SURVEY FOR HAEMATOPHAGOUS FLIES AND *TRYPANOSOMA SPECIES* IN CATTLE IN THREE SELECTED DISTRICTS OF GWAGWALADA AREA COUNCIL, FEDERAL,CAPITAL TERRITORY, NIGERIA.

BY

KARNAK PYENKWAM, DANDAM
DEPARTMENT OF VETERINARY PARASITOLOGY AND ENTOMOLOGY
FACULTY OF VETERINARY MEDICINE
AHMADU BELLO UNIVERISITY,
ZARIA, NIGERIA

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ABSTRACT

Biting flies remain important medically, Veterinary and by implication, economically. Beside the menace of biting caused by the members of haematophagous group, they also serve as vectors for several important pathogens. The study took a dry (February) and wet (August)season survey ofhaematophagous flies occurring in Dobi, Paikon-kore and Ibwa districts of Gwagwalada Area Council, Abuja, Nigeria. A total of 15Biconical and 15 Nzi traps were strategically set to trap flies within 48 hours period in each season within the three districts. Blood samples were collected from cattle in the areas where traps were set. A total of 243 cattle were sampled. The sampling (trapping of flies and collection of blood from cattle) were done in February and August. Flies caught in the traps were harvested, identified and counted. From the study, a total of 605 flies were caught during the sampling periods, and Paikon-kore had the highest number of trapped flies in August 186(75.6%) while Ibwa had the highest number of trapped flies for February 79(37.8%).The highest number of flies were caught in August (430) (71.1%) while in February, the number of flies caught was 175 (28.9%). From the study, *Stomoxysspecies* was the predominant fly in February and August, with highest occurrence of 79.1% in August while *Culex species* had the lowest occurrence of 0.2% in August. In February, *Stomoxysspecies* also had the highest occurrence of 64.6% while the lowest occurrence was by *Chrysomyaspecies* with 0.6%. In both February and August, Biconical trap was observed to be more efficient in trapping flies; catching more flies 71.6% while Nzi trap accounted for 28.4 % of the total of flies caught. There was significant association (p=0.001) between the flies occurrence and the two months of the year, but no statistical association (p= 0.000) between the number of flies caught and the trap used. Although *Glossina*spp were not caught, 2.5% of the sampled cattle were infected with *Trypanosomavivax.* In the analysed blood samples, other haemoparasites were identified which

includes; *Theileriamutans* 16(19.4%), *Babesiabovis(14.8%),Theileriavelifera (12.6%), Anaplasmamarginale(11.1%), Babesiabigemina (8.7) and Anaplasmacentrale(4.9%).*

From the findings it was concluded that *Glossina species* were absent or very scanty in the study area, *Stomoxyscalcitrans* was the dominating flies in all Districts. Biconical traps were more efficient than Nzi traps. The presence of *T. vivax* even though very low can pose a health risk to animals in the study area.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background of the Study

Insect-borne diseases exact a high public health burden and have a devastating impact on livestock, crops and humans. One of such diseases that have plagued sub-Saharan Africa is caused by protozoan African trypanosomes (the *Trypanosoma species*) and transmitted cyclically by a member of haematophagous fly (*Tsetse fly*) and mechanically by other members like *Stomoxys* (Aksoy and Rio, 2005). According to Lehane *et al.* (2003), tsetse flies transmit African trypanosomosis, leading to half a million cases annually and that the disease known as 'Nagana' in animals, remains the cause of massive losses on African Agricultural development. Over the past 100 years, a lot of efforts have been initiated, aimed at controlling tsetse fly. There has however been limited impact in terms of reducing the losses (Anon, 2002). Some of the areas where tsetse fly populations were considerably reduced have become re-infested due to, in most cases, lack of consolidation of achievements (Oluwafemi *et al.,* 2001). Many species of Haematophagous dipterans are responsible for the spread of diseases in man and his livestock (Gruvel, 1980). They are categorized as those transmitting cyclically (Molyneaux and Ashford, 1983), mechanically (Dirie *et al.*, 1989) or by regurgitation (Coleman and Gerhardt, 1988; Straif *et al.,* 1990).

Haematophagous flies are large biting flies in Africa which live by feeding on the blood of vertebrate animals. They are similar to other large flies such as the housefly (*Musca domestica),* but can be differentiated by two visually distinctive features such as the forward projecting proboscis and the discal cell on the wing, shaped like a cleaver and referred to as the 'hatchet cell' which lies between the fourth and fifth wing veination (Buxton, 1955).

The Tsetse fly belongs to the genus *Glossina*, a name described by Wiedemann (1830) and monotypic family *Glossinidae*. They have existed in the modern morphological form for at least 34 million years since Fossil tsetse were recovered from the Florissant Fossil Beds in Colorado (Cockerrel, 1917).

1.2 Statement of Research Problem

Haematophagous flies, especially tsetse fly, are among several factors that constrain livestock sector development in Nigeria and sub- Sahara Africa in general. They are vectors of trypanosomosis, a disease which lowers productivity in livestock, reduces cattle density by 70%, sale of meat and milk by 50%, calving rates by 20% and increase calf mortality by 20% (Swallow, 2000). Tsetse flies, through the cyclical transmission of trypanosomosis to both humans and their animals, greatly influence food production, natural-resource utilization and the pattern of human settlement throughout much of sub-Saharan Africa (FAO, 2000). It is estimated that the annual direct production losses in cattle alone amount to between US$6billion and $12billion, while animal deaths is about 3 million (FAO, 1993). Trypanosomosis is a devastating disease that affects livestock and brings economic hardship on the livestock owners not only through low production of milk, meat and work performance but also from financial losses on drugs. According to Lehane *et al.* (2003), tsetse flies transmit African trypanosomosis leading to half a million cases annually and that the disease known as 'Nagana' in animals remains a massive brake on African Agricultural development. Trypanosomosis is an important constraint to livestock farming and mixed cropping in tropical Africa. More than a third of the land area across Africa is infested with tsetse flies (8.7 million km^2), where at least 46 million cattle are exposed to the risk of contracting tsetse-borne trypanosomosis, as are millions of sheep, goats, donkeys, camels and horses (Reid *et al*., 2001). African livestock producers are administering an estimated 35 million curative and preventive treatments annually (Geerts and Holmes, 1997).

According to them a price of approximately $1 per treatment, the disease is costing producers and governments at least $35 million per year and assuming an average of two treatments per animal annually, this implies that 17.5 million animals are treated each year for the disease, or 38% of those at risk thereby generally constraining farmers from the overall benefits of livestock to farming sufficient nutrient cycling, less access to animal traction, lower income from milk and meat sales, less access to liquid capital. The effects of trypanosomosis on these animals range from anaemia, immunosuppression, retarded growth, low milk production, low meat quality, and weight loss as well as infertility, abortion, stillbirth and depression of reproductive performance and reduced capacity to work (Snow *et al*., 1996; Sekoni, 1994). These damaging effects and the manpower and material resources committed to the disease result in financial loss of more than $23.8 million annually in Nigeria (Milligan and de Leeuw1983). This study was therefore, initiated to obtain the prevalence of haematophagous flies and trypanosomes in three districts of Gwagwalada Area Council of the Federal Capital Territory Abuja, Nigeria, to obtain reasonable information about haemoparasitic infection in the livestock in the study area as a useful parameter to design a strategy for haemoparasitic disease control.

1.3 Justification

The presence of haematophagous flies excludes livestock from large areas of considerable agricultural potential, by virtue of the severity of the diseases caused by tsetse-transmitted trypanosomes. Animals have a central role in most African societies, and they provide milk, meat, manure, hides and skins as well as valuable draught power. Livestock represent a means of accumulating and distributing wealth, and have great social significance. Control of *Trypansonosoma* spp and Haematophagous flies may be aimed against either the flies or the trypanosomes. At the farmer's level, trypanocides provide a way for the individual to take action to control the disease. With few exceptions, throughout Africa, governments have lacked the

resources to continue to provide effective Veterinary Services to control trypanosomosis, among other diseases. Survey of Haematophagous flies and trypanosomes will be a step in the right direction in the strategy to control the vector and aetiologic agent of trypanosomosis in the process it will lead to improve animal production in Nigeria and help to reduce the economic waste in treating the disease by the livestock owners. Gwagwalada Area council is situated in the Savanna area which is tsetse endemic and a favorable settlement area for cattle nomads (Ohaeri and Eluwa 2007). Therefore, there is a need to undertake this study to ascertain the presence of Haematophagous flies and trypanosomes infection in the area and then suggest haematotophagous flies control measures.

1.4 Aim of the Study

To survey for Haematophagous flies and *Trypanosoma spp* in cattle in three (3) districts of Gwagwalada Area Council, FCT, Nigeria.

1.5 Objectives of the Study

The objectives of the study were to:

1. Investigate the presence and distribution of Haematophagous flies in three (3) districts of Gwagwalada Area Council through trapping.
2. Compare the trapping efficiency Biconical and Nzi traps on Haematophagous flies.
3. Determine the occurrence of *Trypanosoma* species in sampled cattle in the 3 districts of Gwagwalada Area Council, F.C.T. Nigeria.

1.6 Research Questions

1. Are there haematophagous flies in Gwagwalada Area Council?
2. Which of the two traps used (Biconical or Nzi) is more efficient in trapping these heamatophagous flies?
3. Are there *Trypanosoma spp* infecting cattle in Gwagwalada Area Council?

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Historical Perspective

Hematophagous flies are among those species belonging to the families *Glossinidae* and *Tabanidae* and the genus *Stomoxys*. Prominent in this group is *Glossina* a vector of trypanosomes which has existed for 200 million years and the trypanosome has existed for 300 million years (Pieter, 2002). Sleeping sickness was recorded earliest from the upper Niger during the 14th century when Ibn Khaldoun, an Egyptian historian wrote on the disease in his account of the history of North Africa (Lambrecht,1985). The disease in West Africa caused visible swollen lymph gland and was known as winterbottom's sign after the description of Winterbottom in 1803. This symptom was readily recognized by slave traders who avoided trading and buying slaves with such signs. Forde and Dutton in 1902 found for the first time, a trypanosome infection in man and called it "trypanosome fever" (WHO, 2005). The presence of trypanosomes in cerebrospinal fluid taken from a sleeping sickness patient was observed by Castellani which he identified a year later as the cause of sleeping sickness (Pieter, 2002). The relationship between tsetse flies, the trypanosomes and the diseases was discovered in 1895 (Grev, 2002) , by Bruce who correctly recognized that trypanosomes were the causative agent of sleeping sickness transmitted to humans by tsetse flies and that "trypanosome fever" and "sleeping sickness" both thought to be different diseases at the time were in fact the same.

2.2 Tsetse Fly (*Glossina spp*)

Tsetse flies are important medical and veterinary vectors of the African trypanosomes, the agent of sleeping sickness in human and animal trypanosomosis, also known as Nagana (Batista *et al*., 2011). Tsetse flies are blood sucking dipterans, both males and females feed exclusively on the blood of vertebrates for few minutes every 2-3 days (Syed and Guerin, 2004). The flies seek cover from high temperature to conserve energy and plants provide shelter in all the biotypes. In

some cases, the flies take cover in plantation and under invasive bush in many areas of tsetse fly belt of Africa and flies from such refugia are implicated in sleeping sickness epidemics (Syed and Guerin, 2004).

Trypanosomes replicate in the tsetse fly and are transmitted by the bite of the tsetse fly when the fly feeds on animals and man.

2.2.1 Classification of tsetse fly

Kindom: *Animalia*, Phylum: *Arthropoda,* Class: *Insecta* Order: *Diptera,* Sub section: *Calyptratae,* Super family: *Hyppoboscoidea,* Family*: Glossinidae (Theobald, 1903),* Genus: *Glossina* (Wiedemann, 1830)

2.2.2 Habitat and distribution

There are 30 species and subspecies of tsetse fly all in the genus Glossina, with three (3) major species groups of tsetse flies responsible for transmission of trypanosomes. The genus *Glossina* occupies over some 11 million km^2 of Africa.The Northern limit extends across 14^oN but in Somalia it is only about 4^oN. The Northern limit corresponds closely to the Southern edges of the Sahara and Somalia Deserts. The Southern limit is less well defined. In the South West it ranges between $10^o$and 20^oS, corresponding closely to the Northern edges of the Kalahari and Namibian Deserts, whereas in the South East, it is generally at about 20^oS but extends as far as 29^oS along the East African littoral (Jordan, 1986), with the mid point of infestation at about 7^oS (Davies, 1977). In Nigeria, tsetse flies occupy about 75% of the landmass from latitude 4^o to 13^oN and longitude 2^o to 15^oE, an area covering all the six agro-ecological zones of the country (Onyiah, 1997).

Twenty two species of *Glossina* exists, and are divided into three distinct species groups based on habitat preference (Ford, 1970) and morphological differences in the structure of the male

genitalia (Pollock, 2000). These species are: The *Morsitans* species group (Morsitans) these include seven species and subspecies namely:- *Glossina longipalpis; G. pallidipes; G. morsitans morsitans; G. morsitans submorsitans; G. morsitans centralis; G. swynnertoni; G. austeni.*

The Palpalis groups (Nemorhina) consists of a total of nine species and subspecies namely: - *Glossina palpalis palpalis; G. palpalis gambiensis; G. fuscipes fuscipes; G. fuscipes martini; G. fuscipes quanzensis; G. tachinoides; G. pallicera pallicera; G. pallicera newsteadi; G. calliginea.*

The Fusca species group consists of fourteen species and subspecies namely: - *Glossina fusca fusca; G. fusca congolensis; G. nigrofusca nigrofusca; G. nigrofusca hopkinsi; G. fuscipleuris; G. haningtoni; G. schwetzi; G. tabaniformis; G. nashi; G. vanhoofi; G. medicorum; G. severini; G.brevipalpis; G. longipennis.*

The geographical distribution of tsetse flies is determined by temperature (with mean annual temperature of between 20^{o}C and 28^{o}C), relative humidity (ranging between 50% and 85%), rainfall; sunshine and vegetation cover (Glasgow, 1970). In higher altitudes of over 1800m (6000ft), tsetse flies are not found (Davies, 1977).

2.2.3 The morsitans group

These are exceptionally good vectors of trypanosomes; all the seven species are potential vectors of both human and animal trypanosomosis. All species in this group inhabit the Savanna woodlands that surround the two major blocks of lowland rainforests in Africa, and are referred to as 'game or savanna flies' (Davies, 1977). In wetter areas, the flies are observed to disperse more widely over the woodland, but in drier areas their movements are restricted to the mesophytic vegetation of the watercourses, particularly during the severe dry season (Nash, 1937; Jordan, 1986). In Eastern and Southern Africa where *Glossina morstans morsitans* is the

primary vector of human and animal trypanosomosis, the 'Miombo' woodland (Brachystegia – Jilbernardia) that extends from Mozambique to Tanzania, as well as the 'Mopane' woodlands (Colophospermum mopane) in Zambia and Zimbabwe are typical habitats. *Glossina morsitanscentralis* dominates northwards from Botswana and Angola into southern Uganda. *Glossina morsitans submorsitans* has an east to west distribution from Ethiopia to Senegal in 'doka' woodland where the vegetation is dominated by *Isoberlinia doka* species, and can be sporadically found to occur in the Southern Guinea Savanna vegetation zone as well as the drier Sudan zone (Jordan, 1986). *Glossina swynnertoni* is restricted to a small area between Tanzania (Serengeti) and southern Kenya (Masaimara) where Acacia- commiphora vegetation can be found, with abundant wild life. *Glossina longipalpis* and *Glossina pallidipes* both have a much wider range of possible habitats displaying versatility by existing in different vegetation types. *Glossina longipalpis* occurs in the narrow savanna belt just north of the rainforest in West Africa, from Guinea to Cameroon while *Glossina pallidipes* occurs in East Africa from Mozambique to Ethiopia over a relatively wide range of climatic and vegetation conditions. Finally, *Glossina austeni* occupies secondary shrub, thickets and islands of forests along the East African coast from Mozambique to Somalia. Its distribution is discontinuous, rarely being found at altitudes over 200m or more than 250km inland from the coast (Jordan, 1986). In Nigeria, the two morsitans groups' species present are: - *Glossina morsitans submorsitans* found in the North where annual rainfall is as low as 635mm (25in) with a dry season of seven months and further South, with an annual rainfall of 1400mm (55in) and a dry season of four months. *Glossina longipalpis* is found in the Southern guinea and derived Savanna zones in the West and small localities in the East, with annual rainfall not less than 1150mm 45in) or more than 2300mm (90in) (Davies, 1977).

2.2.4 The palpalis group

There are nine species in the palpalis subgenera, with only five palpalis and fuscipes subspecies as vectors of both human and animal trypanosomosis. Although flies in this group are continuously found in the lowland rainforest, some are known to extend out of the Savanna region, particularly along rivers and streams. The habitat of the palpalis flies is mainly the drainage systems leading to the Atlantic or the Mediterranean ocean which extends from the wet mangrove and rainforests along the coastal regions of West Africa to the drier Savanna areas at Northern part of the rainforests. The palpalis group is less tolerant to the wide range of climatic conditions of the Savanna belt, and are therefore restricted to the ecoclimate of the water courses from where they derived their label as the 'riverine species'. Many of the *G. Palpalis species*, like the *Glossina palpalis palpalis* in Cote d'lvoire prefer peri-domestic conditions and have been observed to maintain close association with villages (Baldry, 1980). The advancement of *Glossina tachinoides* in stern Cote d'lvoire and Togo have been attributed to intense agricultural development of the rapid human population growth around the plantation (Hendrick, and Napala, 1997). In general, most of the Palpalis group flies are less suited to desiccating conditions, and therefore survive in thick riverine forests with enough shelter from wind and heat. This is especially the case for the three fuscipes subspecies which are confined to hygrophytic habitats rarely far from open water lacustrine or riverine habitats. *Glossina tachinoides*, although typically a riverine species, were found in Northern Nigeria to extend into human-inhabited savanna woodlands during the wet season, displaying strong adaptation to peri-domestic habitats (Kuzoe, 1985; Ahmed, 2004).

2.2.5 The fusca group

All the tsetse flies in the Fusca group are found in West African forests except *Glossina brevipalpis* and *Glossina longipennis*. None of the species in the Fucsa group is a vector of human trypanosomosis. However both *Glossina fusca* and *Glossina medicorum* are efficient vectors of trypanosomosis to livestock (mainly *Trypanosoma vivax*) causing considerable economic losses (Jordan, 1986). Distribution of the Fusca group depends primarily on forest vegetation and climatic factors. Fusca group species inhabit moist, evergreen habitats either in riverine forests with Savanna (*Glossina medicorum*) or in dense and wet rainforests (*Glossina tabaniformis* and *Glossina nigrofusca*) except *Glossina longipennis,* in contrast to the rest, *Glossina longipennis* lives in one of the driest habitats inhabited by tsetse flies (Jordan, 1986). Due to its pupal adaptation to dry conditions, their primary habitats consisting of dry deciduous acacia bush- are discontinuously spread throughout East Africa (Glasgow, 1963).

2.2.6. Description of *Glossina*

Glossina species have been widely studied because of their economic importance as major transmitters of animal and human trypanosomiasis (Ndams, 1987). Tsetse flies are robust insects measuring about 6-15mm in length. They are readily distinguished from other biting flies morphologically by the combinations of useful features such as widely separated compound eyes which are dark brown, and able to detect moving objects at 137meters (150yards) (Pollock, 1982), with forwardly projecting proboscis which sticks out horizontally from the front of the head (Jordan,1993). They differ from most biting flies and non cyclorrhaphous insects because when at rest, the wings are held over the other such that they overlap like the blade of a closed pair of scissors, thus revealing part of the abdomen (Davies, 1977). They also posses a characteristic wing veination where the discal medial cell of the wing is shaped like a butcher's cleaver (referred to as the 'Hatchet cell') and a distinctive row of branched hairs on the arista of the antenna (Service, 1980; Jordan, 1993). Tsetse flies range in color from yellowish or grayish

to dark or almost blackish-brown, sometimes there is a slight pink or sandy red tinge (Davies, 1977). The dorsal surface has a pattern of dark brown stripes and patches making the insect difficult to see when settled on tree bark, rock and soil. They have seven visible abdominal segments (Pollock, 1982).

2.2.7 Dependence of tsetse species on various hosts.

See the table below for the distribution of Glossina based on Host

Table 1: Glossina species and their animal hosts

Glossina longipalpis	*Glossina morsitans*
(a) Bushbuck *	(a) Warthog
(b) Buffalo	(b)Ox, buffalo, kudu and man
(c) Red river hog	
Glossina pallidipes	*Gossina swynnertoni*
(a) Bushbuck *	(a) Warthog *
(b) Warthog	(b) Buffalo, giraffe, rhinoceros
(c) Bushpig	
(d) Buffalo	
Glossina austeni	*Glossina palpalis*
(a) Bushpig *	(a) Man, reptiles
(b) Ox	(b) Bushbuck, ox
(c) Duiker	
Glossina fuscipes	*Glossina tachinoides*
(a) Reptiles, bushbuck	(a) Man
(b) Man	(b) Ox
	(c) Porcupine
Glossina fusca	*Glossina fuscipleuris*
(a) Bushbuck *	(a) Bush pig
(b) Red river hog,	(b) Giant forest hog aardvark
c) Ox, hippopotamus	
Glossina tabaniformis	*Glossina brevipalpis*
(a) Red river hog *	(a) Bush pig, hippopotamus
(b) Porcupine	(b) Bushbuck, buffalo
Glossina longipennis	
(a) Rhinoceros *	
(b) Buffalo, elephant .	

* The host that accounts for more than half of the total feeds. Source: Pollock (1982)

2.2.8 Host groups and their relative importance

Some species of wild animals are very important as hosts for tsetse, and others are less important. The type of host that is important may depend on the availability of the host animal, the species of tsetse, and on the season (FAO, 2000).

2.2.8.1 The pig family (*Suidae*)

The pig family contains the warthog, bushpig, red river hog and the giant forest hog, as well as the domestic pig. Members of the pig family are very important as hosts of tsetse.

i. *Glossina swynnertoni*: feeds mainly on warthog (60–70%).
ii. *Glossina austeni*: feeds mainly on bushpig (50–60%).
iii. *Glossina fuscipleuris*: feeds mainly on bushpig and giant forest hog (65%).
iv. *Glossina tabaniformis:* feeds mainly on the red river hog (70%).
v. *Glossina morsitans* gets from one third to nearly half (30–45%) of its food from warthog.
vi. *Glossina fusca* may take 15% of its meals from the red river hog.
vii. *Glossina brevipalpis* may take as much as 40% of its meals from the bushpig, but this varies considerably according to the area *Glossina palpalis, G. fuscipes* and *G. tachinoides* generally do not feed on wild pigs very much (about 3%); but *G. tachinoides* and *G. palpalis* near to villages in southern parts of their range may get much of their food from the domestic pig (FAO, 2000).

2.2.8.2 *The antelope, buffalo and cattle (ox) family* (*Bovidae*)

The bovids include a large number of important species, especially antelopes such as bushbuck, kudu and eland, as well as buffalo and cattle. In general, the larger members may be important hosts, but the smaller ones are much less important.

i. *Glossina pallidipes, G. longipalpis* and *G. fusca* get most of their blood meals (55–90%) from the bushbuck.

ii. *Glossina morsitans* takes 25–40% of all its feeds from bovids, especially kudu, buffalo, bushbuck and eland. Cattle are very readily fed upon.

iii. *Glossina brevipalpis* takes about one quarter of all its meals from bovids (buffalo and bushbuck).

iv. *Glossina austeni* takes about one third of its meals from bovids; duiker (various species) may be important to this fly (10%).

v. *Glossina palpalis, G. fuscipes and G. tachinoides* take about 20–40% of their meals from bovids. The exact percentage, and the species fed upon, varies greatly according to local conditions. Cattle are fed upon if they come into infested areas.

vi. *Glossina longipennis* takes about 20% of its feeds from bovids. (FAO, 2000).

2.2.8.3 Primates, including man

The primates are a group of mammals into which scientists place man for the purpose of comparison with other animals. The monkey and the baboon come into the same group.

i. *Glossina palpalis, G. fuscipes and G. tachinoides* all feed on man; the percentage of feeds may be between 8–40% according to local conditions. Flies take a high proportion of their meals from people using watering places and river points.

ii. *Glossina morsitans* may take 7–18% of its meals from primates (mainly man).

iii. Other tsetse species take 5% or less of their meals from primates (FAO, 2000).

2.2.8.4 Some mammals used as hosts.

i. *Glossina longipennis* is unusual in that about 60% of its feeds come from rhinoceros. Elephant is an important but variable host (up to 12% of all feeds).

ii. *Glossina brevipalpis* may take up to 36% of its food from hippopotamus.

iii. *Glossina fuscipleuris* may take up to 20% of its food from hippopotamus.

iv. *Glossina fusca* takes about 12% of its meals from the aardvark.

v. *Glossina swynnertoni* feeds on the giraffe to some extent (about 8%).

vi. *Glossina tabaniformis* takes up to one quarter of its food from the porcupine.

vii. *Glossina tachinoides* may take over 7% of its food from the porcupine (FAO, 2000).

2.2.8.5 Some Mammals seldom used as hosts.

The following common wild game animals are not fed upon by tsetse under natural conditions: Zebra, wilderbeast, many small antelopes. The following common wild game animals are fed upon only quite rarely: Waterbuck , Impala , Hartebeest (except that *Glossina morsitans*has been recorded as using hartebeest for about 2% of its meals).

In general, animals smaller than a porcupine or a duiker are not fed upon by tsetse flies in the wild. Very active animals such as monkeys are not much fed upon (FAO, 2000).

2.2.8.6 Birds

Birds are not used very much by tsetse flies as a rule.

i. *Glossina longipennis* may take up to 7% of its meals from the ostrich.

ii. *Glossina morsitans submorsitans* may take about 6% of its meals from birds other than the ostrich.

iii. *Glossina palpalis and G. fuscipes* may take about 10% of their meal from waterside birds such as cormorants (FAO 2000).

2.2.8.7 Reptiles.

The main reptiles fed upon are monitor lizards (*Varanus*) and crocodile. These animals are very important as hosts for palpalis group flies.

i. *Glossina palpalis and G. fuscipes* may take about a quarter to a third of their food from these hosts, possibly more in quiet places uninhabited by man.

ii. *Glossina tachinoides* takes about 10% of its food from these reptiles. (FAO 2000).

2.2.9 Sex determination

A tsetse fly can be identified as a male or a female by examining the posterior end of the abdomen. The male has a lump on the underside (ventral side) of the abdomen at the posterior end. This is the hypopygium; just in front of the hypopygium is a plate with strong black hairs (hectors). The hypopygium and the plate with hectors are not present in the female fly. These structures can be seen with the naked eye, and more clearly by using an x10 hand lens. This is the best way of distinguishing the sexes of tsetse flies. The female fly is often slightly larger than the male. In a collection of dead flies of one species, the larger flies can be quickly picked out, and these will be found to be mainly females. But because some females are smaller than some of the larger males, size is not a very reliable indicator of sex (Pollock, 1982).

2.2.10 *Glossina* reproduction

Tsetse flies are adenotrophic viviparous (Hagan, 1951) because the egg and larva stages develop within the fly. The egg contains sufficient yolk for embryonic development and the larva in the uterus is nourished by special maternal organs. The result of viviparity is that only a small number of fully developed larvae can be produced, a free-larval stage is practically eliminated and both adults and immature stages are dependent upon the same source of food (blood) (Saunders, 1960). Female tsetse flies mate within a day after emergence from their pupal case; mating usually takes place near or on host animals (Pollock, 1982). Male and female generally meet when the female is about to take the first blood meal (Jordan, 1986). The sexes remain in copulo for an hour or two and during this time a spermatophore is formed within the uterus of the female. At the end of the copulating period, the male jerks vigorously and at this time the sperms

are ejaculated into the spermatophore (Pollock, 1982). In the next few hours, the sperms are slowly released from the spermatophore and move into the spermathecae. Active and viable sperms can remain in the spermathecae throughout the life of the female fertilizing each egg as it is produced from the ovaries. This enables the female to breed throughout her life time (Davies, 1977). Female tsetse flies usually mate only once in their lives but some may mate more than once males can mate several times. Older males are better able to mate successfully than very young ones (Pollock, 1982).

2.2.11 Egg stage

Eggs are fertilized immediately it enters the uterus by sperm from the spermathecae coming into contact with and penetrating the anterior part of the egg. The fertilized egg remains lying in the uterus for about four days, while development of the first instar larva takes place inside. The egg is about 1.6 mm long (FAO, 2000).

2.2.12 Larval stages

The larva in *Glossina* passes through several stages or instars, as it grows (Davis,1977). There are three larval instars in *Glossina* referred to as first, second and third larva instars. The larva has a mouth at the anterior end, and two posterior spiracles. The unusual feature of the *Glossina* life history is that the larva spends practically all its time, and does all its feeding within the body of the female fly (FAO, 2000).

i. First instar larva; This is the stage that emerges from the egg. The first instar grows to 1.8 mm (*G.morsitan)* before changing to the next stage by getting rid of its old tegument and lasts for 1 day.

ii. Second instar larva; This stage has rapid growth and development. There is swelling at either side of the posterior spiracles. The second instar lasts two days, and the larva grows to a length of 4.5 mm (*G. morsitans*). (FAO, 2000)

iii. Third instar larva; There is rapid growth and development at this stage, fully grown larva has a pair of large black swellings at the posterior end with presence of polypneustic lobes, which carry many small holes for breathing. The third instar lasts for over two days and the larva grows to a length of 6–7 mm (*G. morsitans)* (FOA, 2000).

2.2.13 Survival of the larva.

Apart from the food already in the egg, all the food of the three larval instars comes from the milk gland of the mother fly. The milky secretion of this gland is poured out of the duct of the gland, at the head end of the larva. The larva sucks up this secretion and passes it straight to the midgut. In the midgut, the food is slowly digested and assimilated. The air supply to the larva depends on air entering the vulva of the female and then passing into its posterior spiracles or polypneustic lobes.With fully grown larva in the female *Glossina*, it flies around looking for a suitable area to drop the larva. This is usually at a place where there is a patch of loose sandy soil, sheltered by an overhanging rock, branch or twig. The female tsetse settles down either on the ground or on the overhanging object. The larva burrows into the ground and hides. The female flies away. The larva becomes barrel-shaped, darkens and may then be called a pupa. There is no feeding by the larva after it is dropped by the female this process is call larviposition (FAO, 2000). Abortion occurs when a larva fails to reach its full size and is expelled from the uterus before the usual time of larviposition. This is called an abortion and the aborted larva dies. Abortions can be caused by the mother fly not obtaining sufficient food, and may also occur when the fly is carelessly handled, or when it comes into contact with insecticide. The egg may also be aborted for the same reasons, (Pollock, 2000)

2.2.14 Pupa

It is a dark brown rounded object. At its posterior end are the polypneustic lobes the shape of which helps to distinguish the tsetse pupa from the pupae of other flies. The pupa is slightly shorter than the larva that produces it. The food still remained in the midgut is digested and assimilated.The pupal stage usually lasts about four to five weeks, depending on the temperature. Higher temperatures shorten the pupal period while lower temperatures lengthen the pupal period (to more than 50 days in some climates). Too high or too low a temperature will cause the death of the pupa (FAO, 2000). At the end of this period, the adult fly is ready to emerge.

2.2.15. The mature fly.

The body of the adult fly emerges from pupacase and also gets through the surrounding soil by using the ptilinum. The young fly struggles to the top of the soil and comes out into the open air (Pollock, 1982). The body at this stage is very soft and the wings are not fully developed. From the time the fly emerges to the taking of its first meal, the young fly is called a teneral fly (FAO, 2000). After the first blood meal has been taken, the underside of the abdomen appears creamy yellow and transparent, the thorax feels firmer and harder, because of the greater development of muscles in it. At this stage, ptilinum cannot easily be everted. The fly is referred to as a non-teneral fly at this atage. Once mated, a female can produce larvae for the rest of her life. At a temperature of about 25°C a female fly will produce a mature larva every 9–10 days, except for the first one which may take 18–20 days from the time of emergence of the fly from the puparium. Lower temperatures give a lower rate of breeding (FAO, 2000).

2.2.16. Disease transmission by tsetse fly

Tsetse fly is the vector of the protozoan parasite of the Genus *Trypanosoma*, the agents of human and animal trypanosomosis in sub-Saharan Africa (Hu and Askoy, 2006). In West Africa, the

human trypanosomosis caused by *Trypanosoma brucei gambiense* is transmitted by *Glossina palpalis* and *Glossina tachinoides*. It is described as Gambian sleeping sickness (Dutton, 1902; Abenga and Lawal 2005), whereas animal trypanosomosis caused by *Trypanosoma brucei brucei* is transmitted by *Glossina submorsitans* and *Glossina longipalpis*. In East Africa, both the human and animal trypanosomiasis caused by *Trypanosoma brucei rhodesiense* and *Trypanosoma brucei brucei* respectively are transmitted by *Glossina morsitans, Glossina pallidipes* and to some extent *Glossina palpalis* (Willet, 1970; Grev, 2002*).* The East African form described as Rhodesian form (Stephen and Fantham, 1910) is a more acute, lasting for weeks or months (Garcia *et al*., 2006). Trypanosomes exist as trypomastigote in the blood and lymph in infected animals (Vreysen, 1995).

When trypanosomes are ingested by tsetse fly, they undergo a cycle of development within the fly. In the gut, they transform into the trypomastigote form and move into the mouthpart (labrum and/or hypopharynx) where they develop into the epimastigote form. They later reproduce by binary fission to produce the metacyclic form which is the infective form. This final binary fission takes place in the proboscis, midgut or salivary gland depending on the species of trypanosome (Jordan, 1986). However, transmission of trypanosomes can also occur from one mammalian host to the other in or on the mouthparts of various species of haematophagous flies such as *Tabanus, Stomoxys, Chrysops* etc including tsetse. This process is known as mechanical transmission (Jordan, 1986). This cycle of development varies in duration depending on the species of trypanosome, species of tsetse fly, temperature, reservoir host, age, sex. For *Trypanosoma vivax*, *Trypanosoma congolense* and *Trypanosoma brucei brucei*, it varies from 5 – 13 days at 22ºC - 29ºC; 12 -53 days and 11 – 60 days respectively (Davies, 1977; Jordan, 1986), Various authors have reported that *Trypanosoma vivax* have the highest infection rate, followed by *Trypanosoma congolense* and then *Trypanosoma brucei brucei* (Onyiah, 1997;

Omotainse *et al*., 2000). Temperatures increase the rate of breeding. However, temperatures that are too high or too low will cause breeding to stop altogether.

2.3 *Stomoxys calcitrans.*

Stomoxys calcitrans is the commonest specie of this genus and is known as the stable fly or biting housefly. It occurs all over the world and the host includes most animals and man (Zumpt, 1973).

2.3.1 Morphology of *Stomoxys calcitrans*.

Superficially, *S. calcitrans* resembles the housefly *M. domestica*, being similar in size and grey with four longitudinal dark stripes on the thorax. Its abdomen however is shorter and broader than *Musca* with three dark spots on the second and third abdominal segments. The proboscis is conspicuous and forward projecting which differentiates from Musca and other genera of non-biting muscid-flies. Stable fly can be distinguished from biting muscid flies of the genus *Haematobia* by its bigger size and the shorter pulp of stable flies (Urquhart, 1996).

2.3.2 Life cycle

Stomoxys sometimes lays its eggs on horse manure, but prefers decaying vegetable matter like straw and hay, especially when contaminated with urine. The female lays batches of 25-50 eggs, resembling those of house flies. Eggs hatch in 1-4 days, or longer in cold weather, and the larva are mature in 6-30 days. After emergence the adult female require several blood meals before the ovaries mature and egg laying can start. The complete life cycle from egg to adult fly may take 12-60 days depending mainly on temperature (Urquhart, 1996).

2.3.3 Feeding and habitat

When feeding, the proboscis swings downwards and skin penetration is achieved by the rasping action of fine teeth on the end of the labium. Approximately three minutes is required for a blood

meal and feeding is often interrupted, thus allowing mechanical transmission of micro-organisms. The flies are most abundant in summer and autumn and live about a month under natural conditions. They enter buildings only in autumn or during rainy weather. They are swift flies; but do not travel along distances (Soulsby, 1982).

2.4 Tabanus (Horse Fly)

These insects, commonly known as 'horseflies', are also called breeze flies, are large robust flies with wings and large eyes (Soulsby,1982).The pain caused by their bites leads to interrupts feeding and as consequence, flies may feed on succession of hosts and are therefore important in the mechanical transmission of pathogens such as trypanosomes (Urquhart, 1996). During feeding, horseflies transmit to hosts pathogenic viruses (rinderpest), bacteria (haemoragic septicaemia), Protozoa (trypanosomosis) and helminthes (loaiosis) (Foil, 1989). The painful bites of tabanids also influence parameters of fitness in farm animals. These include reduction in: weight gain, milk production, feeding and feed conversion rates, blood, and traction power. Moreover, distraction and irritation by bites predispose animals to accidents and predation (Soulsby, 1982) .There are many genera of tabanids, but only three are of veterinary significance, namely; *Tabanus, Haematopota* and *Chrysops*. Since the three genera are closely related in behaviour and pathogenic significance they are consisered as a group.

2.4.1 Distribution and host

Distribution is worldwide although certain genera are absent from large area for example, there are no *Haematopota* species in Australia or North and South America (Urquhart *et al*.,1996).The tabanid flies have been recovered from localities at sea-level and at altitude of up to 10,000 feet

(Nnochiri,1974). The host is generally large domestic or wild animals and man, but some small mammals and birds may also be attacked (Urquhart, 1996).

2.4.2 Morphology

These are medium to large biting flies, up to 2,5cm in length, with wing spans of up to 6.5cm.They are generally dark coloured, but may have various stripes or patches of colour in the abdomen or thorax which varies from brown and red to yellow. They have large coloured compound eyes which occupy a wide area of an equally semi lunar head. The eyes are dichoptic in the female and holoptic in the male. The wings are broad and are characterized by their marginal cells. The coloration of the wings is useful in differentiating the three major genera: thus *Tabanus* has clear or brownish wings, while there are often dark bands across the wings in Chrysops. In contrast, *Haematopota* has ruffled or speckled wings (Urquhart *et al*., 1996) They have a short, stout, interiorly projecting antennae consisting of three markedly differentiated segments (Hewitt ,1914).The first segment of the antennae is small, the second may be expanded, and the third is marked by annulations that make tabanid antennae appear to consist of many more than three units.

The antennae are also useful in generic differentiation. In species of the Genus *Chrysops* the first and second segments of the antennae are long: the third (terminal) segment has four annulations. While in species of the genus *Haematopota* the first segment of the antennae is large and the second segment narrower, while the terminal segment has three annulations (Soulsby, 1982).

The mouthparts, which are adopted for slashing/sponging, are short and strong and always point downwards. The short labium which is grooved dorsally to take the other mouth parts, collectively termed the biting fasicicle is most prominent. The labium is also expanded terminally as paired large labella which carry tubes called pseudotrachea through which the blood or fliud from wounds is aspirated, The biting fascicle, which create the wound, consist of

six elements, the upper sharp labrum, hypopharynx with its salivary ducts paired rasp like maxillae on pared boad pointed mandibles. Male flies have no mandibles and therefore cannot feed on blood (Urquhart, 1996).

2.4.3 Life cycle.

After a blood meal the female lays batches of several hundred creamy-white or grayish cigar-shaped eggs, 1.0-2.5mm long, on the underside of vegetation or on stones, generally in muddy or marshy areas. The larvae hatch after four to seven days and drop into the water, or mud, into which they disappear. They are maggot-like and the body has 11 segments, besides the cephalic portion of which is conspicuous. Each segment has weight fleshy tubercles. The mouth parts are prehensile and masticatory. The larvae are carnivorous. There are three jointed antennae and the large lateral tracheae open on the penultimate segment, which also bears a retractile siphoned tube. The larvae feed on small crustaceans, or even on one another, and grow for two or three months, performing several ecdyses. Finally, they pass through a quiescent stage and then pupate. The pupa is brown and sub cylindrical; the abdominal segments are movable and in the anterior part the appendages of the Imago can be distinguished. These stages last for about 10-14 days. The whole life-cycle takes four to five months under favorable conditions, but low temperature prolongs development and the larvae may hibernate (Soulsby, 1982).

2.4.4 Feeding and habitat

Unlike the males which feed only on vegetable sugars (nectar), female tabanid flies attack animals and in addition, feed on plant juices. Feeding occurs during day light hours especially early in the morning and late in the afternoon. A blood meal is usually necessary for the development of the ovaries. With the exception of *Chrysops silacea* which feeds indoors, most

tabanids are outdoor feeders. Some feed mainly (Ostrolenk and Welch, 1942) on the underside of the abdomen around the navel or on the legs; others bite also on the neck and withers. They bite at a number of time in different places before they are replete (Soulsby, 1982). The flies feed about every three days. After feeding they rest for two hours on the under-side of leaves or on stones or trees.

2.4.5 Pathogenic significance

The bites of the *tabanidae* are painful and may give rise to weals in soft-skinned animals. Horses and cattle are restless when attacked by these flies and may become unmanageable if they are in harness; their feeding is often disturbed and they act as an efficient mechanical vectors of the organism responsible for such diseases as anthrax, pasteurellosis, trypanosomosis, anaplasmosis and the human filarial disease, loaosis (Urquhart, 1996).

2.5 Economic Importance of Haematophagous Flies

2.5.1 Tsetse flies

Tsetse fly and the diseases it transmits are one of the most severe medical and veterinary problems in Africa, infecting more than 500,000 people, with a mortality of 50,000 people and three million livestock annually (FAO, 2002; Okhoya, 2003); thereby, preventing the development of sustainable and productive agricultural systems (Vreysen, 2000). Both male and female tsetse flies are blood-suckers.Tsetse fly and trypanosomosis is widely recognized as one of Africa's greatest constraints to socio-economic development, causing death, debility, diminished productivity in man and domestic animals, massive economic loss and reduction of revenue from tourism (Anon, 2004). The burden imposed by tsetse and trypanosomosis remains one of the greatest causes of hunger, poverty and immense suffering to communities in Africa.

The occurrence of tsetse flies in national parks and game reserves where natural fauna and flora are protected provide ideal habitat for the flies and the pathogenic trypanosomes, and as such, domestic livestock grazing around the periphery of parks are permanently exposed to the risk of trypanosomosis and, if trypanosomes pathogenic to man occur in the local tsetse, staff and tourist frequenting the parks stand the risk of contracting sleeping sickness (Jordan, 1986). An initiative known as the Pan African Tsetse and Trypanosomiasis Eradication Campaign (PATTEC) was developed and a decision was passed by the African Heads of State and Government, urging Member States (since tsetse-transmitted disease is trans-boundary) to collectively embark on the campaign in order to rid Africa of the threat and burden of tsetse-transmitted disease. The PATTEC initiative seeks to employ an area-wide approach and appropriate fly suppression methods, to eradicate tsetse from areas of tsetse infestation, progressively and to ultimately create tsetse-free zones (Kabayo, 2002). This initiative was borne out of the successful elimination of tsetse from the Island of Zanzibar in early 1990s through the sterile insect technology (SIT) approach (Vreysen, 2001).

2.5.2 Stomoxys

Both males and females are blood-suckers, attacking man, horse, cattle and other mammals, and even birds and reptiles.*Trypanosoma evansi* (the causative agent of Surra of equines and dogs) and *T. equinum* (causative agent of Mal de caderas of equines, cattle, sheep and goats) are transmitted mechanically by *Stomoxys*. The species may also mechanically transmit *T. gambiense* and *T. rhodesience*, the causative agents of human trypanosomosis in Africa, and *T. brucei* and *T. vivax*, which cause nagana of cattle, sheep, goats and equines of Africa. It also serves as intermediate host of the nematode *Habronema majus*, a nematode parasite of the stomach of the horse.

The role of *S. calcitrans* in the transmission of equine infectious anemia is still under debate (Steelman, 1976). However, the fly is responsible for the mechanical transmission of septicaemia infections such as anthrax (Stock, 1997).

2.5.3 **Tabanus** (Horse fly)

Due to their frequent tendency to bite mammals, horse flies are known vectors of disease like Hog Cholera Virus (HCV) in livestock and anthrax in humans. Due to the pain caused by their bites, mammals react by removing the pest, and tabanids are free to infest other host with the blood of the first host still on their mouthparts. The diseases transmitted by this fly can cause decline in livestock having a negative consequences for human and agriculture thus causing significant economic loss. (Askew, 1971; Carn, 1996).

2.5.4 Chrysomya

Chrysomya bezziana causes an African Traumatic myiasis similar to cochliomyiasis, and those due to *Lucilia cuprina* and related species. Hypodermyiasis (warble fly infestation) and oestrosis (nasal bot fly infestation in sheep) still cause major economic losses in domestic animals, justifying their inclusion in control campaigns. The same applies to stomach bot flies of the family Gasterophilidae (Villeneuve, 1994).The economic damage generally associated with *Chrysomya rufifacies* is the cutaneous myiasis commonly inflicted on livestock. In Australia it is known to strike sheep but it is a secondary invader and acts as a scavenger on necrotic tissue and as a predator on other maggots. This species may be considered beneficial since it has been used in the treatment of osteomyelitis.

2.6 Trypanosome

Trypanosomes are protozoan parasites which cause a disease complex known as trypanosomosis. This protozoan parasite develops cyclically in the vector tsetse fly. Trypanosomes are classified in the phylum Sarcomastigophora, the order Kinetoplastida and the family Trypanosomatidae

(WHO, 1998). The trypanosome consists of a single cell varying in size from 8 to over 50 µm. There are distinct differences in appearance, shape and size between the various species of trypanosomes, allowing specific identification (Uilenberg, 1998). The salivaria group of trypanosomes may or may not have a free flagellum, the kinetoplast is terminal or sub-terminal, and the posterior end of the body is usually blunt. They develop as trypomastigotes within the mammalian host and are usually pathogenic (Mulligan, 1970).

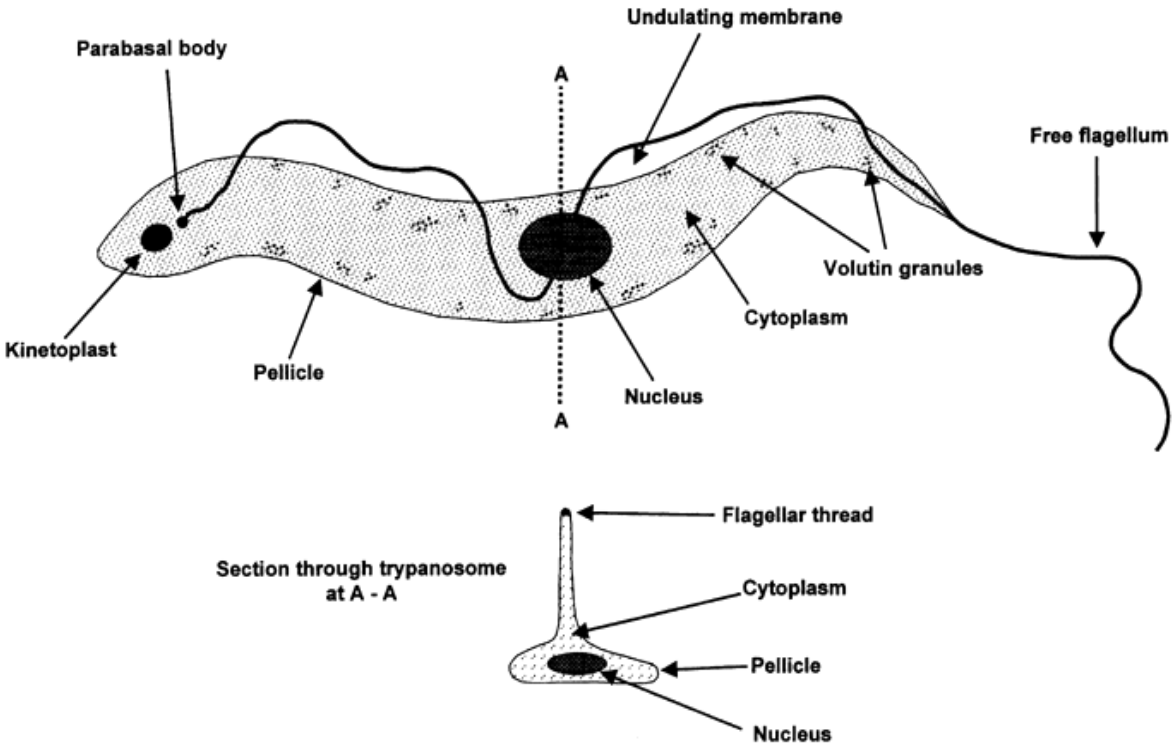

Plate 1: A labelled diagram of typical Trypanosome parasite

Source: http://www.fao.org/docrep/006/x0413E02.1

As trypanosomes progress through their life cycle they undergo a series of morphological changes as is typical of trypanosomatids. The life cycle often consists of the trypomastigote form in the vertebrate host and the trypomastigote or promastigote form in the gut of the invertebrate host. Intracellular lifecycle stages are normally found in the amastigote form. The trypomastigote morphology is unique to species in the genus *Trypanosoma*.

Trypanosomosis has been ranked the fourth most important disease of cattle in Nigeria after Rinderpest, contagious bovine pleura pneumonia (CBPP) and dermatophillosis (Streptothricosis) (Ademosun, 1973). The disease is wide spread and endemic in Nigeria, occurring in all the areas infested by tsetse (Onyiah *et al.*, 1983) and including the arid tsetse-free areas of the North that is infested by other biting flies.

The disease, trypanosomosis, is one of the most important disease of livestock, which hampers agricultural production in sub-saharan Africa including Ethiopia (Awoke, 2000). In other parts of the world, the transmission of trypanosomes pathogenic to animals is believed to be non-cyclical, or mechanical, and is effected mainly by blood-sucking arthropods. The essential difference between cyclical and non-cyclical transmission is that, in the former case, tsetse flies form a reservoir of infection, a factor which complicates the epidemiology and control of the African animal trypanosomoses. In tsetse-infested areas, trypanosomosis is a herd problem, and against the background of reduced productivity, other animal health problems occur. Trypanosomosis cannot therefore be viewed in isolation. In many parasitic conditions, host and parasite establish equilibrium, and the presence of the parasite does not seriously disadvantage the host. This is true in animal trypanosomosis and is best exemplified by the wild animal hosts of tsetse which forms a huge reservoir of trypanosomes.

The trypanotolerant breeds of domestic animals also display an ability to control trypanosomal infections, and they are often to be found in areas where other breeds cannot survive. Even so, there is evidence that so-called trypanosusceptible animals can establish a delicate balance with potential pathogens. Trypanosomal infections are often protracted, and chronic trypanosomosis is much more common than the acute disease in enzootic area (Nantulya, 1990). In all animals, disturbance of the equilibrium precipitates or exacerbates disease. The disturbance takes the form of stress, which may be due to the occurence of another disease, malnutrition, water deprivation,

heat stress or a production stress such as pregnancy, lactation or work. The significance of this point is that management determines, to a large extent, the well-being of animals and may do much to mitigate the effects of trypanosomosis. However, the pathogenicity of the different species of trypanosomes in the different species of livestock varies, and within a trypanosome species there is a range of virulence. The current threat of African animal trypanosomosis is rank among the top 10 cattle diseases on sustainable livestock production and mixed farming, couple with failure of vector control as chemotherapy/ chemoprophylaxis to control the present resurgence of the disease, presently a major constraint in the development of the African continent (Perry *et al.*, 2002; Abanga *et al*., 2002; Samdi *et al.*, 2010a). These constitute a major threat in attaining food security in several parts of sub- Sahara Africa and Nigeria (Samdi *et al*., 2010a). It is currently estimated that about 60 million people and 48 million cattle are at risk of contracting African trpanosomosis from the 23 species and 33 sub species of tsetse flies infecting 10 million Km^2 of Africa land stretching across 40 countries (Kristjanson *et al*., 1999; Samdi *et al*., 2010a). The trypanosome species of economic importance in cattle are: *Trypanosome congolence*, *T. vivax,* and *T. brucei*. Tsetse transmitted African trypanosomosis is responsible for 55000 human, 3 million livestock deaths annually and hinders mixed farming through reduced work efficiency of draft animals (Abanga *et al*., 2002; Samdi *et al*., 2010a). The loss in livestock production and mixed agriculture alone is valued at 5 billion US dollars yearly, In Africa however, effective and sustainable control measures can result in up to 3 fold increase in the current estimated livestock population in Nigeria (Onyiah, 1997).The decrease in national and international funding for research and surveillance of trypanosomosis has resulted in insufficient information on the current status of the disease (Maikaje *et al*., 2009).

2.6.1 Morphology of trypanosome.

Trypanosomes are slightly curved elongated protozoa measuring about 10 - 35µm with each one having a single nucleus and possess a single flagellum which originates near the posterior end of

the body and extends forward to the body by an undulating membrane. Near the base of the flagellum is the dark-staining kinetoplast. The size and shape of the body, position of the nucleus and kinetoplast, length and form of the undulating membrane and flagellum are the diagnostic characters of various species of Trypanosomes (Jordan, 1986).

The form of trypanosomes in the tsetse fly can be identified to subgenus on morphological grounds and on their sites of development within the fly. In the subgenus Duttonella is the *Trypanosoma vivax*; a group of trypanosomes with large terminal kinetoplast, distinct free flagellum and an inconspicuous undulating membrane. *Trypanosoma vivax* is a large (18 - 26µm long) monomorphic organism that is very active in wet mount blood smears. Their development within the tsetse fly is restricted to the proboscis (labrum or hypopharynx). In the subgenus Nannomonas is the *Trypanosoma congolense* which is a small trypanosome with medium – sized marginal kinetoplast, no free flagellum, and a poorly developed undulating membrane. Trypanosoma congolense is a heamatic trypanosome found only in the blood vessels of the animal they infect. They develop within the proboscis (labrum or hypopharynx) and midgut of the tsetse fly. In the subgenus: Trypanozoon is *Trypanosoma brucei brucei*. This group is an extremely polymorphic; while some trypanosome occur as short, stumpy organisms without flagella, others are found to be long slender organisms with distinct flagella, and there are intermediate forms that are usually flagellated (Moulton, 1976). These trypanosomes develop within the proboscis, midgut and salivary glands. However, when trypanosomes are found only in the labrum, they are regarded as immature form of all the subgenus mentioned, whereas if they are found in the labrum and midgut, it indicates immature form of the congolense group (Davies, 1977).A sound knowledge of the basic features of the various trypanosomes enables the identification of each species and so the exact cause of the disease.

2.6.2 Trypanosome species in Nigeria with their vectors.

There are five (5) economically important animal trypanosome species in Nigeria. These include: *T. congolense,T. vivax, T. simiae, T. brucei, T. suis, T. evansi,Trypanosoma equiperdum* (Omotainse, 2004). Trypanosome species with some of their *Glossina* vectors as classified by (Hoare, 1972, Hunt, 2004) in animals.

2.6.3 *Trypanosome species* and their *Glossina* vector

T. brucci brucei, Glossina vectors *includes; Glossina swynartoni, G. Pallidipes, G. palpalis, G. tachinoides.* For *T. congolenseGlossina* vectors include; *G. palpalis, G. longipalpis, G. tachinoides, G. Morsitans* and *T. Simiae Glossina* vector *includes; G. tachinoides, G. longipalpis, G. fusca, G. tabaniformis* (Hunt, 2004).

2.6.4 Clinical trypanosomosis.

Pathogenic trypanosomes cause disease in all species of domesticated livestock throughout many of the tropical and subtropical regions of the world. In Africa, *Trypanosoma brucei, T. vivax* and *T. congolense* occur wherever the tsetse fly vector is found. *Trypanosoma evansi*, which is transmitted mechanically by several different species of haemotophagous biting flies, is found in North Africa, the Near East, the Far East and Central and South America. *Trypanosoma vivax* also occurs in South America where it, too, is transmitted mechanically by biting flies. The clinical signs of the disease caused by these organisms vary according to the *Trypanosoma species*, the virulence of the particular isolate and the species of host infected. Acute disease is characterized by anaemia, weight loss, abortion and, if not treated, possibly death. Animals that survive are often infertile and of low productivity. In some instances, infected animals show no overt signs of disease but can succumb if stress for example, by work, pregnancy, milking or adverse environmental conditions (Luckins, 1988).

2.6.5 Transmission of trypanosomes.

Stercorarian trypanosomes infect the insect, most often the triatomid kissing bug, develop in its posterior gut and infective organisms are released in the feces and deposited on the skin of the host. The organism then penetrates and can disseminate throughout the body. Insects become infected when taking a blood meal.

Salivarian trypanosomes develop in the anterior gut of insects, most importantly the Tsetse fly, and the infective organisms are inoculated into the host by the insect bite before it feeds. Both male and female flies carry trypanosomes and it is assumed that all species of *Glossina* can act as vectors. The pathogenic *Trypanosoma species* (*T. brucei*, *T. congolense*, *T. simiae*, *T. godfreyi*, *T. vivax*) are transmitted to the host during feeding. Within the fly, trypanosomes undergo cycles of development and multiplication, involving different parts of the alimentary tract and salivary glands depending on the trypanosome species. The complex lifecycle in the fly may take from a few days for *T. vivax* to a few weeks for *T. brucei*. However, the trypanosome has then gained the advantages of (i) the amplification of a typically scanty parasitaemia in the host, (ii) the potential transfer of infective forms every time the fly bites a new host and (iii) in addition, at least in *T. brucei* has the opportunity for sexual reproduction in the fly (Jenni and Ahmed, 2004).

In their mammalian hosts, trypanosomes replicate in the extracellular environment in the blood and express a thick surface coat that undergoes antigenic variation (variant surface glycoprotein, VSG). Already in the mammalian bloodstream, some trypanosomes undergo differentiation from long-slender to short-stumpy forms, apparently in preparation for the rigors of life in the fly. Importantly, rather than the sole dependence on glycolysis for energy metabolism demonstrated by long-slender forms, these trypanosomes develop a functional mitochondrion as they differentiate, and cease division until reactivated by transfer to the fly (Vickerman, 1965).

When trypanosomes are taken in with a bloodmeal as the tsetse feeds, they undergo a sudden and drastic change and need to adapt to cooler temperatures, new food sources, hostile digestive enzymes and immune molecules. The two key stages in the development of *T. brucei* or *T. congolense* within the tsetse are the initial establishmentof infection in the midgut and its subsequent *maturation* to produce infective metacyclics either in salivary glands or in the oesophagus, respectively.

To survive in the midgut, trypanosomes must radically transform their metabolism and begin multiplying (Aksoy *et al.*, 2005). At around 3 days at 25^{o}C a process of attrition is evident, leading to the complete elimination of the infection in a proportion of flies (Dipeolu and Adam, 1974). In successful infections, the trypanosomes invade the ectoperitrophic space after about 5 days and establish infections where they continue to divide and move anteriorly (Dipeolu and Adam, 1974; Robertson, 1913). After 6–8 days the trypanosomes begin to congregate in the proventriculus region in large numbers (Van den Abbeele *et al*., 1999). Here, they apparently cease division (Lewis and Langridge, 1947; Robertson, 1913; Van den Abbeele *et al*., 1999) and grow into long, extremely motile proventricular forms, which migrate anteriorly. Again, how the trypanosomes get from the ectoperitrophic space to the gut lumen is uncertain, but there is evidence from light and electron microscopy that they penetrate through the soft peritrophic matrix in the proventriculus where it is produced (Fairbairn, 1958; Steiger, 1973). The migrating forms in the oesophagus and mouthparts are scanty and not culturable in vitro (Van den Abbeele *et al*., 1999).

In the salivary glands of the fly, the epimastigotes attach to the epithelium via outgrowths of the flagellar membrane and sheath (Tetley and Vickerman, 1984; Vickerman, 1985), and multiply to populate the whole salivary gland. In the progression from epimastigotes to metacyclics (maturation), the mitochondrion becomes less branched; only the nascent and mature metacyclics

have a VSG coat and no dividing forms of these stages are observed (Tetley and Vickerman, 1984). Metacyclics have a small repertoire of VSG genes (Pays *et al*., 2001) and on inoculation into a host in the saliva, they multiply for a fewdays at the site of fly bite, before invading the bloodstream and lymphatics.

2.6.6 Antigenic variationof trypanosomes

Trypanosomes are unicellular, eukaryotic parasites that live extracellularly in a wide range of mammals, including humans. They have a surface coat, composed of variant surface glycoprotein (VSG), which probably is essential and acts as a defence against general innate immunity and against acquired immunity directed at invariant surface antigens. In effect, the VSG is the only antigen that the host can target, and each trypanosome expresses only one VSG. To counter specific antibodies against the VSG, trypanosomes periodically undergo antigenic variation, the change to expression of another VSG. Antigenic variation belongs to the general survival strategy of enhanced phenotypic variation, where a subset of 'contingency' genes of viruses, bacteria and parasites hypermutate, allowing rapid adaptation to hostile or changing environments (Pays *et al.,* 2001). A fundamental feature of antigenic variation is its link with the population dynamics of trypanosomes within the single host. Antigenic variants appear hierarchically within the mammalian host, with a mixture of order and randomness. The underlying mechanisms of this are not understood, although differential VSG gene activation may play a prominent part. Trypanosome antigenic variation has evolved a second arm in which the infective metacyclic population in the tsetse fly expresses a defined mixture of VSGs, although again each trypanosome expresses a single VSG. Differential VSG expression enhances transmission to new hosts, in the case of bloodstream trypanosomes by prolonging infection, and in the metacyclic population by generating diversity that may counter existing partial immunity in reservoir hosts. Antigenic variation employs a huge repertoire of VSG genes. Only one is

expressed at a time in bloodstream trypanosomes, as a result of transcription being restricted to a set of about 20 bloodstream expression sites (BESs), which are at chromosome telomeres. Only one BES is active at a time, probably through transcriptional elongation being inhibited in the silent BESs. Although transcriptional switching between BESs can affect a VSG switch, the most prolific switch route involves homologous recombination of deoxyribonucleic acid, usually by the copying of a silent gene into a BES. Hierarchical expression of VSGs may be dictated in part by the different types of locus occupied by VSG genes. The VSG genes expressed in the metacyclic population also occupy telomeric sites, which appear to be derived from BESs but have a simpler structure. Their differential expression is achieved by random transcriptional activation; the detailed story requires direct study of the metacyclic stage itself. Available evidence suggests that the VSG originated as a surface receptor, and it can be proposed that a number of selective events have contributed to the evolution of the complex, multisystem phenomenon that antigenic variation has become (Vickerman, 1985).

2.6.7 The subgenus Nannomonas

This is the smallest of the pathogenic trypanosomes, with a length of 8-24 μm. The bloodforms are monomorphic, in that they lack a free flagellum. Generally, two variants are to be seen, a shorter form (9-18 μ), the typical congolense type and a longer form (up to 25 μ), with individuals intermediate in length between the two (Hoare, 1972). There is evidence which indicates that strains with the longest forms, the so-called 'dimorphic' strains, cause a more severe form of trypanosomosis (Uilenberg, 1998). In stained specimens of *T. congolense* the cytoplasm stains a diffuse, even, pinkish colour and is seldom granular (Uilenberg, 1998). The nucleus is centrally placed. The Kinetoplast is of medium size and is usually situated at the margin of the body, just in front of the posterior extremity (marginal and subterminal). The undulating membrane is poorly developed and inconspicuous (Hoare, 1972). Recent studies have resulted in a subdivision of the T. congolense species in several 'types', which can be

distinguished by isoenzymatic differences and molecular techniques. These are designated as *T. congolense* savannah type, *T. congolense Tsavo* type, *T. congolense* forest type, *T. congolense Kilifi type (*Majiwa *et al.,* 1985*;* Majiwa *et al.,* 1993*).*

Trypanosoma simiae species are polymorphic, with a length of 15-19 μm. In typical cases, some individuals are with and others without a free flagellum. The Kinetoplast is of medium size, marginal and subterminal, as in *T. congolense* (Hoare, 1972). *Trypanosoma gogfreyi* has been separated recently from *T. congolense* in the Gambia, on the basis of isoenzymatic and DNA differences, but also because the disease it causes is different. It is pathogenic for pigs, but the disease is more chronic than the one caused by *T. simiae*. Morphologically, it is similar to *T. congolense*, with a length of 9-22 μm, but the undulating membrane is described as being usually conspicuous (Hoare, 1972).

2.6.8 The subgenus Duttonella

Trypanosoma vivax was named because of the vigour of its activity under the microscope when examined in fresh preparations. The parasite moves rapidly across the field of view. *A*s seen in the blood of mammals it is also essentially monomorphic, with a free flagellum. Its length, including the free flagellum, varies from 18 to 31 μm. The kinetoplast is large and terminal or almost so. It is much larger than in any of the other pathogenic species, and this is a distinguishing feature. The nucleus is centrally placed. The posterior extremity is swollen and blunt. The undulating membrane is inconspicuous. *Trypanosoma uniforme* are small trypanosomes (from 12 to 20 μm), otherwise they are similar to *T. vivax* (Molyneux and Ashford, 1983).

2.6.9 The subgenus Trypanozoon

The subgenus Trypanozoon is the most homogeneous group of Salivarian trypanosomes, represented conventionally by species which are morphologically indistinguishable but differ in

biological features. This group comprises five members*: T b. brucei, T. b. gambiense, T. b. rhodesiense, T. evansi and T. equiperdum. Trypanosoma brucei* is polymorphic, with three main forms, all of which have a small kinetoplast and a conspicuous undulating membrane: these forms include; (a) Long slender forms (23-30μm in length) with a free flagellum, which may be up to one half of the length of the organism. The posterior end is pointed and the nucleus is central. The kinetoplast is placed up to 4μm in front of the posterior extremity. (b) Short stumpy forms (17-22μm in length) normally without a free flagellum, but in which there may occasionally be individuals with a short free flagellum. The kinetoplast is usually subterminal. The position of the nucleus varies greatly and it is in some cases in the posterior part of the cell, sometimes so far posterior that the kinetoplast is anterior to it (so-called postero-nuclear forms) (Vickerman, 1985). (c) Intermediate forms, varying in length between the two previously mentioned types. A free flagellum, of varying length, is always present. The nucleus is centrally placed. The posterior end is somewhat variable in shape, but usually bluntly pointed. The kinetoplast is close to the posterior extremity (Hoare, 1972).

2.6.10 Course of the infection

During the course of the infection, there is a change in the trypanosome population from the long thin forms, through the intermediate, to the short stumpy forms, and this altered appearance is accompanied by a change in the type of respiration, as the trypanosome prepares for its period within the tsetse fly. The short stumpy forms are adapted to living and developing in the tsetse, while long thin forms are the true mature blood forms which die in the gut of the insect (Hoare, 1972). *Trypanosoma evansi* is typically represented almost exclusively by thin trypomastigotes comprising slender and intermediate forms corresponding to those in *T. brucei*. The slender forms have along free flagellum and a narrow posterior extremity, which may be rounded or truncated, with the kinetoplast situated at some distance from the tip. The intermediate forms

have a shorter free flagellum and a short, frequently pointed, posterior extremity, with the kinetoplast lying near this end (Hoare, 1972). *Trypanosoma equiperdum* is morphologically indistinguishable from *T. evansi*. Like the latter species, it is typically monomorphic, being represented by thin (slender and intermediate) trypomastigotes possessing a free flagellum. The size of *T. equiperdum* is likewise within the range of that of *T. evansi*.Furthermore, as in the case of *T. evansi*, it is liable to become pleomorphic, exhibiting typical and postero-nuclear stumpy forms (Hoare, 1972).

2.7 Other Blood Prozoans

Babesiosis and anaplasmosis are tick transmitted diseases caused by obligate intraerythrocytic hemoparasites. It has been estimated that at least 500 million cattle are exposed to babesiosis and anaplasmosis worldwide. Following acute infection, both diseases establish persistent infections that confer lifelong immunity to the host. The existence of a subclinical carrier state in cattle poses a risk when animals are moved from endemic to nonendemic regions. For this reason, these diseases are currently classified in List B of the Office International des Epizootics (OIE, 2004a, 2004b). These tick-borne hemoparasitic diseases of greatest global economic importance in cattle are babesiosis, anaplasmosis, theileriosis and cowdriosis (Uilenberg, 1996). Of these, only anaplasmosis, caused by *Anaplasma marginale* and babesiosis, caused by *Babesia bigemina* and *Babesia bovis*, are intimately associated with erythrocytes Tick-borne protozoan diseases pose important problems for the health and management of domestic cattle in the tropics and subtropics (Jongejan and Uilenberg, 1994). Cattle recovering from babesiosis retain a latent infection lasting six months to several years (De Vos *et al*., 2004). Unlike anaplasmosis, recrudescence of parasitemia occurs at irregular intervals (Mahoney, 1977). At least 5 mechanisms have been identified which might contribute to evasion of host immunity by *Babesia* organisms (Allred, 2003). These include: (1) antigenic variation of a variant erythrocyte

surface antigen (VESA1), (2) cytoadhesion/sequestration, (3) binding host IgM to the surface of infected erythrocytes, hiding the parasite from immune recognition, (4) monoallelic expression of different members of multigene families, and (5) induction of transient immunosupression (Allred, 2003). Maintenance of persistent infections confers lifelong immunity to the host. The carrier state is also important in endemic areas as it contributes to endemic stability of the disease. Under these circumstances young animals are exposed to infection before passive protection from maternal antibody wanes. Consequently, serious disease outbreaks do not occur even though the causal organism is widely distributed in the population (Sergent, 1965; Mahoney, 1977).

The most widespread and malignant *Theileria* species is *Theileria annulata*, causing tropical theileriosis, which occurs around the Mediterranean basin, in the Middle East, and in Southern Asia. The other malignant *Theileria* species is *T. parva*, which occurs in East and Southern Africa and causes East Coast fever. In addition, benign forms of theileriosis are caused by *T. mutans*, *T. velifera*, and *T. taurotragi*, which are mainly located in Africa, whereas parasites of the *T. sergenti-T. Buffeli-T. Orientalis* group, referred to below as the *T. orientalis* group, occur worldwide (Uilenberg, 1998).

2.8 Laboratory Techniques for Detecting Haemoparasites

Several techniques for detection of these hemoparasites have been developed separately for each species (Figueroa *et al.*,1992) For the *T. orientalis* group; several sensitive PCR assays based on the gene encoding the major piroplasm surface protein have been developed (Kawazu *et al* 1992). A similar approach using the Tams1 gene was followed for *T. annulata* (d'Oliveira *et al*., 1995). Sensitive PCR methods are available for *B. bovis* (Calder *et al.*, 1996) and *B. bigemina* but not yet for *B. divergens*. Assays to differentiate *Theileria* species on the basis of their rRNA genes (Figueroa *et al.*,1992) have been described by Allsopp *and Hursey,* 2004.

These assays did not include all *Theileria* species. Moreover, these assays were developed merely for the differentiation of species rather than for detection in carrier animals. To date, screening for the presence of benign *Theileria* species, such as *T. mutans, T. velifera,* and *T. taurotragi,* in cattle depends on the immunofluorescent antibody test and may give rise to cross-reactions (Papadopoulos, 1996). Moreover, since serodiagnosis does not detect the parasite itself, the animal may have already cleared the pathogen but remained seropositive. The first integrated approach to sensitive detection and differentiation of *B. bigemina*, *B. bovis,* and *Anaplasma marginale* in a 16S–18S rRNA gene multiplex PCR was reported by Figueroa et al., 1992.

Despite the usefulness of these assays, it would be very practical to have a universal test to simultaneously detect and differentiate all protozoan parasites that could possibly be present in the blood of carrier cattle.

2.9 Trapping of Haematophagous Flies

A range of designs for haematophagous fly traps were developed in the 1960s and earlier but these included a number of features that made them generally difficult or impractical to use. Many, for instance, were large, cumbersome, and difficult to transport. With the development of the biconical trap by (Challier and Laveissiére, 1973) mainly for tsetse fly, a trap became available that was relatively cheap, collapsible, so that many can be carried in a vehicle, and quickly and easily assembled The biconical trap is particularly effective for species of the palpalis group of tsetse including G. palpalis and G. tachinoides, and less so for other tsetse, but widely used for sampling (Lancien and Gouteux, 1987). Different species of tsetse occur under and are adapted for different environmental conditions. The best design of trap depends upon the species of tsetse fly and it is important to match the trap to the species. Following the development of the biconical trap, a range of different traps have been designed. For riverine species such as Glossina palpalis or *G. fuscipes*, the best traps are the biconical or pyramidal

traps. For tsetse living in savanna habitats, e.g. *G. morsitans* and *G. pallidipes*, the best traps have been shown to depend on the locality, or where they are to be used. In East Africa, the Ngu or Nzi traps appear to be the best, whereas in southern Africa the Epsilon trap has produced better results. For G. brevipalpis, the H-trap is preferred whereas the Ngu and Epsilon traps have been used successfully to catch G. longipennis in Kenya and Somalia. There is a wide variety of traps that have been specifically designed to catch tsetse. The 'best' design depends upon which species you are trying to catch - and thus it is important to match the trap to the species.

For riverine species such as *G. palpalis* or *G. fuscipes*, use biconical (Challier and Laveissiere, 1973) or pyramidal (Lancien and Gouteux, 1987) traps. Examples of other modifications of the pyramidal trap designed specifically for riverine species of tsetse found in West Africa can be seen at the sleeping-sickness.com website.

For savanna flies, such as *G. morsitans* subspp. and *G. pallidipes*, the 'best' trap seems to depend on where you are. In east Africa, the Ngu (Brightwell *et al.*, 1987) or Nzi (Mihok, 2002) traps seem to be the best, whereas in southern Africa the Epsilon (Hargrove and Langley, 1990) is better.

For the fusca species *G. brevipalis*, the H-trap (Kappmeier, 2000) is best whereas the Ngu and Epsilon traps have been used successfully to catch *G. longipennis* in Kenya and Somalia.

2.9.1 Biconical (Challier - Laveissiere) trap

This trap catches certain species of all three species groups. It is less efficient for catching crepusclar species (those species active at dawn and dusk). It is fairly cheap and is collapsible so that many can be carried in a single vehicle; even a man can carry several at once (Challier and Laveissiere, 1973). It consists of two fabric cones, approximately equal in size, joined base to base and supported by a vertical pole passing through the middle. The bottom cone of blue cotton fabric is divided into four segments by dark cloth partitions, visible through four slits in the blue cloth. The upper cone is of strong mosquito netting (Kappmeier, 2000). At the top of the vertical pole is mounted a small cone made of a wire frame and covered with mosquito netting. This small cone is open at the base and has a snail exit hole at the top. It acts as a non-return device, and leads to a collecting cage at the top of the trap. Flies attracted to the trap enter through the dark slits at the base, and move up into the collecting cage at the top (Brightwell *et al.,* 1987).

The whole trap can be fixed firmly in the ground by the vertical pole, or can be suspended by a greased wire from a branch or tripod. The lower part of the pole or wire can be smeared with grease to prevent ants getting to the caught flies. The trap stands about shoulder height (Challier and Laveissiere, 1973).

2.9.2 The Nzi trap

"Nzi" is one of the Swahili words for fly. The Nzi trap was developed in Kenya for savanna species of tsetse such as Glossina pallidipes (Mihok, 2002). It is also a very effective trap for stable flies (Stomoxys spp.) and horseflies (Tabanidae) (Torr *et al.,* 2005). The Nzi trap is a simple, safe and economical cloth trap for the capture of biting flies (tsetse flies, horse flies, deer flies, stable flies). It was developed by Steve Mihok at ICIPE in Kenya as an environment-friendly alternative to the use of insecticides, following many years of research on appropriate and sustainable technology for African farmers. It is a passive killing device that works through

the attraction of flies to large blue and black objects (Torr, 2003). Flies simply die from exposure after entering into an innovative configuration of cloth and netting. The trap is made from simple shapes for economy, and for ease of assembly (Torr, *et al.,* 2004). The layout is triangular with all pieces cut to the width of the material (e.g. one metre or one yard). The trap walls are formed by a square piece of netting at the back and by two black cloth rectangles at the sides. The body is closed at the top front by a vertical blue shelf (Mihok, 2002). Two blue rectangular wings extend out at an angle from the front. A trapezoidal piece of netting extends horizontally half-way into the body from the bottom of the blue shelf (Brightwell *et al.,* 1987). The top is closed by a cone made by cutting a wedge out of a square piece of netting and sewing up the sides (a tetrahedron results: a 4-sided shape with equal triangular sides)(Torr,2003).

2.9.3 Basic principles of haematophagous traps

Haematophagous flies (especially Tsetse) has a high metabolic rate and feed exclusively on vertebrate blood. Their survival therefore depends on detecting and encountering suitable hosts on which to feed (Hargrove and Langley,1990). This principle can be exploited in the design of traps and targets which mimic key features of the normal host animals, attracting flies in such a way that they can then be captured or killed. With traps, the captured flies can be identified and counted, useful in sampling and monitoring tsetse populations (Bouyer *et al.,* 2005). Flies targets simply use insecticide-treated surfaces to kill the tsetse by contact and are of little use in population sampling or monitoring. Both targets and traps are exposed to damage and stealing and these methods require active participation from rural communities. As techniques for tsetse control, both traps and targets function by removing individuals from the tsetse population. Their efficiency depends on the length of time the devices remain operational, and the likelihood that an individual fly will encounter the device and be killed by it (Kuzoe and Schofield, 2004).

2.10 Control of Haematophgous Flies

2.10.1 Tsetse flies.

Several methods have been developed and put into practice to keep the disease under control, some less effective than others (Grev, 2002). The control strategies over the years have been directed against both the parasites and the vectors (Onyiah, 1997). Hand-catching has been the initial methods of tsetse control comprising of Bush clearing and Game destruction (Glasgow, 1970), Settlement of people and the use of chemicals as insecticides (Davies, 1977). The use of traps and screens (Challier and Laveissiere, 1973) and the Sterile Insect Technique (SIT) are the more recent method of tsetse control. Other interventions aimed at eliminating the parasites have been by chemotherapy, chemoprophylaxis and promotion of trypanotolerant breeds of cattle (Grev, 2002).

a. Hand-catching

Hand-catching is the most ancient method of insect control and was first tried in about 1913 against *G. palpalis* in the Portuguese Island Principe, and against *G. palpalis* (*G. fuscipes*) by the Germans on an island, Riamugasire, on Lake Victoria. Hand-catching is absolutely specific and has been found effective against some species, however they are more expensive than chemicals because it always require a large labour force especially if large areas were to be attacked, thus it fell into disuse (Glasgow, 1970).

b. Bush burning/Clearing

This method was performed based on the knowledge of the biology of tsetse fly, by cutting down dense vegetation, thus destroying both the adult fly and pupae due to decrease in humidity (Nagel, 1995). Bush clearing can be total or ruthless when all vegetation is totally cleared and partial when it involves the destruction of only a portion of the vegetation to render the environment unsuitable for tsetse fly. Partial clearing could be discriminate clearing when woody vegetation in known tsetse concentration sites is destroyed or selective where

only certain components of the vegetation forming the fly habitat are removed, which may be removal of only the under storey, leaving the tall trees untouched or removing particular species of shrubs and trees (Jordan, 1986).

The history of the development and application of methods of partial clearing for tsetse control have been extensively reviewed by a number of authorities (Buxton, 1955). In Nigeria, the record of partial clearing was described by Moiser (1912) in Geidam, Borno State when the population of *Glossina tachinoides* was controlled by vegetation clearing. Significant control was also achieved by Nash, (1940) on *Glossina tachinoides* at Gadau and Anchau in Kano state.

The major short-coming of this method lie in the limited size for which they can be economically deployed relative to the total size of tsetse affected area (Agyemang, 2001) since it requires economically unacceptable destruction of vast area of bush and forest. Bush clearing also results in soil degradation and deforestation and as such was no longer in use around 1970 (Nagel, 1995).

c. Game Destruction

The concept of game destruction was developed following the great rinderpest epizootic at the end of the nineteenth century, which resulted in the death of many game animals and thus the disappearance of tsetse flies and trypanosomiasis (Jordan, 1986). This method was used many years in Zimbabwe, Zambia, Mozambique, Botswana and Uganda to eliminate a wide range of the population of savanna species of *Glossina* and trypanosomiasis due to its close association with game animal. McLennan (1981) reported the ineffectiveness of game destruction on riverine species because they feed on other hosts besides wild game e.g. man, domestic animals, crocodiles and reptiles. In West Africa, savanna species of tsetse population thrives on very low game densities and as such, this method of control against tsetse did not succeed

in Nigeria. This method ceased because, animals migrated to area once cleared from larger mammals, making it possible for tsetse to recover, and also because it became unacceptable to public opinion (Grev, 2002).

d. Trapping Method

Trapping method for tsetse fly originated from the Island of Principe, where farm workers wore on their back a dark – colored piece of cloth, covered with glue, to reduce biting nuisance of the flies (Maldonado, 1910) alongside with vegetation clearing and pig elimination to control tsetse on the Island (Da Costa *et al.*, 1916). The first real tsetse trap constructed by Harris was used to eliminate *Glossina pallidipes* from Zululand, and other various modified versions were introduced such as the Animal trap (Morris and Morris, 1949); Biconical trap (Challier and Laveissiere, 1973); Monoconical trap (Lancien, 1981); Nitse trap (Omoogun, 1994) etc. In Nigeria, Biconical and Nitse traps have been extensively used for tsetse sampling, ecological studies and control (Omoogu, 1994; Dede *et al.*, 1974). Traps were initially made for control purposes but were later used for sampling and ecological studies due to their poor efficiency (Omoogun, 1994).

The interest in controlling tsetse flies by trapping declined rapidly with the introduction of synthetic insecticides, but was renewed in the seventies, mainly due to increased public awareness of environmental pollution by excessive use of insecticide (Grev, 2002). In West Africa, traps were widely used to control riverine species and was suggested to be better for community – based operations because local people can see flies being caught and killed, and can also see the catch decline as the operation progresses. Trapping method is simple, inexpensive, and harmless to the environment and is currently being used as an integrated part of the arsenal of tsetse control method where fly population is suppressed by trap and other

conventional methods such as bait and target. However, reinvasion remains a reoccurring problem (Hargrove, 2002).

e. Use of Pheromones

Most work undertaken on pheromones (Langley *et al.,* 1975; Huyton *et al.,* 1980; Offor *et al.,* 1981; Carlson *et al.,* 1984) was based on the identification of the sex recognition pheromone, in the cuticle of female *Glossina* species which induces copulatory behavior in males of the same species, but not of other species, upon contact. This method aims at mass producing the compound(s) that constitute the sex recognition pheromones for the purpose of attracting flies to impregnated traps or screens to thus effect control, but because of the lack of volatility of these pheromones, they could not be exploited as attractant (Jordan, 1986).

f. Bait Technology

The potential of bait technology for the control of tsetse fly was appreciated in the first half of the 20th century. The strategy of the technology was to improve bait design (live or artificial) by the careful analysis of basic responses of tsetse to baits, and using the knowledge to improve the design of devices used in the field (Vale and Torr, 2004; De Deken, 2004) Esterhuizen et al., (2006) reported the use of artificial baits to control fly and reduce trypanosomiasis at lower cattle density and stressed that insecticide – treated cattle are more effective than stationary bait in area with higher cattle densities. The work of various authors (Ndams, 1987; Bossche, 1997; Brightwell *et al.,* 2001; Esterhuizen *et al.,* 2006) has suggested the efficiency of this method for the control of tsetse flies.

Even though improved traps and bait technology (targets) rapidly became the standard control method throughout Africa in late 1980s and 1990s, bait technology had pitfalls in its application which include tackling too small an area and the variability in costs and benefits relating to community – based action (Vale and Torr, 2004).

g. Use of Insecticides

The use of insecticides as control methods against tsetse fly commenced in the mid-1940s and is still today a major technique used in large scale (Grev, 2002), and almost all method now used depend on insecticide. Two chemical groups have been widely used in tsetse control: The organochlorides (DDT, dieldrin, endosulfan):- DDT was the first chemical insecticides used against tsetse fly, after which dieldrin became popular because of its high lethal characteristics in more humid conditions. These two chemicals were then displaced by endosulfan for its toxicity and better solubility in spray solvent (Vreysen, 1995). DDT is cheap, has low mammalian toxicity, persists long in the environment and is effective against tsetse fly, while dieldrin on the other hand is expensive, has longer persistent rate than DDT (Davies, 1977). However, both fall victim of international ban due to their environmental side effect (Allsopp and Hursey, 2004). The spray of endosulfan against tsetse was reported to cause significant fish mortality (Douthwaite *et al.,* 1981). The synthetic pyrethroids (deltamethrin, alpacypermathrin and betadyfluthrin) (Mangwiro *et al.,* 1999), are the most potent insecticides used against tsetse fly. Deltamethrin have been widely used for both spraying and for Impregnating traps and targets. The major disadvantages of the synthetic pyrethroids are their high cost (Vreysen, 1995).

In Nigeria, the use of insecticides to control tsetse fly started in 1955 (Davies, 1977) at Kamadugu Gana river system which later extended to Kiyawa-Jama'are Katagun system, both area lying within Kano, Borno and Bauchi States. Persistent insecticides (DDT) were applied from the ground on the tsetse resting sited by means of pneumatic knapsack pressure sprayers (Davies, 1977; Maclennam, 1967). Aerial spraying started in 1971 in the northern guinea vegetation zone, extended to the southern guineas savanna zone (Spielberger *et al.,* 1977). Spraying and re-spraying activities reclaimed large area of land of about 399,551km² in the

Sudan and northern guinea vegetation zones from tsetse fly (Bature, 1985). In Botswana, an area of 16,000km² in the Okavango Delta was reclaimed from *Glossina morsitans centralis* using aerial spraying with deltamethrin applied at 0.26 – 0.3g/ha and 12,000 deltamethrintreated targets (Kgori *et al.*, 2006).

h. Sterile Insect Technique (SIT).

The application of the sterile male technique received considerable attention in the 1980s and is one method that seems feasible for the eradication of tsetse flies from the continent of Africa. Economic feasibility of which is grater in the area-wide approach (Feldmann, 2004). This approach was applied on an area-wide basis to eradicate the New World Screw worm *cochiiomyia hominivirax* in the U.S.A, Mexico and Central America. Since then very effective programmes integrating the SIT have been mounted against tropical fruit flies (Klassen and Curtis, 2005) and some species of tsetse flies on pilot trials at Lake Kariba, Zimbabwe (vs. *Glossina morsitans morsitans*), at Tanga, Tanzania vs. *Glossina morsitans morsitans*), in Burkina Faso (vs. *Glossina palpalis gambiense, Glossina tachinoides and Glossina morsitans submorsitans*) in Plateau state, Nigeria (vs. *Glossina palpalis palpalis)* and in Zanzibar, Tanzania (vs.*Glossina austeni*) (Feldmann, 2004). The success in Zanzibar demonstrated the technical feasibility of fighting the disease through the sterile insect technique approach (Kabayo, 2002).

The concept of sterile insect technique involves the mass production, sterilization – by exposure of males to short burst of gamma radiation from a cobalt-60 source (Okhoya, 2003) and sequential release of sterile males to the target species to compete with the wild male population. Mating between released sterile males and the wild females produces unviable progeny and the population is reduced over several generations to unsustainable levels (Abila *et al.*, 2003). The obvious constraints of sterile insect technique are the high cost associated

with mass rearing, low competitiveness of released sterile males (Whitten and Mahon, 2005), low reproductive rate and low rate of re-infestation (Feldmann, 2004). The technique which can only be employed realistically when density of target population is low is impractical for use against high density population. It is therefore used only in an integrated approach with other control methods such as traps and targets (Jordan, 1986). However, sterile insect technique still remains an exceptionally promising pest control method in terms of efficacy and environmental compatibility (Nagel and Peverling, 2005).

2.10.2 Stomoxys.

The fly is most troublesome in localities where suitable breeding places are readily found. Control measures should therefore be directed toward destroying breeding-places by regular removal of most bedding, hay and faeces from stables and yards, and food waste from feeding troughs, and by preventing the accumulation of heaps of weeds, grass cuttings and vegetable refuse. Regular application of pyrethrins, synergized pyrethins, pyrethroids, coumaphos, stirofos, or dichlorvos is indicated. Application of insecticides to areas where they habitually rest (Urquhart, 1996).

2.10.3 Tabanus.

This poses a special problem since breeding places are both diffuse and difficult to detect (Urquhart, 1996). Where drainage is possible the breeding place may be destroyed by these methods. Since the flies have the habit of skimming over water and occasionally dipping their bodies in to it, the practice of pouring kerosene into water which kills the flies when they dip into it can be utilized. Animals should be kept away from places where the flies abound during hot part of the day.

For general flies control insecticidal spray with a residual effect are used in animal houses and on the animals themselves. There is also the possibility of dark panels with sticky adhesive as

drapes and there are a number of electrocution grids which may prove useful in animal's houses (Urquhart *et al*., 1996)

2.10.4 Chrysomya.

Pyriproxyfen, an insect growth regulator is recommended for the fly control by the world health organisation (WHO, 2006). The primary effect is to prevent metarmorphosis of pupae into adult, although, it also has embryogenic and reproductive effects (Invest and Lucas, 2008). Improvement of Sanitation of the area where people and animals live is necessary as many types of decaying matter serve as food source and should be cleared out of the area. Adult flies can be controlled by the use of insecticides.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area:

Three randomly selected districts in Gwagwalada Area Council were used for this study. Gwagwalada, a Capital town of Gwagwalada Area Council of the Federal Capital Territory, Nigeria, with an area of 1,043 km² and a population of 157,770 as at the 2006 census.

The vegetation of these districts is typical of the Northern Guinea woodland Savannah. However, because of the effects of annual bush fires, there are now both the Northern and the derived Savanna zones found in the districts. Gwagwalada Area Council falls within the *Glossina tachinoides* and *G.palpalis* infestation belt (Omoogun *et al*., 1991).Settlement patterns are mainly hamlets and farm compounds. There are seven different prominent languages spoken in this Area Council: namely Gbagi, Gade, Bassa, Koro, English, Fulani and Hausa which is the general spoken language. The main occupations of the people are civil service, farming, fishing and trading. Mixed farming of crop cultivation and animal production is the usual practice. The Area is known for its yam and maize production and supply to several parts of Nigeria. During the dry season, many of the herdsmen go on transhumance to southern States with their animals to get better pastures for their livestock and return at the end of the dry season. Infrastructural development is poor, with some villages like Kasanki, Dadabiri, and Old Kaida being inaccessible during the rainy season.

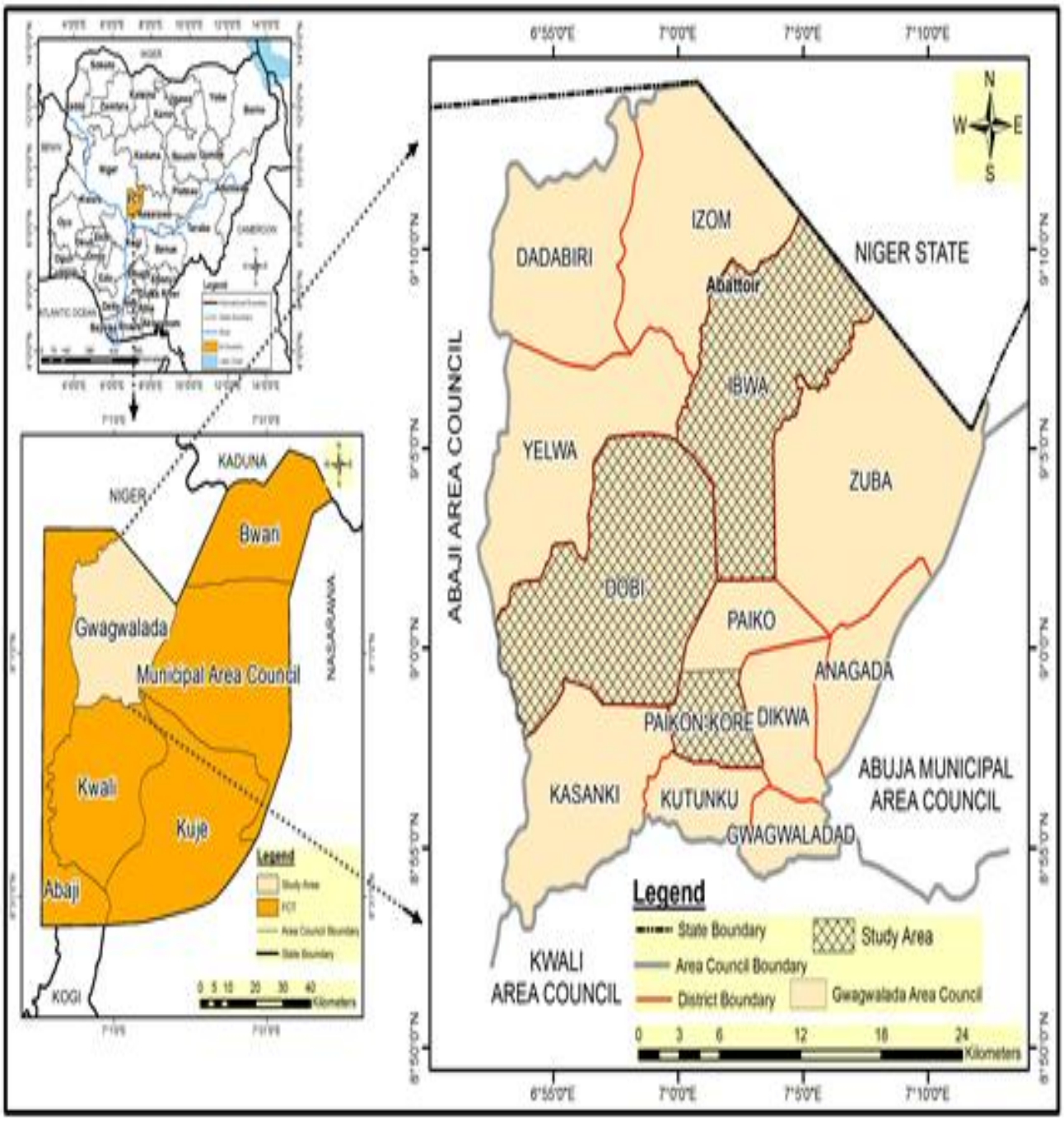

Fig. 2.1 Map of Gwagwalada Area Council showing the sampled Districts

Source: AbujaGeographicInformationSystem

3.2 Sample Size

Sample size was determined using the formula described by Thrushfield (1997). Thus:

$N=ZP^2q/d^2$

Where

N= Sample size

Z= Standard normal deviation for 95% confidence interval (1.96)

P= Expected prevalence 15.8% by (Ahmed, 2007)

D= desired absolute precision (0.05)

q= 1-p

$n= \frac{1.96^2 \times 0.157 \times (1-0.157)}{0.05^2}$

= 203

203 cattle were sampled in this study. However 243 cattle were sampled to increase precision.

3.3 Trapping of Haematophagous Flies

The sampling was undertaken during August for the wet / rainy season sampling and during February for dry season sampling. Both flies and blood samples were collected from cattle in the study area during these periods.

During each sampling season, a total of 30 traps: 15 Biconical traps (Challier and Larvessiere, 1973) and 15 Nzi traps were deployed to selected points along transects in order to cover large areas in each district .Traps were placed at about 100 - 150 meters apart and the coordinate of each point was taken . Five each of Biconical and Nzi traps were set at each of the three districts; Paikon- kore district, Dobi district and Ibwa district. The trapped flies were harvested 48 hours after deployment of the traps, and put in polythene bags and transported to the Entomology Laboratory, Department of Veterinary Parasitology and Entomology, Ahmadu Bello University

(A.B.U.). Zaria for identification and counting as described by Murray and Mc Intyre, (1977) and Mungube et al. (2012).

3.4 Blood Sampling

A simple random sampling without replacement was used to obtain a total of 243 blood samples from cattle of different ages. Cattle below one year were considered as young calves, and one year and above were regarded as adults. Two milliliters of blood were taken from each animal from the jugular vein into specimen bottles containing ethylene diaminetetra acetic acid (EDTA) as an anticoagulant and dispensed at one milligram permilliliter (mg/ml) of blood and conveyed in cold box on ice packs tothe Protozoology Laboratory of the Department of Veterinary Parasitology and Entomology A B U Zaria for analysis.

3.5 Processing of the Blood Samples

The techniques described below were carried out on the blood samples collected.

3.5.1 Thick blood smear preparation

Three drops of blood samples were placed on the center of a clean grease-free glass slide. The corner of a second slide was used to place the three drops of blood and with a circular motion; the blood was spread to cover an area of about 2cm in diameter, the smear was allowed to air-dry.

Procedure for staining with Giemsa stain.

The dried slide was then transfered to a jar of tap water for 5 second to lyse the Red blood cells, the water was then poured off. The staining jar was then filled with 10mls stock Giemsa stain plus 90mls of Buffer solution Ph 7.0. It was left in the staining solution for 25 – 30 minutes and rainsed with clean tap water.The slide was allowed to air dry on a wooden block.

The slide was examined using ×100 objective lens using oil immersion for presence of blood parasites.

3.5.2 Thin blood smear preparation

A pin-head drop of blood was dropped on one end of a clean grease-free glass slide, a spreader was firmly held in horizontal position so that an angle of about 30^{o} was formed between the spreader and the glass slide. The drop of blood was touched with the spreader in that position so that the blood ran along the line of contact between the two slides; the spreader was pushed gently but firmly along the surface of the horizontal slide and the blood was dragged behind the spreader to form the film with a feathered edge.

Procedure for staining with Giemsa stain.

The smear was dried, and then it was fixed in methyl alcohol for 3 minutes and transferred to a staining jar. Staining jar was filled with 1:10 dilution of Giemsa stain (10ml stock Giemsa stain plus 90mls of Buffer solution Ph 7.0), stained for 25–30 minutes. The stain was poured off and rinsed several times in running distilled water and rinsed once with buffer distilled water Ph 7.0 and left to air dry vertically in a wooden slide block.

The slide was examined using ×100 objective lens using oil immersion for presence of blood parasite.

3.5.3Haematocrit centrifuge techniqueprocedure

Each blood sample was mixed thoroughly and filled into two capillary tubes to approximately three quarter of its volume; the empty ends were sealed with a Bunsen flame and the tubes were placed in a micro haematocrit centrifuge in opposite direction to balance, with their sealed ends facing outwards.

They were centrifuged for three minutes at 12 revolution per minute (rpm)

The spun capillary tubes were placed on a glass slide and oil immersion applied on the buffy coat area and examined under a microscope using x10 objective lens (low power) for trypanosomemovement at the buffy coat region.

3.6 Data Analysis

Prevalences of haemoparasites were calculated and expressed as percentages.

a) Fisher's Exact Test test was used to test for association between prevalence of flies and difference in prevalence with respect to season.

b) The level of significance was determined at $p<0.05$ at 95% Confidence Interval (C.I)

For flies:

The prevalence of flies was determined by dividing the number of flies caught by traps from or in the district by the total number of flies caught in the study, multiplied by 100.

Mathematical expression;

$$\frac{number\ of\ flies\ caught\ by\ traps\ or\ in\ a\ district}{total\ number\ of\ flies\ caught} \text{ x } 100 \qquad \text{(Thrushfield, 1995)}$$

For blood samples/parasites:

The prevalence of parasites (*Trypanosoma species* inclusive) was determined by dividing the number of parasites detected from the total number of samples collected.

Mathematical expression;

$$\frac{number\ of\ parasites(trypanosome\ inclusive)detected}{total\ number\ of\ samples\ collected} \text{ x } 100 \qquad \text{(Thrusfield, 1995)}$$

CHAPTER FOUR

4.0 RESULTS

4.1 Distribution of Haematophagous Flies Caught in the Study Area in Febuary and August

A total of 430 flies were caught during August. Paikon Kore had the highest occurrence of flies with 186 (43.3%) followed by Ibwa 130(30.2%) and Dobi 114(26.5%). *Stomoxys species* were the dominating flies in all the districts with Paikon Kore, Dobi and Ibwa having 150(80.6%), 102(89.5%) and 88(67.7%), respectively. Other flies caught in the study included *M. domestica* (17.9%), *Tabanus species* (1.4%) *Haematopota spp* (0.5%), *Anthophila spp* (0.7%), *Culex spp* (0.2%), Butter fly larvae (0.2%)There was a statistically significant association between the location and the species of flies that were captured during the wet season ($p < 0.001$) (Table 4.1.)

A total of 175 flies were caught during February. Ibwa had the highest occurrence of flies with 79(45.1%) followed by Paikon Kore and Dobi having 60 (34.3%) and 36(20.6%) respectively. *Stomoxys calcitrans* was the dominating flies in all the districts with Paikon Kore, Dobi and Ibwa having 33(55.0%), 27(75.0%) and 53(67.1%) respectively it was followed by *M. domestica* (29.1%), *Tabanus species* (2.3%) *Caelifera spp* (2.3%), *Anthophila spp* (1.1%) and least by *Chrysomya spp* at 0.6%.There was no statistically significant association between the location and the species of flies that was captured during the dry season ($p > 0.05$) as shown in Table 4.2. *Chrysomya Spps* were only caught during February just as *Haematopota spp* and *Culex Spp* and Butterfly were only in August.

Table 4.3 shows the association between seasons with the number of flies caught in the 3 districts of Gwagwalada area council. A total of 560 flies were caught in the districts. Paikon Kore has the highest prevalence of flies with 246(40.7%) followed by Ibwa 209(34.5%) and then Dobi 150(24.8%). The number of flies caught was significantly higher ($X^2 = 13.523$; df=2;P = 0.001) during August than during February.

Table 4.1: Occurence of flies in the three districts of Gwagwalada Area Council of Abuja during August.

District	**Total (%)**	**Species of fly; (%)**						
		Stomoxys calcitrans	*Musca domestica*	*Tabanus Spp*	*Haematopota Spp*	*Anthophila spp (Bees)*	*Culex Spp*	Butter fly larvae
Paikon kore	186 (43.3)	150 (80.6)	35 (18.8)	0 (0.0)	1 (0.5)	0 (0.0)	0 (0.0)	0 (0.0)
Dobi	114 (26.5)	102 (89.5)	11 (9.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.9)
Ibwa	130 (30.2)	88 (67.7)	31 (23.8)	6 (4.6)	1 (0.8)	3 (2.3)	1 (0.8)	0 (0.0)
Total	430(100.0)	340 (79.1)	77 (17.9)	6 (1.4)	2 (0.5)	3 (0.7)	1(0.2)	1 (0.2)

Fisher's exact test = 31.132; d f = 2; p = 0.000

Table 4.2: Ocurence of flies in three districts of Gwagwalada Area Council of Abuja during February

		No. Species of fly					
District	Total (100%)	***Stomoxys species***	***Musca domestica***	***Tabanus Spp***	***Chrysomya Spp***	***Anthophila Spp***	***Caelifera Spp***
Paikon kore	60 (34.3)	33 (55.0)	22 (36.7)	2 (3.3)	1 (1.7)	2 (3.3)	0 (0.0)
Dobi	36 (20.6)	27(75.0)	5 (13.9)	2(5.6)	0 (0.0)	0 (0.0)	2 (5.6)
Ibwa	79 (45.1)	53 (67.1)	24 (30.4)	0 (0.0)	0 (0.0)	0 (0.0)	2 (2.5)
Total	175(100.0)	113 (64.6)	51 (29.1)	4(2.3)	1(0.6)	2 (1.1)	4 (2.3)

Fisher's exact test = 12.130; df = 2; p = 0.156

Table 4.3: Occurrence of flies in August and February season in the three districts in Gwagwalada Area Council.

District	Total no. of flies	Season	
		Wet; No. (%)	Dry; No. (%)
Paikon kore	246(40.7)	186(75.6)	60 (24.4)
Dobi	150(24.8)	114 (76.0)	36 (24.0)
Ibwa	209(34.5)	130 (62.2)	79 (37.8)
Total	605(100)	430 (71.1)	175 (28.9)

$X^2 = 13.523$; d f = 2; p = 0.001

4.2 Distribution of Flies Caught in Gwagwalada Area Council Based on the Type of Trap

Of the total 433 flies caught by bicornical trap during the study, Paikon kore had the highest with 183(42.3%) catch, followed by Ibwa, 159(36.7%) and Dobi 91(21.0%). *Stomoxys calcitrans* was the dominating fly in all the districts with 148(80.9%) in Paikon Kore, 84(92.3%) in Dobi and 102(64.2%) in Ibwa as shown in Table 4.3.

Similarly, of 172 flies were caught by Nzi traps in the study, Paikon Kore had the highest with 65(37.8%) catch, then Dobi 63(36.6%) and least Ibwa, with 44(25.6%). *Stomoxys calcitrans* dominated flies caught in all the districts having 42(65.6%), 48(76.2%) and 33(75%) in Paikon Kore, Dobi and Ibwa, respectively as shown in Table 4.4.

Table 4.5 shows the association between the number of flies caught and the type of trap. There was a statistically significant association ($p < 0.0001$) between the type of trap and prevalence of flies in the three districts of Gwagwalada Area Council. The distribution sites of the traps as to coordinates and elevations are presentated in Appendix I.

Table 4.4: Efficiency of Biconical trap in the three districts of Gwagwalada Area Council.

District	Total (%)	Species (types) of fly							
		Stomoxys calcitrans	*Musca domestica*	*Tabanus Spp*	*Culex Spp*	*Caelifera spp*	*Anthophila Spp*	*Chrysomya spp*	Butter fly Larva
Paikon kore	183 (42.3)	148 (80.9)	31(16.9)	2 (1.1)	0 (0.0)	0 (0.0)	1 (0.5)	1 (0.5)	0 (0.0)
Dobi	91(21.0)	84(92.3)	5(5.5)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (2.2)
Ibwa	159(36.7)	102 (64.2)	50(31.4)	0 (0.0)	1 (0.6)	1 (0.6)	5(3.1)	0 (0.0)	0 (0.0)
Total	433(100.0)	334(77.1)	86 (19.9)	2 (0.5)	1 (0.2)	1 (0.2)	6(1.4)	1 (0.2)	2(0.5)

Fisher's exact test =39.510; d f = 2; p = 0.000

Table 4.5: Efficiency of Nzi trap in the three districts of Gwagwalada area council.

District	Total (%)	Species (types) of fly						
		Stomoxys calcitrans	*Musca domestica*	*Tabanus Spp*	*Culex Spp*	*Caelifera Spp*	*Anthophila Spp*	*Haematopota Spp*
Paikon kore	65 (37.8)	42 (64.6)	21 (32.3)	0 (0.0)	1 (1.5)	0 (0.0)	1 (1.5)	0 (0.0)
Dobi	63 (36.6)	48 (76.2)	13 (20.6)	1 (1.6)	0 (0.0)	1 (1.6)	0 (0.0)	0 (0.0)
Ibwa	44 (25.6)	33 (75.0)	4 (9.1)	6 (13.6)	0 (0.0)	0 (0.0)	0 (0.0)	1 (2.3)
Total	172(100.0)	123 (71.5)	38 (22.1)	7 (4.1)	1 (0.6)	1 (0.6)	1 (0.6)	1 (0.6)

Fisher's exact test =24.824; d f = 2; p = 0.001

Table 4.6: Comparison of the efficiency of the traps in Gwagwalada Area Council.

District	Total	Types of trap	
		Biconical	Nzi
Paikon kore	248	183(73.8)	65 (26.2)
Dobi	154	91 (59.1)	63 (40.9)
Ibwa	203	159 (78.3)	44 (21.7)
Total	605	433 (71.6)	172 (28.4)

$X^2 = 25.193$; d f = 2; p = 0.000

4.3 Occurrence of *Trypanosoma species* in Gwagwalada Area Council.

Table 4.7 shows the occurrence of *Trypanosoma spp* in cattle in the study area. Only 2 (2.5%) out of the total of 81 samples collected in Paikon Kore were positive for *Trypanosoma spp*. While 163 blood samples collected from Dobi and Ibwa districts were negative of *Trypanosoma species.* Other haemoparasites were however obtained as shown in Table 4.8. Where *Theileria velefera* with the highest incidence of (8.0%) followed by *Babesia bovis* 12(6.0%), *Theileria mutans* 10(5.0%), *Anaplasma marginale* 9(4.5%), *Babesia bigemina* 7(3.5%) and then *Anaplasma bovis* 4(2.0%).

Table 4.7: Ocurrence of *Trypanosome* spp in cattle in Gwagwalada Area Council.

District	No. of samples	No. of positives	Prevalence (%)
Paikon kore	80	2	2.5
Dobi	82	0	0.0
Ibwa	81	0	0.0
Total	243	2	2.5

Table 4.8: Prevalence of blood parasites detected in cattle in the three districts of Gwagwalada area councils

Parasites	Paikon kore		Dobi		Ibwa
	No. of Samples	No. positive (%)	No. of Samples	No. positive (%)	No. of Samples
T. Velefera	81	5(6.2)	82	3(3.7)	80
B. bigemina	81	2(2.5)	82	2(2.4)	80
B. bovis	81	6(7.4)	82	4(4.9)	80
A.Centrale	81	1(1.2)	82	2(2.4)	80
A.marginale	81	3(3.7)	82	4(4.9)	80
T.mutans	81	7(8.6)	82	6(7.3)	80

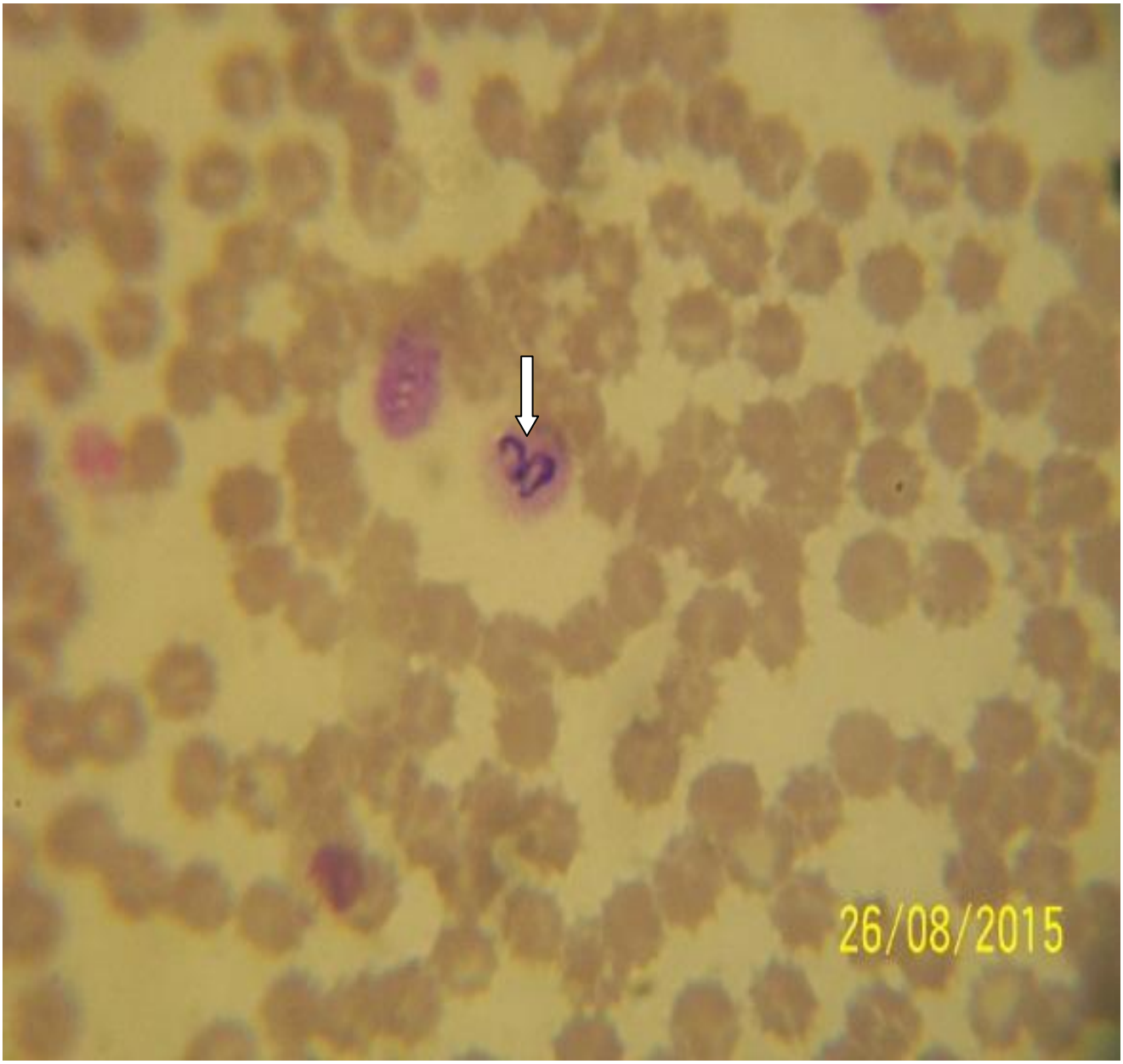

Plate 3: *Babesia bovis* x100

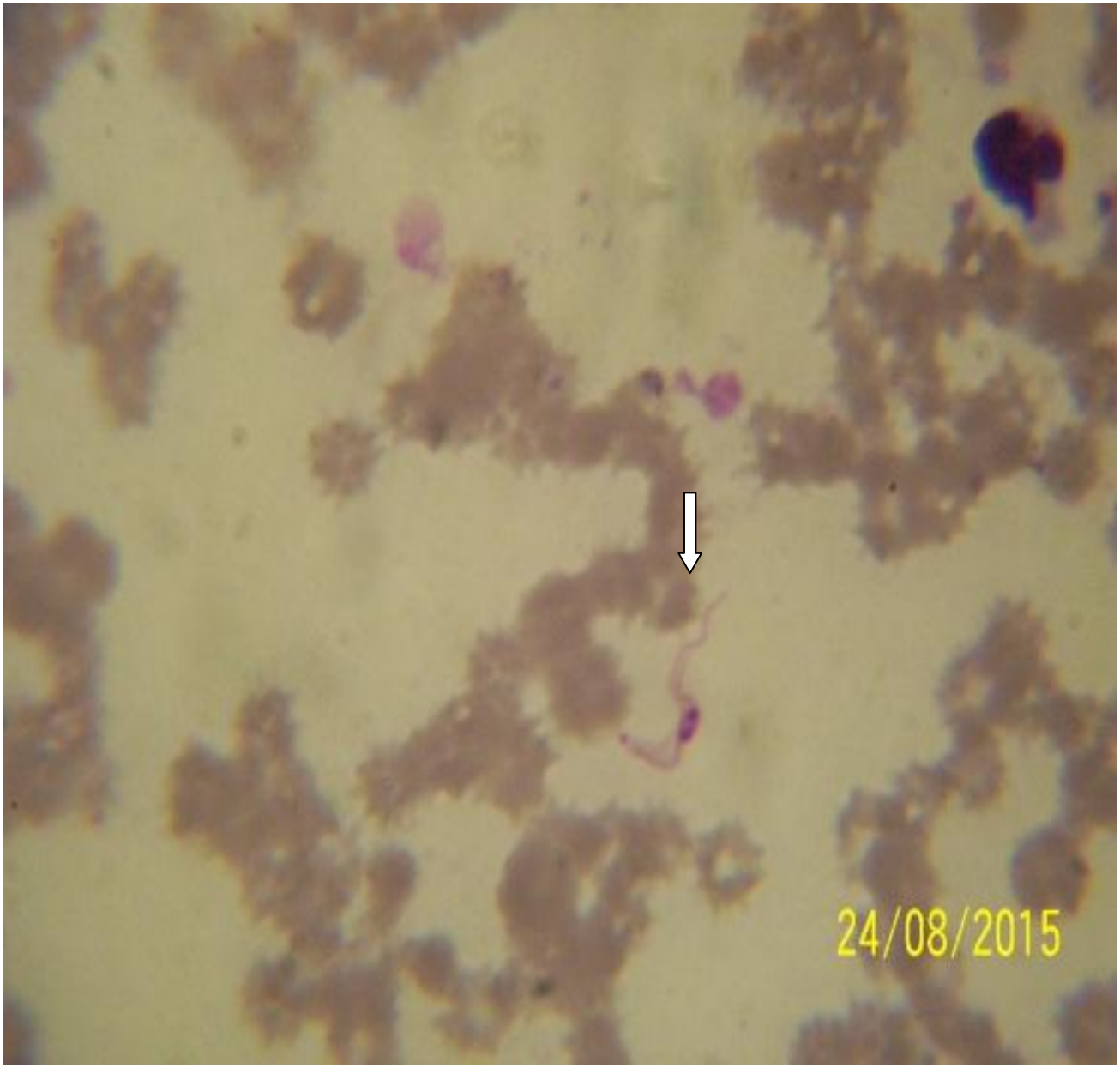

Plate 4: *Trypanosome vivax* x100

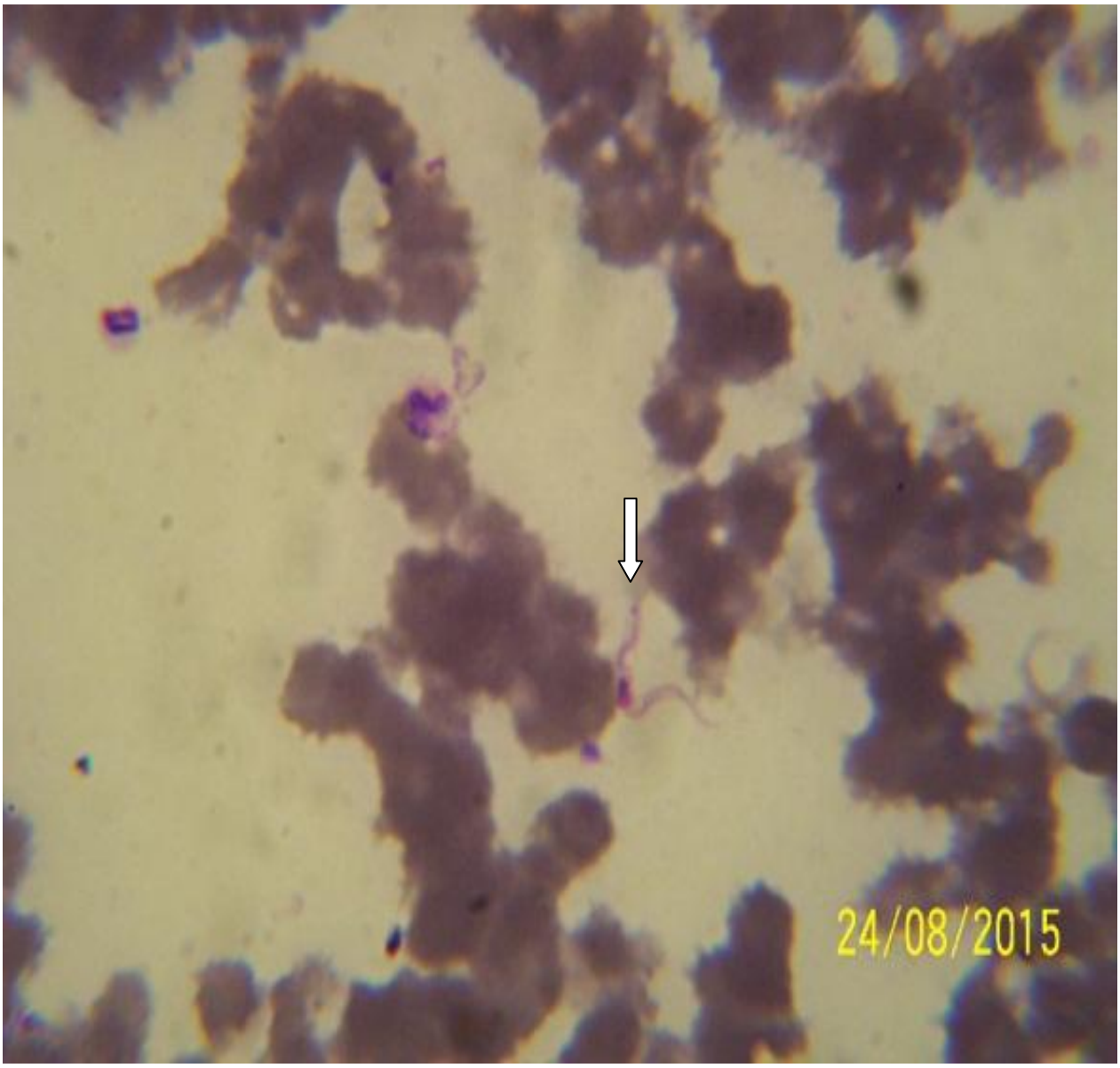

Plate 5*: Trypanosoma vivax* x100

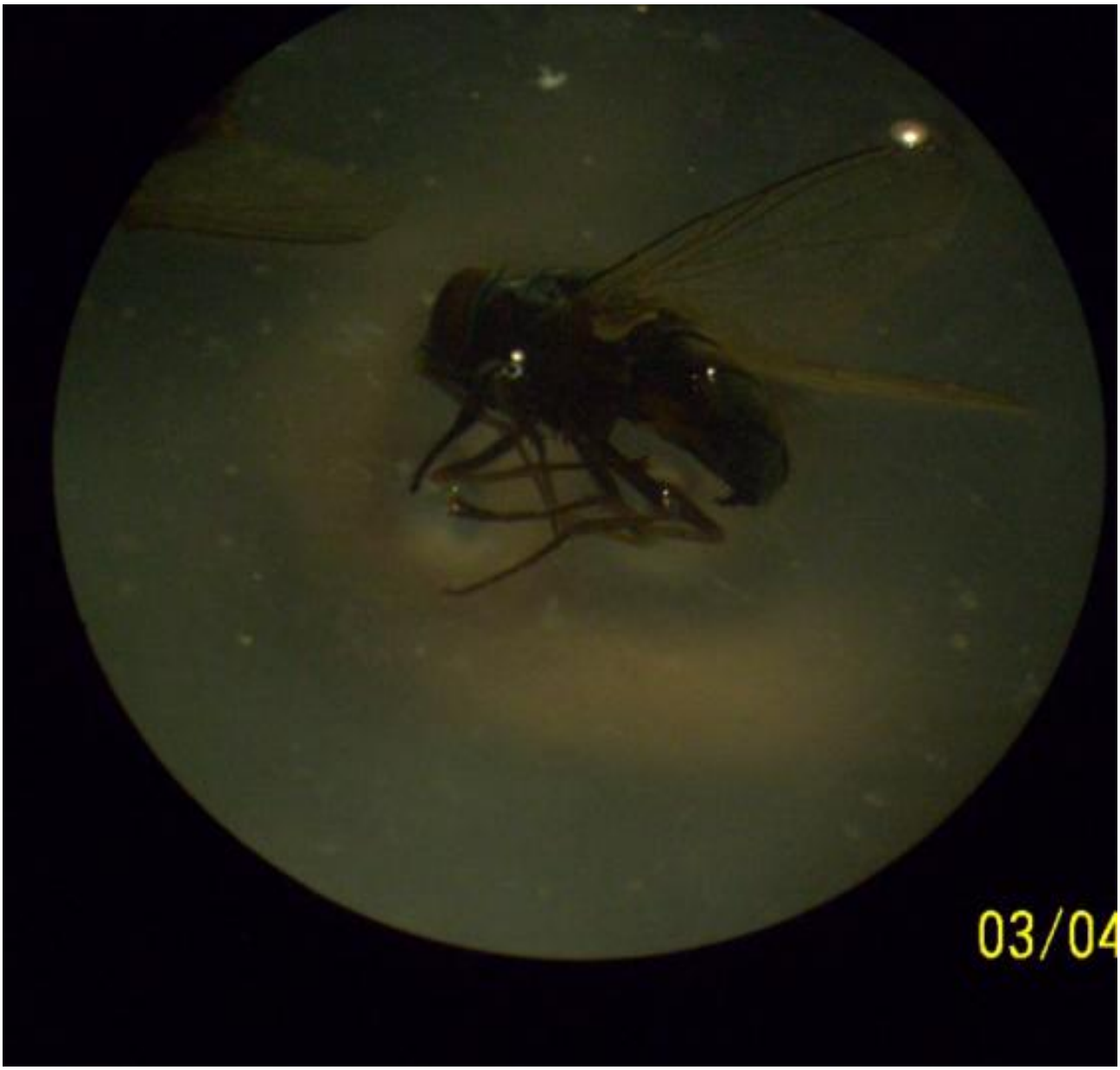

Plate 7: *Stomoxys specie* x100

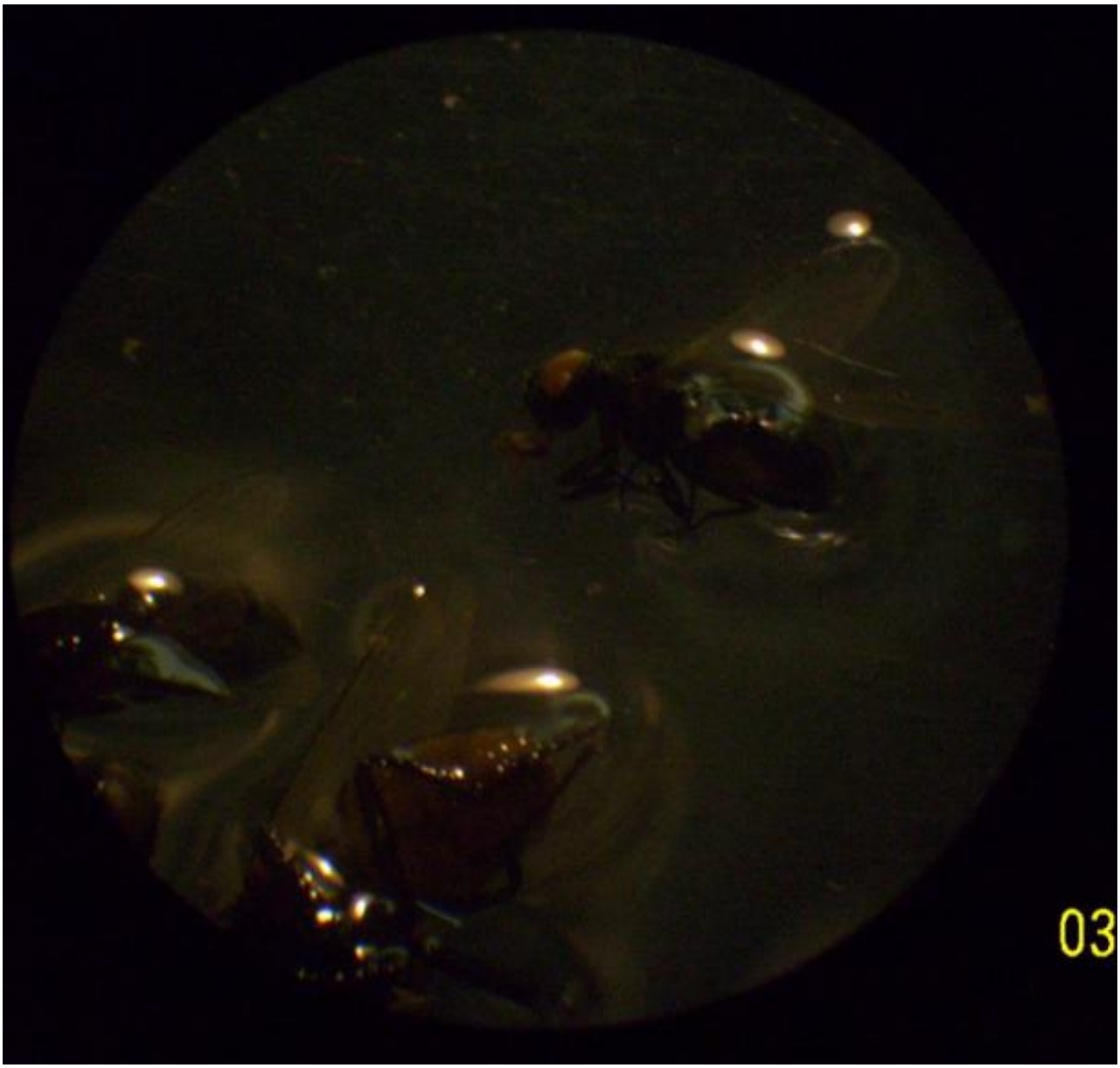

Plate 8*: Musca domestica* x100

CHAPTER FIVE

5.0 DISCUSSION

In the present study, no *Glossina* spp. was caught in the sampling areas using traps in all the three districts of Gwagwalada Area Council. Despite the fact that the study area falls within the Guinea Savanna vegetation zone of the country, in which the Savanna species of *Glossina* had been identified. The increasing extent and intensity of both farming and urbanization within this region has greatly changed the pattern of vegetation and land use, and inevitably led to an overall reduction in wildlife species, and in many places brought about their local extinction. Thus both natural habitats and hosts of tsetse, the vectors of trypanosomoses, have declined, which in turn has brought about a widespread reduction in the flies' distribution. The two Savanna species, *G. morsitans* and *G. longipalpis*, which typically have high trypanosome infection rates, have been most severely affected by the changes taking place within the subhumid zone, and their overall distribution has contracted. This explains why there was no *Glossina* spp caught in the study area.

Oluwafemi (2008) reported that forest areas along the rivers in some places in Nasarawa State have given way to farming activities and has led to moderate and low tsetse fly population in these areas. Even though the riverine and woodland vegetation are thick in the wet season, it is possible that dispersal could have been responsible for the absence of the fly due to low humidity. This observation is supported by the findings by Rogers and Robinson, (2004). The reduction of wild animals in the study area, especially as a result of poaching activities may have also contributed for the absence of tsetse flies, particularly due to lack of blood meal; and basically because they are game flies, they probably have followed the few animals into the interior of the bush. The observation supports the finding of Omoogun *et al.* (1991) and Hadis *et al*. (1995), who reported a decline in tsetse fly and attributed it to increased human activities such as poaching which depleted the wild life population.

During the study, total number of flies caught was 605. They were significantly higher than each other in numbers especially in Paikon kore district with the highest number of flies caught. This may probably be due to local concentration of breeding sites along the district; since these districts are characterized by sparse vegetations and thicket (particularly at Dobi) which provide poor breeding locations for the flies.

The most prevalent haematophagous fly in the study areas was *Stomoxys calcitrans* in all the districts during the two seasons followed by *Tabanus* spp fly although with lower prevalence (2.4 % and 1.4 % in the wet and dry seasons, respectively). The relative abundance of *Stomoxys calcitrans* may be associated with the differences in vegetation along the districts and probably due to the behavioral pattern of the fly, since *Stomoxyx calcitrans* is known to inhabit thickets and areas with close-canopy vegetation, particularly where light penetration is minimal (Berry, 1985). Similar findings were reported by Dipeolu (1977) and Ahmed *et al.* (2005). During their investigations on 29 livestock establishments in different towns across the country, Dipeolu (1977) also noted the dominance of *Stomoxys specie* over other haematophagous dipterans collected. This dominance may account for Dipeolu's (1974) earlier description of *Stomoxys* as a plague of cattle in Nigeria. Ahmed *et al*. (2005) reported that of the 15 species of blood sucking flies caught, the genus *Stomoxys* represented by *S. nigra* and *S. calcitrans* were most abundant, occurring throughout the year in large numbers.

The widespread presence of haematophagous dipterans (especially *Stomoxysspecies*) in the study area suggest that they could be playing a greater role in disease transmissions than previously thought. High prevalence of *Stomoxys* had been associated with transmission of anaplasmosis, trypanosomosis, bacterial diseases and viral diseases in animals (Najla *et al.,* 2011).

This study shows that there is significantly higher number of flies during the wet season than during the dry season. The prevalence of flies caught showed a general increase in relative abundance during the August and a decline in the Febuary. This could be due to availability of more suitable breeding sites in wet season where there are more green vegetations and high humidity. The finding of the study agrees with those of other researchers such as Ahmed *et al*., 2005 in Southern Kaduna. The low number of flies caught in the dry season could be due to bush burning leading to destruction of breeding sites and even burning of the insects and their eggs and larvae.

For the traps, Biconical traps caught more flies than its counterpart, the Nzi either district-wise and season-wise. This shows that Biconical traps may be more effective than the Nzi traps.

Trypanosoma vivax was detected only at Paikon kore which was found to be 2(2.5%), while no prevalence of *trypanosoma specie* was detected either in Ibwa or Dobi. Since no tsetse flies were caught in the study, the presence of *Trypanosome vivax* must have been as a result of mechanical transmission by other haematophagous flies. It could also probably be due to migration of infected animals from other districts into the study area (Paikon kore).

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions.

From the findings of this study, we conclude that:

1. There is possibility that *Glossina species* are absent or very scanty in the study area as no *Glossina species* was caught using the two types of traps (Biconical and Nzi traps) in the three districts of Gwagwalada Area Council.
2. *Stomoxys calcitrans* was the dominating fly in all the districts with 150(80.6%) in Paikon Kore, 102(89.5%) in Dobi and 88(67.7%) in Ibwa
3. Flies caught differ significantly within districts and seasonal variation with August having higher number, 430 (71.1%) of flies than dry season, 175 (28.9%).
4. Biconical traps were more efficient in catching of flies, 433 (71.6%) than Nzi trap, 172 (28.4%)
5. The presence of the *Trypanosoma vivax* even though very low (2.5%) can impose a health risk to animals in the study area due to the pathogenic potentials in cattle.
6. Of the other haemoparasites observed, *Theilera mutans* had the highest prevalence of 16 (8.0%) followed by *Babesia bovis* 12 (6.0%), *Theilera velifera* 10 (5.0%), *Anaplasma marginale* 9 (4.5%), *Babesia bigemina* 7 (3.5%) and *Anaplasma centrale* 4 (2.0%).

6.2 RECOMMENDATIONS

1. Further investigation of the entire districts in the Area Council is suggested in order to understand the actual status of the flies in the Area Council and the Federal Capital Territory, since the study area is in the Savanna belt where tsetse flies should ordinarily be found.
2. A broader investigation into nearby Area Councils and states should be considered as there may be possibilities of infected animals migrating into the study area.
3. The presence of trypanosomes, though at a very low rate poses a health risk to animals in the districts. The study recommends that proper control or eradication of Haematophagous flies is suggested in the study areas .
4. Biconical trap should be used in the control of biting flies as it was found to be more efficient than Nzi trap.

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APPENDIX I

COORDINATES AND ELEVATIONS OF THE TRAPS.

DOBI DISTRICT:
TRAPS WITH THEIR COORDINATES AND ELEVATIONS.

SNO.	TRAP	COORDINATE	ELEVATION
01	B1	09^0 0111.6" N $007^0$00'$00^0$2 E	215m
02	B2	09^0 01'16.7" N $006^0$59'58.0 E	209m
03	B3	09^0 01'16.7" N $006^0$59'51.6" E	216m
04	B4	09^0 01'17.0" N $006^0$59'47.9" E	210m
05	B5	09^0 01'19.2" N $006^0$59'46.4" E	205m
06	N5	09^0 01'23.6" N $006^0$59'46.0" E	199m
07	N1	$09^0$01'14.2" N $006^0$59'58.7" E	212m
08	N2	09^0 01'15.3" N 006^0 59'.55.0" E	214m
09	N3	$09^0$01'15.2"N $006^0$59'48.2" E	209m
10	N4	$09^0$01'18.1" N $006^0$59'48.2" E	211m

PAIKON-KORE DISTRICT:
TRAPS WITH THEIR COORDINATES AND ELEVATIONS.

SNO.	TRAP	COORDINATE	ELEVATION
01	B6	09^0 0112.7”N $006^0$59’54.1”E	220m
02	B7	09^0 00’50.1”N $006^0$59’58.4” E	244m
03	B8	09^0 00’45.1”N $006^0$59’55.3” E	250m
04	B9	09^0 0040.1” N $006^0$5960.3” E	247m
05	B10	09^0 0035.1” N $006^0$5965.6” E	248m
06	N6	09^0 0030.1” $006^0$5970.1” E	235m
07	N7	09001 0.1”N 0065990.7” E	205m
08	N8	009 15.1”N 006 000.3” E	215m
09	N9	090000.1” N 006000.3”E	224m
10	N10	081959.1” N 0065995.3” E	315m

IBWA DISTRICT:
TRAPS WITH THEIR COORDINATES AND ELEVATIONS.

SNO.	TRAP	COORDINATE	ELEVATION
01	B11	090025.1” N 0665975.1” E	224m
02	B12	090020.1”N 0065980.3” E	218m
03	B13	09 0015.1”N 006598.1” E	216m
04	B14	080085.1” N 0065985.4” E	215m
05	B15	080080.1” N 005980.3” E	230m
06	BN11	080075.1” N 005975.4” E	234m
07	N12	080090.1”N 0055990.4” E	210m
08	N13	080085.1”N 006598.5” E	225m
09	N14	080080.1” N 006598.0” E	222m
10	N15	080075.1” N 006597.5” E	205m

APPENDIX II

DRY SEASON SAMPLES (THIN BLOOD SMEAR) Cattle

S/NO	SLIDE NO	PARASITE SEEN	DISTRICT
01.	C1	NPF	Dobi
02.	C2	NPF	"
03.	C3	NPF	"
04.	C4	NPF	"
05.	C5	NPF	"
06.	C6	NPF	"
07.	C7	NPF	"
08.	C8	NPF	"
09.	C10	NPF	"
10.	C11	NPF	"
11.	C12	NPF	"
12.	C13	NPF	"
13.	C14	NPF	
14.	C15	NPF	
15.	C16	*Theileria mutans* seen	
16.	C17	*NPF*	
17.	C18	NPF	PAIKO KORE
18.	C19	NPF	
19.	C20	NPF	
20.	C21	NPF	
21.	C22	NPF	
22.	C23	NPF	
23.	C24	NPF	
24.	C25	NPF	

S/NO	SLIDE NO	PARASITE SEEN
25.	C26	*NPF*
26.	C27	NPF
27.	C28	*Theleria mutan* seen
28.	C29	NPF
29.	C30	NPF
30.	C31	NPF
31.	C32	NPF
32.	C33	NPF
33.	C34	NPF
34.	C35	NPF
35.	C36	NPF
36.	C37	NPF
37.	C38	NPF
38.	C39	NPF
39.	C40	NPF IBWA
40.	C41	NPF
41.	C42	NPF
42.	C43	NPF
43.	C44	NPF
44.	C45	NPF
45.	C46	NPF
46.	C47	NPF
47.	C48	NPF

S/NO	SLIDE NO	PARASITE SEEN
48.	C49	NPF
49.	C50	NPF

50.	C51	*Babesia bovis* seen
51.	C52	NPF
52.	C53	NPF
53.	C54	NPF
54.	C55	*Anaplasma bovis* seen
55.	C56	NPF
56.	C57	NPF
57.	C58	*Babesia bovis* seen
58.	C59	NPF
59.	C60	*Babesia bovis* seen
60.	C61	NPF
61.	C62	NPF
62.	C63	NPF
63.	C64	NPF
64.	C65	NPF
65.	C66	*Babesia bovis* seen
66.	C67	NPF
67.	C68	NPF
68.	C69	*Anaplasma bovis* seen
69.	C70	*Babesia bovis* seen

THIN BLOOD SMEAR CATTLE (DRY SEASON).

S/NO	SLIDE NO	PARASITE SEEN
70.	C71	NPF
71.	C72	NPF
72.	C73	NPF
73.	C74	*Theileria mutans* seen
74.	C75	NPF
75.	C76	NPF
76.	C77	NPF
77.	C78	*Theleria mutan* seen
78.	C79	NPF
79.	C80	*Theleria mutan* seen
80.	C81	NPF
81.	C82	NPF
82.	C83	NPF
83.	C84	*Theleria mutan* seen
84.	C85	NPF
85.	C86	NPF
86.	C88	NPF
87.	C89	NPF
88.	C90	*Anaplasma bovis* seen
89.	C91	NPF
90.	C92	*Theleria mutan* seen

THIN BLOOD SMEAR CATTLE DRY SEASON.

S/NO	SLIDE NO	PARASITE SEEN
91.	C93	*Theleria mutan* seen
92.	C94	NPF
93.	C95	NPF
94.	C96	NPF
95. 96 97. 98 100 101 102 103	C97 C98 C99 C100 C101 C102 C103 C104	NPF NPF NPF NPF *Theleria mutan* seen *Theleria mutan* seen NPF NPF

DRY SEASON SAMPLES (THIN BLOOD SMEAR)

S/NO	SLIDE NO	PARASITE SEEN
01.	S1	*Theileria ovis* seen
02.	S2	NPF
03.	S3	NPF
04.	S4	*Theileria ovis seen*
05.	S6	*NPF*
06.	S7	NPF
07.	S8	NPF
08.	S9	NPF
09	S10	NPF
10.	S11	NPF
11.	S12	NPF
12.	S13	NPF

13.	S14	NPF
14.	S15	NPF

THIN BLOOD SMEAR

S/NO	SLIDE NO	PARASITE SEEN
15.	S16	NPF
16. 19. 20.	S17 S18 S19	NPF *Theileria ovis* seen *Theileria ovis* seen

THIN BLOOD SMEAR DRY SEASON

S/NO	SLIDE NO	PARASITE SEEN
01.	G1	NPF
02.	G2	NPF
03.	G3	NPF
04.	G4	*Anaplasma marginale* seeen
05.	G5	NPF
06.	G6	NPF
07.	G7	NPF
08.	G8	*Anaplasma marginale* seeen
09.	G9	NPF
10	G10	NPF
11.	G11	
12.	G12	*Anaplasma marginall* seen
13.	G13	NPF
14.	G14	NPF
15.	G15	NPF
16.	G16	NPF

17.	G17	NPF
18.	G18	NPF

DRY SEASON SAMPLES (THICK BLOOD SMEAR) CATTLE

S/NO	SLIDE NO	PARASITE SEEN
01.	C1	NPF DOBI
02.	C2	NPF
03.	C3	NPF
04.	C4	NPF
05.	C6	NPF
06.	C7	NPF
07.	C8	NPF
08.	C9	NPF
09.	C10	NPF
10.	C12	NPF
11.	C13	NPF
12.	C14	NPF
13.	C15	NPF
14.	C16	NPF
15.	C17	NPF PAIKON KORE
16.	C18	NPF
17.	C19	*Theileria mutans*

DRY SEASON SAMPLE THICK BLOOD SMEAR

S/NO	SLIDE NO	PARASITE SEEN
18.	C20	*Theileria mutans*
19.	C21	*Babesia bigemina seen*
20.	C22	*NPF*
21.	C23	*NPF*
22.	C27	*Theileria mutans*
23.	C28	NPF
24.	C29	NPF
25.	C31	*Theileria mutans* seen
26.	C32	NPF
27.	C33	*NPF*
28.	C34	NPF
29.	C35	NPF
30.	C37	*Theileria mutans* seen
31.	C38	*Theleria mutans* seen
32.	C39	*Theileria mutans* seen
33.	C40	*Theileria mutans* seen IBWA
34.	C41	NPF
35.	C42	*Theileria mutans* seen
36.	C43	*NPF*
38.	C44	*NPF*
39.	C45	*NPF*
40.	C46	*NPF*

THICK BLOOD SMEAR (DRY SEASON)

S/NO.	SLIDE NO.	PARASITE SEEN
41.	C47	*Theileria mutans* seen
42.	C48	NPF
43.	C49	NPF
44.	C50	*NPF*
45.	C51	*Babesia bovis seen*

THIN BLOOD SMEAR)

S/NO	SLIDE NO	PARASITE SEEN
19.	C52	NPF
20.	C53	NPF
21.	C55	NPF
22.	C56	*Anaplasma marginall* seen
23.	C57	*Babesia bigemina* seen
24.	C58	*NPF*
25.	C59	NPF
26.	C60	*Theileria mutans seen*
27.	C61	NPF
27.	C61	NPF
28,	C62	*Anaplasma marginale* seen
29.	C63	NPF
30.	C64	NPF
31.	C65	NPF
32.	C66	*Babesia bigemina* seen
33.	C67	NPF
34.	C68	NPF
35.	C69	NPF
36.	C69	NPF

37.	C70	*Babesia bovis* seen
38.	71	NPF
39.	C72	NPF
40.	C73	NPF
41.	C74	*Theileria mutans* seen
42.	C75	NPF

S/NO	SLIDE NO	PARASITE SEEN
43.	C76	NPF
44.	C77	NPF
45.	C78	NPF
46.	C79	NPF
47.	C80	*Theileria mutans* seen
48.	C81	NPF
49.	C82	NPF
50.	C83	NPF
51.	C84	*Theileria mutans* seen
52.	C85	NPF
53.	C86	NPF
54.	C87	NPF
55.	C88	NPF
56.	C89	NPF
57.	C90	*Theileria mutans* seen
58.	C91	NPF
59.	C92	*Theileria mutans* seen

60.	C93	*Theileria mutans* seen
61.	C94	NPF
62.	C95	NPF
63.	C96	NPF

S/NO.	SLIDE NO.	PARASITE SEEN
64	C97	*Theileria ovis* seen
65.	C98	*Theileria ovis* seen
66.	C99	NPF
67.	C100	*Theileria ovis* seen
68.	C101	NPF
69.	C103	*Theileria ovis* seen
70.	C104	NPF

THICK BLOOD SMEAR (DRY SEASON).

S/NO	SLIDE NO	PARASITE SEEN	DISTRICT
01.	S1	*Theileria ovis* seen	DOBI
02.	S2	*Theileria ovis* seen	
03.	S3	*Anaplasma ovis ,Theleria ovis* seen	
04.	S4	*NPF*	
05.	S5	*NPF*	
06.	S6	NPF	
07.	S7	NPF	
08.	S8	NPF	
09.	S9	NPF	
10.	S10	*NPF*	
11.	S11	NPF	
12.	S12	*NPF*	
13.	S13	NPF	PAIKO KORE
14	S14	NPF	
15.	S15	*Theileria ovis* seen	
16.	S16	*Theileria ovis* seen	
17.	S17	NPF	
18.	S18	NPF	
17.	S19	NPF	

THICK BLOOD SMEAR DRY SEASON .

S/NO	SLIDE NO	PARASITE SEEN
01.	G1	NPF
02.	G2	NPF
03.	G3	NPF

04.	G4	*Theileria ovis* seen
05.	G5	NPF
06.	G6	NPF
07.	G7	NPF
08.	G8	NPF
09.	G9	NPF
10	G10	*Theileria ovis* seen
11.	G11	*NPF*
12.	G12	NPF
13.	G13	NPF
14.	G14	NPF
15.	G15	NPF
16.	G16	NPF
17.	G17	NPF
18.	G18	NPF

WET SEASON SAMPLES

THIN BLOOD SMEAR WET SEASON (CATTLE)

S/NO.	SLIDE NO.	PARASITE SEEN	DISTRICT
01.	C1	NPF	DOBI
02.	C2	NPF	
03.	C3	NPF	
04.	C4	NPF	
05.	C5	*Anaplasma bovis* seen	
06.	C6	NPF	
07.	C7	NPF	
08.	C8	NPF	

09.	C9	NPF
10.	C10	*Babesia bovis* few seen
11.	C11	NPF
12.	C12	NPF
13.	C13	NPF
14.	C14	NPF
15.	C15	NPF
16.	C16	NPF
17.	C17	NPF
18.	C18	*NPF*
19.	C19	NPF
20.	C20	*NPF*
21.	C21	NPF
22.	C22	*Babesia bovis* seen
23.	C23	NPF
24.	C24	NPF
25.	C25	NPF

S/NO.	SLIDE NO.	PARASITE SEEN PAIKO KORE
26.	C26	*NPF*
27.	C27	NPF
28.	C28	*Theileria mutans seen*
29.	C29	NPF
30.	C30	NPF
31.	C31	*Trypanosoma spp*ⓛseen
32.	C32	NPF
33.	C33	*NPF*
34.	C34	NPF
35.	C35	NPF

36.	C36	NPF
37.	C37	NPF
38.	C38	NPF
39.	C39	*Babesia bovis* seen
40.	C40	NPF
41.	C41	NPF
42.	C42	NPF
43.	C43	*NPF*
44.	C44	NPF
45.	C45	NPF
46.	C46	NPF
47.	C47	*NPF*
48.	C48	NPF
49.	C49	NPF

THIN BLOOD SMEAR WET SEASON (CATTLE)

S/NO.	SLIDE NO.	PARASITE SEEN	DISTRICT
50.	C50	NPF	IBWA
51.	C51	*Babesia bovis*② seen	
52.	C52	*NPF*	
53.	C53	NPF	
54.	C54	NPF	
55.	C55	*Babesia bigemna* seen	
56.	C56	NPF	
57.	C57	NPF	
58.	C58	NPF	
59.	C59	NPF	
60.	C60	*Theileria mutans seen*	
61.	C61	NPF	
62,	C62	NPF	
63.	C63	NPF	
64.	C64	NPF	
65.	C65	*Babesia bigemna* seen	
66.	C66	NPF	
67.	C67	NPF	
68.	C68	NPF	
69.	C69	*Babesia bigemna* seen	
70.	C70	*Theileria mutans seen*	

THIN BLOOD SMEAR WET SEASON.

S/NO.	SLIDE NO.	PARASITE SEEN	DISTRICT
01.	S1	NPF	DOBI

02.	S2	NPF
03.	S3	*Theileria ovis* few seen
04.	S4	NPF
05.	S5	*NPF*
06.	S6	*NPF*
07.	S7	*NPF*
08.	S8	NPF
09.	S9	*Theileria ovis* few seen
10.	S10	NPF

S/NO.	SLIDE NO.	PARASITE SEEN
11.	S11	*NPF*
12.	S12	*NPF*
13.	S13	NPF

THIN BLOOD SMEAR WET SEASON.

S/NO.	SLIDE NO.	PARASITE SEEN	DISTRICT
01.	G1	NPF	DOBI
02.	G2	NPF	
03.	G3	NPF	
04.	G4	*Anaplasma marginale*few seen	
05.	G5	NPF	
06.	G6	NPF	
07.	G7	NPF	
08.	G8	*Anaplasma marginale*few seen	
09.	G9	NPF	
10.	G10	NPF	
11.	G11	NPF	
12.	G12	*Anaplasma marginale*few seen	

13.	G13	NPF
14.	G14	NPF

WET SEASON SAMPLES

THICK BLOOD SMEAR WET SEASON (CATTLE)

S/NO.	SLIDE NO.	PARASITE SEEN	DISTRICT
01.	C1	NPF	DOBI
02.	C2	*NPF*	
03.	C3	NPF	
04.	C4	NPF	
05.	C5	NPF	
06.	C6	NPF	
07.	C7	NPF	
08.	C8	NPF	
09.	C9	NPF	
10.	C10	*Babesia bigemina* seen	
11.	C11	NPF	
12.	C12	NPF	
13.	C13	NPF	
14.	C14	NPF	
15.	C15	NPF	
16.	C16	NPF	
17.	C17	NPF	
18.	C18	*Babesia bovis* few seen	
19.	C19	NPF	
20.	C20	*Babesia bovis* seen	
21.	C21	NPF	
22.	C22	NPF	

23.	C23	NPF
24.	C24	NPF
25.	C25	NPF

S/NO.	SLIDE NO.	PARASITE SEEN PAIKO KORE
26.	C26	*Babesia bovis* seen
27.	C27	NPF
28.	C28	*NPF*
29.	C29	NPF
30.	C30	NPF
31.	C31	*Trypanosoma spp*①seen
32.	C32	NPF
33.	C33	*Babesia bovis* few seen
34.	C34	NPF
35.	C35	NPF
36.	C36	NPF
37.	C37	NPF
38.	C38	NPF
39.	C39	*NPF*
40.	C40	NPF
41.	C41	NPF
42.	C42	NPF
43.	C43	*Theileria mutans* few seen.
44.	C44	NPF
45.	C45	NPF
46.	C46	NPF
47.	C47	*Babesia bovis*②seen
48.	C48	NPF

49.	C49	NPF

THICK BLOOD SMEAR WET SEASON (CATTLE)

S/NO.	SLIDE NO.	PARASITE SEEN DISTRICT
50.	C50	NPF IBWA
51.	C51	*Babesia bovis*②seen
52.	C52	*NPF*
53.	C53	NPF
54.	C54	*Babesia bovis*seen
55.	C55	*Babesia bovis*seen
56.	C56	*NPF*
57.	C57	*Babesia begemina* seen
58.	C58	NPF
59.	C59	NPF
60.	C60	*Theileria mutans* few seen
61.	C61	NPF
62,	C62	*Anaplasma marginale* seen
63.	C63	NPF
64.	C64	NPF
65.	C65	NPF
66.	C66	NPF
67.	C67	NPF
68.	C68	NPF
69.	C69	NPF
70.	C70	*Theileria mutans* few seen

THICK BLOOD SMEAR WET SEASON.

S/NO.	SLIDE NO.	PARASITE SEEN DISTRICT
01.	S1	NPF DOBI
02.	S2	*Theileria ovis* few seen
03.	S3	*Theileria ovis* few seen
04.	S4	*Theileria ovis* few seen
05.	S5	*Theileria ovis* several seen
06.	S6	*NPF*
07.	S7	*NPF*
08.	S8	NPF
09.	S9	NPF
10.	S10	*Anaplasma ovis* few seen

S/NO.	SLIDE NO.	PARASITE SEEN
11.	S11	*Theileria ovis* seen
12.	S12	*Theileria ovis* seen
13.	S13	NPF

THICK BLOOD SMEAR WET SEASON.

S/NO.	SLIDE NO.	PARASITE SEEN DISTRICT
01.	G1	NPF DOBI
02.	G2	NPF
03.	G3	*Theileria ovis* few seen
04.	G4	*NPF*
05.	G5	NPF
06.	G6	*Theileria ovis* few seen
07.	G7	NPF
08.	G8	NPF

09.	G9	NPF *Theileria ovis* few seen
10.	G10	NPF
11.	G11	*Theileria ovis* few seen
12.	G12	NPF
13.	G13	NPF
14.	G14	NPF

Appendix III

Summary of positive slides

		Parasitological Techniques				
	District	Thin blood smear	Tag no.	Thick blood smear	Tag no.	Animal tested
	Dobi			Theleria mutans	C19, 20,27, 31	Cattle
				Babesia bigemina	C21	
	Paik on-kore			Theleria mutans	C37, 38,39	
				Babesia bovis	C51, 60	
				Babesia bigemina	C58, 66	
	Ibwa			Babesia bovis	C70	
				Theleria mutans	C74, 80,84,90, 92,93,40, 42	
	Dobi	Theleria mutans	C16, 28			
		Babesia bovis	C26			

	Paikon-kore	Babesia bovis	C51, 60			
		Babesia bigemina	C58, 66			
	Ibwa	Babesia bovis	C70			
		Theleria mutans	C74, 80,84,90, 92,93,101, 102			
	Dobi			Theilera ovis	S01, 02,04, 13	Sheep
	Paikon-kore			Anaplasma ovis	S3	
				Theilera ovis	S3	
	Ibwa			Theilera ovis	S15, 16	
				Babesia bigemina	C56, 57	
				Theleria mutans	C60, 70	
				Anaplasma marginale	C62	
	Dobi	Theilera ovis	S01, 04			
	Paikon-kore	Theilera ovis	S18, 19			
	Ibwa	Theilera ovis	S19			

	Dobi			Theileraovis	G04	Goats
	Paikon-kore			Theileraovis	G10	
	Dobi	Anaplasma marginale	G04			
	Paikon-kore	Anaplasma marginale	G08			
	Ibwa	Anaplasma marginale	G12			
WET SEASON						
	Dobi			Babesiabovis	C21,18,20,26	Cattle
				Babesiabigemina	C10	
	Paikon-kore			Trypanosome sp.	C31	
				Baberiabovis	C33,47,51,54	
				Anaplasma bovis	C39	
				Theileramutans	C43	
	Ibwa			Babesiabovis	C55,65,69	

				Babesia bigemina	C56, 57	
				Theileramutans	C60, 70	
				Anaplasma marginale	C62	
	Dobi	Anaplasma bovis	C5			
		Babesia bovis	C10			
	Paikon-kore	Theileramutans	C28			
		Trypanosome sp.	C31			
		Babesia bovis	C22			
	Ibwa	Babesia bovis	C39, 51			
		Babesia bigemina	C55, 65,69			
		Theileramutans	C60, 70			
		Anaplasma marginale	C62			
	Dobi			Theileraovis	S2,3, 4,5	Sheep
				Anaplasma ovis	S3	
	Paikon-kore			Babesia ovis	S10	
				Theileraovis	S12	

	Ibwa			Theileraovis	S15	
	Paikon-kore	Theileraovis	S3			
	Ibwa	Theileraovis	S9			
	Dobi			Theileraovis	G3,6	Goats
	Paikon-kore			Theileraovis	G11	
	Ibwa			Theileraovis	G9	
		Anaplasma marginale	G04			
		Anaplasma marginale	G08			
		Anaplasma marginale	G12			