012) which Indicates that chemical sprayed remain on animal skin for at least two weeks and the efficacy decrease overtime. This is also consistent with the report of Damian *et al* (2014) and Jemaler *et al*. (2009). This was also agreed with a trial carried out in Ethiopia in which monthly application had resulted in a 98% decrease in apparent density of G. pallidipes (Leak *et al*., 1995).

The study result was also agreed with Vale *et al*. (1999) when chemical agents are used to combat tsetse flies; it must be taken into account that tsetse flies spend about 50% of their lifespan under the ground as pupae. Therefore, either insecticide must be used, which remain active for at least the maximal pupal period, or repeated treatments with a short-acting product must be foreseen (Vale *et al*., 1999.)

This method is simpler, cheaper and less damaging techniques. In particular, there has been renewed interest in the use of devices that can be used to kill or sterilize tsetse in the field, in principle, and if properly carried out, these methods should allow the eradication of tsetse without the need to remove hosts or vegetation and without depositing large quantities of toxic chemicals in the environment (Hargrove, 1988). Initial interest was in traps, then in insecticide treated ‘targets. The that modern pyrethroids were better suited

for use as sprays or dips for cattle than previous generations of insecticides, such as the chlorinated hydrocarbons (Hargrove, 1988)

In the early 1900s, sticky traps worn by plantation workers were successfully deployed on the Island of Principe to eradicate *G. palpalis*. Since then, trapping techniques have been greatly enhanced by development of designs that mimic the fly's perception of vertebrate hosts (Torr *et al*., 2007). These generally use blue and black cloth in a shape that attracts the flies and then funnels them upwards into a netting trap usually in the form of a mono conical (pyramidal) or bi conical shape (Bouyer *et al*., 2009).

For tsetse control, a simpler and cheaper device involves a suspended screen of blue and black cloth (often known as a tsetse target) impregnated with a biodegradable pyrethroid insecticide such as Deltamethrin, flies are attracted by the blue segments and land on the black segment, quickly succumbing to the insecticide, the effectiveness of traps and targets can be greatly enhanced by addition of an appropriate odor bait, are usually short-chain aromatic compounds such as acetone or Octenol (Bouyer *et al*., 2001)

Host resistance protection Trypano-tolerant animals are being used to establish ranches in areas where tsetse challenge is not too heavy, but they have not been readily accepted in some countries, supposedly because they are smaller in size and they produce less milk than other indigenous breeds and crosses with exotic breeds (Radostitis *et al*., 2007). They are infected by tsetse flies but do not show clinical disease, however, these breeds have not been readily accepted because they are small in size and low in milk producing (OIE, 2013). In the study of Eshetu and Begejo (2015), four Ethiopian cattle breeds in aspects are related to trypano-tolerance breeds are Abigar, Gurage, Horro and Sheko.

2.6. Impact of Trypanosomosis and Tsetse Problem in Ethiopia

2.6.1 Trypanosomosis Problem in Ethiopia

Trypanosomosis has long been recognized as a massive constraint on animal husbandry, livestock production and mixed farming in vast areas of rural sub-Saharan Africa including Ethiopia The disease Trypanosomosis, locally called "Ghendi" was reported to be the most and the first important livestock constraint affecting animal health, (Oluwafemi, 2014). Ethiopia is known with its large and diverse livestock resource endowments, livestock is primarily kept on small holdings where it provide drought

power for crop production, manure for soil fertility and fuels, serves as a sources family diet and sources of cash income (from livestock and livestock products) (Abebe *et al*., 2004).

Despite large livestock population, Ethiopia fails to optimally utilize this resource due to different constrains facing the livestock subsector (Abebe *et al*., 2004). Trypanosomosis is serious constraints to livestock production and agricultural development in Ethiopia. A total of 14.8 million cattle, 6.12 million sheep and goats, 1 million camels and 1.23 million equine are at risk of contracting trypanosomes (Abebe *et al*., 2004).

Multiple previous studies conducted in cattle in different parts of Ethiopia reported a wide range of prevalence ahigh prevalence of 22% was reported from Arba Minch,Southern Ethiopia by Zekarias and Zeryehun (2012),prevalence of 12.5% was reported by NTTICC (2007). From Meda Jalala, Western Ethiopia, Abebe and Jobre (1996) reported a prevalence of 17.67% form tsetse infested areasof Ethiopia.

Prevalence of 17.20% was reported from Pawe, Northwest Ethiopia by Afework (1998), 12.3% from Bedele, Southwestern Ethiopia by Regaassa *et al.,*(2015), 17.20% from Metekel district by Yohanes (1997), 19.01%from Goro district, Southwest Ethiopia by Abiy (2002), 16.10% in and around Bahir Dar by Adane(1995)and 16.93% from Guduru district, Horo Guduru Wollega by Amante *et al.,* (2014*)*. Recently, Leta *et al.,* (2015) conducted a comprehensive Meta-analysis of bovine Trypanosomosisin Ethiopia and they estimated the prevalence of bovine Trypanosomosis in Ethiopia to be 8.12%. According to Leta *et al*., (2015),

2.6.2 Tsetse Problem in Ethiopia

Several reports made in Ethiopia revealed that tsetse fly occupy over 66,000 km2 areas based on a 1500 m.a.s.l. breeding limit in the Southern and Southwestern valleys of the country. Langridge (1976) has reported that some 98,000 km2area 1600 m.a.s.l. However, due to the advancement of tsetse flies into formerly free areas reaching 130,000km2 to 150,000 km2 (Silnbergh, 1992) based on 1700 m.a.s.l.and recently 220,000 km2 areas is estimated to be affected by tsetse flies (NTTICC, 2004) based on 2000 m.a.s.l.

Currently five regional states are directly affected by the tsetse problem. These are Amhara, Oromia, Southern Peoples Nations and Nationalities Regional State, BeneshangulGumz and Gambella (Abebe, 2005; Dagnachew *et al*., 2005; Cherent *et al.,*

2006; Fikru *et al*., 2012). Tsetse flies in Ethiopia are confined to Southwestern and Northwestern regions between longitude 33o and 38o E and latitude 5oand 12oN covers an area of 220000 km² (NTTICC, 2004). Tsetse infested areas lie in the low lands and also in the river valleys of Abay (Blue Nile), Baro, Akobo, Didessa, Ghibe, and Omo (Megersa *et al*., 2019).Consequently, new areas are being invaded and settled communities are being continually evicted by the advancing tsetse, five species of *Glossina* (*G. m. submorsitans, G. pallidipes, G. tachinoides, G. f. fuscipes and G. longipennis*).

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3. MARTIAL AND METHOD

3.1. Description of the Study area

The study was conducted from October 2009 to April 2010 at Fura Kebele from Mirab Abaya and Ganta Kancham kebele from Arba Minch Zuria Woreda. They are among the woreda in the Southern Nations, Nationalities, and Peoples' Region of Ethiopia, a part of Gamo Zone in the Great Rift Valley, located between 5° and 37°E longitude lines and at an altitude of 1100-2900 meter and 1200-3310 meter above sea level Mirab Abaya and Arbaminch Zuria respectively. Mirab Abaya is located at about 445 km south of Addis Ababa and Arbaminch Zuria is located at about 505 kilometers south of Addis Ababa (Diresigne 2016)

Study areas also include known two lakes namely Abaya and Chamo. Nechisar National Park is located between these lakes. According to CSA (2007) the area has bimodal rainy season with long rainy season from March to May and short rainy season from end of August to mid of October with mean annual rain fall of 900-1000mm and mean annual temperature of 23°c. The area has sub humid climate with moderately hot temperature climatically, three ecological zones are found i.e. 14% Dega, 53% Woina-dega & 33% Kola CSA (2007). Animal population of the study area is cattle 121460, equine 8544, sheep 24689, goat 72610, poultry 216150 CSA (2013).

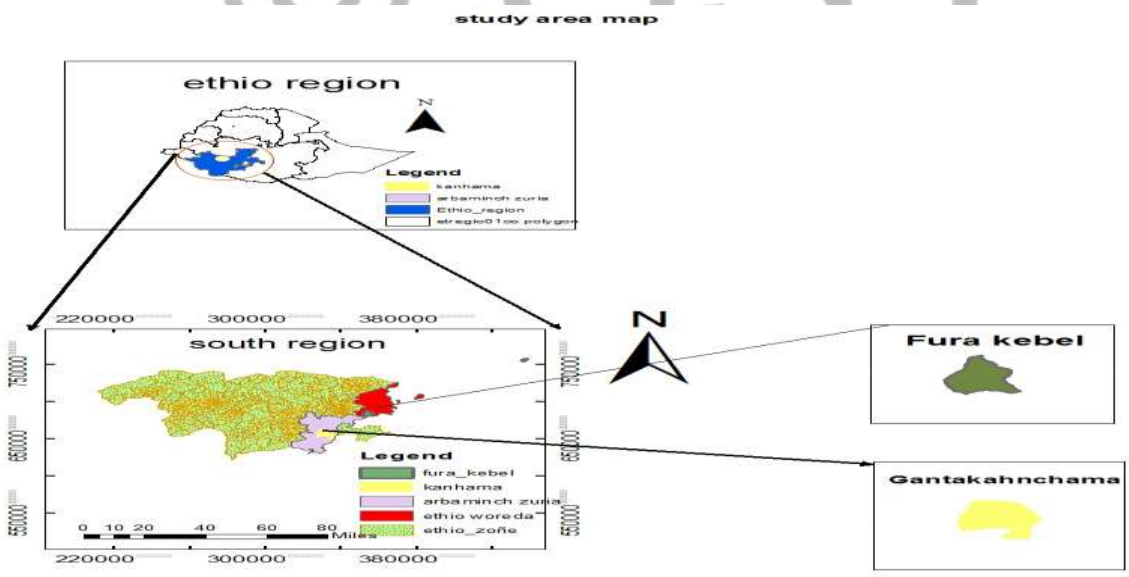


Figure 2: Study Area map (CSA, 2007)

3.2. Study animals

The Animals used for the study were zebu cattle usually keep under extensive husbandry system were selected from Fura and Ganta kanchama Kebele and they were ear tagged, de wormed regularly and farmers were agreed voluntarily not to sell the cattle included in the study.

3.3. Study Design

Pre intervention phase was done by cross-sectional study and intervention phase was done by longitudinal study. Pre-intervention phase comprises of baseline parasitological and entomological data collection to determine the existing situation just before the intervention. It was conducted at beginning October 2019. Following this, an intervention phase started by application of test insecticide (Deltamethrin) in the Fura and Kanchama Kebele. Monitoring of the intervention phase was in operation monthly from October 2019 to April 2020 in each study area.

3.4. Sampling Method and Sample Size Determination

Multistage sampling method was implemented to conduct the study. The two district were selected purposively based on tsetse density of Gamo zone, the study kebeles were selected purposively based on accessibility to transport from tsetse infested areas. The herd of cattle selected randomly from each village of the study areas. The study animals were selected based on simple random sampling method after owners bring their animals to the sampling site for free pour on treatment. The cattle owner were systematically selected by numbers on the cards distribute to them. To determine sample size, the prevalence of 22% was considered from previous study in Arbaminch Zurea by Abraham and Tesfahewet (2012). As result the sample size was 264 cattle's. From two kebeles 108 and 156 cattle were sampled based on proportion of their total cattle population 1939 and 2801 in Fura and Kanchama Kebele respectively. Absolute desired precision of 5% at confidence level of 95% were used for two districts the formula for estimating sample size (Thrusfield, 2018)

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

Where, n= required sample size; $P_{exp=}$ expected prevalence; d= desired absolute precision

$$n= \frac{(1.96)^2\ 0.22(1-0.22)}{0.05^2} = \ 264$$

22

3.5. Study Techniques

The cattle were treated with delthamitrene 1% monthly at Ganta kanchama and quarterly base at Fura Kebele from October 2019 to April 2020. According to the proportion of cattle population the blood sample was taken from 156 cattle in Kanchama and from 108 cattle in Fura Kebele, for parasitological and PCV examination. Tsetse fly surveys from selected sites were conducted monthly in study areas. Traps were deployed 200 up to 250 meters interval considering the tsetse habitat (FAO, 2009) and these sites were monitored for 72 hr.

3.5.1. Pre-intervention phase entomological data.

Entomological data collection was carried out by deploying a total of 20 NGU traps (10 in Fura and 10 in Kanchama).

The NGU traps constructed from locally made blue and black cloth with white mesh on the top was baited with 3-week-old bovine urine (Brightwell *et al*., 1990). Traps were set at approximately 200–250 m apart. All trap positions were geo-referenced (using hand-held GPS), and the altitude and vegetation type recorded. It was attempted to include different vegetation types such as bush land (BUL), wooded grassland and (WGL), for trapping. Collection of trapped flies took place 72 hr after deployment.

3.5.2. Pre-intervention phase *parasitological data.*

Blood samples were collected from marginal ear veins using micro-haematocrit capillary tube from a total of 264 and sealed on one side with cristaseal (Hawskely Ltd.). The capillary tube was then transferred to a haematocrit centrifuge and spun for 5 min at 1200 revolutions per minute. The centrifuged capillary tube was measured for PCV values on the haematocrit reader. It was then cut 1 mm below the buffy coat and the contents of the tube expressed on to a slide, mixed and covered with a 22 mm 22 mm cover slip. This slide was then examined under 40 objective using phase contrast (Abebe *et al*., 2004) or dark field microscopy to examine for the presence/absence of motile trypanosomes.

3.5.3. Intervention phase Pour on application

Cattle were treated with delthamethrin 1% monthly in Kanchama and quarterly in Fura Kebele, during intervention period. This was done by spraying Deltamethrin 1% (w/v) pour-on ready-for-use formulation (Appropriate Applications Ltd., USA) at a dose rate of

10 ml per 100 kg body weight. The cattle Deltamethrin 1% pour-on ready-for use formulation were used to apply the insecticide along the line starting from in front of the shoulder running back to behind the hip to cattle which has age group greater than one year (STEP, 2012)

Figure 3: Pour on application

3.5.4. Intervention phase entomological survey

To assess the apparent density of tsetse fly in relation to different Deltamethrin1% application regime in treated cattle of the study area, sampling was done in selected sites of the study area tsetse flies were collected by 20 NGU traps in different positions of the study areas before sun rise in the morning (7:00- 9:30 AM). The odor baits used contained, cow urine in order to release the necessary amounts of attractants. All odors were on the ground about 30 cm upwind of the trap. The density and species of tsetse flies were assessed using odor-baited NGU traps deployed at 200-250 m. Traps were allowed to stay at the deployment site for a period of 72 hrs. Before collection, in watering and grazing points in which the animal and the vector were believed to have frequent contacts (FAO, 2009) The altitude levels and vegetation types were recorded during the sampling period.

Fly catch per trap per day (F/T/D) was determined to calculate the fly density and distribution (Leak *et al*., 1987).The species of the dominant tsetse fly were determined following the standard procedures and biting flies according to their morphological characteristics such as size, color, wing venation structure, and proboscis at the genus level (FAO, 2009).The apparent density of tsetse flies were determined based on the daily mean number of flies captured in NGU traps and recorded as fly per trap per day (F/T/D) (Leak *et al*., 1987).

Figure 4 Entomological data collection by NGU Trap

3.5.5. Intervention phase Parasitological

Blood samples were examined by dark ground (DG) buffy coat microscopic method to detect the presence of trypanosomes (Murray *et al.*, 1982). Blood samples were obtained by puncturing of the ear vein with a lancet and collected directly into a capillary tube, which was treated with heparin sealed one end with Cristaseal (Hawksely), the capillary tubes were placed in micro haematocrite centrifuge with the sealed and outer most, after screwing the rotary cover and closing the centrifuge lid, the specimens were allow to centrifuge at 12,000 rpm for five minutes (Radostits, 2007).

The tubes were placed in hematocrite reader and expressed the reading as percentage of packed red cells to the total volume of whole blood (Taking the PCV value >24 as normal for zebu cattle (Radostits, 2007). Haematocrite tubes were cut by diamond tipped pencil few millimeters below the junction of the buffy coat / plasma level and the erythrocyte, the contents homogenized on to clean slide and covering with a 22 x22 mm cover slip, then the slides were examined under a microscope using x40 objective and thin blood smears were done small drop of blood from a micro haematocrit capillary tube to the slide was dropped to clean slide and spread by using another clean slide at angle of 45^0. The smear were dry by waving it in air and fixed for 2 minutes in methyl alcohol, flooded with Giemsa stain (1:10 solution) for 30 minutes and washed using distilled water allowed it to dry by standing up right on the rack and examined under the microscope (x100) oil immersion objective lens (Murry *et al.,* 1982).

3.6. Data Management and Analysis

At the time of sampling the owner's name, animal age, animal sex, body condition score and color coat were recorded using the recording format, the data were recorded during sample collection, parasitological examination, PCV measurement and entomological data into excel spread sheet 2010 to create a database and import to STATA version 14.2 software for analysis. The pre-intervention entomological data (trap catches) were calculated as apparent density and expressed as catch off tsetse flies per trap per day. The point prevalence during pre-intervention was determined by dividing positive cattle to total examined. Deltamethrine 1% effect (Monthly and quarter) application regime in reduction of trypanosome infection and tsetse density, parasitological and entomological data were analyzed by logistic regression and Poisson regression test for longitudinal data (repeated measures). In all analyses the confidence interval (CI) is 95% and P value $p < 0.05$ was consider as significance. PCV were categorized as anemic if it is less than 24% and normal if it is greater than 24% (Uilenberg, 1998).

4. RESULTS AND DISCUSSION

4.1 Results

4.1.1Pre-Intervention Entomology results

A total of 20 NGU traps were deployed for 72 hour in the two study sites. Out of 20 NGU, 10 were in Fura Kebele and the remaining 10 were in Kanchama Kebele. Pre intervention entomology results as indicated on table 2 below, there was total 63 and 532 tsetse flies were cached in Fura and Kanchama Keble respectively. From the total 11 male, 52 female flies Fura Kebel whereas in kanchama Kebele from the total 179 male and 353 were female. *Glossina pallidips* was the only species of tsetse detecte

Table 1: pre interventionn entomological result

Kebele	Number of traps	Tsetse species	Cache			Catch/trap/day
			Male	Female	Total	
Fura	10	*G.paladips*	11	52	63	2.1
Kanchama	10	*G.paladips*	179	353	532	17.73

4.1.2. Pre-Intervention Parasitological results

Parasitological examination was done by taking blood sample from the ear vine of randomly tagged cattle.

Table 2: Pre intervention parasitology results

Kebele	Cattle				Prevalence %
	Examined	Infected			
		TC	TV	Total	
Fura	108	6		6	5.5
Kanchama	156	13	2	15	9.6
Total	264	19	2	21	7.9

4.1.3. Pre-Intervention Hematology results

Hematological test result In table 4 was indicates PCV value of study cattle, in Fura Kebele a total of 51 cattle have PCV value less than 24, from those 6 cattle were positive and 45 cattle were negative. Similarly, in Kanchama Kebele a total of 75 cattle were PCV value less than 24% from them 13 cattle were positive 62 cattle were negative. There was no positive result seen in Fura from the cattle with PCV value greater than 24% (57cattle) but there were two positive test results in Kanchama Kebeles from the cattle with PCV value greater than 24% (81). As indicated below in table 5 the mean PCV of parasitemic cattle was 20.2 % and 21.5 and aparasitemic cattle was 21 % and 22.7% in Fura and Kanchama Kebele respectively.

Table 3: Pre intervention frequency distributorship in PCV with infection status

Number of cattle	PCV<24		PCV>24	
	Fura	Kanchama	Fura	Kanchama
infected	6	13	0	2
Non infected	45	62	57	79
Total	51 (47.2%)	75 (48.1%)	57 (52.8%)	81 (51.9%)

Table 4: The mean PCV with infection status

kebele	Mean PCV in preintervation phase		Mean PCV
	parasitemic	Aparasitemic	all cattle
Kanchama	21.5 %	22.7 %	22.58
Fura	20.2 %	21 %	20.95

4.2. Intervention results

4.2.1. Parasitological results

The test results indicated that 42 positive test result in Fura keble 8 in October , 5 in November , 6 in December , 7 in January , 3 in February , 8 in March, and 5 in April. As indicated in tabel 5 there was no significant difference in incidence *Trypanosoma* at preintervation to last the monitoring *p (0.*385*)* and in kanchama Kebele total of 36 tested positive, 9 in October, 7 in November, 6 in December, 4 in January, 4 in February, 3 in March, and 3 in April there was significant difference in the incidence Trypanosoma preintervation to last the monitoring *P (0.006).*(Table 5, Table 6, Tabele 7 and Figure 5).

Figure 5: Incidence of Trypanosomosis during monitoring period**.**

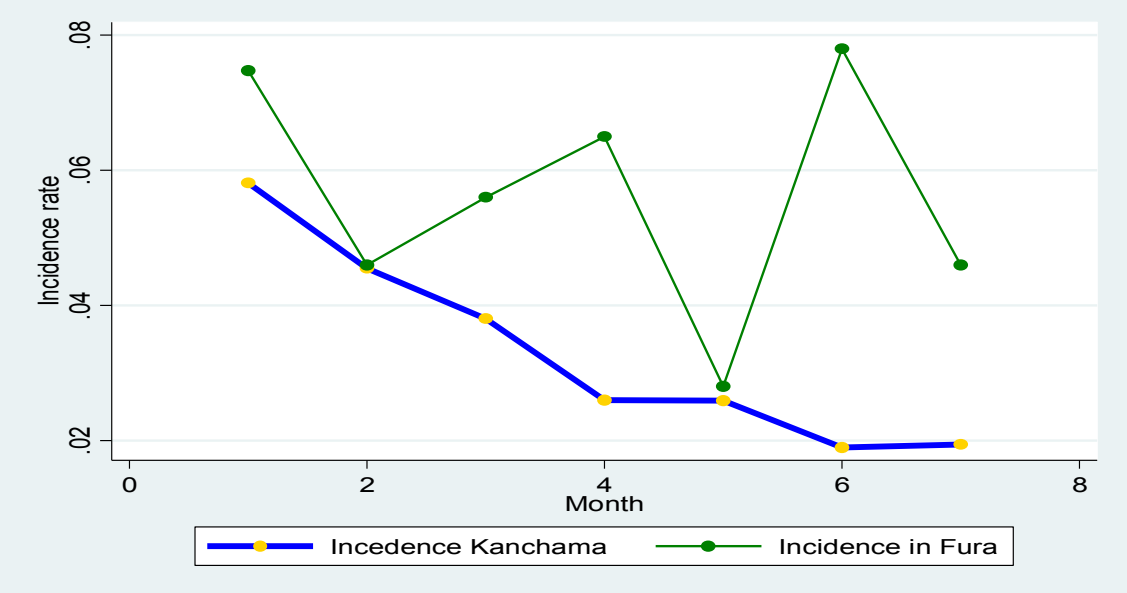


Table 5: Incidence of Trypanosomosis in Kanchama kebel

Month	cattle examined	At risk	Infected	Incidence
1	155	156	9	0.0581
2	154	156	7	0.0455
3	156	156	6	0.038
4	156	156	4	0.026
5	154	156	4	0.0259
6	156	156	3	0.019
7	155	156	3	0.0194

Table 6: Incidence of Trypanosomosis in Fura kebe

Month	cattle examined	At risk	Infected	Incidence
1	107	108	8	0.0748
2	108	108	5	0.046
3	108	108	6	0.056
4	108	108	7	0.065
5	108	108	3	0.028
6	108	108	8	0.078
7	107	108	5	0.046

Tabele7:logestic regression result of trypanosomosis infection

kebele	Trypanosomosis infection	Coef	Std. Err	Z	p>|z|	(95%conf.Interval)	
Kanchm	intervention	-2.103096	.761743	-2.76	0.006	-3.596085	-.6101069
	Conc.	-2.24071	.2715858	-8.25	0.000	-2.773008	-1.708411
Fura	intervention	-.5096127	.587205	-0.87	0.385	-1.660513	.641288
	Conc.	-2.515678	.3675609	-6.84	0.000	-3.236084	-1.795272

The test result of pre intervention and subsequent monitoring have shown that the incidence of Trypanosomosis in Fura declined from 5.5% at pre intervention to 4.7 % at final monitoring, this finding confirms that there was a reduction with a slow trend. Also the test result in Kanchama indicates that incidence of Trypanosomosis declined from 9.6% at pre intervention to 3.2 % at final monitoring. This results were indicates that monthly application of Deltamethrine 1% has more effect on reduction of trypanosomes infection. This agrees with the result of Leak *et al.* (1995), 77.4% and 70%, overall prevalence reduction, respectively. Van den Bossche *et al*. (2004) reported monthly incidence of Trypanosomosis being negatively correlated with the time elapsed since the start of Cyfluthrin applications in the control of *G. m. morsitans* in Zambia. Leak *et al*. (1995) also reported a rise in PCV of cattle from a mean of 23.8% at pre-control to 26.8% following tsetse control trial with Cypermethrin .The result also agreed with the overall reduction in prevalence estimated by computing from the incidence record as derived by (Thrusfield 2018).

4.2.2. Hematology results

Table 8: the mean PCV value in Fura and Kanchama kebele.

Month	Number of cattle examined		Mean PCV	
	Fura	Kanchama	Fura	Kanchama
October	107	155	25.3	24.27
November	108	154	25.41	25.74
December	108	156	25.34	25.2
January	108	156	25.1	26.4
February	108	154	25.4	26.5
March	108	156	25.6	27.7
April	107	155	26.01	29.6

Tabele 9: the mean PCV with infection status in Fura and Kanchama kebele

kebele	Mean PCV in intervention phase	
	parasitemic	aparasitemic
Kanchama	23.9 %	26.7 %
Fura	22.3 %	25.63 %

During the present study there were marked difference was noticed in PCV according to the infection status of animals, where parasitaemic animals had generally lower PCV than non-parasitaemic ones. The level of anaemia or the PCV usually gives a reliable indication of the disease status (Van den Bossche *et al*. 2000)) or delayed recovery of the anemic situation after current treatment with trypanocidal drugs.

The Present study indicated the mean PCV-value for the parasitemic cattle was 22.3 and 23.89 in Fura and Kanchama Kebele, respectively which falls in the range of anemia, While the mean PCV of parasitological negative cattle were higher than positive cattle which was 25.63 and 26.7 in Fura and Kanchama Kebele respectively. This test result was in agreement with the report of Sinishaw (2004), Bitew *et al*. (2011) and Dagnachew *et al*. (2011) parasitemic cattle PCV value is less than twenty four. Similarly, the report of Rowlands et al. (1993) in Ghibe valley at South Western Ethiopia and the previous result of Garoma (2009) were reported the average PCV parastiologically negative animals were significantly higher than the average PCV of parasitological positive animals *($p<0.05$)*.

The study also revealed that almost 16.7 and 19.4 have PCV value greater than 24, and they react positively to Trypanosomosis infection. This may be due to early infection with Trypanosomosis. This result agrees with the previous result of Garoma (2009**)** who reported that the occurrence of positive animals with PCV of greater than 24 might be thought of recent infections of the animals. Similarly, Murray *et al.* (1977) and Afework.(1998) report were indicates that occurrence of positive animals with PCV of greater than 25 this may have occurred due to the inadequacy of detection method used.

According to test result the mean PCV value in kanchama Kebele increase from pre intervention period to last monitoring (22.58 to 29.7) and the result in Fura kable indicates the mean PCV value was almost similar from ferest intervention to last monitoring 20.95 to 26.01).The result seen in kanchama kebele was agreed with Jemaler

(2009), Van den Bossche *et al*. (2004), Leak *et al*. (1995) also reported rise in PCV of cattle from a mean of 23.8 at pre-control to 26.8 following tsetse control trial

4.2.3. Entomological results

Table 10: Poisson regression result of tsetse fly catch.

kebele	Testse catch	Coef	Std.Err	Z	p>|z|	(95%conf.Interval)	
Kanchama	intervention	-.994365	.071765	-13.86	0.000	-1.135022	-.8537082
	Conc.	4.275276	.0372937	114.6	0.000	4.202182	4.348371
Fura	intervention	-0.0336166	.2048366	-0.16	0.870	-.435089	.3678558
	conc	1.586965	.1507557	10.53	0.000	1.291489	1.882441

Table 11: the effect of monthly and quarterly application of Deltamethrin

kebele	Testse catch	Coef	Std.err	Z	p>|z|	(95%conf.Interval)	
Kanchama	deltamethri	-.7117627	.0748627	-9.51	0.000	.-.8584978	-.5650416
	Conc.	3.992681	.0429537	92.95	0.000	3.908493-	4.076869
Fura	deltamethri	-.119708	.104444	-1.15	0.252	-.3244145	.0849985
	Conc.	1.58412	.0716115	22.12	0.000	1.443764	1.724476

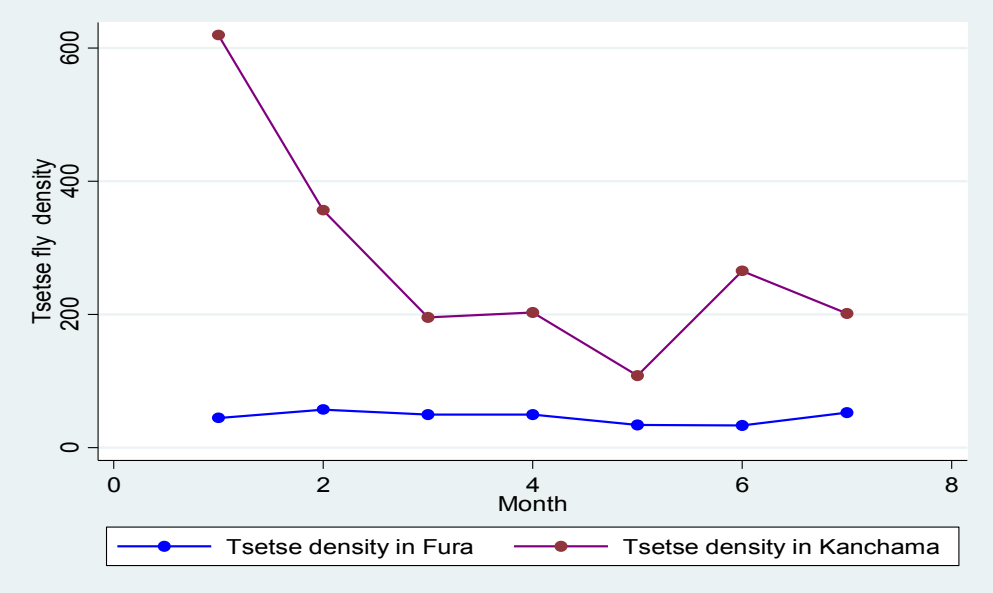
Table 12: Apparent density of tsetse flies per month in Fura kebele

Month	Cache/trap/day	Total catch	Number of male fly	Number of female fly
October	1.47	44	14	30
November	1.9	57	16	41
December	1.63	49	10	39
January	1.63	49	15	34
February	1.13	34	8	26
March	1.1	33	8	25
April	1.73	52	15	37

Table 13: Apparent density of tsetse flies per month in Kanchama Kebele

Month	Cache/trap/day	Total catch	Number of male fly	Number of female fly
October	20.63	619	225	394
November	11.87	356	137	219
December	6.5	195	55	140
January	6.77	203	75	125
February	3.6	108	27	81
March	8.83	265	92	173
April	6.7	201	80	121

Figure 6: Tsetse fly densities during monitoring period



Different technologies have been introduced and applied in the field work for two decayed to control tsetse flies. These have provided a suitable solution to the problems of animal trpanosomiosis (Terzu, *et al.,* 2004). The use of insecticide treated cattle (application of Deltamethrin 1% pour-on formulation to cattle) has adequately reduced tsetse fly population and Trypanosomosis incidence in cattle (Jemere, *et al*., 2009)

The apparent densities of tsetse flies caught in pre intervention phase were 2.1 flies per trap per day and 17.73 flies per trap per day in Fura and Kanchama Kebele respectively. During the study period Deltamethrin 1% pour-on monthly applied in kanchama Kebele and quarterly applied in Fura kebele. At the end of intervention, the apparent densities of tsetse flies per trap per day were 1.73 in Fura and 6.7 in Kanchama Kebele. Also the result indicated in table 12 shows that during monitoring period tsetse fly catch per day were almost similar in Fura Kebele. Whereas, Tsetse flies catch per day decrease from the first monitoring to last period of intervention in Kanchama Kebele as indicated in figure6, the reduction tsetse flies density from the first monitoring to last month (April) in Kanchama Kebele indicates that monthly application of Deltamethrin 1% has significant effect in tsetse flies density reduction *p (0.000)* than quarterly application *p (0.252*). This result agreed with the study of Vale and Torr (2004) they reported that monthly application of pyrethroid was the most effective method to kill tsetse flies .Also Alemu *et*

al. (2013) which revealed that the insecticide effectively works through week 5 but start declining at week 6 and totally fails on week 7. The test result also agreed with Hargrove *et al*. (2012) which Indicates that chemical sprayed remain on animal skin for at least two weeks and the efficacy decrease overtime. This is also consistent with the report of Damian *et al* (2014) and Jemaler *et al*. (2009). This was also agreed with a trial carried out in Ethiopia in which monthly application had resulted in a 98% decrease in apparent density of *G. pallidipes* (Leak *et al*., 1995).

The study result was also agreed with, Barry and Ejigo (2006) Survey result in konso distract indicates application of delametrin with four week interval reduce tsetse density. Similarly, the test results of Damian kajuguri *et al*. (2014) were indicated that the insecticide is only effective for 1-3 weeks. Also the findings of Thomson *et al* (1991) which showed that pour-on persisted for only about 40 days at Rekomitjie

The study result was also agreed with Vale *et al*. (1999) when chemical agents are used to combat tsetse flies; it must be taken into account that tsetse flies spend about 50% of their lifespan under the ground as pupae. Following the previously published work in Burkina Faso (Bauer *et al*., 1992) and Zimbabwe (Mangwiro and Wilson, 1993) it has commonly been accepted that Spot On persists for about 100 days. Present work emphasizes that the persistence can often be much less, in accord with the findings of Vale *et al*., (1999) and Thomson *et al*. (1991) it was considered that the insecticide was always re-applied when daily knockdown approached 50% .

According to study result of Fura and Kanchama Kabele, the proportion of females and male flies, Female fly accounted for 72.95 % and 62.6 % catch during this study in both kabele respectively. This result was somewhat approached to the report of Bekele *et al*.(2008) and Bancha (2001) where this indicated about 63.2% and 60% catch of female, respectively. This finding complies with finding of Efrem *et al.* (2013) in Lalo kile district, Kellem Wollega zone, which stated that the female tsetse flies physiologically necessitated feed more animal blood during pregnancy than males which exposes it to trapping than male tsetse flies**.** According to the report of Lovemore and Phelps (1994) higher catches of female *G.pallidipes* to be attributable due to their longer life span (average of 8 weeks) than males living about 4 weeks, so that more catch of females were observed. The test result were indicates the dominant Glossina species during pre

intervention and during intervention period was *Glossina pallideps*, this result agree with the previous study by Muturi *et al*. (2000).

Table 14: The effect of season in the effectiveness of Deltamethrin

Deltamethrin	Coef	Std.err	Z	p>|z|	(95%conf.Interval)	
session	-0.0121912	0.0394081	-0.31	0.757	-0.0894297	0.650473
Conc.	3.645161	.0594627	10.53	0.000	3.528617	3.761706

The test result indicated in Kanchama Kebele the tsetse fly density were decrease over the monitoring month, but there was long rain season were seen during study this was indicates that Deltamethrin 1% has similar effect during the seasons *p>0.05)*. This result was agreed with Gimonneau *et al.* (2016) reported that no significant difference in the mean knockdown in the dry and wet session *p (0.22*) and also Vale *et al*. (1999) the mean daily rain was no significant effect on mean knockdown Tsetse fly ($P > 0.1$) and the persistence period of insecticide was more constant, averaging around 80% in season.

monthly application of Deltamethrin 1% has effect in tsetse flies density reduction as indicate in figure 6 and table 10 tsetse flies significantly decrease from month one to seven *p<0.05*.

5. CONCULOSION AND RICOMENDATION

Application of deltamethiren 1% on cattle in monthly interval was adequately reduces tsetse fly population and trypanosomisis incidence with resultant improvement in PCV. Conversely, the use of quarterly application of delthametrin 1% results less when compared to motherly application. Quarterly application offers a slow reduction in trypanosomisis incidence and has slight variation in tsetse flies density during the first intervention when comparing to last monitoring time. So, the current study suggested that there was marked difference among the two. In another hand, practical lessons were taken from the use of pour on application on cattle was found easy to apply, requires less lobar and highly appreciated by user community. There was long rain season were seen during study but the tsetse fly density were decrease over the monitoring month in Kanchama kebel, this was indicates that Deltamethrin 1% has similar effect during the seasons.

Based on the above conclusion the following recommendation was given

- Deltamethrin application to tsetse fly control by pour on should be applied within monthly interval. Thus, when this activity integrated with other methods in the tsetse fly suppression activities may give good result with in short period.
- Community participation and awareness creation should be strengthened for the proper implementation and sustainability of control programs.
- Veterinary service should be expanded for the proper application of the control programs
- Further research should be done on the effect of season in control of tsetse fly by using Deltamethrin 1% pour on.

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7. APPENDIXE

Annex 1 Animal blood Sample Collection Format

No	ID	Owners Name	Age	Sex	Coat color	BCS	PCV	Parasitemia
1								
2								
3								
4								
5								
6								

Annex 2Entoomology Data Collection Formate

No/	Latitude	Longitude	Altitude	Veg[1]	St[2]. date	St time	End date	End time	Tsetse fly				
									Spp[3]	M[4]	F[5]	Und[6]	Total
1													
2													
3													
4													
5													
6													

[1] Vegetation, [2] Set, [3]species [4]Male, [5]Female [6] Undefined,

Annex 3 During Entomological Work

In kanchama Kebel

In kanchama Kebel

In Kanchama Kebel

In Fura Kebel

Annex4 during Deltamethrin 1% pour on

In Kanchama kebele

Annex5 during Blood Collection

In Fura kebele