FACTORS AFFECTING PUPARIA DISTRIBUTION AND MORTALITY IN A NATURAL POPULATION OF *GLOSSINA PALLIDIPES* AUSTEN (DIPTERA: GLOSSINIDAE) AT NGURUMAN, KENYA

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by

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DECLARATION BY CANDIDATE

This thesis is my own original work and has not been presented for a degree in any other University.

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DEDICATION

To my entire family especially my parents, my wife Farida and baby Judith for their perseverance was a source of encouragement and their love and understanding made this task worthwhile.

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ABSTRACT

Glossina pallidipes Austen is an important vector of both human and animal trypanosomiasis in Kenya. In an effort to generate information that could be useful for its control, studies were carried out on its ecology to investigate the factors affecting the distribution and mortality of *G.pallidipes* puparia at Nguruman, Kenya.

It was revealed by these studies that natural puparia mortalities were caused by developmental and emergence failures (29.0 %), pupal tissue degeneration (16.1 %), puparia parasitism (4.4 %) and fungal infections (50.5 %). Regression analysis of k-values of each submortality against the total mortality (K) showed that fungal infection was the most significant factor ($r= 0.55$, $P< 0.05$) causing puparia mortality in the field.

It was also revealed by these studies that submersion of puparia in water for more than 6 hours caused death probably through asphyxiation indicating that waterlogging or flooding could result in death of tsetse puparia.

These studies also showed that puparia parasites of *G.pallidipes* comprised of two diptera species of the

family Bombyliidae, namely, *Exhyalanthrax lugens* Lw and *E. beckerianus* Bezzi. Mortality caused by these parasites was highest (35-100 %) during the short rains.

When the weights of field collected tsetse puparia were determined, it was found that the mean weight of ecloded puparia of *G.pallidipes* was 33.30 ± 0.6 mg. Puparia whose weight was less than 28.3 mg were recovered when they had died before eclosion. It was also shown that parasitised puparia could not be distinguished from unparasitised puparia on the basis of weight, although the former puparia on average were lighter than the latter puparia.

There were also significant ($t = 13.2$, $P < 0.01$) seasonal variations in the mean weight of puparia collected during the dry and rainy season. Puparia collected during the rainy season were heavier (34.40 mg,) than those that were collected during the dry season (32.80 mg,).

It was also revealed by these studies that there was a significant positive correlation ($r = 0.7$, $P < 0.01$) between puparia weight and relative humidity in the previous month. This indicated that relative humidity in the previous month was an important factor that influenced puparia weight probably because it was the vital climatic factor experienced by the parent female flies.

These studies also showed that there were differences in the relative abundance and distribution of puparia recovered from different vegetation types. This was probably due to a seasonal shift in larviposition sites from the lowland riverine vegetation to the valley woodland vegetation during the rainy season.

Studies were also carried out to determine the effect of adult tsetse suppression control on the abundance and distribution of *G.pallidipes* puparia. It was revealed that there was a decline in the live puparia number per site in the suppression area which coincided with a decline in the mean catches of adult flies within the area, suggesting that the distribution and abundance of puparia was to a large extent dependent on the distribution and abundance of adult female flies.

From these studies it was also revealed that the most important predators of *G.pallidipes* puparia at Nguruman belonged to two families namely, formicidae (Order: Hymenoptera) and gryllidae (Order: Orthoptera). The most abundant formicid ants belonged to the genus *Pheidole* while those of gryllids belonged to the genus *Gryllus*. It was also revealed that there were seasonal fluctuations in the numbers of formicid ants and gryllids caught in larviposition sites. The predators were most abundant during the short rainy season and

during the cold-dry season. Low numbers of the predators were found during the long rainy season probably due to flooding of larviposition sites.

Field experiments were also conducted to determine the rates of tsetse puparia predation buried at different densities and soil depths. It was shown that predation of puparia buried at different soil depths varied significantly ($F= 15.7$, $P< 0.05$). Puparia left on the soil surface were the most damaged compared to those buried within the soil. It was also revealed that there was no significant relationship ($P> 0.05$) between overall predation rate and puparia density, indicating that puparia predation was not a density dependent factor.

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CHAPTER ONE

INTRODUCTION

1.1 The biology of *Glossina* species

Tsetse belong to the order Diptera and the genus *Glossina*. The genus consists of several species which occur only in Africa between latitudes 15° N and 28° S. Tsetse flies are the vectors of trypanosomes that cause African trypanosomiasis. These parasites affect man and his livestock causing sleeping sickness or human trypanosomiasis and nagana or animal trypanosomiasis, respectively. Tsetse flies and the disease they transmit occur in an area of about 10 million square kilometres spanning 37 sub-Saharan countries (Anon., 1979).

The pathogenic human trypanosomes transmitted by tsetse flies are *Trypanosoma brucei gambiense* Dutton 1902 and *T. brucei rhodensiense* Stephens and Fantham 1910 while those of cattle are *T.brucei brucei* Plimmer and Bradford 1899, *T.congolense* Broden 1904 and *T.vivax* Ziemann 1905 (Jordan, 1986). Transmission of these pathogenic trypanosomes is brought about when a fly bites an infected host and then picks up the parasites in the blood. These become infective after undergoing

morphological and physiological changes in the gut or proboscis of the fly and are transmitted to a susceptible host during the next bite. Once a fly becomes infective it remains so for life and may infect any animal on which it feeds (Jordan, 1986).

Glossina are adenotrophically viviparous and larviposit at the third instar larval stage about nine days after the egg is dropped in the uterus (Jordan, 1986). Only one larva is deposited at a time. Once the larva is deposited, usually in the afternoon or late evening, being negatively phototactic it burrows into the soil to a depth of about 2- 4 cm where it pupates and becomes enclosed in a black case, the puparium (Jordan, 1986). At the end of the incubation period, which is usually 20-40 days, depending on weather conditions, a teneral fly emerges from the pupa. Larviposition occurs throughout the year but may be depressed and more dispersed in the rains during which time puparia may be difficult to find.

1.2 Tsetse and trypanosomiasis

Human trypanosomiasis has been singled out as being one of the six major world diseases that should be controlled (Anon., 1979). From this observation and the fact that tsetse are resident in the tropics it is

apparent that the future of tropical Africa depends to a large extent on the efficient control of the flies. Fortunately, human trypanosomiasis does not occur throughout the tsetse belt of Africa (Cunningham, 1979; Jordan, 1986). It tends to occur in limited areas where unfortunately it has been the cause of millions of deaths. Severe epidemics of the disease have occurred in the past, for instance between 1902 and 1905 in Busoga (Uganda) some 200,000 people died from human trypanosomiasis (Cunningham, 1979). More recently during the 1980's similar epidemics of human sleeping sickness have been reported in the same area (Mulamberi, 1989). In Zaire it was estimated that an epidemic of sleeping sickness during the 1970's claimed some half a million lives (Cunningham, 1979). Today it is recognised that constant vigilance is a necessity for the prevention of severe outbreaks of sleeping sickness (Cunningham, 1979; Jordan, 1986).

However, in economic terms, trypanosomiasis of domestic animals (nagana), rather than sleeping sickness, is a much more important disease (Cunningham, 1979)). The disease kills large numbers of cattle and in this way it is a serious constraint to rural development by way of preventing rearing of livestock, especially cattle, over large areas of Africa (Jordan, 1986). Nagana is a particularly difficult disease to

control as it is a zoonosis (Soltys, 1971). This implies that wild game acts as a reservoir of infection although they do not themselves exhibit symptoms of the disease (Soltys, 1971)). Tsetse flies can thus acquire their infections when feeding on wild game and then pass them onto domestic animals and in certain cases to man as well (Soltys, 1971).

1.3 The distribution of tsetse in Kenya

Several species of tsetse flies occur in Kenya. These include *G.pallidipes*, *G.swynnertoni* Austen 1923, *G.austeni* Newstead 1910, *G.brevipalpis* Newstead 1910, *G.longipennis* Corti and *G.fuscipes* Newstead 1910. Of these species *G.pallidipes* and to a lesser extent *G.fuscipes* are known vectors of human sleeping sickness. On the other hand *G.pallidipes*, *G.swynnertoni*, *G.austeni* and *G.brevipalpis* transmit nagana to domesticated animals. These species occur either singly or co-exist with other species. In these studies *G.pallidipes* was chosen for investigations because of its being a major vector of both sleeping sickness in man and nagana in domestic animals in Kenya (England and Baldry, 1972). *G.pallidipes* may be found existing sympatrically or allopatrically with other species in its ecological range. For example, in Coast

Province, *G.pallidipes* co-exists with *G.austeni*, *G.brevipalpis* and *G.longipennis* in thicketed woodland or forest grassland interface and in large areas of semi-arid acacia - commiphora thorn bush (Snow, 1980). The species also co-exists with *G.brevipalpis* in dense primary and secondary thickets dominated by *Lantana camara* L (Snow, 1980).

At Kibwezi in Machakos district a low density of *G.pallidipes* co-exists with *G.brevipalpis* in woodland thickets (Owaga, 1985).

In Meru National park in Eastern province *G.pallidipes* is confined to areas where acacia-combretum are the prominent vegetation communities (Lambrecht, 1980). At Nguruman in the Rift valley province, where the current investigations were conducted, *G.pallidipes* co-exists with *G.longipennis* and occupies most of the vegetation habitats although it is somewhat restricted to riverine thickets during the dry seasons (Etten, 1981). During rainy seasons, it tends to spread into other vegetation habitats including the open plains, mixed woodland and hilly valley sites (Etten, 1981).

In Lambwe valley (Nyanza province) *G.pallidipes* is found in continous hill thickets and thicket clumps at the bottom of the valley and in woodland. However, the extensive grasslands with occasional trees is little

used by this species except as a wet season dispersal area (Allsopp and Baldry, 1972). Turner (1981) reported that in addition to the thickets, exotic coniferous plantations bordered by *Euphorbia tirucalli* L. constituted a suitable habitat for the species in the area. In this region *G.pallidipes* is of both medical and veterinary importance because in addition to animal trypanosomiasis a low grade transmission of *T.rhodensiense* exists in the human settlements around the Ruma game area (Otieno pers comm.).

1.4 Tsetse and trypanosomiasis control in Kenya

Tsetse control in Kenya started in the 1940s (Moggridge, 1949b). The selective application of residual insecticides like DDT and dieldrin to tree trunks, lower branches of trees and other resting sites was the main method of tsetse control in Kenya (Burnett *et al.*, 1957). This method resulted in successful elimination of *G.pallidipes* in some areas in Nyanza province and of *G.fuscipes* in some areas along the shores of Lake victoria (Glover *et al.*, 1960; Baldry, 1971).

Attempts have been made since 1980 to eliminate trypanosomiasis by controlling *G.pallidipes* in the Lambwe valley (Turner, 1984; Wellde *et al.*, 1989).

According to Wellde *et al.*, (1989), initial tsetse control measures at Lambwe valley consisted of applications of dieldrin to the periphery of the Ruma game reserve. This had a marked effect on the prevalence of the disease. However, concern about the use of dieldrin caused the cessation of this method and justified an aerial application of endosulfan and later pyrethrum both of which were unsuccessful. Since then the method of control has been ground spraying using cypermethrin and limited bush clearing in areas of rugged terrain (Turner and Brightwell, 1986). While temporary control has been achieved through the sequential spraying method, total elimination of the flies has not been achieved due to the constant re-invasion and/or resurgence of the population in the treated area. More recently insecticide-impregnated targets have been installed and preliminary observations indicate that the population in the area is being reduced (Opiyo pers comm).

In an attempt to throw more light on some aspects of the tsetse biology, there is real need to gain a full understanding of the ecology of *G.pallidipes*, its behaviour and the epidemiology of the disease it transmits. It is deemed that a sound knowledge of these aspects is a fundamental pre-requisite for determining

the prospects for successful tsetse and trypanosomiasis control.

It is against this background that the ICIPE Nguruman tsetse and trypanosomiasis research project was initiated in 1983 with the objective of developing new approaches to tsetse and disease control through a greater understanding of tsetse population dynamics and disease epidemiology.

One area which was identifield as requiring a considerable research effort was puparia ecology. This was commenced by Adabie (1987), who succeessfully identified dry season larviposition sites and monitored seasonal changes in puparia numbers. More work however remained to be carried out. Firstly, although loss rates between puparia and tenerals was shown to be density dependent (Adabie, 1987), it was not known whether this was caused by predation, parasitism, flooding or by death due to fungal infections. Secondly, it became necessary to determine weights of puparia as soon as they were found in order to ascertain when they died. Thirdly, it became important to know whether tsetse suppression using trapping resulted in population decline of puparia in larviposition sites within the tsetse suppressed area. All these aspects required incessive studies. It is against this background that the studies reported here

were conducted to investigate the factors affecting the distribution and mortality of *G.pallidipes* puparia at Nguruman.

1.5 Summary of major objectives of the study

(a) To determine the species composition, distribution and abundance of *G.pallidipes* predators at Nguruman

(b) To determine the rates of *G.pallidipes* puparia predation buried at different densities and soil depths

(c) To determine the factors affecting mortality of *G.pallidipes* puparia at Nguruman.

(d) To determine the effects of adult tsetse trapping on the distribution and abundance of *G.pallidipes* puparia within and outside the suppression area.

CHAPTER TWO

LITERATURE REVIEW

2.1 The larviposition sites of *Glossina pallidipes*

Since the studies reported here were carried out in larviposition sites of *G.pallidipes* it became necessary as a prerequisite to examine past work on this aspect to assist in designing appropriate methodologies for various experiments that were carried out.

The larviposition sites of several species of tsetse flies including *G.pallidipes* have been studied in the past (Carpenter, 1920; Zumpt, 1936; Swynnerton, 1936; Harley, 1954; Buxton, 1955; Atkinson, 1971a; Challier, 1982; Laveissierre *et al.*, 1984a, b; Kaminsky 1984; Okoth, 1985; Adabie, 1987). Tsetse flies do not larviposit at random within their geographic ranges, but usually select the most favourable parts of the habitat to deposit their larvae. Forest and riverine tsetse species tend to larviposit in areas with narrowly defined characteristics and puparia are widely scattered. Savannah species which are exposed to great seasonal changes in climate and vegetation use many types of breeding places which tend to be localized (Nash, 1939; Vanderplank, 1948a; Buxton, 1955; Jewel,

1958; Glasgow, 1961; Nash, 1969; Nash and Trewern, 1972).

Early ecologists, notably Nash (1939) and Buxton (1955) described the general characteristics of larviposition sites of *Glossina* species. The sites described were mainly found in shaded protected spots with loose soils. The puparia of *G.pallidipes* have been found in a wide variety of sites within the considerable geographic range of this species. In South Africa, for example, Swynnerton (1936) and Du Toit (1954) found puparia of *G.pallidipes* under leaf litter in thickets. In Mozambique, Swynnerton (1936) also recorded puparia of this species in sites found under fallen logs from which also the puparia of *G.morsitans* were recovered. Swynnerton (1936) also studied the larviposition sites of *G.pallidipes* in Zimbabwe and regarded thickets, game and waterpools as ecological requirements for the satisfactory breeding of this species in that country. Similarly, Phelps *et al.*, (1966) and Phelps and Vale (1978) found that in Zimbabwe *G.pallidipes* larviposited in riverbeds during the cool dry seasons and in animal burrows during the hot dry seasons.

In East Africa, Swynnerton (1936) found the puparia of *G.pallidipes* in evergreen thickets, under fallen logs, rocks and leaning trees. Parsons (1954) recovered puparia of *G.pallidipes* in evergreen shrubs

of *Craibia* sp. on the Kiangini river, Makueni, in Machakos district. Turner (1981) also recovered puparia of *G.pallidipes* in exotic coniferous plantations in Lambwe valley. Adabie (1987), working on the ecology of *G.pallidipes* at Nguruman reported that the larviposition sites of this species were found in riverine thickets and in dense patches of woodland savannah. She (Adabie, 1987) reported that most of the sites had loamy-sandy soils which were mildly acidic or alkaline. She further observed that soil temperatures in the sites always ranged between 1 - 5 ^{0} C lower than the ambient temperature indicating that puparia in the soil never experienced high fatal temperatures. Puparia of this species occurred to a depth of 5 cm but the majority of puparia were found between 1 and 3 cm of soil depth.

A number of ecologists have also reported a shift in the larviposition sites of *Glossina* species according to seasons (Nash, 1937, 1939, 1941; Burtt, 1952; Parker, 1956; Bursell, 1960a; Jackson and Phelps, 1967; Baldry, 1970; Atkinson, 1971; Jordan, 1974; Davies, 1977). The shift in sites may be brought about by changes in climatic factors such as a rise in soil temperature or soil moisture (Buxton, 1955). At Nguruman in Kenya, Adabie (1987) reported that *G.pallidipes* shifted its larviposition sites from the riverine thicket sites to the hilly valley sites during

the wet season. The shift coincided with the onset of the long and heavy rains indicating that the female flies probably changed sites to avoid flooding and consequent mortality of its puparia.

Several researchers have also reported the existence of an association of certain vegetation types with larviposition sites of different tsetse species (Buxton, 1955; Langridge, *et al.*, 1963; Gruvel, 1974b; Turner, 1981). *G.pallidipes* was recorded in forests fringing permanent surface water and lakes as well as in vegetation of banks of seasonal pools, rivers and streams that are dry for most of the year (Nash, 1937). Nash (1937) reported further that the species was also associated with thicket patches, some evergreen trees and secondary shrubs on banks of seasonally dry streams and rivers. The flies were attracted more to woody vegetation than to herbaceous plants (Nash, 1937, Buxton, 1955).

In any habitat the choice of a specific larviposition site in which the larva is extruded is determined by the behaviour of the pregnant females (Nash, 1930; Jordan, 1986). The responses involved in the selection of the exact larviposition sites within a habitat have been investigated by a number of workers (Lamborn, 1915; Swynnerton, 1936; Lewis, 1934; Burtt, 1952; Parker, 1956a; Finlayson, 1967; Phelps and Jackson, 1971; Atkinson, 1971a; Davies, 1977; Rowcliffe

and Finlayson, 1981). In northern Nigeria Parker (1956a) demonstrated that a variety of black objects were strongly attractive to pregnant females of *G.palpalis*. There was also a strong preference for rough surfaced soils, while soils with moisture content of 25% or more were avoided in favour of those whose moisture content was at equilibrium with that of the atmosphere. Since adult flies are negatively phototactic at high temperatures (Davies, 1977), this factor could also be important in the selection of a larviposition site. On the other hand humidity involvement might only be circumstantial as its effects have not been determined. Nash *et al.*, (1976) also suggested that there was a larval pheromone produced at the time of pupariation which acted as an attractant for the pregnant tsetse to larviposit in a particular site resulting in aggregation of puparia. Whilst Rowcliffe and Finlayson (1981) found no evidence for Nash *et al.*,'s (1976) hypothesis, experiments conducted recently (1990) at ICIPE suggested the possible involvement of larval pheromone in attracting gravid females to larviposit in certain sites leading to aggregation of puparia (Lennard, pers comm.).

2.2 Mortality factors affecting puparia in larviposition sites

Mortality factors affecting puparia of *G.pallidipes* and indeed other species of tsetse in larviposition sites could be grouped into two categories: density independent and density dependent factors (Buxton, 1955). Density independent factors killing puparia of the species were identified as being the extremes of temperature (very low and very high) and flooding of pupal sites (Nash, 1938, 1939; Buxton, 1955). Adabie (1987) reported that high rainfall resulting in the flooding of larviposition sites probably caused mortality of *G.pallidipes* puparia at Nguruman. She further reported that climatic factors such as relative humidity and soil temperatures were probably also involved in causing seasonal variations of puparia numbers in larviposition sites.

Density dependent mortality factors of puparia resulted from predation and parasitism (Nash, 1970). Numerous instances of predation and parasitism on tsetse puparia have been reported by several tsetse workers and literature on parasites and predators of *Glossina* was extensively reviewed by Buxton (1955) and Mulligan (1970). Lists and bibliographies of records of natural enemies of puparia are provided by Saunders (1960), Jenkins (1964), Anon., (1974) and Laird (1977). There is therefore substantial evidence in the literature indicating that the tsetse puparia are attacked by numerous natural enemies which presumably

contribute more or less to the natural regulation of the tsetse population. Recently Adabie (1987), showed that puparia loss rate estimated from the relative densities of puparia and teneral flies was significantly density dependent.

2.2.1 The predators of tsetse puparia

Many vertebrate and invertebrate predators have been reported or suspected of preying on tsetse puparia since they have been observed burrowing, scratching or wandering in larviposition sites of tsetse (Swynnerton, 1936; Nash, 1970; Laird, 1977; Challier, 1982). For example, Swynnerton (1936) observed traces of scraping, footprints and excreta of mongoose (*Herpestes* sp.) and the African shrew (*Petrodromys tetradactylus* L.) in larviposition sites of *G.morsitans* and *G.austeni* and concluded that these mammals were involved in the destruction of large numbers of puparia. He also observed several species of birds including fowls (*Numida* spp.) and bush fowls (*Francolinus* spp.) wandering and scratching the soil in larviposition sites and concluded that they were predators of the tsetse puparia.

In Tanzania, Carpenter (1912) reported that the species *Euponera senaarensis* Mayr and *paltothyreus tarsalis* (Gerstaecker) (Hymenoptera: Formicidae),

devoured the puparia of *G.morsitans* and *G.palpalis*. Similarly, Ford (1940) observed ants of the genus *Pheidole* (Hymenoptera: Formicidae) carrying puparia of *G.swynnertoni* into their nests and because of this, he regarded them as being efficient predators of tsetse puparia. Similar observations were reported in Uganda by Rogers (1974) who showed that puparia mortality of *G.f.fuscipes* was mainly caused by *Pheidole* ant species.

In Uganda, Fiske (1920), reported that the adults and larvae of the coleopteran families Carabidae, Elateridae and Cicidelidae preyed upon and destroyed a portion (7 % of 9000) of puparia of *G.palpalis*. Nash (1933a, b; 1939, 1970), observed that the larvae of *Melyris pallidiventris* L. (Coleoptera: Melyridae) devoured tsetse puparia in Tanzania. Challier (1971a, b) found large crickets in larvipositon sites of *G.palpalis gambiensis* in Burkina Faso and considered that they were occasional predators of puparia. Recently Adabie (1987), confirmed through serological analysis of gut smears that crickets belonging to the species *Phaeophillacris* and *Gryllus* were important predators of *G.pallidipes* puparia at Nguruman.

2.2.2 Parasites of tsetse puparia

Twenty three species of Hymenoptera and over ten species of Diptera (family Bombyliidae) were listed by

Jenkins (1964) as important parasites of tsetse puparia, although they rarely parasitize tsetse puparia exclusively. Among the hymenopterans, the genera *Syntomosphyrum* (Eulophidae) and *Mutilla* (Mutillidae) were the most important parasites of tsetse puparia (Nash, 1947). *Syntomosphyrum glossinae* Waterston and *S.albiclavus* Kerrich were first reported in puparia of *G.fuscipes fuscipes* from the shores of Lake Victoria by Waterson (1915a, b) while their distribution was reported by Saunders (1960, 1961); Potts,(1970b) and Baldry (1979).

S.glossinae has been recorded in Kenya, Uganda, Tanzania, Malawi, Senegal, Liberia, Nigeria, and Zimbabwe (Potts, 1970b) while *S.albiclavus* Kerrich was recorded only in East and South Africa (Potts,1970b). Both species were reported as being abundant in nature although *S.glossinae* received more attention since Lamborn (1916) reported favourably on its short life cycle, high fecundity and its easiness of being reared in a variety of dipterous puparia including those of *Glossina*, *Sarcophaga* and *Musca* species. However, attempts to control *G.morsitans centralis* Westwood by releasing this parasite at Kikori in Tanzania were not largely successful in that during the first month of its release 9.9 % parasitization was recorded which declined drastically to 0.8 % some eight months later (Nash, 1933a).

Three species of the hymenopterous family Mutillidae (*M.glossinae* (Turner), *M.auxiliaris* (Turner) and *M.benefactrix* (Austen) have been reared from puparia of *Glossina* (Laird, 1977). Most records of *Mutilla* species parasitic in tsetse are from South Africa, where high parasitism rates have been recorded from tsetse puparia (Potts,1970b). *M.glossinae* (Turner) and *M.auxiliaris* (Turner), have been found to be important parasites of *G. morsitans* and *G.pallidipes* in Zambia, Malawi and Zimbabwe (Heaversedge, 1969a, c; Nash, 1970).

Evidence for seasonal variations in the rate of parasitism has been reported. In Zimbabwe, Chorley (1929) found that the rates of parasitism of *G. m. morsitans* puparia by *M.glossinae* were lowest in the cold dry months, but were highest up to 20 % in the hot dry months. It was also observed that, the rates of parasitism fell as the rains began (Laird, 1977). In his search for parasites for control of tsetse, Markham (1984) carried out an extensive collection of tsetse puparia from Zimbabwe, Zambia, and Malawi. The puparia collected from Zimbabwe failed to yield mutillid parasites although puparia cases from Malawi showed 18 % mutillid parasitism. He (Markhan, 1984) also reported that the *mutillid Chrestomutilla ? glossinae* developed in both *Sarcophaga* argyrostroma L. and *G.morsitans* puparia in 45- 50 days.

Ten species of the genus *Exhyalanthrax* (family Bomyliidae) have been recorded as the only Diptera parasitic on tsetse puparia (Mulligan, 1970). According to this author all the 10 species were reported as being widely distributed in East and Central Africa, but reports on *Exhyalanthrax* from West Africa are scarce. However *E.argentifrons* (Austen) was recorded in puparia of *G.m.submorsitans* Newstead in Nigeria (Lester, 1931; Taylor, 1932), while *E.beckerianus* Bezzi was found in *G.tachinoides* puparia in Chad (Gruvel, 1974a).

Analysis of the incidence of *Exhyalanthrax* species in field collected puparia indicated that there existed important variations between different areas and different seasons of the year (Taylor, 1932; Nash, 1942, 1970; Hursey, 1970; Gruvel, 1970a). For example in Gadau, northern Nigeria, Taylor (1932) recorded a high parasitization rate of *G.tachinoides* and *G.morsitans* puparia by *E.beckerianus* in the warmer drier months than at other times of the year. Similarly, Nash (1942) observed variations in the rates of puparia parasitization by *Exhyalanthrax* species in relation to vegetation types. For example, whereas in *Berlina globifera* L. and *Acacia robusta usambarensis* (Taub.) Brenan woodland the parasitization rate of *G. morsitans* puparia was 15.7 %, the rate fell to 7.8 % in *Brachystegia microphylla* De wild woodland.

Hursey (1970) recorded a high parasitization rate of 46.6 % caused by *E.abruptus* Lw in puparia of *G.pallidipes* near the lesser Kiboko river in Machakos, Kenya. Other species of the same genus which have been reported in puparia of *G.pallidipes* are *E.alliopterus* Hesse and *E.brevifacies* Hesse (Laird,1977). *Exhyalanthrax* species reported in other tsetse puparia species are *E.burtis* Hesse and *E.transciens* Bezzi (Potts, 1955, 1970b; Hursey, 1970; Mulligan, 1970).

The penetration by *Exhyalanthrax* larva into a tsetse puparium is dependent on a number of factors (Laird, 1977). Among them are the coincidence of oviposition sites of host and parasite, the nature of the soil at the sites, the depth at which the puparia occurs, the density of the soil, mobility, longevity of the planidia and some aspects of the climate. The size of the emerging adult parasite has also been reported to be dependent on the size of the host puparium in which it developed (Heaversedge, 1968a). He (Heaversedge, 1968a) observed that puparia of *G.pallidipes* produced larger *E.abruptus* than those of *G. morsitans*. Recently Adabie (1987) reported that *G.pallidipes* puparia were parasitised by *Exhyalanthrax lugens* (Lw) and *E.beckerianus* Bezzi. The author also showed that parasitization rates of the puparia by these parasites was higher during the rainy, rather than during the dry season. Other hymenopteran species

have also been recorded as being parasites of tsetse puparia. In Uganda, Kangwangye (1971) reported that puparia of *G. pallidipes*, *G.f.fuscipes* and *G.fuscipleuris* were parasitized by *Trichopria capensis robustior* Silv (Hymenoptera: Diapriidae). Chalcids have also been recorded as parasites of tsetse puparia but their importance was not well documented (Laird, 1977). The full range of the parasites that attack the puparia of *G.pallidipes* at Nguruman has not been established and part of the studies reported here were aimed at obtaining this information.

2.2.3 Pathogens of tsetse puparia

Although many micro-organisms have been reported to affect adult tsetse flies only a few affect puparia (Laird, 1977). Among the pathogenic micro-organisms only bacteria and fungi have been reported to cause mortality in tsetse puparia. For example, Trelliard and Roubald (1935) isolated a Coccobacillus from the puparia of *G.morsitans* and described it as *Bacterium mathisi* Roubald. This bacteria was shown to be lethal to adult tsetse following ingestion. Vey (1971) isolated two fungi species *Absidia repens* Hagem and *Penicillium lilaceum* Thom. from puparia of *G.fusca congolensis* Newstead and Evans and showed that they were primary pathogens of puparia and not contaminants.

At Nguruman, fungi belonging to *Penicillum* and *Rhizopus* species were isolated from puparia of *G.pallidipes* (Adabie,1987). However, she (Adabie,1987) did not establish as to whether they were pathogenic or not. In these studies further investigations were therefore carried out to identify if any other species of fungi attacked the puparia of *G.pallidipes* and also establish their pathogenicity.

2.3 Mode of puparia mortality from predators

The lack of critical experiments to investigate density dependent mortalities, coupled with the general reluctance to believe in them, led to the view that climate both controlled and regulated population size (Buxton,1955). However, tsetse population resurgence to changes brought about by insecticides (Tarimo *et al.*, 1970; Rogers and Randolph, 1984; Turner, 1984; Turner and Brightwell, 1986) suggested some kind of interference with some density related processes. The few experiments previously carried out to assess the level and mode of puparia mortality from predators will now be described.

In Kakoma, Tanzania, Jackson (1937) buried puparia in larviposition sites and left them in the field over a period of time. Out of the 188 puparia buried, 54 (29%) were lost or taken by predators, but all the

others emerged which indicated that no mortality was due to disease or failure to develop or emerge. He, however, dissected another group of puparia as they were found and recorded high natural mortality.

In other studies, Ford (1940) attempted a quantitative estimation of predation of ants upon *Glossina swynnertoni* puparia in Tanzania by comparing the decrease in numbers of puparia that were artificially distributed according to a definite plan in a natural forest and savannah habitat. He recorded estimated predation rates of 11-18 % in the forest habitat and 25-44 % in the savannah habitat. He further observed members of the genus *Pheidole* carrying puparia into their nests and concluded from these observations that the ants were very efficient puparia predators. Ford's (1940) experiments were later repeated by Kemp (1951) who buried some tsetse puparia individually at marked spots and dug for them 14 days later. The results showed that the intensity of predation by *Pheidole* ants was higher in the absence of other food sources.

Using similar field experimentation, Rogers (1974) quantified puparial losses from predation on puparia of *Glossina f. fuscipes* Newstead in Busoga forest in Uganda. He showed that most puparia mortality was caused by *Pheidole* ants, which took a constant number of puparia at lower densities but an increasing

percentage at higher densities indicating a density-dependent process.

Little is known about the density relationships between predator density and functional responses of the predators to changes in puparia numbers due to the difficulties in observing actual contact between puparia and their natural enemies (Rogers, 1974). Futhermore, there is no evidence to show any specific action of any of the predators upon tsetse puparia. There is therefore a great need for quantitative evaluation of puparia predation in order to first, understand natural regulatory proccesses, and secondly, to identify those predators that could possibly be incorporated or manipulated into an ecologically safe and rationale tsetse control strategy. Some of the studies reported here were aimed at obtaining this information.

Adabie (1987) initiated similar studies through experimental burying of puparia at different densities. Using a latin square design, she (Adabie, 1987) buried six densities of puparia (i.e 1, 4, 9, 16, 25, and 36 per m^2) and left them in the field for two weeks. In these investigations similar studies using a different experimental design were carried out to elucidate further or not predation was a density dependent factor.

CHAPTER THREE

DESCRIPTION OF THE STUDY AREA

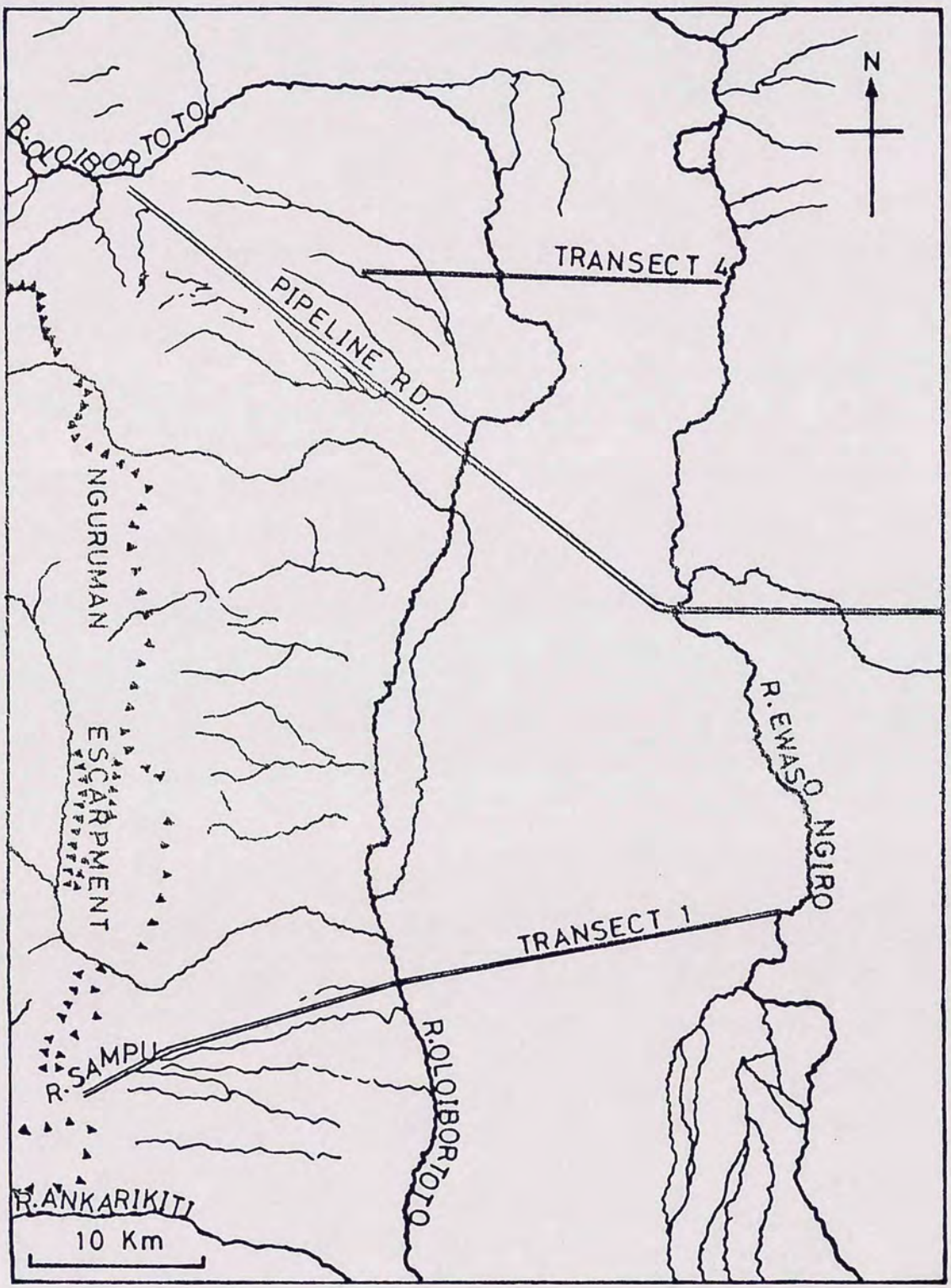
3.1 INTRODUCTION

All the field studies reported here were carried out at Nguruman in the Kajiado district of the Rift valley province in South western Kenya. The area is situated in a generally semi-arid zone and was formerly classified as a Maasai reserve area. A wide variety of game animals are abundant in the area. Today the area is inhabited mainly by the indigenous Maasai pastoralists who keep cattle, sheep, goats and donkeys. These animals are moved East of the Ewaso Ngiro river during the rains to conserve pasture and minimize tsetse challenge. During the dry season however, they are moved progressively closer to the woodland at the base of the escarpment resulting in higher tsetse challenge. Chemotherapy is widely used at this time of the year but losses can be high especially if the cattle are malnourished. In addition to the Maasai population, other ethnic groups have settled in the area and practice irrigation farming, producing maize and a variety of fruits and vegetables.

The area has considerable potential for development. Firstly, the rangeland could be used for

Figure 1: A sketch map of the study area showing Transect 1, pipeline road and Transect IV.

(From: Dransfield et al., (1990).



improved animal production for the benefit of the local Maasai community and for the nation as a whole. Secondly, the irrigation scheme could possibly be extended to increase crop and vegetable production. Finally, the abundant game could be a tourist attraction if tourist facilities were to be developed in the area. One of the reasons why the area has not been fully exploited for these resources has been the presence of large numbers of tsetse flies, specifically *Glossina pallidipes* and *G.longipennis*. Apart from the fact that tsetse flies can be a nuisance to man when numbers are very high, the species found in the area are vectors of nagana. Considerable numbers of Maasai cattle have been reported to regularly die from the disease especially during drought periods when the resistance level of the animals is very low and animals expose themselves to higher tsetse challenge by moving into the denser woodland in search of food. Therefore the control of tsetse flies and nagana would contribute enormously to the development of the area. It was because of this existing need that studies reported here on the puparia ecology of *G.pallidipes* were conducted at Nguruman.

3.2 Location of the study area

Figure 1 shows the location of the study area. The area, which forms part of the alluvial plains of the Rift valley, is located between latitude 1^{o} 50 'N and longitude 35^{o} 56 'E and lies at about 700 m above sea level.

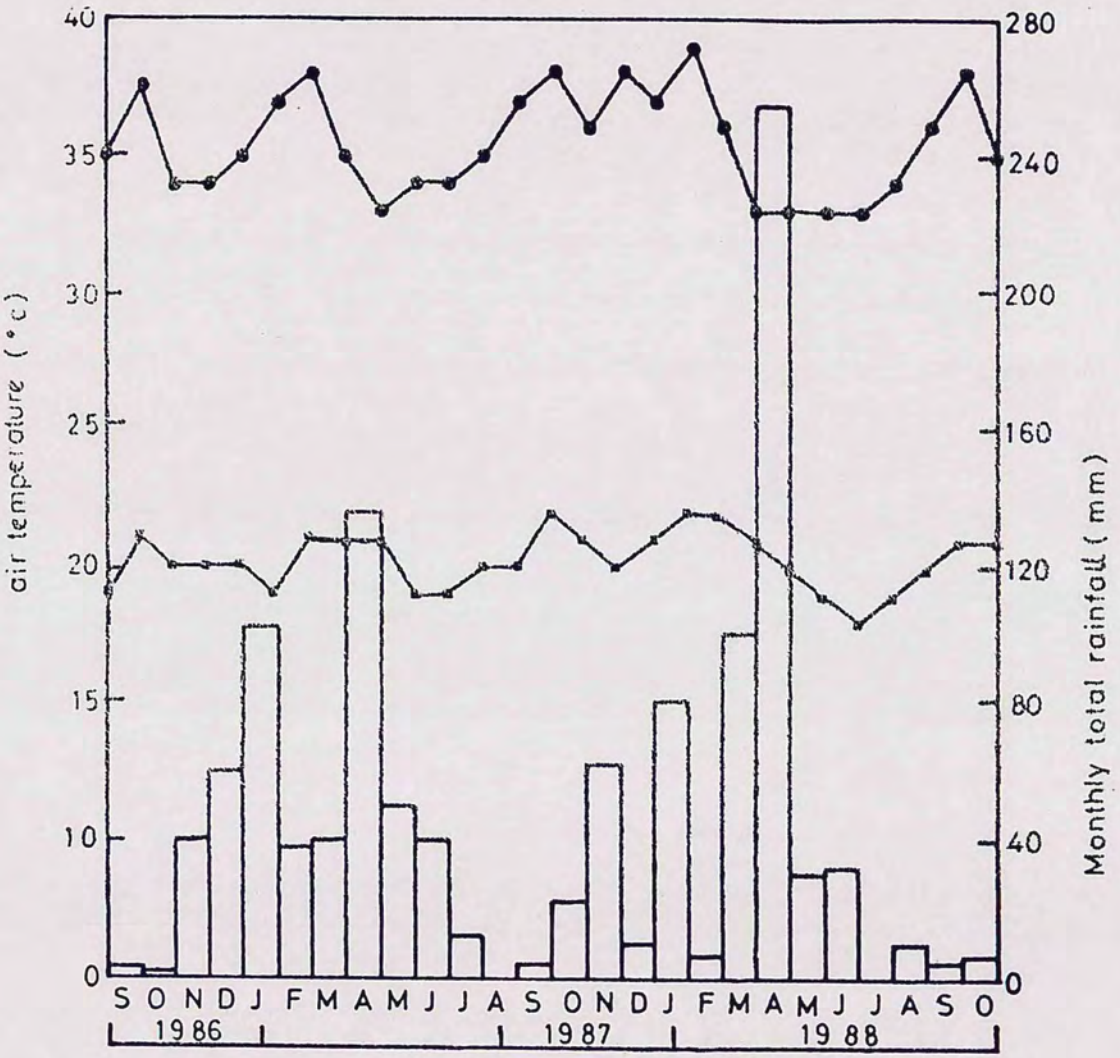
The major landmarks are the Ewaso Ngiro river to the East, the Nguruman Escarpment rising to about 3000 m in the West and Mount Shompole to the South. The area is drained by the permanent Ewaso Ngiro river and the semi-permanent Lenkutoto, Entosopia, Oloibortoto, Enkarikiti, Sampu and the Lentore river systems which drain Eastwards eventually joining the Ewaso Ngiro swamp to lake Natron. During the main rainy season these rivers are swollen with water from the escarpment which results in extensive flooding of the area.

3.3 Climate of the study area

The climatic data for the duration of this study are shown in Figure 2. The Nguruman area generally experiences a mean annual rainfall of about 500-700 mm. Generally the annual mean monthly maximum temperatures ranges from 31-39 ^{o} C while the mean monthly minimum temperature ranges from 18-22 ^{o} C.

The rainfall generally occurs in two seasons in a year; short rains occur more or less in November and December and the long rains from March till May. These

Figure 2: Mean Monthly maximum and minimum temperatures (upper and lower lines) and total rainfall histogram at Nguruman over the study period.



are separated by hot dry months from January to early March and cooler dry months from June to September. During the rains, grasses and shrubs grow rapidly and food is generally abundant for both domestic and wild animals. In the dry seasons, most grasses die leaving the ground bare, deciduous trees and bushes loose their leaves and there is a general scarcity of vegetation for animals. During this period cattle are moved into thicker woodland at the base of the escarpment in search of good pasture.

3.4 Vegetation of the study area

The vegetation in the Nguruman area has been described by Lewis (1934), Sayad and Sayad (1980), Van Etten (1981) and Dransfield *et al.*, (1986). In moving from the Ewaso Ngiro river to the base of the escarpment one traverses five main vegetation types as follows:

(a) The Open Plains

These plains start immediately after the wooded fringes of the Ewaso Ngiro river. The vegetation in this area consists mainly of grassland with isolated acacia trees and various shrubs. During the dry season the ground is patched and bare except for some isolated

tussocks of grass. In the rainy season however, the area is covered with tall grasses and herbs and wild game graze in this area.

(b) The Acacia Woodland

This type of vegetation is dominated by thorny acacia species including mainly *A. tortilis radiana* (Savi) Brenan., *Acacia albida* Del., and *A.seyel fistula* (Schweinf.) Oliv. There are more shrubs than are found in the plains, with a good grass cover.

(c) The Riverine Thicket

This type of vegetation is found along the courses of rivers and streams. The thickets have a mosaic of vegetation types dominated by tall trees such as *Ficus* sp., *Euphorbia candelabrum* Kotschy, *Acacia pennata* L. and *Mystroxylon aethiopicum* (Thub.) Loes with an undergrowth of shrubs and herbs comprising of *Scutia myrtina* (Burm. f.), *Scolopia* spp, *Aloe* spp, lianes and thorny bushes. This vegetation type is characterised by densely shaded thickets which form the main larviposition sites for *G.pallidipes* during the dry season. During the long rainy season some of this area is flooded and tsetse shift their larviposition sites

to the hilly valley sites at the base of the escarpment.

(d) The Lower and Upper woodland

This vegetation type consists of close stands of thick shrubs and scattered acacia and other trees. The dominant shrubs include *Boscia coriacea* Pax and *Aloe* spp. This area provides animals with bushes for grazing during the dry season. The vegetation becomes thinner as the area rises to the rocky ground towards the base of the escarpment. The tall trees give way to shorter scattered xerophilic *Acacia mellifera* (Vahl) Benth and thorny bushes. The grasses here are shorter and the ground is generally more open than the previous type.

(e) The valley Woodland

This consists of a mosaic of vegetation types dominated by tall trees. The dominant trees in this area include fig trees of *Ficus* and *Albizia* spp. The dominant shrubs include *Salvadora persica* L., *Cordia sinensis* Lam.and *Vanqueria rotundata* Robyns, while the dominant grasses include *Sporobolis consimilis* Fresen, *Setaria sphacelata* Stapf and C.E. Hubbs, *Thermeda triandra* Forsk. and *Cynodon dactylon* (L.) Pers.

3.5 Macrovertebrates of the study area

A wide range of both domestic and wild animals form the macropopulation of Nguruman. The domestic animals kept by the Maasai include cattle, goats, sheep and donkeys.

A variety of wild animals are encounterd as one moves from the plains into the woodland. Generally, the open plain game include the Grants Gazelle (*Gazelle granti* Brooke), Zebra (*Equus zebra* L.), Wildebeest (*Connochaetes taurinus*) and, less commonly, Hartebeest (*Alcelaphus buselaphus* Thomas). The Zebra and Wildebeest migrate into the area during the long rains. Also found in the open plains are the Eland (*Taurotragus oryx* (Pallas)), Waterbuck (*Kobus ellipsiprymmus* (Ogilby) and Warthog (*Phacochoerus aethiopicus* L.). Lion (*Panthera Leo* (L)), and Cheetah (*Acinonyx jubatus* (Schreber)) may also be encountered in the open plains as well as scavengers such as the black backed jackal (*Canis adustus* Sundevall) and the spotted hyena (*Crocuta crocuta* Erxleben). Packs of African hunting dogs (*Lycaon pictus* (Temmick)) are also found rarely. The Cape Hare (*Lepus capensis*) is a very common sight on the plains and Aadvaark (Orycteronyx afer (Pallas)) are also found there. Impala (*Aepyceros melampus* Lichtenstein) and Giraffe (*Giraffa camelopardalis* (L.) are encountered more frequently at

the edge of the woodland with giraffe being more frequent in the acacia woodland.

Large numbers of Baboon (*Papio anubis* (Fischer)) are a common sight between the plains and the edge of the woodland whilst the Vervet (*Cercopithecus* sp.) and Colobus (*Colobus* spp.) monkeys are found in the thick woodland. In the thickest parts of the woodland are found Buffalo (*Sycerus caffer* Sparrman) which tend to graze near river beds in the morning and late evening and hide in the heavy shade of bushes in the hot afternoon. Bushbucks (*Trangelaphus scriptus* Pallas) and Dik dik (*Madoqua guentheri* Thomas) are also found in the area. Occasional sightings of Leopard (*Panthera pardus* (L)) and Porcupine (*Hysterix* sp.) have been reported.

A large number of bird species are found in this area including Ostrich (*Struthio camelus* L.), Kori bastard (*Ardeotis kori* L.), and the secretary bird (*Sagittarius serpentarius* L.). Francolins (*Francolinus* spp.) and Guinea fowls (*Numida* spp.) are abundant in the acacia and open woodland. Most of these macrovertebrates are hosts of the tsetse *G.pallidipes*.

3.6 Tsetse control in the area

The Nguruman tsetse control project was initiated by ICIPE in 1983. Its main objective was to develop

improved methods of trypanosomiasis control through a better understanding of tsetse population dynamics and development of more appropriate control technologies. The use of traps without insecticides was considered the most feasible option taking into consideration that insecticides have the problems of polluting the environment, the possibility of resistance developing in tsetse and the dangers of contamination of inexperienced staff. Brightwell *et al.*, (1987) described the initial experiments conducted leading to the development of the NG2B prototype trap and its transformation into an operational control version including the identification of the optimum dose rates of the odour baits. The optimum dose rate of acetone was found to be 150 mg/hr dispensed from a medicine bottle with a 0.2 cm diameter hole punched in the cap. Cow urine was dispensed from a 1 Kg used cooking fat tin with the open end covered with polythene and a 2 x 4 cm slot cut below the rim; this was found to give a dose rate of about 1000 mg/hr. Trial deployment of odour baited NG2B traps to suppress the local population of *G.pallidipes* commenced in February 1987 (Brightwell *et al.*, 1987).

The operation was limited to the main study area which included transect I and covered about 100 km^2 and hence termed the" suppression area" (see Fig.1). Although not completely isolated from other tsetse

infested areas, it was hoped that immigration could be reduced by setting up barrier traps along the invasion routes. Based on information on the natural mortality rates and estimates of the population size of *G.pallidipes* it was estimated that an average trap density of one trap/ km^2 plus additional barrier traps would reduce the population to very low levels.

The traps were deployed over several days in the first week of February 1987. Each trap was placed in a moderately shaded area to avoid direct exposure to sunlight which can cause rapid destruction of the nylon netting of the trap. The sites were cleared of surrounding vegetation to improve visibility of the trap to flies. To the North of the suppression area, a higher density of traps was placed along the narrow strip of vegetation leading to transect 4 (an area hence called" non-suppression area"), which was considered to be one of the main sources of immigration. To the South of the area lay more open woodland except for a narrow neck of thicker vegetation along the banks of the Oloirbortoto river within which several traps were set at 1 km intervals.

Early in 1988 the area was extended slightly upto the pipe-line road and additional traps were put in along the pipe-line road to strengthen the barriers.

CHAPTER FOUR

EFFICIENCY OF ADULT TSETSE SUPPRESSION CONTROL TECHNIQUE AS DETERMINED BY DISTRIBUTION AND ABUNDANCE OF PUPARIA OF *GLOSSINA PALLIDIPES* AT NGURUMAN

4.1 INTRODUCTION

It has been established that the overall distribution and abundance of tsetse puparia in a habitat is directly related to the distribution and abundance of the adult tsetse population (Laveissiere, 1979). Thus it is probable that changes in fly population could lead to corresponding changes in distribution and abundance of its puparia. The broad objective of the studies reported here was to gather information on whether there are any changes in distribution and abundance in tsetse puparial population when the adult fly is subjected to trapping or suppression by the use of traps.

Traps have been used in many parts of Africa to suppress tsetse population (Vale, 1980, 1982; Vale and Hall, 1985; Dransfield et al., 1986; Vale et al., 1988). As a control method for tsetse, traps have many advantages over other methods (Dransfield et al., 1986). The quantity of "trappable" flies has been used as a basis for judging the efficiency or effectiveness

of the type of trap used (Dransfield et al., 1986, 1990). It then follows that if changes occur in puparial population as a result of intensive fly suppression, then a second method to ascertain the effectiveness of the traps would be based on the assessment of puparial density in the habitat. Reported here are studies designed to obtain this information. Other factors studied were effects of meteorological elements on puparia changes.

4.2 MATERIALS AND METHODS

4.2.1 Determination of adult fly and puparial changes in abundance and distribution in an area under fly suppression and in an area without suppression

The effects of suppression through trapping were monitored in two ways. Sampling for one week each month with baited biconical traps was continued both inside and outside the suppression area (see Figure 1). In addition, 20 representative NGU traps were designated monitoring traps, greased to keep ants out and emptied 2-3 times each week and the flies counted. The flies were then sexed to determine the numbers of female and male flies caught on each sampling ocassion.

There was one main puparia sampling area in the study area in which the fly was subjected to

suppression. This area was designated as Transect 1 (Figure 1). Samples were also recovered from an area in which the fly was not under any suppression identified as Transect IV (Figure 1).

Within the suppression area two vegetation types, namely, riverine thickets and valley woodland were sampled for tsetse puparia. Eleven sites were sampled for puparia in the riverine thickets which were liable to flooding while only two sites were sampled for puparia in the valley woodland which were not liable to flooding. In Transect 1V sampling took place in 9 sites in open woodland and near seasonal streams. The time constant hand searching method was used to sample puparia in the foregoing defined sites between 09.00 hr-18.00 hr. on each sampling occasion. The number of puparia and empty puparia cases found from each site and vegetation type were counted and recorded. During the rainy season puparia numbers were corrected by a factor of two since it has been demonstrated that searching efficiency was reduced due to wet soil conditions.

Climatic conditions were monitored in two sites for five days each month for two years. Ambient temperatures and relative humidities at the sites and in the general breeding area were recorded by means of minimum-maximum thermometers and thermohygrographs, respectively. Soil temperatures at the sites were

recorded using minimum-maximum thermometers buried horizontally in soil at 2 and 4 cms. The amount of rainfall was recorded using a rain guage. The correlation between log puparia density and the corresponding climatic indices of the same month or the previous month was investigated by regression analysis.

4.3 RESULTS

4.3.1 Determination of fly and puparial changes in abundance and distribution in an area under fly suppression and in an area without suppression

The mean catches per day of adult *G.pallidipes* in suppression and in non-suppression areas is shown in Table 1. The data shows that mean adult catches in suppression area were lower (132.4) than catches in the non-suppression area (332.3). When t-test was applied to the two sets of data, it was revealed that there was a significant difference between mean adult catches in suppression and in non-suppression areas ($t = 2.42$, $P < 0.05$). The coresponding number of puparia per site recovered from the suppression and non-suppression areas is presented in Table 2. Table 2 shows that the mean number of puparia per site recovered in the suppression area was lower (0.4) than that recovered in the non-suppression area (1.2). When t-test was applied

Table 1: Monthly apparent catches of adult female *G.pallidipes* in suppression and non-suppression areas at Nguruman over a 2 year period, 1986-88

		Mean female catches per day	
Year	Month	Suppression area	non-suppression area
1986	Sept.	456.2	355.3
	Oct.	348.3	269.7
	Nov.	419.5	253.7
	Dec.	546.5	343.0
1987	Jan.	470.4	374.5
	Feb.	81.3	431.7
	Mar.	88.0	630.8
	Apr.	262.0	302.7
	May	87.4	804.0
	June	117.6	989.0
	July	139.3	201.3
	Aug.	12.0	80.2
	Sept.	8.2	249.7
	Oct.	1.7	173.3
	Nov.	13.3	391.3
	Dec.	7.3	255.5
1988	Jan.	16.7	246.7
	Feb.	4.7	233.5
	Mar.	0.7	174.8
	Apr.	21.0	529.0
	May	92.8	569.2
	June	62.8	187.0
	July	36.3	129.0
	Aug.	10.7	59.2
	Sept.	6.0	74.3
	Average	132.4	332.3
	t-test = 2.42, P< 0.05		

Table 2: Monthly abundance of puparia of *G.pallidipes* per larviposition site within the suppression and non-suppression areas over a 2 year period, 1986-88 at Nguruman.

Year	Month	Mean number of puparia per site	
		Suppression area	non-suppression area
1986	Sept.	3.2	-
	Oct.	3.0	-
	Nov.	1.5	-
	Dec.	0.5	-
1987	Jan.	0.2	1.7
	Feb.	0.2	2.0
	Mar.	0.5	2.7
	Apr.	0.4	1.3
	May	0.1	0.9
	June	0.2	0.8
	July	0.2	0.9
	Aug.	0.8	1.3
	Sept.	0.0	0.3
	Oct.	0.1	3.1
	Nov.	0.1	3.2
	Dec.	0.1	2.3
1988	Jan.	0.0	0.9
	Feb.	0.0	0.6
	Mar.	0.0	1.6
	Apr.	0.0	0.0
	May	0.0	0.0
	June	0.0	0.0
	July	0.2	0.7
	Aug.	0.1	1.2
	Sept.	0.1	0.6
	Oct.	0.1	0.6
	Average	0.4	1.2
	t-test = 4.30, P< 0.01		

to data collected significant difference ($t=4.30$, $P< 0.001$) existed between the mean number of puparia per site in suppression and non-suppression areas.

The changes in the adult population levels of *G.pallidipes* over a 2-year study period in the suppression and non-suppression areas are depicted in Figure 3. It is evident (Figure 3) that while the mean catches of adult flies were quite high in the non-suppression area corresponding catches declined rapidly within the suppression area. Shown in Figure 4 are corresponding changes in the number of puparia per site within the suppression and non-suppression areas. Figure 4 shows that while the puparia number in the non-suppression area were high (especially in April and December 1987) corresponding puparia number in the suppression area declined rapidly and after October 1987 very few puparia were found in the area.

The relationship between puparia density and apparent density of adult flies in the suppression area is given in Figure 5. Figure 5 shows that there was a significant positive correlation ($r= 0.7$, $P< 0.01$) between puparia density and adult flies within the suppression area indicating that the distribution and abundance of puparia was to a large extent dependent on the density of the adult flies in the suppression area.

The seasonal fluctuations in the number of puparia in the two vegetation types studied within the

Figure 3 : Changes in apparent densities of female G. pallidipes in the suppression and in non-suppression area.

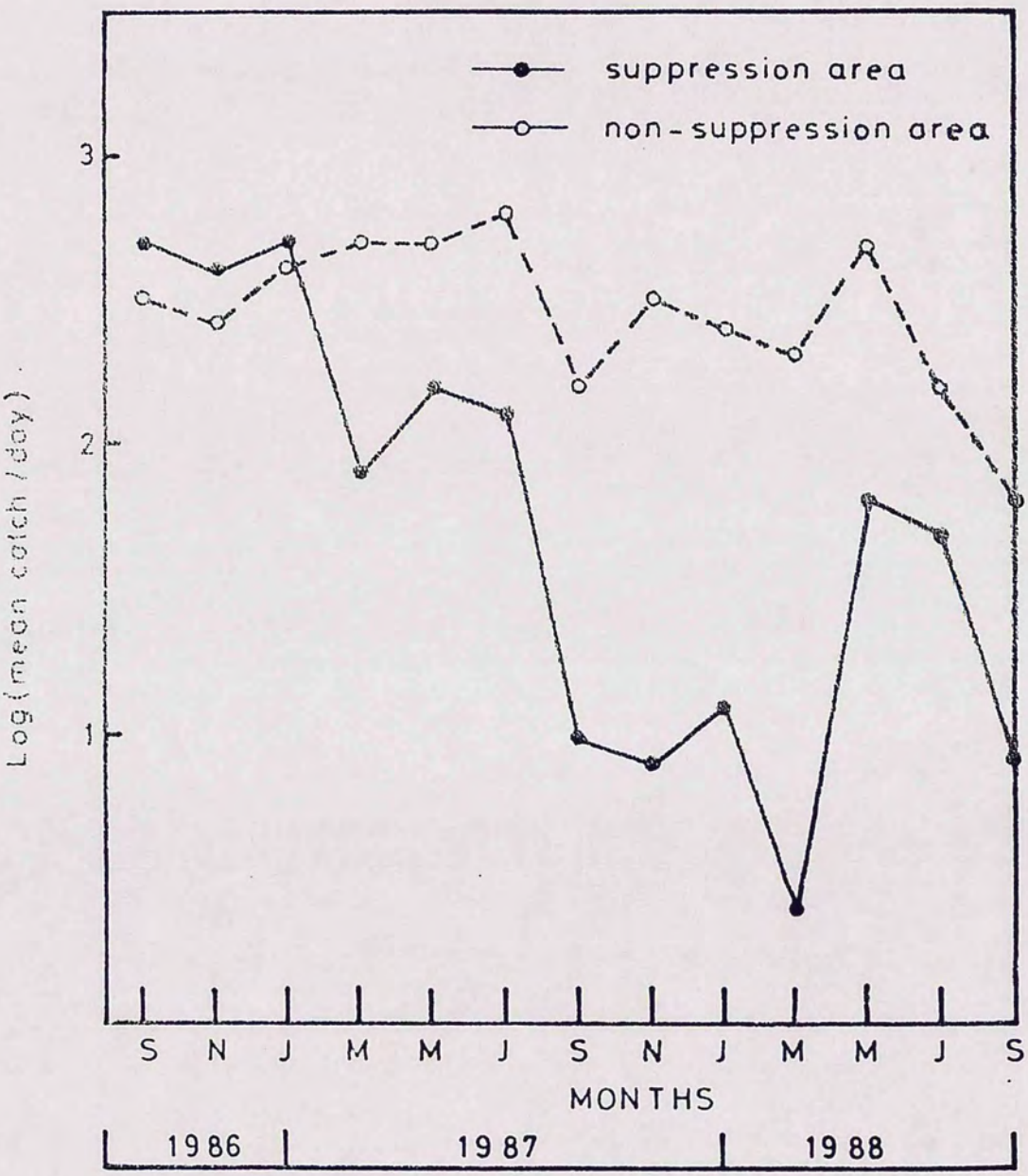
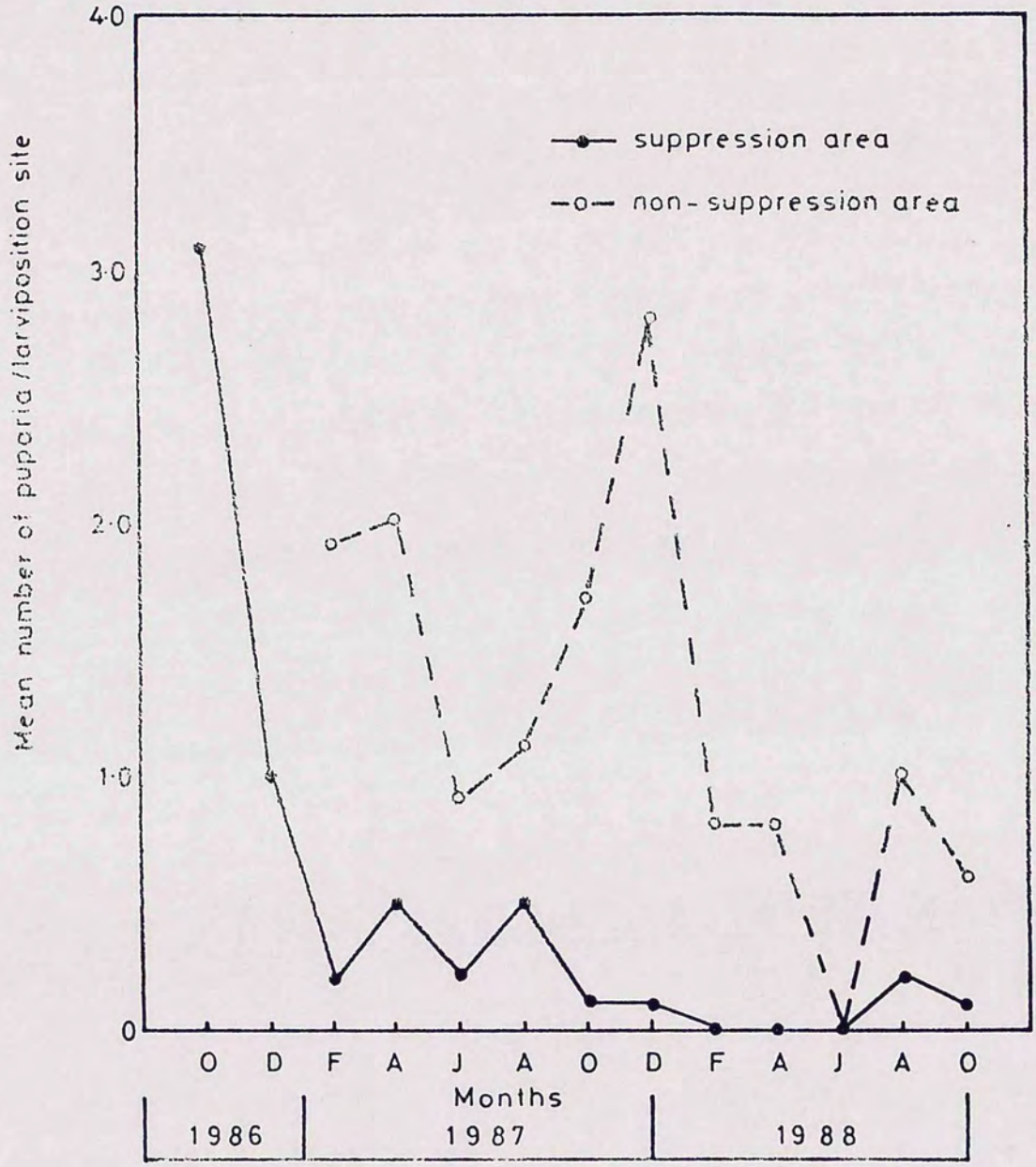


Figure 4: Mean puparia number per site in suppression and non-suppression area



suppression area is shown in Figure 6. Figure 6 shows that the rate of larviposition varied widely in the two vegetation types with more puparia being recovered in the riverine thickets than in the valley woodland. For example, from 1986-87 puparia were found each month in the riverine thickets but with low occurance in the short (December-January) and long (April-May) rains. In the valley woodland however, puparia were only found during or shortly after the rainy seasons (April in 1987 and July in 1988). This suggested a possibility of there having been a seasonal shift in larviposition sites of *G.pallidipes* from riverine thickets which were liable to flooding to the valley woodland sites which were less likely to be flooded.

The mean number of puparia per site collected from 13 different sites within the suppression area is shown in Figure 7. Figure 7 shows that the number of puparia per site within the suppression area varied between 0.1-2.4 among different sites, a 24-fold difference in puparia number between sites. This observation suggested that the female tsetse flies perhaps had a large range of sites in which to larviposit within the suppression area.

The climatic data collected during these studies are presented in Figure 8 and Appendices 1, 2i and 2ii. The highest monthly temperature recorded in the general tsetse area was 38.5^{o}C (February, 1988) and the

Figure 5 : Relationship between puparia density and apparent density of reproductive females in the suppression area

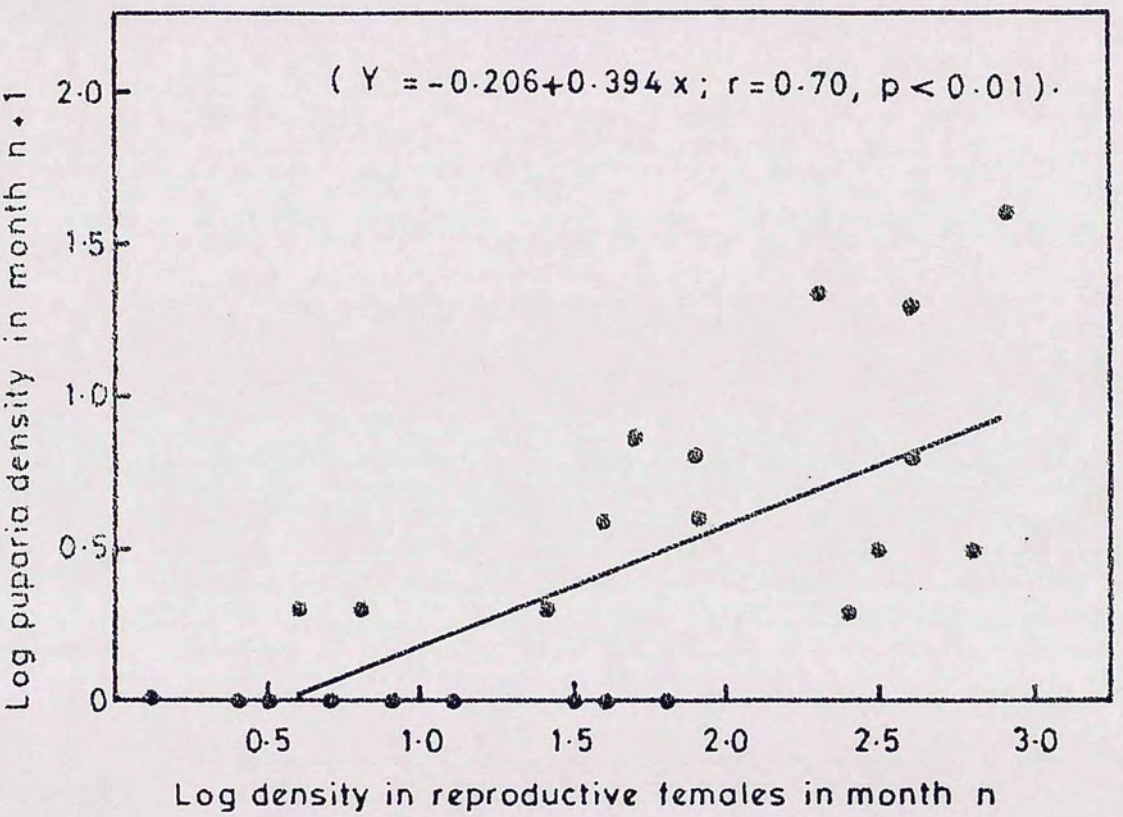


Figure 6 : Seasonal fluctuation in puparia abundance and distribution in the suppression area. (transect 1) Showing shift in larviposition sites of G. pallidipes.

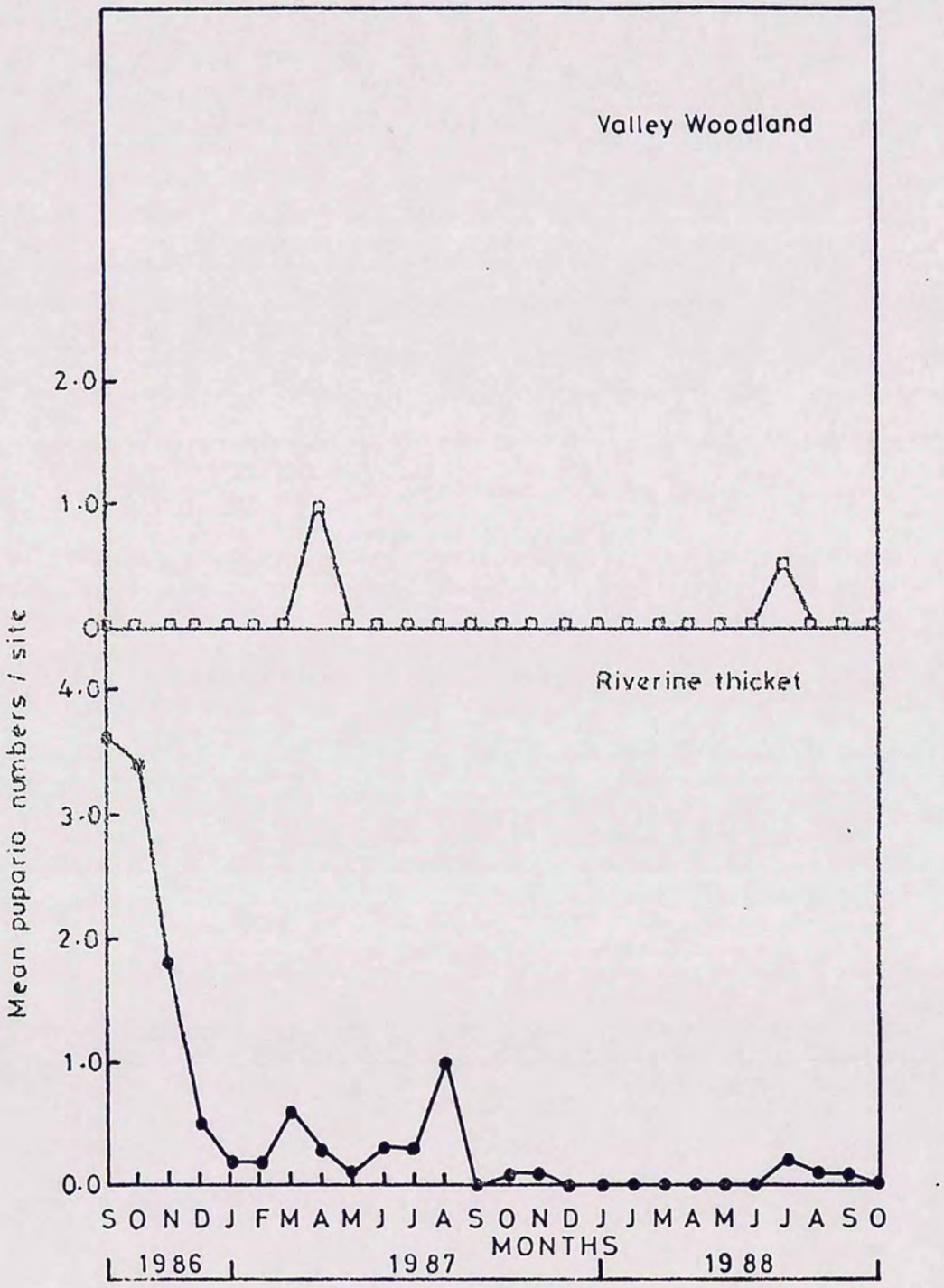


Figure 7 : Overall site utilization by G. pallidipes in suppression area during the study period between October, 1986–October, 1988.

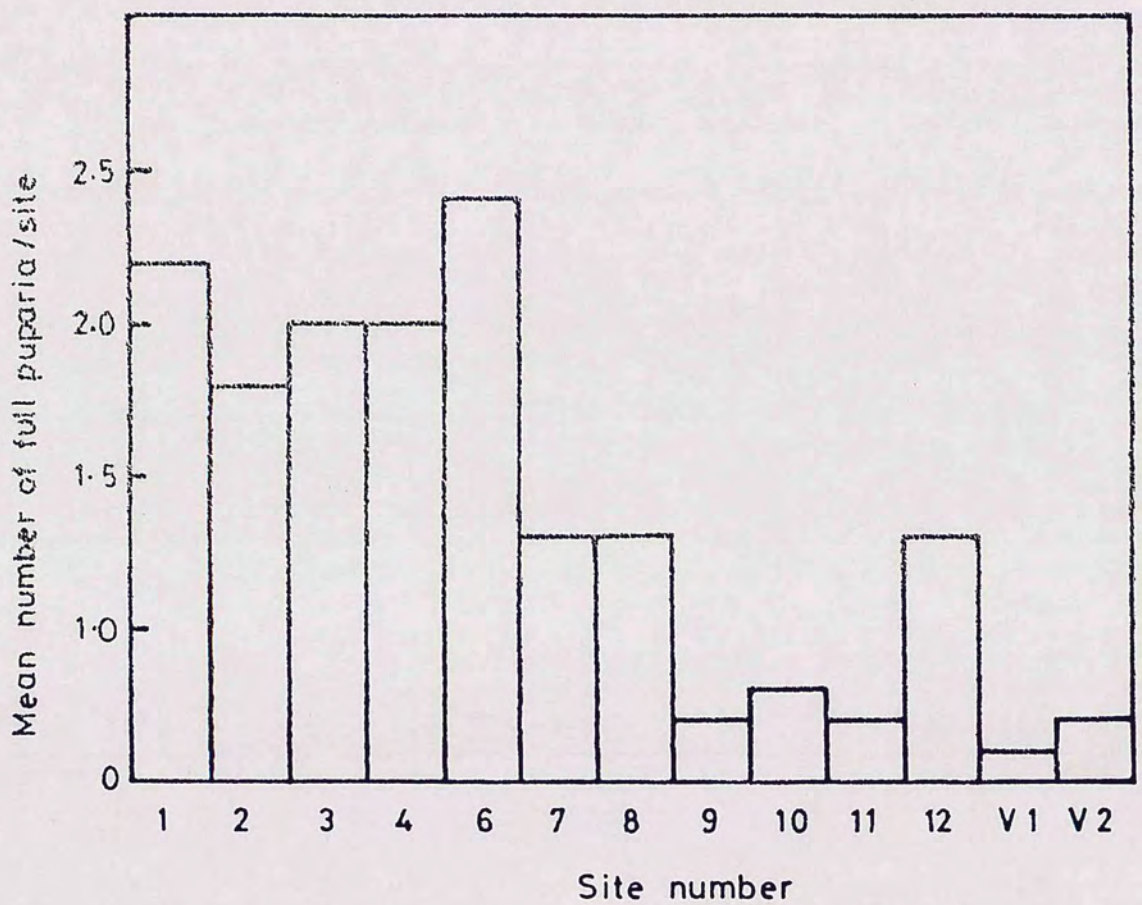
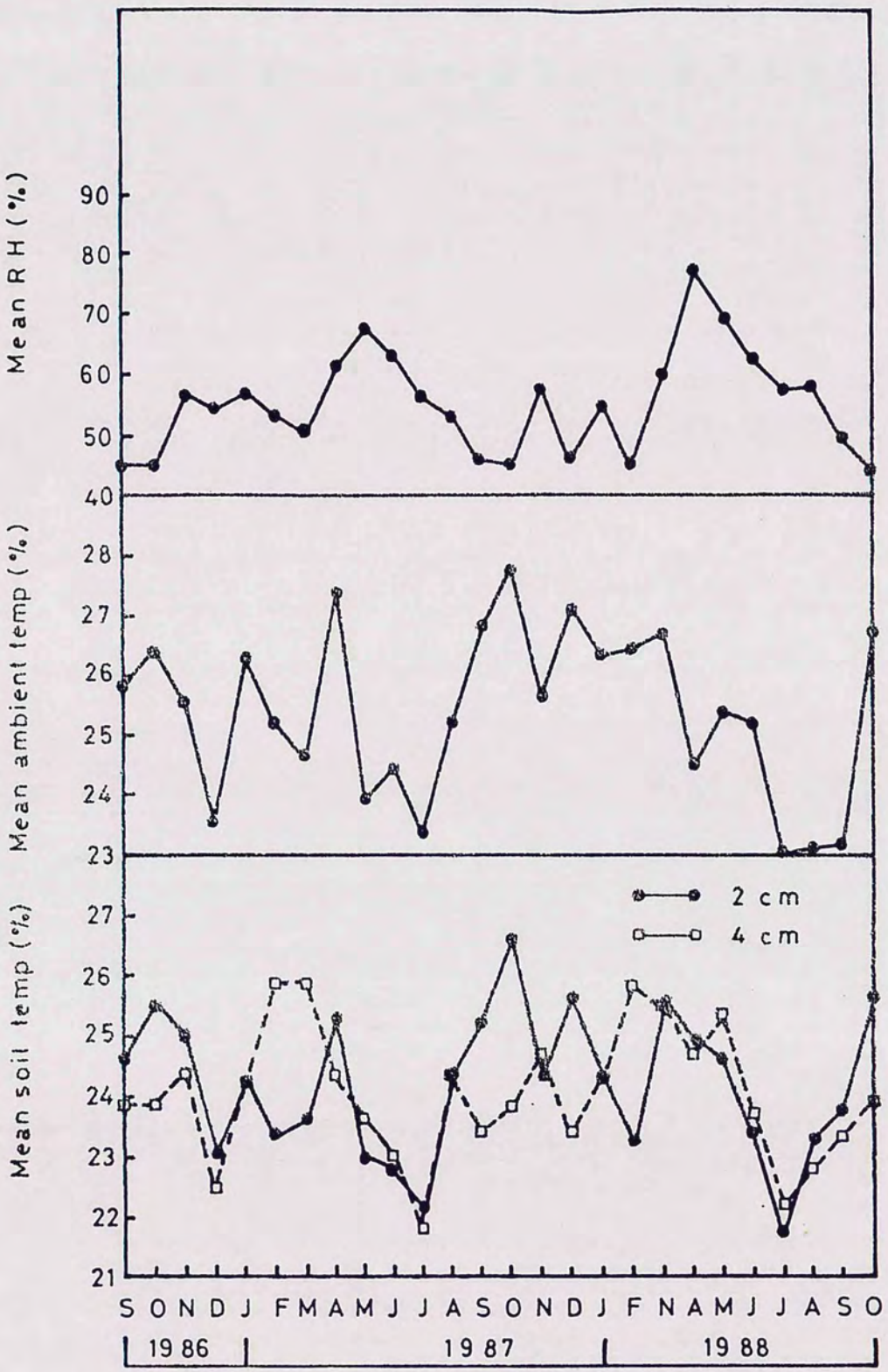


Figure 8 : Variations in climatic factors in larviposition sites of G.pallidipes at Nguruman.



corresponding temperature for the larviposition site was 36.4°C (December,1987). The lowest monthly minimum temperature for the tsetse general area and larviposition sites was 18.2°C and 15.2°C, respectively (both recorded in July, 1988). The monthly mean air relative humidity in the sites was always higher than 40 % as compared to 25 % recorded in the general open area. The highest rainfall (average 194.7 mm) was recorded in April of both 1987 and 1988.
The maximum mean soil temperatures in larviposition sites at 2cm and 4cm were 29.2°C and 27.4°C respectively (recorded in October 1986 and 1987.), while the minimum mean soil temperatures at 2cm and 4cm were 19.1°C and 20.3°C respectively (recorded in July, 1987 and 1988) (Appendix 2ii).

When regression analysis of the log puparia number against meteorological conditions in the general tsetse area and in larviposition sites were performed, none of the climatic factors investigated (i.e maximum temperature, mean temperature, relative humidity and rainfall), in either the same or previuos month, was correlated to puparia numbers. This was probably due to the trapping exercise which was operational in course of these studies.

4.4 DISCUSSION

The distribution and abundance of *G.pallidipes* adults and puparia was monitored in both the suppression and in non-suppression areas. There was a rapid decline in puparia number in the suppression area which coincided with a decline in the number of adult flies within the area. This indicated that trapping reduced the population of the reproductive female flies leading to a decline in puparia numbers within the suppression area.

There were seasonal fluctuations in the number of puparia found in different vegetation types within the suppression area. During the rainy season the flies appeared to shift their larviposition sites from the riverine thickets to the valley woodland sites. This corresponded with a drop in puparia number in the thickets and an increase in number in the valley woodland sites. Seasonal shift in larviposition sites by *Glossina* has been reported by other tsetse workers (Nash, 1937; Parsons, 1954). At Nguruman, the shift in site usage by *G.pallidipes* from the lowland riverine thickets to the sloping sites with good drainage in the valley woodland coincided with the onset of the long and heavy rains. This indicated that the reproductive female flies probably changed their larviposition sites to avoid flooding and consequent puparial mortality.

There was evidence that within the same vegetation type larviposition sites showed considerable variations as indicated by the number of puparia found in them. This suggested that some sites were perhaps more favoured than others within the same habitat.

There was no relationship between puparia density and the climatic factors investigated within the suppression area. This observation was perhaps brought about by the trapping exercise which was operational in course of these studies.

CHAPTER FIVE

FACTORS AFFECTING THE WEIGHT OF *GLOSSINA PALLIDIPES* PUPARIA AT NGURUMAN

5.1 INTRODUCTION

Variation in size of the larval stage of an insect usually reflects the favourability of the environment experienced by its parent (Norris, 1933). In the case of tsetse where the larval stage develops in utero, such variations also reflects the favourability of the environment for the parent female. Puparia weight depends on the nutritional state of the parent female fly (Bursell, 1958). For instance, during the dry season, the female flies are under high level of nutritional stress and consequently produce low weight puparia from which emerge small teneral flies (Bursell, 1960). Studies on the weight of tsetse puparia are therefore of great biological significance as this could be correlated with the size of the newly emerged teneral, its physiology and longevity (Van der Vloedt, 1970). Such studies are also important because one may be able to predict whether puparia collected from the field were alive, dead or parasitised at the time of collection (Van der Vloedt, 1970).

Puparia weight is affected by certain climatic factors. Included among these are temperature and relative humidity (Bursell, 1958). For instance, Bursell (1958) found that puparia maintained at high temperatures and low humidity lost their weight more rapidly than those maintained in moist conditions.

There are also seasonal variations in puparia weight of *Glossina* species. For instance, Van der Vloedt (1970) found that seasonal variability in mean weight of puparia of *G.m.morsitans* existed among the different seasons in Zimbabwe. He (Van der Vloedt, 1970) found that puparia collected in the rainy season had higher weight than those collected in the dry period of the year.

Death of puparia from parasites or pathogens results in a sharp decline in puparia weight. Hursey (1970) recorded puparia weight loss in *G.pallidipes* whenever *Exhyalanthrax abruptus* parasitised them. The same occurred whenever fungal pathogens infected the puparia (Vey, 1971). Studies on the puparia weight of *G.pallidipes* have not as yet been undertaken under field conditions in Kenya. Such studies could be used as an independent measure of size of emerging tenerals. Therefore the objectives of the studies reported in this chapter were:

(á) to determine the normal limits of live puparial weight in the field, so that live puparia can be

distinguished from dead or parasitised puparia; and,

(b) to determine seasonal changes in puparial weight of *G.pallidipes*.

5.2 MATERIALS AND METHODS

5.2.1 Determination of the mean weight of ecloded tsetse puparia

The puparia collected from the field were taken to the laboratory and weighed using a Mettler (PE-360) balance. The weighed puparia were then kept individually in ventilated plastic containers (diameter= 3.0 cm) which were in turn placed in a large metal tray containing sand in which 50 mls of water was added. The tray was kept in an insectary at Nguruman at an average temperature of 26 ^{0} C (range 24-28 ^{0} C) and 60-70 % relative humidity until eclosion. Dates of collections and emergences were recorded. The emerging flies were sexed and recorded. The weights of puparia from which teneral flies emerged were then used to determine live puparia weight.

5.2.2 Determination of the mean weight of dead and parasitised tsetse puparia

The puparia which did not eclode within 2-3 months were assumed dead and their weights were noted and used to determine the mean weights of dead puparia. The weights of puparia from which *Exhyalanthrax* parasites had emerged were used to determine the mean weights of parasitized puparia. Frequency distributions for live, parasitised and dead puparia weights were then determined.

5.2.3 Determination of the mean weight of tsetse puparia during dry and rainy months of the year at Nguruman

The mean weights of puparia collected during the dry months (i.e October, November, December, January, February and March) (mean temperature = 27.9 ^{o}C) and during the rainy months (i.e April, May, June, July, August, September) (mean temperature = 27.2 ^{o}C) were recorded and frequency distributions established. A t-test was used to determine whether there were significant difference between mean puparia weights during the dry and rainy months.

5.2.4 Determination of the meteorological factors influencing seasonal changes in tsetse puparia weights

The climatic factors in the general Nguruman area and in larviposition sites were determined as described in chapter five of this thesis. The weights of ecloded puparia were determined as already described. The relationship between puparia weight and the corresponding climatic indices of the same month or the previous month was investigated by regression analysis to reveal if there was any relationship between the weather factors and tsetse puparia weight.

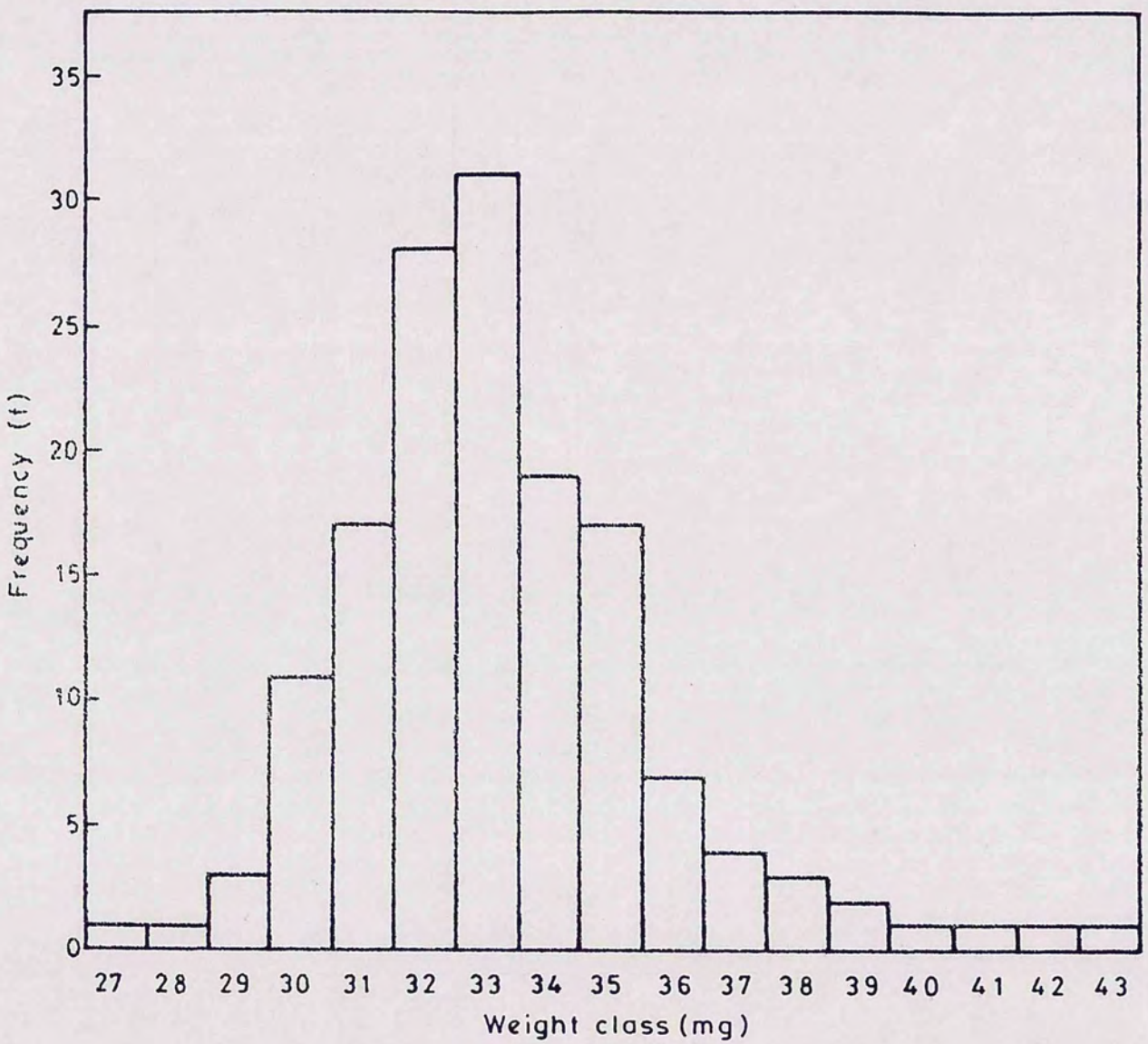
5.3 RESULTS

5.3.1 Determination of the mean weight of ecloded puparia

Figure 9 shows the frequency distribution of the weight of 151 ecloded puparia. The data shows that the weight distribution appproximated a normal curve with the mode at 33.0 mg. The calculated mean weight of ecloded puparia was found to be 33.30 $\pm$ 0.6 mg. When the following formula was applied to data collected:

x = mean $\pm$ (1.96 x standard deviation), where x= 33.30 and 1.96 = the probability value of the mean lying within 95 % confidence limit, it was found that puparia with a mean weight of less than 28.3 mg (i.e 33.3 $\pm$ (1.96 x 2.54) were unlikely to be live (P<

Figure 9 : Frequency distribution of weight of ecloded puparia at Nguruman.



0.05). Thus dead puparia invariably became desiccated and weighed less than 28.0 mg.

5.3.2 Determination of the mean weight of dead and parasitised puparia

Figure 10 shows a frequency distribution of weights of dead puparia. A total of 109 puparia did not eclode and were assummed dead. There were wide variations in the weights of dead puparia ranging from 3.0-40.0 mg (average= 14.0 mg). The majority of dead puparia weighed below 28.3 mg. However, there were dead puparia (32.1 %) weighing more than 28.3 mg.

Figure 11 shows the frequency distribution of the weights of parasitised tsetse puparia. The distribution of the weights approximated a normal curve with most of the parasitised puparia weights occurring in the 28.0 and 29.0 mg weight classes. The calculated mean weight of parasitised puparia was found to be 28.3 $\pm$ 0.3 with a range of 26.0-30.0 mg.

Figure 12 shows a comparison of weight distributions of live, parasitised and dead puparia. The data shows that parasitised puparia could not be distinguished from live puparia on the basis of weight although on average they were lighter than live puparia. The majority of dead puparia were below 28.3 mg weight class. However, dead puparia of higher

Figure 10 : Frequency distribution of weight of dead puparia from Ngurumon.

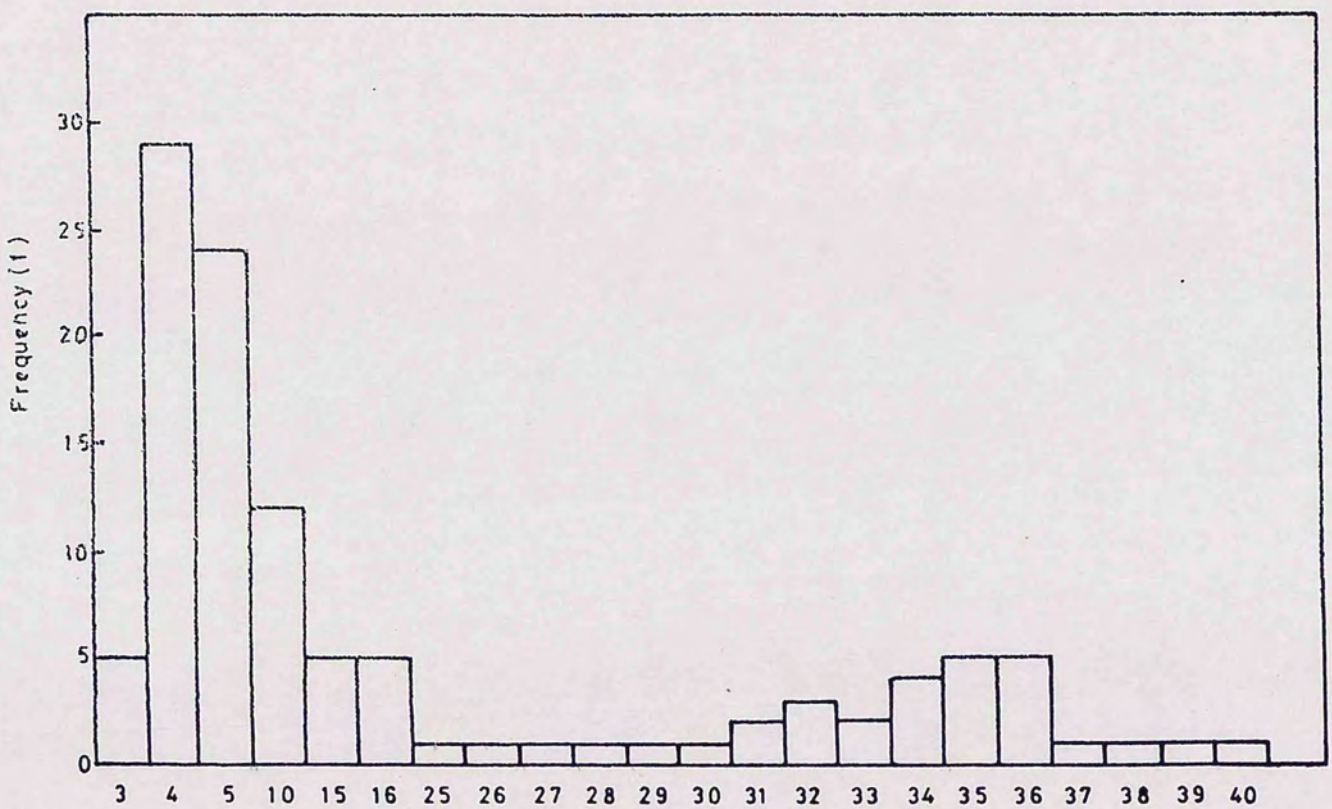


Figure 11 : Frequency distribution of parasitised puparia at Nguruman.

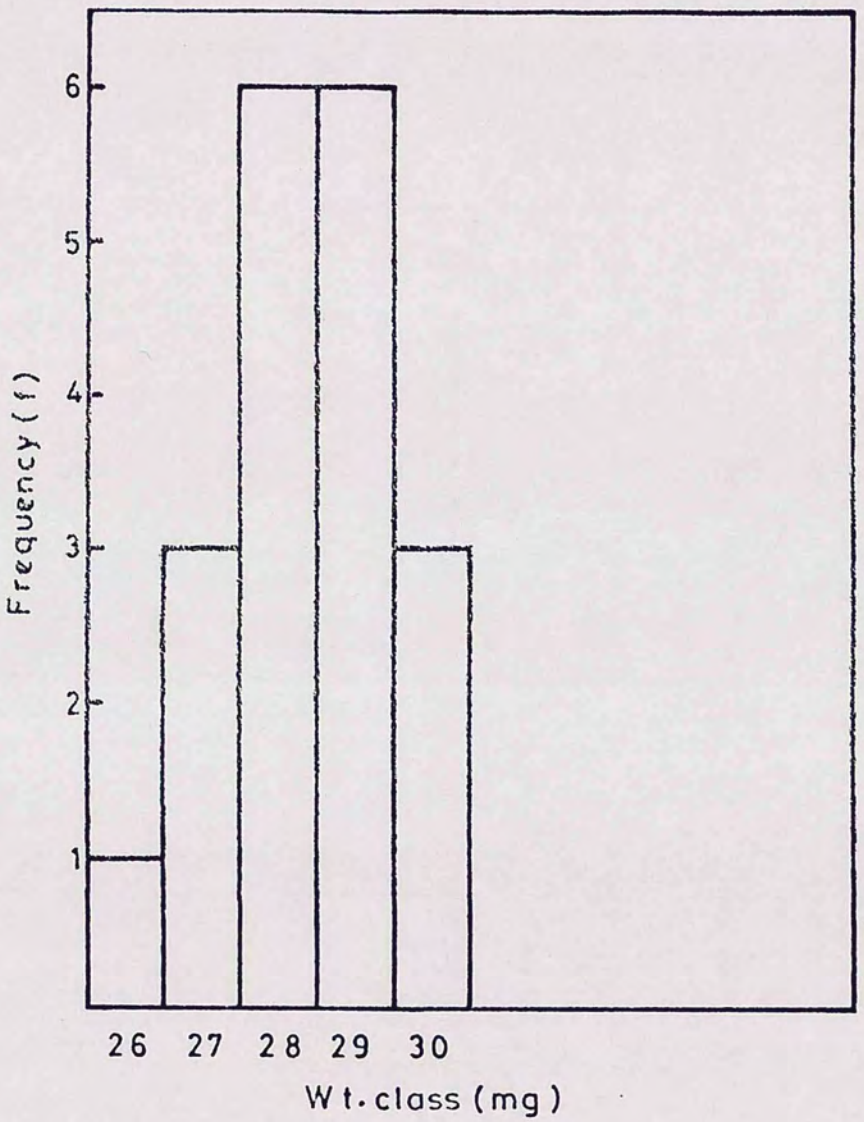
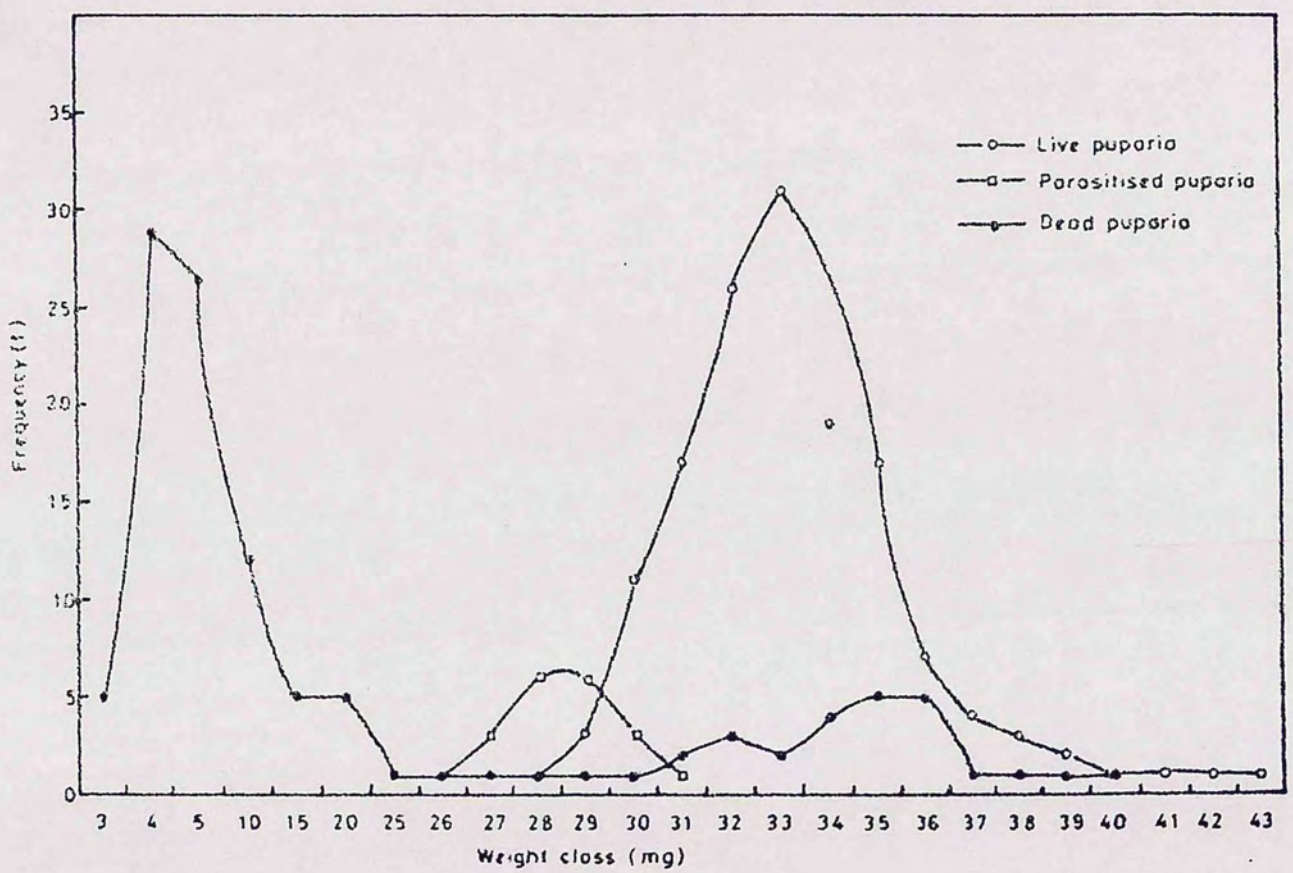


Figure 12 : comparison of weight distribution of live, parasitised and dead puparia



weights overlapped with the weights of live and parasitised puparia.

5.3.3 Determination of the mean weight of live puparia in the dry and rainy months

Figure 13 shows the frequency distributions of weights of puparia collected during the dry and rainy months of the year at Nguruman. The data shows that puparia with low weights occurred in the dry (A) than in the rainy months (B). The mean weight of puparia collected in the rainy months was 34.40 mg while that of puparia collected in dry months was 32.80 mg. When t-test was applied to the two sets of data it was revealed that the difference between mean weights of puparia in dry and rainy months was highly significant ($t = 13.2$, $P<0.001$).

5.3.4 Relationship between puparia weights and meteorological data at Nguruman

Figures 14 and 15 show the changes that occurred in mean puparia weights in relation to relative humidity. From the data collected, it was revealed that a significant correlation ($r = 0.70$, $P< 0.01$) existed between puparia weight and relative humidity of the previous month. On the other hand no relationship was

Figure 13: Frequency distribution of puparia weight in dry and rainy months at Nguruman.

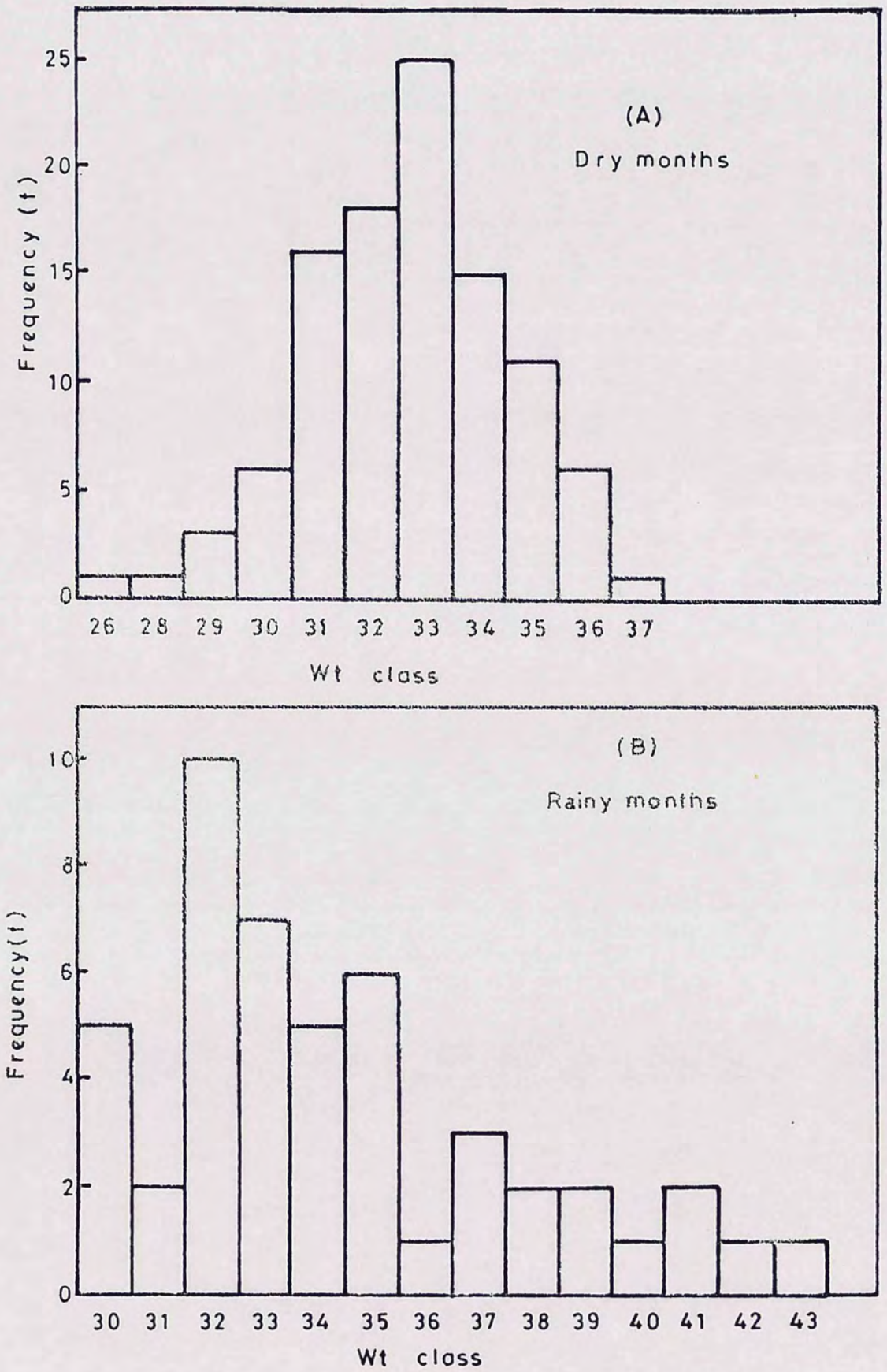


Figure 14 : Relationship between seasonal change in pupal weight and relative humidity the previous month.

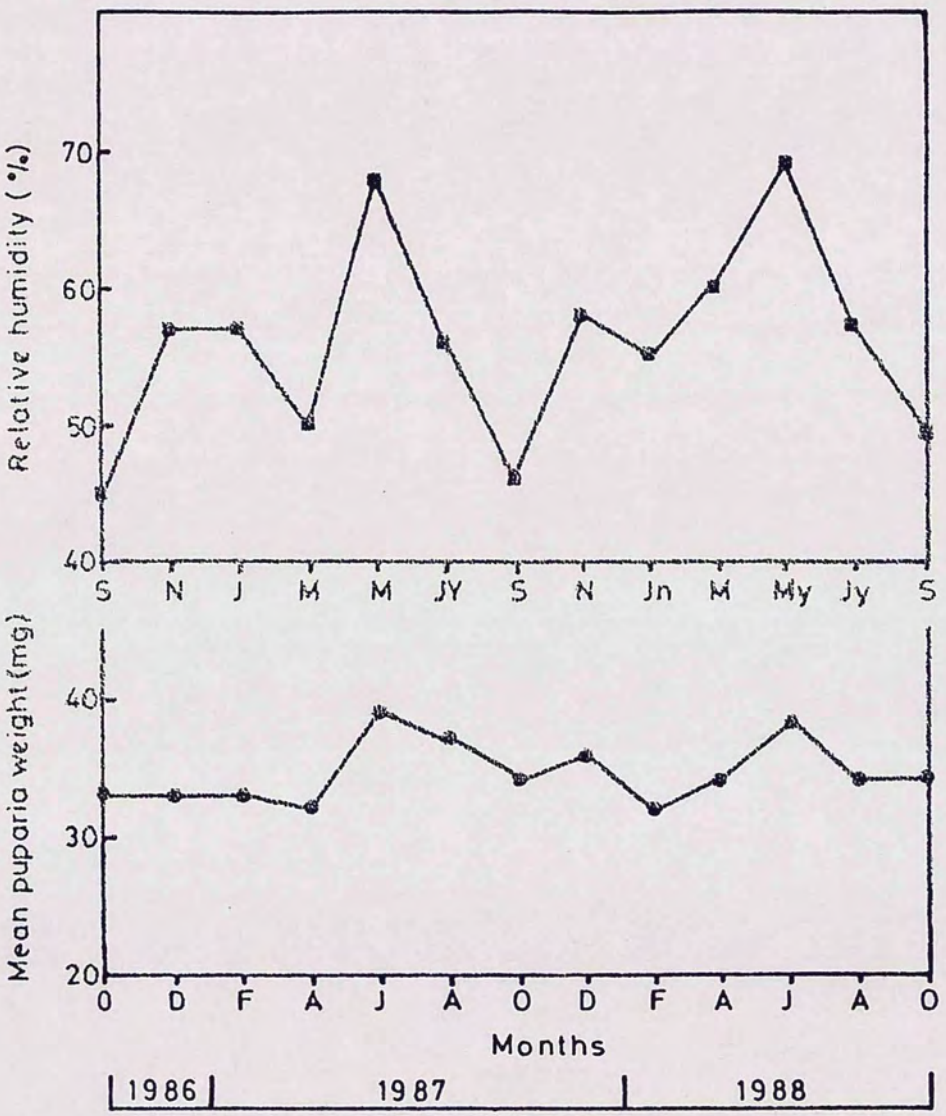
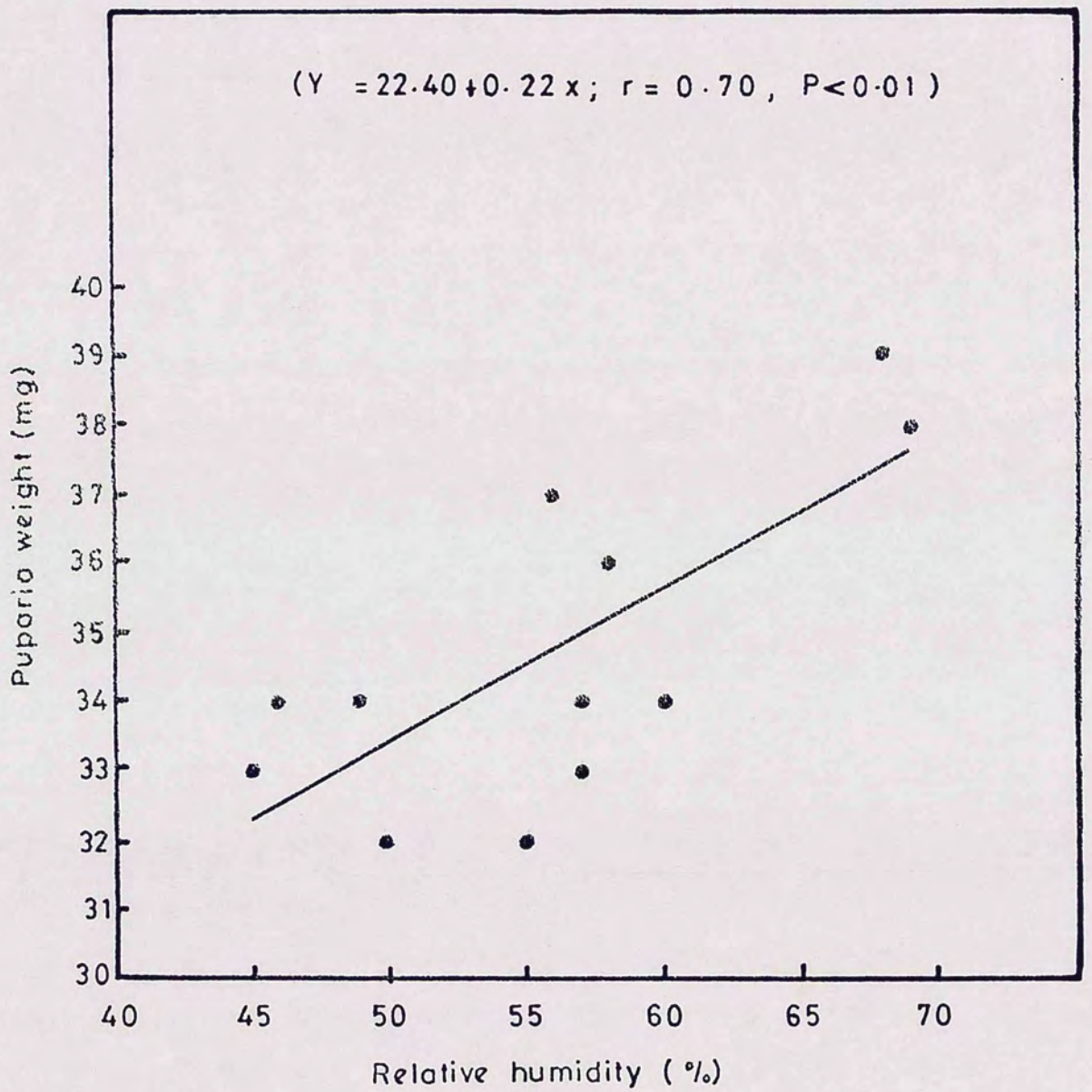


Figure 15 : Relationship between puparia weight and mean relative humidity the previous month.



established between puparia weight and both mean temperature and rainfall of the previous month, indicating that these parameters did not have any influence on puparial weights.

5.4 DISCUSSION

The mean weight of ecloded puparia at Nguruman was 33.30 $\pm$ 0.6 mg. From the distribution of live puparia weights it was found that puparia whose weight was less than 28.3 mg were likely to be dead while those weighing more than 28.3 mg were likely to be alive. Hursey (1970) reported similar observations in his studies in which he found that the mean weight of puparia of *G.pallidipes* collected in the wild was 32.6 mg and puparia weighing less than 23.0 mg were dead. However the factors accounting for the deaths of puparia at Nguruman even when their weights (lowest critical weight = 28.3 mg) were well above 23.0 mg identified as being the lowest critical weight below which puparia died in Hursey (1970) studies, were not established. The involvement of differing environmental and experimental conditions under which the studies were conducted in causing this apparent disparity was suspected.

These studies revealed that puparia parasitization caused loss in their weight at Nguruman. Similar

observations depicting weight loss of parasitised puparia has been reported by other workers. For example, Van der Vloedt (1970), found that parasitism led to weight loss in puparia of *G.pallidipes* in Zimbabwe. Similarly, Hursey (1970) reported weight loss in *G.pallidipes* puparia parasitised by *Exhyalanthrax abruptus* at Kiboko in Machakos district.

In general, dead puparia were of two categories: light weight dead puparia and high weight dead puparia. Light weight dead puparia were identified as being those puparia which had died for many months if not years before collection and had dried out completely accounting for their very low weights. On the other hand high weight dead puparia were identified as being those puparia that could have recently died in the field or those that might have been alive when collected but died during pupation before emergence.

Several factors could have caused death in high weight dead puparia. For instance, puparia collected immediately after rains could have died due to asphyxiation caused by flooding in larviposition sites or death could have occurred due to attack by pathogens such as fungal infections before emergence. From the foregoing observations, it became apparent that weight was not a suitable basis on which to base categorization of dead puparia.

There were significant seasonal differences in the mean weight of puparia of *G.pallidipes* collected from Nguruman. Puparia collected during the rainy season (April-September) were generally heavier than those collected during the dry period of the year (October-March). Similar observations have been reported in studies of puparia of other tsetse species (Buxton and Lewis 1934; Van der Vloedt, 1970). Seasonal variation in puparia weight has been attributed to nutritional status of the pregnant females (Mellanby, 1937), temperature regimes at which the pregnant females were kept during pregnacy (Jackson, 1939) and loss of water during the free larval period of the puparia stage (Bursell, 1958).

The seasonal changes in puparia weights were correlated to changes in relative humidity of the previous month. This was probably so because these were factors experienced by the parent female flies.

Since low weight puparia occurred during the hot and dry season, it follows that small sized nulliparous flies were produced during this period of the year. This has been reported to be the period when the parent female flies suffer from nutritional stress (Mellanby, 1937; Dransfield *et al.*, 1989); their progeny are small in size and are likely to die before taking their first blood meal. Size-dependent mortality has been reported in nulliparous flies of tsetse flies at Nguruman (

Dransfield *et al.*, 1989). They (Dransfield *et al.*, 1989) reported that small sized flies were less viable and died at the time of emergence since they were unable to fly or feed properly.

Since size-dependent mortality was correlated to relative humidity of two months previously while puparia weights were correlated to relative humidity of the previous month, it would seem that humidity has a significant effect in determining the size of flies and weights of puparia from which the flies emerge. Studies on live puparia weights is therefore of great biological importance because they could be used as an independent measure of size of emerging nulliparous flies.

CHAPTER SIX

FACTORS AFFECTING MORTALITY OF *G.PALLIDIPES* PUPARIA AT NGURUMAN

6.1 INTRODUCTION

In order to control insect pest and vector populations, it is important to identify the key mortality factors so that their regulatory effects could be preserved or their effectiveness deliberately enhanced (Varley and Gradwell, 1960). Insect populations are usually under constant action of density dependent and density independent mortality factors (Smith, 1935). Density independent factors such as adverse climatic and environmental conditions cause seasonal fluctuations in insect populations, while density dependent factors such as competition (intraspecific or interspecific), predation, parasitism and the action of pathogens such as fungal infections are primarily responsible for regulating the population around an equilibrium level (Varley et al., 1973).

Several methods could be applied in studies aimed at the identification of mortality factors of puparia. One technique would be to dissect non-ecloded puparia to identify the cause of mortality. Another method used before is to keep field collected tsetse puparia under

laboratory conditions and determine percentage emergence. This method also enables the determination of percent parasitism and identification of the parasites involved (Bursell, 1960). The method cannot however provide data on loss rates of puparia through predation (Jackson,1949). Once the puparia sub-mortality factors have been identified they could be incorporated in the construction of a life table budget of tsetse population.

Since flooding has been reported to cause puparia mortality in the field (Nash, 1933a; Buxton and Lewis, 1934), some experiments on this aspect as a mortality factor in tsetse puparia at Nguruman were conducted.

The objectives of studies reported here were:

(a) to determine the rate and pattern of emergence of tsetse and parasites from *G.pallidipes* puparia;

(b) to determine other causes of non-emergence of field collected tsetse puparia:

(a) already dead puparia

(b) those puparia which died before emergence

(c) to determine the effect of flooding as a mortality factor on puparia

6.2 MATERIALS AND METHODS

6.2.1 Determination of the rate and pattern of emergence of tsetse and parasites from *G.pallidipes* puparia

Live puparia at the time of collection were kept individually in ventilated plastic containers which were in turn placed in a large metal tray of wet sand. The tray was kept under similar conditions as described in chapter V. Dates of puparia collection and tsetse and parasite emergence were recorded. The frequency of tsetse and parasite emergence at different times (06.00, 09.00, 12.00, 15.00, 18.00 and 21.00 hours) during the day were recorded in order to determine the diurnal pattern of emergence. The emerging flies were sexed while the emerged parasites were kept in vials for later identification. The percentage emergence rate was then calculated using the following formula:

$$\text{Emergence rate (ER)} = \frac{\text{Number of emerged puparia}}{\text{Total number of live puparia}} \times 100$$

6.2.2 Determination of other causes of mortality of field collected tsetse puparia

Both dead puparia when collected in the field and those which died while being incubated before tsetse

emergence were dissected and examined under a binocular microscope to determine the possible causes of non-emergence. Puparia were regarded as being dead at the time of being collected in the field if their mean weight were less than 28.3 mg. When the puparia lasted more than 60 days after field collection without tsetse or parasite emergence they were regarded having died while in incubation. The causes of non-emergence were classified as follows:

(a) developmental abnormalities- included all instances of developmental and emergence failures;

(b) pupal tissue degeneration- included dead puparia which had their cases lined with rotten tissues representing decomposed pupa;

(c) fungal infected puparia- these included puparia infected with fungal mycelia or retained dead adult flies covered with fungal mycelia and,

(d) dead parasitised puparia- some puparia were found to contain dead parasites and were included with emerging parasites for quantification of this mortality factor.

The puparia infected with fungi were kept individually in clean plastic tubes and taken to the

laboratory at ICIPE Research Centre for culturing and identification.

All the sub-mortalities causing non-emergence of puparia were expressed as k-values which were estimated by subtracting log Nt+1 from Nt, where Nt is the number of live puparia on which the mortality factor acted upon and Nt+1 is the number of puparia surviving that mortality factor. Total puparia loss (k) in each month was determined by summing up k-values of all the sub-mortalities, k1, k2, k3, k4, etc or by subtracting log number of adult flies emerging from log number of puparia collected in that month. Varley and Gradwell's method (1960, 1970) was then applied in partial generation key factor analysis. The k-values of each sub-mortalities were then plotted against months to determine the seasonal variations of the sub-mortalities affecting puparia. The relationship between mortality factors and puparia densities was determined by plotting the k-values against the log puparia densities on which they acted. Finally the relationship between k-values from key factor analysis and climatic factors were investigated using regression analysis.

6.2.3 Determination of the effect of flooding as a mortality factor of *G.pallidipes* puparia

Experiments were designed to investigate the effect of flooding on the survival of puparia under waterlogged conditions. Laboratory reared puparia were buried randomly in trays containing waterlogged clay and sandy soils for durations of 3, 6, 9, and 12 hours respectively. Groups of 10 puparia were buried and three replicates were made for each duration. Small pegs painted white and 30 cm long were used to mark the actual positions where puparia were buried. The retrieved puparia were dried with a tissue paper and then kept individually in ventilated plastic tubes which were in turn buried in a large tray of wet sand and kept in an insectary to await emergence. The ecloded puparia were counted and recorded while those puparia which did not eclode within 60 days after submersion were assumed dead. A control experiment on flooding was also set up in which puparia were buried in soils which were not subjected to flooding.

6.3 RESULTS

6.3.1 Determination of the rate and pattern of emergence of tsetse and parasites from *G.pallidipes* puparia

The estimated percent emergence of *G.pallidipes* in the different months during the study period is shown in

Table 3: Monthly percentage emergence in puparia of *G.pallidipes* at Nguruman. Figures in parenthesis are uncorrected estimates of % emergence.

Year	Month	Puparia collected	Live puparia	Emerged flies	Percentage emergence
1986	Sept.	42	25	25	100.0 (59.5)
	Oct.	40	27	17	63.0 (42.5)
	Nov.	20	7	2	28.6 (10.0)
	Dec.	6	6	0	- (-)
1987	Jan.	3	1	1	100.0 (33.3)
	Feb.	7	6	5	83.3 (71.4)
	Mar.	11	3	2	66.7 (18.2)
	Apr.	8	6	1	16.7 (12.5)
	May	2	2	1	50.0 (50.0)
	June	3	1	1	100.0 (33.3)
	July	3	0	0	- (-)
	Aug.	13	10	7	70.0 (53.8)
	Sept.	3	3	0	- (-)
	Oct.	27	24	23	95.8 (85.2)
	Nov.	30	29	25	86.2 (83.3)
	Dec.	2	2	1	50.0 (50.0)
1988	Jan.	8	7	6	85.7 (75.0)
	Feb.	5	3	3	100.0 (60.0)
	mar.	14	13	13	100.0 (92.9)
	Apr.	0	0	0	- (-)
	May	0	0	0	- (-)
	June	0	0	0	- (-)
	July	6	4	3	75.0 (50.0)
	Aug.	12	10	9	90.0 (75.0)
	Sept.	6	5	3	60.0 (50.0)
	Oct.	5	4	4	100.0 (80.0)

Table 4: Seasonal fluctuations in the emergence rate of *G.pallidipes* at Nguruman between 1986-87.

Seasons	Months	Live puparia	Number emerged	Percent (%) emergence
Short rains	Oct/ Dec.	40	19	47.5
Hot/ dry	Jan/ Mar.	10	8	80.0
Long rains	Apr/ June	9	3	33.3
Cold/ dry	Jul/ Sept.	13	7	53.8

Figure 16: Monthly percentage emergence of G.pallidipes at Nguruman between 1986 - 1988.

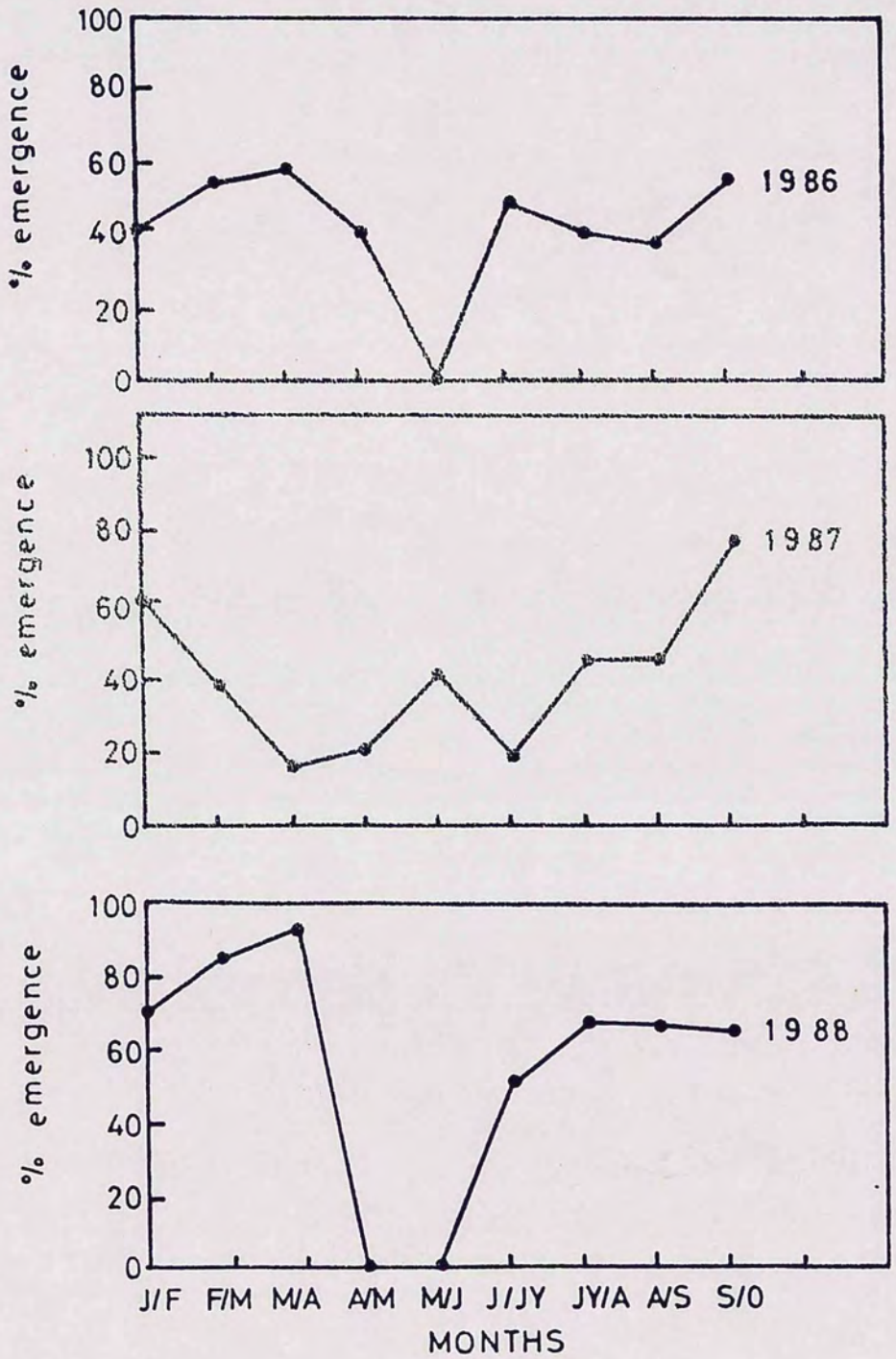


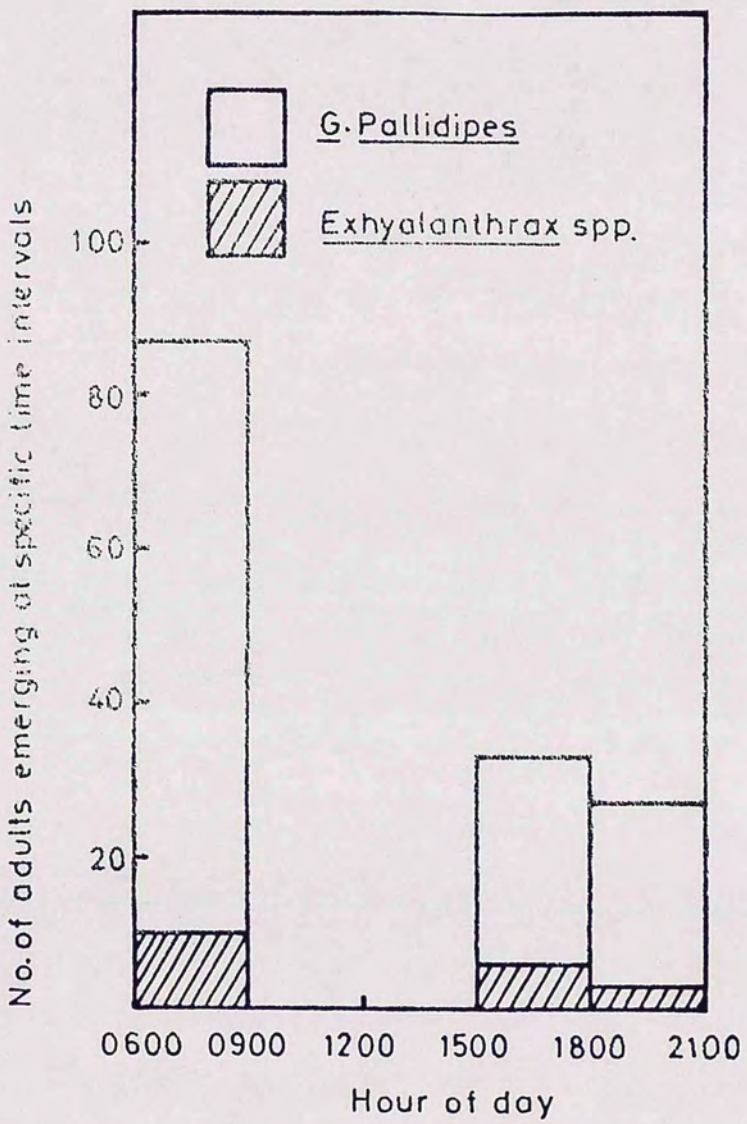
Table 3. Adult tsetse emergence from live puparia at the time of collection ranged from 16.7-100.0 % and averaged 58.5 ± 0.6 (Table 3). Higher rates of emergence occurred in September (100.0%) and October (63.0 %) in 1986; January (100.0 %), February (83.3 %), June (100.0 %) and October (95.8 %) of 1987; February (100.0 %), March (100.0 %) and October (100.0 %) in 1988, while fewer or no emergences occurred in December (0.0 %) in 1986; April (16.7 %) and September (0.0 %) in 1987 (Table 3). The low emergence rates mainly coincided with seasonal transitions of climatic conditions. Figure 16 shows a comparison of two month running means of uncorrected estimates of percent emergence between 1986-88. The data collected shows that there was tsetse emergence throughout the study period except in May and June 1986 and in April, May and June in 1988. The seasonal fluctuations in the rate of emergence between 1986-87 is shown in Table 4. Data in Table 4 shows that the highest emergence rate (80.0 %) occurred during the dry season but dropped during the long rainy season.

The parasites parasitising *G.pallidipes* puparia at Nguruman were identified as belonging to the genus *Exhyalanthrax* (=*Thyridanthrax*) of the family Bombyliidae. The two species found were *Exhyalanthrax lugens* (Lw) and *E.beckerianus* Bezzi. Their seasonal fluctuations in percentage puparia parasitism is shown

Table 5: Seasonal fluctuations in percent puparia parasitism due to *Exhyalanthrax* parasites at Nguruman. Number of parasites in parenthesis.

		1986-87		1987-88	
Month	Season	Number puparia	percent parasitism	Number puparia	percent parasitism
Oct/ Dec.	Short rains	40 (14)	35.0	3 (3)	100.0
Jan/ Mar.	Hot dry	10 (0)	0.0	0 (0)	0.0
Apr/ June	Long rains	9 (0)	0.0	0 (0)	0.0
July/ Sept.	Cold dry	13 (2)	15.4	2 (0)	0.0

Figure 17: Diurnal pattern of adult emergence.



in Table 5. The data presented (Table 5) indicated that puparia parasitism due to *Exhyalanthrax* parasites was highest (35-100 %) in the short rainy season at Nguruman between 1986-88. There was also considerable variations in the rates of puparia parasitism in the different years of study (Table 5). In general, puparia parasitism due to *Exhyalanthrax* parasites was higher in 1987-88 than in 1986-87 at Nguruman (Table 5).

Figure 17 shows the diurnal pattern of emergence of tsetse and parasites from 152 field collected tsetse puparia. It is shown in Figure 17 that there was a bimodal pattern with a major peak occurring in the morning between 06.00 -09.00 hours and a minor peak occurring between 15.00 -21.00 hours in the evening for both tsetse and parasites. No emergence occurred between 09.00 -15.00 hours (Figure 17).

6.3.2. Determination of other causes of non-emergence of field collected tsetse puparia

Other causes of non-emergence of field collected live puparia of *G.pallidipes* were due to developmental and emergence failures, pupal tissue degeneration, and fungal infections. Table 6 shows the seasonal fluctuations in the causes of non-emergence in puparia of *G.pallidipes*:

Table 6: Seasonal fluctuations in causes of non-emergence in puparia of *G.pallidipes* collected in the field between September 1986 and September, 1987. Percentages in parenthesis.

Season	Number dissected	Develop/ emergence failure	Tissue degeneration	Fungal infections	Dead parasites
Short rains (Oct/Dec)	40	10 (25.0)	6 (15.0)	20 (50.0)	4 (10.0)
Hot dry (Jan/Mar)	11	4 (36.4)	2 (18.2)	5 (45.5)	0 (0.0)
Long rains (Apr/Jun)	13	3 (23.1)	3 (23.1)	7 (53.8)	0 (0.0)
Cold dry (Jul/Sep)	29	10 (34.5)	4 (13.8)	15 (51.7)	0 (0.0)
Total	93	27 (29.0)	15 (16.1)	47 (50.5)	4 (4.3)

(a) developmental and emergence failures (Table 6)-

out of 93 dead puparia dissected, 27 (29.0 %) showed developmental and emergence failure; Mortality due to this factor was highest in the dry season (36.4 %);

(b) pupal tissue degeneration (Table 6)- out of 93 dead puparia examined 15 (16.1 %) contained degenerated pupal tissue and the highest (23.1 %) was observed during the long rainy season;

(c) fungal infections (Table 6)- puparia mortality due to fungal infections was quite high being 50.5 % as compared to 29.0 % due to developmental and emergence failure and 16.1 % due to pupal tissue degeneration (Figure 18); fungal mortality was highest in the wet than during the dry seasons; for example, during the long rainy season (April-June) 53.8 % (N= 7) of the tsetse puparia died due to fungal infections while during the short rainy season (Oct-Dec) 50.0 % (N= 20) casualties were due to fungal infections (Table 6); when the isolated fungal spores were cultured in the laboratory the complex

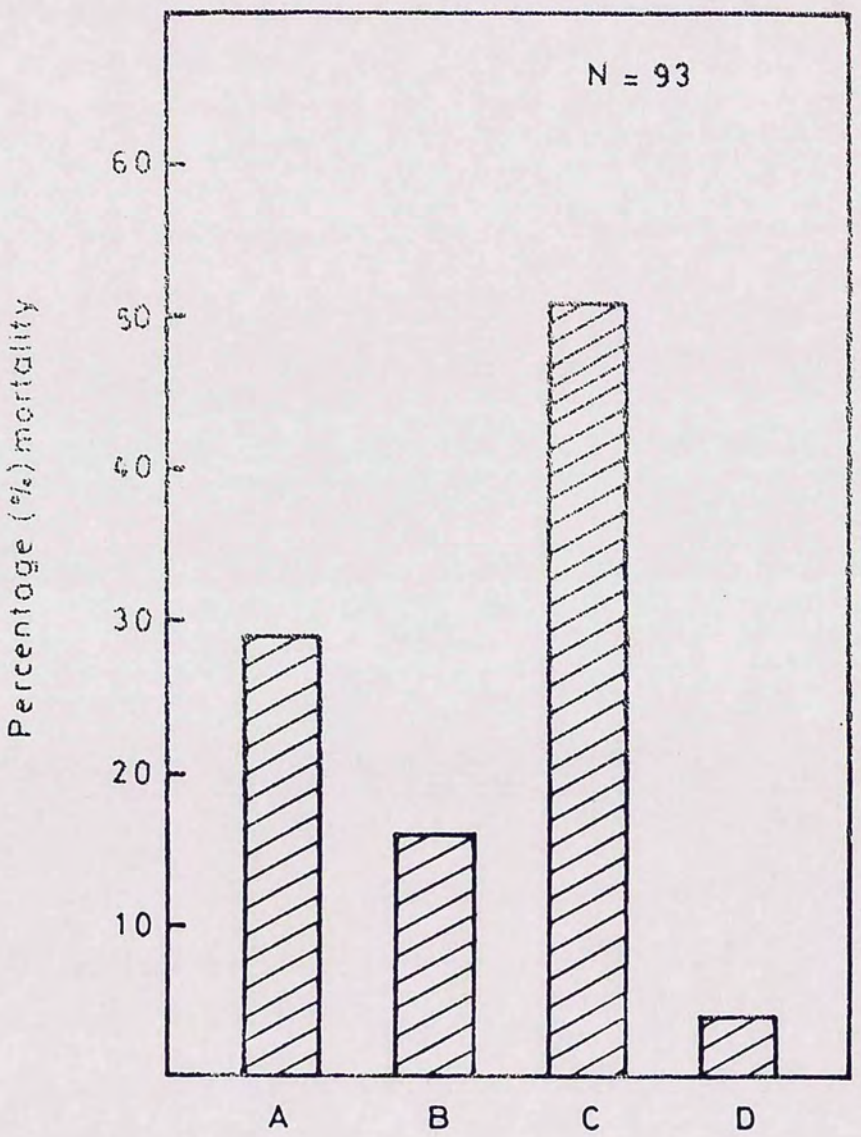
Figure 18: Overall percentage mortality in puparia of G. pallidipes due to

(A) developmental and emergence failure,

(B) pupal tissue degeneration,

(C) fungal infections,

(D) dead parasites.



of fungi species identified as infecting tsetse puparia at Nguruman were:

(i) *Aspergillus flavus* Link;

(ii) *Aspergillus niger* Link;

(iii) *Fusarium* spp

(iv) *Penicillium* spp;

(v) *Trichoderma* spp; and

(vi) *Rhizopus* spp;

(d) puparia containing dead parasites (Table 6)- only 4 (4.2 %) out of 93 puparia dissected contained dead parasites.

The life table data (Tables 7a and b) for *G.pallidipes* puparia assumes that mortality factors due to developmental and emergence failures (k1), pupal tissue degeneration (k2), parasitized puparia (k3) and fungal infections (k4) acted sequentially. Tables 7a and b show the uncorrected partial life table data of *G.pallidipes* puparia between 1986-87. The data shows that fungal infections were the major cause of puparia mortality (k-value = 0.22) during 1986-87 (Table 7a). The other causes of mortality included developmental and emergence failures (k-value = 0.08); pupal tissue degeneration (k-value = 0.06) and puparia parasitism (k-value = 0.08). Table 7b shows the corrected partial life table data of *G.pallidipes* puparia. The data

Table 7a: Uncorrected partial life table data for *G.pallidipes* at Nguruman. (N= No.observed, k=k-value, 1986-87).

Year	Month		Puparia collect.	Develop & emerg. failure	Pupal tissue degeneration	Parasitised puparia	Fungal infection	Adults emerged	Stage mort.
1986	Sept	N=	42	7	1	1	9	24	
		k		0.08	0.01	0.01	0.14		0.24
	Oct	N=	38	1	3	10	7	17	
		k		0.01	0.04	0.15	0.17		0.37
	Nov	N=	20	6	2	6	4	2	
		k		0.15	0.07	0.03	0.48		0.73
	Dec	N=	6	1	1	0	4	0	
		k		0.08	0.10	0.0	0		0.48
1987	Jan	N=	2	0	0	0	2	0	
		k		0.0	0.0	0.0	0.0		0.0
	Feb	N=	2	1	0	0	0	1	
		k		0.30	0.0	0.0	0.0		0.30
	Mar	N=	7	3	2	0	1	1	
		k		0.25	0.30	0.0	0.30		0.85
	Apr	N=	5	0	2	0	3	0	
		k		0.0	0.22	0.0	0.48		0.70
	May	N=	1	0	0	0	0	1	
		k		0.0	0.0	0.0	0.0		0.0
	June	N=	3	2	0	0	0	1	
		k		0.48	0.0	0.0	0.0		0.48
	July	N=	3	1	1	0	1	0	
		k		0.18	0.30	0.0	0.0		0.48
	Aug	N=	11	2	1	1	2	5	
		k		0.09	0.05	0.05	0.15		0.34
	Sept	N=	0	0	0	0	0		
		k		0.0	0.0	0.0	0.0	0	0.0
Total		N=138		24	13	18	33	52	
		k		0.08	0.06	0.08	0.22		0.44

Table 7b(ii): Corrected partial life table data for *G.pallidipes* at Nguruman.
N= Number of live puparia determined by weight, K= k-value.

Year	Month	Live puparia	Develop.& emerg.failure	Pupal tissue degeneration	Parasitised puparia	Fungal infection	Adults emerged	Stage mort.
1986	Sept.	N=25	0	0	0	0	25	
		K	0.0	0.0	0.0	0.0		0.0
	Oct.	N=27	0	0	9	1	17	
		K	0.0	0.0	0.17	0.03		0.2
	Nov.	N=7	0	0	5	0	2	
		K	0.0	0.0	0.55	0.0		0.55
	Dec	N=6	2	0	0	4	0	
		K	0.18	0.0	0.0	0.60		0.78
1987	Jan.	N=1	0	0	0	0	1	
		K	0.0	0.0	0.0	0.0	0.0	0.0
	Feb.	N=6	0	0	0	1	5	
		K	0.0	0.0	0.0	0.08	0.08	0.16
	Mar.	N=3	0	0	0	1	2	
		K	0.0	0.0	0.0	0.18	0.30	0.48
	Apr.	N=6	1	1	0	3	1	
		K	0.08	0.10	0.0	0.06	0.12	0.36
	May	N=2	0	0	0	1	1	
		K	0.0	0.0	0.0	0.30	0.0	0.30
	June	N=1	0	0	0	0	1	
		K	0.0	0.0	0.0	0.0	0.0	0.0
	July	N=0	0	0	0	0	0	
		K	0.0	0.0	0.0	0.0	0.0	0.0
	Aug.	N=10	0	0	1	2	7	
		K	0.0	0.0	0.05	0.10	0.85	1.0
	Sept.	N=3	0	1	1	1	0	
		K	0.0	0.18	0.0	0.0	0.0	0.18
	Total	N=97	3	2	16	14	62	
		K	0.02	0.01	0.08	0.09		0.20

presented (Tables 7i, ii) confirmed earlier findings that fungal infections were the main cause of puparia mortality (k-value = 0.09) during 1986-87 period of study. Figure 19 summarises the seasonal variations in puparia mortality factors in which the sub-mortalities expressed in k-values were plotted against months. The pattern of change of k4 due to fungal infections was very similar to that of total mortality (K) from September, 1986 to September, 1987, indicating that fungal pathogens were the key factors contributing to total puparia mortality during the study period.

The role of each sub-mortality was evaluated by calculating the regression coefficient of the k-values of each submortality against the total mortality. The analysis showed that mortality due to fungi (k4) was the only significant mortality factor (r= 0.55, P< 0.05). The submortalities (k1) due to developmental and emergence failures (r = 0.43, P>0.05); (k2) due to pupal tissue degeneration (r = 0.08, P>0.05) and (k3) due to puparia parasitism (r=0.25, P>0.05) were not significant.

Figure 20 depicts the relationship between k-values and the puparia densities on which they acted (Table 7ii). Of all the mortality factors investigated none had any significant relationship with puparia densities on which they acted upon, indicating that none of them were density dependent.

Figure 19 : Seasonal variations in key factor submortalities of puparia (september, 1986 - september, 1988). (from corrected life table).

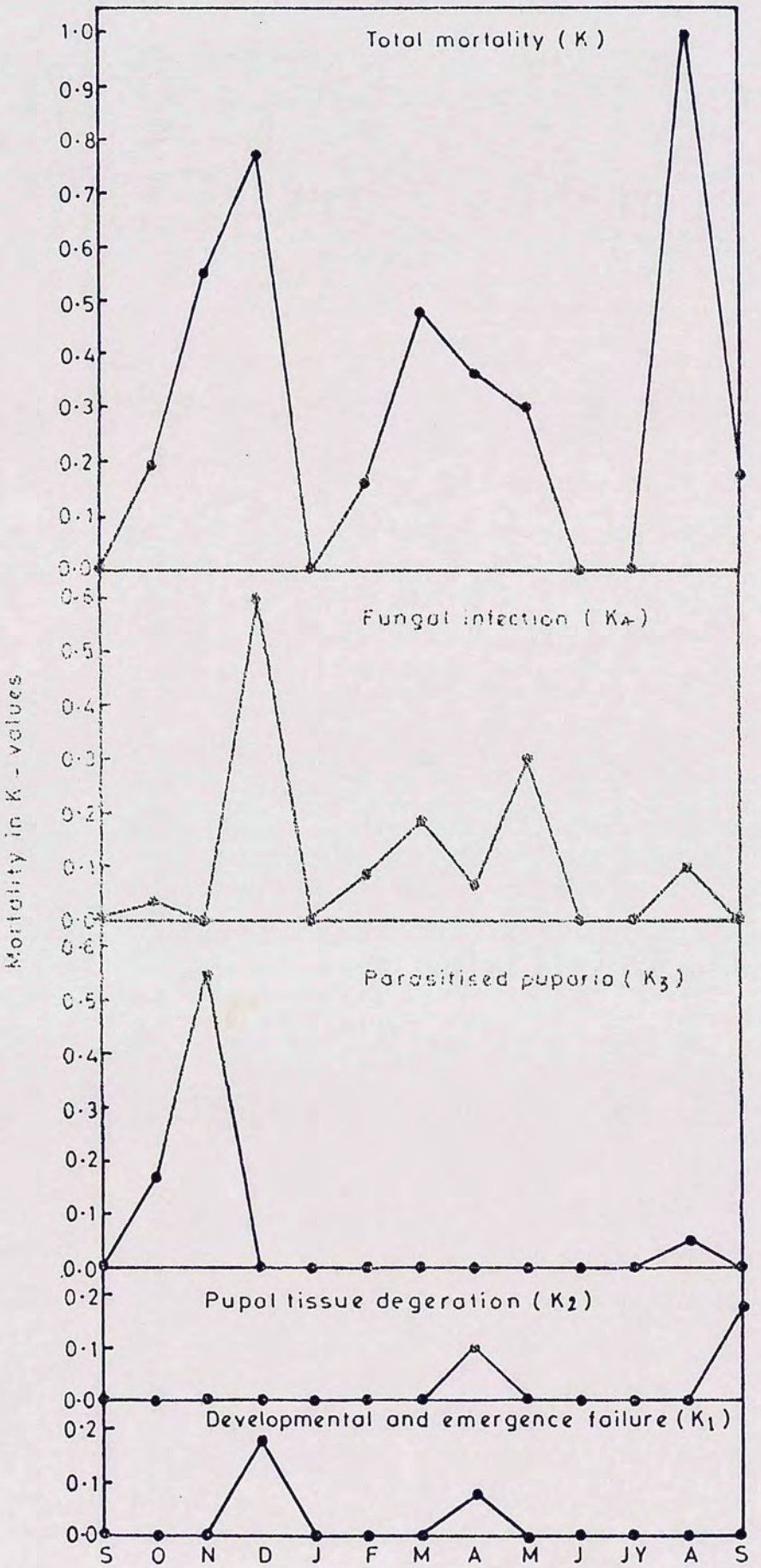
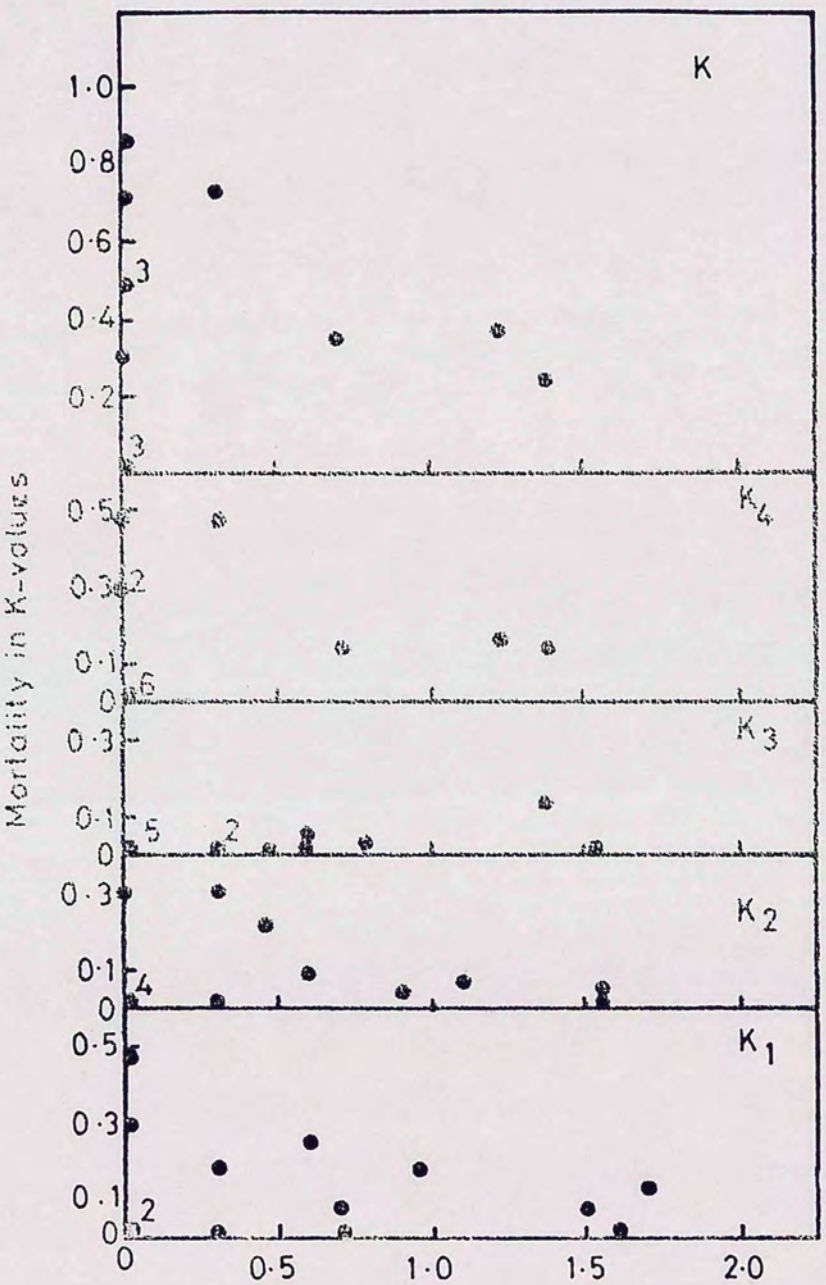


Figure 20: Relationship between K-values of submortalities and puparia densities on which they acted.



Of all climatic factors investigated (relative humidity: r = 0.02, P>0.05); rainfall: r= 0.01, P>0.05) and temperature: r= 0.05, P>0.05) none of them was significantly correlated with puparia mortality estimated from key factor analysis.

6.3.3 Determination of the effect of flooding on puparia survival

Table 8 gives the mean number of puparia alive after submersion in waterlogged clay and sandy soils. The data shows (Table 8) that 3 and 4 puparia survived after 3 hours of submersion in clay and sandy soils, respectively, while 2 and 3 puparia survived after 6 hours of submersion in clay and sandy soils, respectively. However, no puparia survived after 9 and 12 hours of submersion in both the soil types. The difference in puparia mortality in the two soil types as compared by the chi-squared test, was not significant (x^2 = 3.40, P> 0.05). All puparia buried in dry clay and sandy soils (controls) and kept under insectary conditions emerged indicating that flooding caused death to puparia and was therefore identified as one of the mortality factors of puparia in the field.

6.4 DISCUSSION

Table 8: Mean number of puparia alive after submersion for various time intervals in two different soil types and a chi-squared test (x^2)

Soil type	Mean no. of puparia alive after submersion period. 3	6	9	12 hours
Clay	3	2	0	0
Sandy	4	3	0	0

Variation in puparia mortality due to flooding in different soil types.

Soil type	No. buried	No. alive	No. dead	% mortality
Clay	30	15	15	50.0
Sandy	30	21	9	30.0
Total	60	36	24	

$X^2 = 3.40ns, P>0.05$

There were great variations in the rate of emergence of *G.pallidipes* from puparia collected at Nguruman. The emergence rate was higher in the dry season but fell just before or at the beginning of the rainy season. The reason for the higher emergence rate during the dry season was perhaps due to the high temperatures that prevailed during that season. Recorded temperatures during the season ranged between 33.6-38.5 ^{0} C (Dransfield, *et al.*, 1986). The foregoing observations were not peculiar in view of similar findings reported by earlier workers notably Jackson (1948). In his studies Jackson (1948) showed that the emergence rate of *G.morsitans* dropped drastically just before the rainy season and was attributed to low temperatures that came with the rainy season. It was shown that flooding of puparia while in the soil killed them. This factor helps to further explain why low tsetse emergence was observed during the rainy season.

The pattern of emergence of *G.pallidipes* puparia showed a bimodal pattern with a major peak in the morning and a minor peak in the afternoon. This observation is similar to the findings of earlier researchers on *G.pallidipes* (Jaenson, 1978, 1981; Van Etten, 1981). However in other species studied elsewhere, it was reported that most adults emerged in the afternoon. For example, in Burkina Faso, Challier

(1973) observed that the emergence rate of *G.palpalis gambiensis* was very low in the morning with most of the adult flies emerging in the afternoon. Laveissiere *et al.*, (1984a) reported a similar pattern of emergence in *G.tachinoides* puparia in the humid savannah in West Africa.

Exhyalanthrax lugens (Lw.) and *E.beckerianus* were found to be the most important parasites of *G.pallidipes* puparia at Nguruman. These species have also been found to parasitize other tsetse species. For example, *E.beckerianus* was found parasitising *G.tachinoides* and *G.morsitans* in Gadau northern Nigeria (Taylor, 1932); *G.tachinoides* in Chad (Gruvel, 1970a, b); and *G.swynnertoni* in Tanzania (Mulligan, 1970). *E.lugens* is also widely distributed in Kenya, Zimbabwe, Tanzania, Malawi, Zambia, Namibia and in South Africa and has been found parasitizing *G.brevipalpis*, *G.longipennis*, *G.morsitans*, *G.pallidipes* and *G.austeni* (Hess, 1956).

The mean percent parasitisim of *G.pallidipes* puparia by *Exhyalanthrax* spp. at Nguruman was 6.5 $\pm$ 4.0 %. This was a low level of parasitism as compared to 27.5 % recorded for *E.abruptus* Loew. for the same tsetse species at Kiboko, Machakos (Hursey, 1970). However, the low level of parasitism due to this parasite at Nguruman was comparable to parasitism levels recorded by Saunders (1960) and Minter (1971) in

other tsetse species including *G.pallidipes* in other parts of East Africa.

The low levels of parasitism due to *Exhyalanthrax* spp. could probably be due to, first, lack of coincidence of larviposition sites of tsetse and the parasites; secondly, due to the inability of the parasites to locate puparia buried deep in the soil; and thirdly, due to the inability of the parasites larvae to penetrate tsetse puparia after complete sclerotization. It is, moreover likely that parasitism rates reported in these studies were overestimated since the developmental period of the parasite is longer than that of tsetse (Hursey, 1970).

It was also revealed in these studies that parasitization rates by these parasites were highest at the beginning of the short rainy season at Nguruman. This was probably because parasitised puparia had accumulated in the dry season sites and the tsetse changed larviposition sites at the start of the rains in November; hence only parasitised puparia were left in these sites giving very high apparent parasitism rates.

There were considerable variations in the rate of puparia parasitism due to *Exhyalanthrax* parasites in the different years of study. For example, the rates were higher in 1987 as compared to those obtained in 1986. Similar fluctuations in puparia parasitism due to

these parasites were reported by other workers notably, Hursey (1970). For instance, Hursey (1970) found considerable annual variations in *G.pallidipes* puparia parasitism due to *E.abruptus* at Machakos.

It was also revealed that there were two categories of puparia which did not emerge. First, those puparia which were already dead at the time of collection (i.e those below the critical weight of 28.3 mg). These puparia had fungal infections after dissections indicating that they were attacked by saprophytic fungi. Secondly, there were those puparia which died in incubation before emergence. The causes of non-emergence of these puparia were due to developmental and emergence failures, pupal tissue degeneration, and fungal infections. Identical causes of non-emergence of puparia in other tsetse species have been reported in literature (Buxton, 1955; Mulligan, 1970).

Fungal infection which accounted for 50.5 % was the major cause for non-emergence of *G.pallidipes* puparia at Nguruman and its action was more enhanced during the rainy season probably because the season provided optimal conditions for fungal growth. Similar observations were reported for the puparia of *G.tachinoides* in Nigeria (Nash, 1933a, 1939) and *G.palpalis* in Ghana (Pomery, 1930). The fungi isolated from *G.pallidipes* puparia belonged to Fusarium,

Aspergillus, *Penicillium*, *Trichoderma* and *Rhizophus* species. Most of the fungal species isolated were saprophytic in nature and perhaps set in after puparia had been killed by other causes.

The key factor analysis carried out on causes of non-emergence gave the main causes of puparia mortality and the way in which they acted. Fungal mortality was confirmed as the key factor causing population change in puparia densities at Nguruman. None of the causes of non-emergence were density dependent, indicating that they played no regulatory role in puparial density.

CHAPTER SEVEN

STUDIES ON FIELD PREDATION OF *GLOSSINA PALLIDIPES* PUPARIA AT NGURUMAN

7.1 INTRODUCTION

Although several arthropods occur in larviposition sites of *Glossina*, not all are predators of puparia. The fact that they are regarded as non-specific (polyphagous) probably prejudiced studies on them by early workers and have not as yet received serious attention. One method used to identify predators of arthropods is the use of serological analysis (Dempster, 1958, 1960; O'Rourke, 1958; Rothschild, 1966; Sutton, 1970; Service, 1973; Ashby, 1974; Giller, 1986; Adabie, 1987). Using this technique Adabie (1987) positively identified the predators of *G.pallidipes* puparia at Nguruman. Adabie (1987) reported that several species of crickets and ants preyed on tsetse puparia. It is therefore became important to study the prevalence and distribution of these predators in order to determine the role they play in natural regulation of tsetse pouplations.

One of the major objectives in these studies on predation was to provide a basis for the understanding of natural regulation of tsetse population at Nguruman. This approach has been stressed as being valuable when

studying the regulation of populations of such arthropods (Rogers and Hubbards, 1974).

Predation on tsetse puparia has been reported by several ecologists in the past, and literature on the subject is extensive (see chapter 2). Literature on insect predation reveals that its rate depends on a number of factors including prey density, predator density, characteristics of the environment, the presence or absence of alternate prey and the attack techniques of predators (Leopold, 1933; Southwood, 1966; Holling, 1959b; Hassell, 1966, 1976; Hassell et al., 1976). These factors have been shown to affect the prey-predator interactions and their functional responses to changes in prey density.

Predation studies on tsetse have been very limited and very little is known about puparia predation in Kenya. This is partly due to the fact that puparia are generally more difficult to obtain than adult flies which are easily trapped. These studies were therefore undertaken with that in view so as to generate basic information on some aspects of the predation of tsetse puparia at Nguruman.

Although predation levels are most difficult to determine in the field, attempts have been made by some ecologists to quantify puparia predation under field situations (Ford, 1940; Kemp, 1951; Rogers, 1974). Specific investigations were carried out during these

studies in an attempt to quantify levels of predation at Nguruman.

The specific objectives of the studies were:

(a) to determine the species composition, prevalence and seasonal distribution of the predators of *G.pallidipes* puparia in larviposition sites

(b) to determine the rates of predation of puparia buried at different soil densities; and,

(c) to study the mode of predation of puparia buried at different densities

7.2. MATERIALS AND METHODS

7.2.1 Determination of the species composition, prevalence and distribution of the predators of *G.pallidipes* puparia in larviposition sites

The species composition, prevalence and seasonal distribution of the predators of puparia were monitored for two years in eight sites at Nguruman using unbaited pitfall traps between October 1986 and October, 1988. The pitfall traps which were positioned within the larviposition sites, consisted of 1 litre capacity glass jars sunk in a vertical position into the soil so that their mouths were level with the soil surface. They contained water to which a detergent had been

added to kill arthropods that fell into the traps. In the dry season additional water was added to compensate for evaporation while in the wet season extra detergent and formalin were added to prevent dilution by the rains.

Sampling of the predators took place at monthly intervals and the pitfall traps were allowed to accumulate samples continually for three days each month. All the pitfall traps were then inspected and emptied in the same order on every sampling occasion. The predators were removed with forceps and preserved appropriately in either vials containing 80 % alcohol or pinned for later identification. The identification of the trapped species was performed by using entomological keys in standard textbooks and later confirmed by the Kenya National Museum in Nairobi and the Commonwealth Institute of Entomology (CIE) in the United Kingdom. Counts of predators from all the pitfall traps were combined and used for estimating the relative numbers of the various predators of puparia in different months and seasons.

7.2.2 Determination of the rates of tsetse puparia predation at different soil depths

An experiment was designed in order to investigate the rate of puparial losses and damage due to predation

at three different soil depths in larviposition sites at Nguruman. Sample areas measuring one square metre (1 m^2) each were searched thoroughly to remove all live puparia and empty puparia cases prior to the experiments. Puparia of less than one week old produced by a colony of field collected flies of *G.pallidipes* were arranged in groups of eight and buried randomly at 0, 1.5, and 3.0 cm soil depths respectively. Three replicates were made for each depth. Puparia positions at different soil depths were marked with wooden pegs painted in different colours. The buried puparia were left in the field for 4 weeks after which they were excavated and assessed for survival, predation and complete loss.

7.2.3 Studies on predation of puparia buried at different densities

A latin square design was used to investigate puparial losses and damage due to predation at five densities in five different sites and over a period of five months. Laboratory bred puparia of *G.pallidipes* were buried at densities of 1, 2, 4, 8 and 16 per linear metre in five different sites. Pegs were used to locate the positions of the buried puparia within the experimental areas.

The buried puparia were left in the field under a layer of leaf litter and twigs for 4 weeks after which they were excavated and the surviving, predated and missing puparia recorded. The surviving puparia were then kept individually in the laboratory to await emergence.

7.3. RESULTS

7.3.1. Determination of the species composition, prevalence and distribution of the predators of *G.pallidipes* puparia

The species composition of puparial predators of *G.pallidipes* observed in larviposition sites of the fly are presented in Table 9. Identification for most of the predators were only possible up to generic level. The predators belonged to two families: formicidae (order: Hymenoptera) and gryllidae (order: Orthoptera) (Table 9). The formicid ants belonged to the following genera (Table 9):

(a) *Pheidole*;

(b) *Polyrachis*;

(c) *Camponotus*;

(d) *Paltothyreus*;

(e) *Acantholepis*; and

(f) *Viticicola*.

Table 9: Species composition of the predators of *G.pallidipes* puparia caught in larviposition sites at Nguruman

1. Order: Hymenoptera	
family: Formicidae	
genus: *Pheidole*	+++
Polyrachis	++
Camponotus	++
Paltothyreus	+
Acantholepis	+
Viticicola	+
2. Order: Orthoptera	
Family: Gryllidae	
genus: *Gryllus*	+++
Gryllulus	++
Homoeogryllus	+
Liogryllus	+
Phaeophillacris	+
Scapsipedus	+

Key:

+++ = Very abundant

++ = abundant

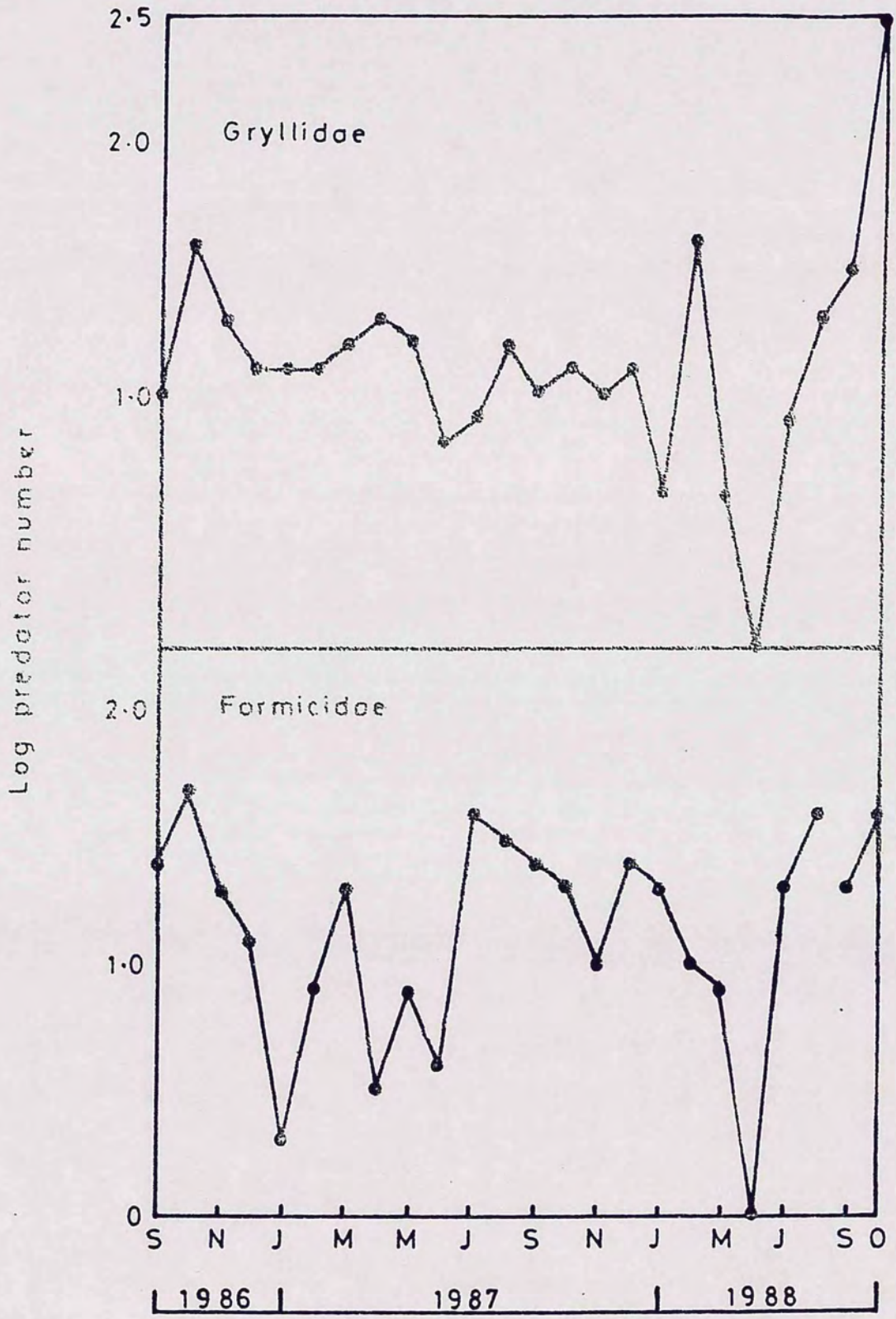
+ = rare

Table 10: Seasonal fluctuations in the number of predators of *G.pallidipes* in larviposition sites at Nguruman between September 1986 and October, 1988

Season	Month	Total number of predators			
		Formicidae		Gryllidae	
		a	b	a	b
Short rains	Oct-Dec.	131	86	110	314
Hot dry	Jan-mar.	24	60	38	80
Long rains	Apr-June	11	11	39	39
Cold dry	Jul-Sept.	118	181	41	85

a- 1986-87, b- 1987-88.

Figure 21 : Monthly fluctuations in population of pupal predators caught in larviposition sites.



The most abundant formicid ants in tsetse larviposition sites belonged to the genus *Pheidole*.

In the Orthopteran family Gryllidae, individuals from six genera were collected and were:

(a) *Gryllus*;

(b) *Gryllulus*;

(c) *Homoeogryllus*;

(d) *Liogryllus*;

(e) *Phaeophillacris*; and,

(f) *Scapsipedus*.

Of these, the most abundant genera was *Gryllus*.

Table 10 shows the seasonal fluctuation in the relative abundance of formicid ants and gryllids caught in larviposition sites during the study period. The data presented in (Table 10) shows that the population of formicid ants and gryllids were highest during the short rains and cold-dry seasons but fell during the long rains. Figure 21 shows the monthly fluctuations in the relative abundance of formicid ants and gryllids caught in larviposition sites between September 1986 and October, 1988. It is shown in Figure 21 that the population of formicid ants and gryllids followed a similar trend with peak incidence of both predators caught during the month of October (formicids = 65, (1986); 50 (1987); 63 (1988) and gryllids 52 (1986); 50 (1987); 288 (1988). No predators were caught during the long rains in April and May in 1988 probably due to

flooding of larviposition sites. On the overall, predator populations were higher in 1987-88 than in 1986-87 (Table 10).

7.3.2 Determination of the rates of tsetse puparia predation at different soil depths

Table 11 gives the mean number of how tsetse puparia were lost to predators at different soil depths in larviposition sites. The data (Table 11) shows that most (7.3 $\pm$ 0.3 puparia) were destroyed when left on soil surface as compared to the situation when they were buried at depths of 1.5 cm (4.0 $\pm$ 1.2 puparia) and 3.0 cm (1.7 $\pm$ 0.3 puparia). When the data collected was put to statistical analysis, it was found that the mean predation at the soil surface was significant ($F= 15.7$, $P<0.05$) as compared to predation at 1.5 cm and 3.0 cm soil depths.

7.3.3. Studies on predation of tsetse puparia buried at different densities in the field

Table 12a gives the mean percent predation of tsetse puparia buried at different densities in the field. The corresponding relationship between puparia density and percent predation is shown in Figure 22. Out of a total of 155 puparia buried, 33.6 % could not

Table 11: Number of predated puparia at different soil depths.

Puparia depth (cm)	Total predation in 3 replicates (i.e 24 puparia)	Mean ± S.E	F-ratio
0	22	7.3 ± 0.3	15.7* (2,6)
1.5	12	4.0 ± 1.2	
3.0	5	1.7 ± 0.3	

*- Significant at 5 % level

Table 12: Average percentage predation in puparia buried at different densities and analysis of variance of data collected at Nguruman. All densities replicated five times.

(a) Percentage predation

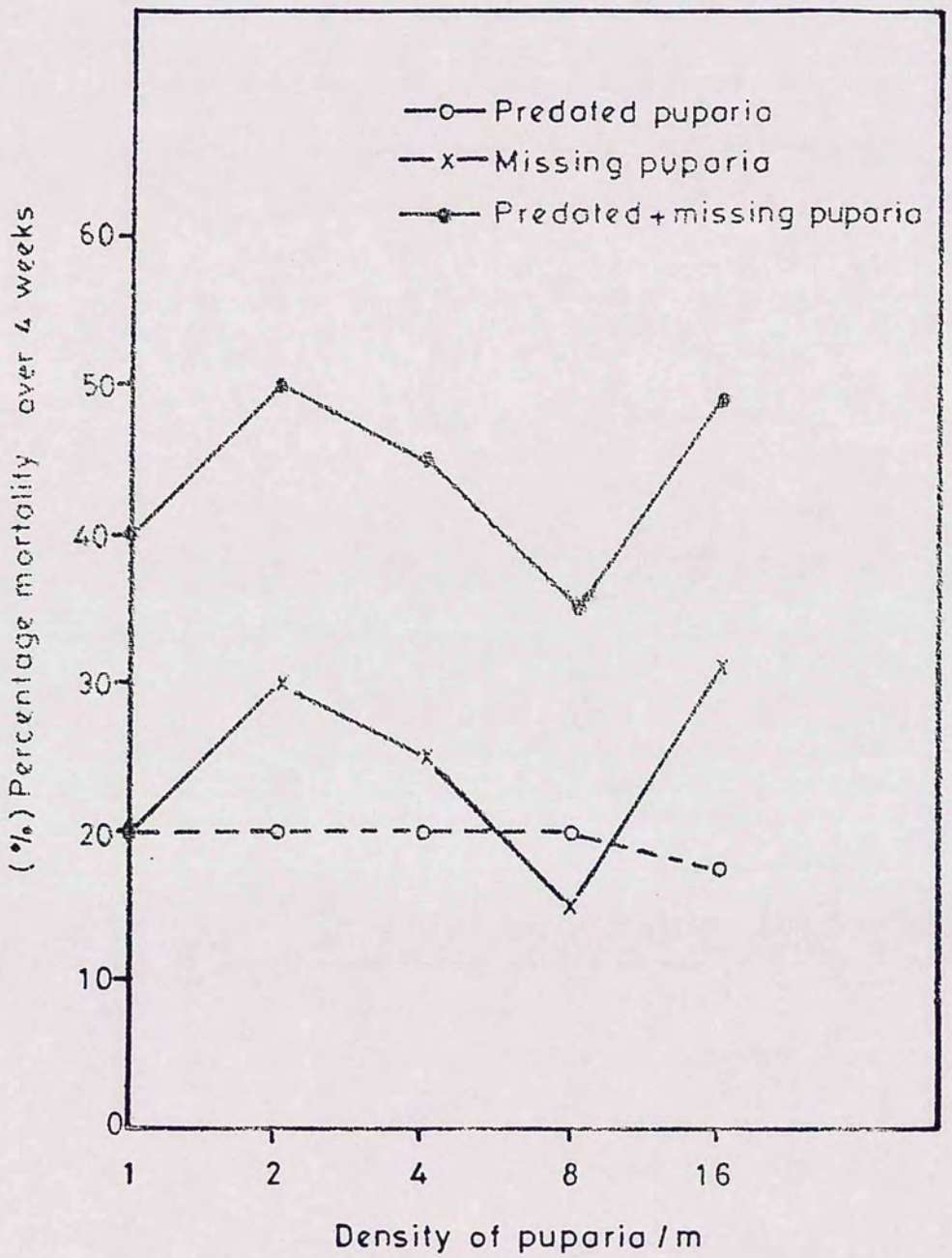
Density	Distance	Mean percent predation (± S.E)		
per meter	between puparia (cm)	No partially eaten	No. Missing	Total predation
1	-	20.0±17.9	20.0±17.9	40.0 ± 15.5
2	100	20.0±12.6	30.0±14.5	50.0 ± 11.1
4	25.0	20.0±8.9	25.0±9.7	45.0 ± 7.9
8	12.5	20.0±6.3	15.0±5.6	35.0 ± 5.3
16	6.25	17.5±4.2	31.3±5.2	49.0 ± 4.0

(b) 5 x 5 Latin square Anova table

Source	Df	SS	MS	F
Sites	4	296.20	74.04	1.78 ns
Months	4	138.12	34.53	0.83 ns
Densities	4	191.76	47.94	1.13 ns
Error	16	665.60	41.60	

ns- not significant at 5 % level

Figure 22: Percent predation in puparia buried at different densities.
(March, 1988 - September, 1988).



be found and were assumed to have been eaten completely; 17.4 % of puparia were partially predated upon while the remaining 49.0 % of puparia were exhumed intact after the exposure period. When these were incubated in the laboratory tsetse fly adults emerged indicating that none of them was parasitized within the time the puparia were buried in the field. The overall mean predation over 4 weeks in which the puparia were buried in the field was 51.0 %.

Latin square analysis of variance of the rate of predation in different sites, months and densities is given in Table 12b. The results indicated that the effects of site (F= 1.78), months (F= 0.83) and density (F= 1.13) were not statistically significant at the 5 % probability level. There was also no significant relationship between the overall predation rate estimates and puparia density (Figure 22). These results indicated that puparia predation was not density dependent.

7.4 DISCUSSION

The predators of puparia caught in larviposition sites of *G.pallidipes* at Nguruman were dominated by formicid ants and gryllids which were found foraging in leaf litter or soil. Among the formicid ants, the genera *Pheidole, Polyrachis, Camponotus, Odontomachus,*

Paltothyreus, *Acantholepis* and *Viticiola* were caught in larviposition sites. This observation is similar to the findings of other researchers in other parts of Africa (Fiske, 1920; Carpenter, 1920; Ford, 1940; Rogers, 1974).

The gryllids caught in larviposition sites of *G.pallidipes* belonged to the genera *Gryllus*, *Gryllulus*, *Homoeogryllus*, *Liogryllus*, *Phaeophillacris* and *Gryllotalpa*. Identical gryllids have been reported in tsetse habitats in other parts of Africa. For example in Burkina Faso, Challier (1971a) noted the presence of large crickets in larviposition sites of *G.palpalis gambiensis* and considered that they were occasional predators of tsetse puparia.

There was significant differences in predation levels of puparia buried at different soil depths. Puparia placed on the soil surface were the most destroyed as compared to those buried in the soil. This observation indicated that the depth at which puparia occurred influenced its risk of predation. Thus puparia found on the soil surface were easily discovered by predators while those buried under the soil were more difficult to discover and therefore less likely to be predated upon.

Observations made in the present studies indicated that puparia predation rate was not density dependent. Thus predation did not increase with increasing puparia

density. This could probably be due to the low puparia densities involved in the experiment. Identical results have been documented for the same tsetse species by Adabie (1987). Adabie (1987) found no density-dependent relationship in puparia predation while using a different experimental design.

CHAPTER EIGHT

GENERAL DISCUSSION

The past and present tsetse control measures have achieved limited successes. This has partly resulted due to the incomplete understanding of the ecology and behaviour of the tsetse species concerned. Over the past few years however, there has been an increasing awareness that sustained control of tsetse and trypanosomiasis is dependent on acquiring a sound knowledge of the vector/trypanosome complex. Progress in the past decade on the development of better control measures of tsetse vectors has been the result of intensive research work on their behaviour and ecology and there is every indication that with a proper understanding of tsetse population dynamics their control is possible.

There has however, been the problem that in most tsetse studies emphasis has been given to the adult population and little or no attention to the puparia. As a result of this bias, there remains lack of knowledge on puparia ecology which is an important aspect in the understanding of tsetse population dynamics. If tsetse control is to achieve its goal, it is vital that proper knowledge of puparia ecology should be obtained on all tsetse species in a given

area. It is in view of this, that studies were carried out to determine the factors affecting the distribution and mortality of *G.pallidipes* puparia at Nguruman.

Puparia mortality is one of the factors that affects the distribution and abundance of tsetse populations. Mortality of puparia may occur as a result of predation, parasitism or through the action of pathogens such as fungal infections. Death can also occur due to extremes of temperature or flooding. At the same time many puparia may be washed away from the sites by torrential rains. These kind of losses would affect the overall distribution and abundance of tsetse puparia. However, in order to sustain their population, adult female tsetse shift their larviposition sites from areas liable to flooding to areas less likely to be flooded.

Predation and parasitism were important mortality factors affecting tsetse puparia in the field. These mortality factors were however not density dependent. The absence of density dependence relationship between puparia loss rate and puparia density could have been due to the fact that most of the puparia predators were polyphagous i.e feeding on any prey which happened to be present when they required a meal. It could have also been due to an evolutionary strategy of the tsetse puparia. Normally the larval stage of tsetse is of very short duration followed by pupation in a tough

puparium. The puparium could have been developed in order to avoid climatic extremes or predation or both. In this tough puparium, the pupa is well protected from predators; Thus, even if the density of puparia is high there would be low predation of tsetse puparia in favour of less protected puparia of other insects.

Fungal infection was found to be the major cause of non-emergence of tsetse puparia collected in the field. Other causes of non-emergence included developmental and emergence failures, pupal tissue degeneration and puparia parasitism. However, none of these causes of non-emergence was density dependent.

Since density dependent mortality was not shown in any of the causes of puparia mortality the question was " At what stage were the mortality factors acting on tsetse population ?". The mortality factors could have acted on the teneral flies immediately after they emerged. This observation was reported by Challier (unpublished data) and by Dransfield *et al.*, (1989). Dransfield *et al.*, (1989) found that mortality occurred at the time of emergence with very small flies being unable to fly or feed properly. Chaudhury (unpubl data) also reported that small flies took longer to spread their wings which made them more susceptible to predation.

What is the relevance of the present studies in the control of tsetse flies ?. The information on

shifting of larviposition sites and seasonal fluctuations in puparia numbers obtained from this study could be relevant in indicating the times, sites, habitats and seasons appropriate for insecticidal or other control measures applications in order to maximise their effects on the proportion of the gravid females entering the sites to larviposit and the emerging teneral flies. For example, in the dry season the flies tended to concentrate in riverine thickets where large numbers of puparia were found. This could be the best time to concentrate control efforts in these habitats in order to achieve maximum control. Since fungal infection was found to be the major cause of non-emergence of tsetse puparia collected in the field, it is possible that if the fungal spores are released from the dead puparia, they may build up in the soil to provide new sources of infection as long as conditions remain favourable to fungal growth. If these spores are applied either singly or integrated with other control measures in larviposition sites they could achieve some control of the larvae and puparia of tsetse.

Since predators were important mortality factors affecting the abundance of tsetse puparia in the field, they could be mass reared and then released at appropriate times in larviposition sites of tsetse. Unfortunately, little progress has been made in

developing expertise in utilising this approach due to mass rearing problems.

SUMMARY

1. A two year study was carried out at Nguruman in the Kajiado district of the Rift valley Province of Kenya to investigate the factors affecting the puparia distribution and mortality in a natural population of *G.pallidipes* Austen.

2. The study area supports an indeginous Maasai population with their cattle, goats, sheep and donkeys. There are a few irrigated farm schemes producing maize, a variety of fruits and vegetables. The area is also rich in game animals, but the presence of large numbers of *G.pallidipes*, *G.longipennis* and other biting insects has rendered the greater part of the area inhospitable for high grade cattle ranching. The climate of the area is divided into two wet and two dry seasons.

3. Trapping of adult tsetse resulted in a rapid decline in the numbers of live puparia in an area subjected to tsetse population suppression.

4. There was a significant positive correlation ($r = 0.7$, $P<0.01$) between adult female density and puparia density within the suppression area suggesting that the abundance of puparia was to a large extent dependent on the density of reproductive female flies.

5. There was a decline in puparia numbers in the riverine thickets and an increase in the valley woodland following heavy rains, suggesting a seasonal shift in sites within the same habitat. This seasonal shift probably evolved to avoid enviromental and climatic catastropes experienced by adult females during the rainy season

6. The mean weight of ecloded puparia of *G.pallidipes* was 33.30 $\pm$ 0.6 mg. Puparia weighing less than 28.3 mg were found when they had died. However, there were wide variations in the weight of dead puparia (range 3.0-40.0 mg)

7. Parasitised puparia could not be distinguished from live puparia on the basis of weight, although on average the dead puparia were lighter than live puparia.

8. There were significant differences ($t = 13.2$, $P<0.01$) in the mean weight of puparia collected in different seasons at Nguruman. Puparia collected during the rainy season weighed more (34.40 mg) than those collected in the dry season (32.80 mg).

9. There was a significant correlation ($r= 0.7$, $P<0.01$) between puparia weight and relative humidity in the

previous month indicating that this was probably the vital climatic factor experienced by the parent female flies.

10. Adult tsetse emergence from live puparia at the time of collection ranged from 16.7-100.0 % and averaged 58.5 $\pm$ 0.6. The emergence rate was higher during the dry than during the rainy season. This was probably because during the dry season high temperatures shorten the pupal period producing a rise in emergence rate.

11. The diurnal pattern of emergence of tsetse and parasites from field collected puparia showed a bimodal pattern with a major peak occurring in the morning between 0600 - 1800 hours and a minor peak occurring in the evening between 1500 -2100 hours.

12. Puparia of *G.pallidipes* were parasitised by two species of parasites namely *Exhyalanthrax lugens* Lw and *E.beckerianus* Bezzi. Mortality caused by these parasites fluctuated seasonally and was highest (35-100 %) at the start of short rains.

13. Natural puparia mortalities were caused mainly by developmental and emergence failures, pupal tissue degeneration, puparia parasitism and fungal infections.

14. Regression analysis of the k-values of each submortality against the total mortality showed that fungal infection was the most significant factor ($r = 0.55$, $P<0.05$) causing puparia mortality in the field.

15. Puparia of *G.pallidipes* survived 6 hours of submersion in water but were killed when submersion exceeded 9 hours.

16. The relative abundance and distribution of the predators of puparia were determined using pitfall traps. The predators belonged to various genera of the families Formicidae and Gryllidae respectively.

17. There was a significant difference ($F= 15.7$, $P<0.05$) in the rate of predation of puparia buried at different soil depths. Puparia placed on soil surface were the most destroyed as compared to those buried in the soil.

18. The levels of predation obtained in the experiments involving burying of puparia at different densities were not density-dependent.

Appendix 1: Climatic data at Nguruman during the period of study (September, 1986- October, 1988).

Year	Month	Ambient temperature(^{0}C) Min	Max	Mean	Mean % Rel. humidity	Total rainfall(mm)
1986	Sept.	18.9	34.7	26.8	45.0	4.3
	Oct.	20.5	37.1	28.8	45.0	3.1
	Nov.	20.1	33.6	26.9	57.0	40.5
	Dec.	20.0	34.2	27.1	54.0	61.3
1987	Jan.	19.6	34.6	27.1	57.0	102.0
	Feb.	19.4	36.6	28.0	53.0	37.8
	Mar.	21.1	37.8	29.5	50.0	41.3
	Apr.	20.9	35.1	28.0	61.0	136.2
	May	20.5	33.1	26.8	68.4	47.8
	June	18.8	33.5	26.1	63.0	37.8
	July	18.5	34.3	26.4	56.0	12.2
	Aug.	19.6	34.5	27.1	53.0	0.2
	Sept.	20.4	37.2	28.8	46.0	5.8
	Oct.	22.1	37.9	30.0	45.0	22.0
	Nov.	21.3	35.6	28.4	58.0	63.4
	Dec.	20.4	38.1	29.3	46.0	9.7
1988	Jan.	21.0	36.5	28.8	55.0	78.2
	Feb.	21.8	38.5	30.1	45.0	5.6
	Mar.	21.8	36.1	29.0	60.0	95.1
	Apr.	21.3	32.7	27.0	77.0	253.8
	May	20.3	33.1	26.7	69.0	29.0
	June	19.2	33.1	26.2	62.0	32.1
	July	18.2	33.4	25.8	57.0	0.6
	Aug.	19.3	33.7	26.5	58.0	18.2
	Sept.	20.3	35.5	28.0	49.0	4.0
	Oct.	21.3	37.7	29.5	44.0	5.4

Appendix 2i: Ambient temperatures in larviposition sites of *G.pallidipes* at Nguruman (September, 1986-October, 1988).

Year	Month	Temperature (^{0}C) Min	Max
1986	Sept.	18.4 ± 0.2	33.2 ± 0.3
	Oct.	18.4 ± 0.2	34.3 ± 0.4
	Nov.	20.6 ± 0.3	30.4 ± 0.5
	Dec.	18.8 ± 0.3	28.1 ± 0.9
1987	Jan.	19.3 ± 0.3	33.2 ± 1.2
	Feb.	18.6 ± 0.6	31.8 ± 1.0
	Mar.	18.9 ± 0.4	30.3 ± 0.9
	Apr.	20.4 ± 0.3	34.4 ± 1.7
	May	19.5 ± 0.2	28.2 ± 0.7
	June	18.0 ± 0.4	30.9 ± 0.5
	July	16.1 ± 0.7	30.5 ± 1.0
	Aug.	19.2 ± 0.6	31.2 ± 0.7
	Sept.	18.5 ± 0.7	35.1 ± 0.7
	Oct.	20.9 ± 0.8	34.7 ± 0.7
	Nov.	20.3 ± 0.1	30.9 ± 0.8
	Dec.	17.7 ± 0.3	36.4 ± 0.5
1988	Jan.	19.3 ± 0.3	33.2 ± 1.1
	Feb.	19.4 ± 0.4	33.4 ± 1.0
	Mar.	19.8 ± 0.3	33.6 ± 1.2
	Apr.	20.2 ± 0.3	28.7 ± 0.9
	May	20.4 ± 0.2	30.3 ± 0.5
	June	19.1 ± 0.4	31.3 ± 0.3
	July	15.2 ± 1.0	30.7 ± 0.3
	Aug.	18.1 ± 0.5	28.1 ± 1.0
	Sept.	15.8 ± 0.3	30.5 ± 0.3
	Oct.	20.2 ± 0.4	33.3 ± 0.7

Appendix 2ii: Soil temperatures in larviposition sites of *G.pallidipes* at Nguruman, Kenya.

Year	Month	Soil temperature 2cm deep Minimum	Soil temperature 2cm deep Maximum	Soil temperature 4cm deep Minimum	Soil temperature 4cm deep Maximum
1986	Sept.	21.3±0.4	27.8±0.5	21.8±0.2	25.8±0.5
	Oct.	21.8±0.4	29.2±0.6	21.8±0.2	25.7±0.9
	Nov.	23.2±0.2	26.7±0.7	23.1±0.2	25.5±0.8
	Dec.	21.7±0.3	24.3±0.4	21.6±0.2	23.1±0.3
1987	Jan.	22.2±0.3	26.3±0.8	23.2±0.4	25.2±0.4
	Feb.	21.2±0.3	25.3±0.3	22.0±0.4	24.2±0.3
	Mar.	21.8±0.3	25.4±0.4	22.3±0.4	24.7±0.3
	Apr.	23.4±0.4	27.1±0.9	23.6±0.4	26.9±0.9
	May	21.9±0.2	24.1±0.2	22.6±0.4	24.5±0.5
	June	20.7±0.4	24.9±0.6	21.9±0.5	24.0±0.5
	July	19.3±0.4	24.8±0.5	20.3±0.4	23.0±0.4
	Aug.	22.2±0.3	26.6±0.5	23.0±0.4	25.5±0.5
	Sept.	22.2±0.3	28.1±1.2	23.3±0.3	26.8±0.8
	Oct.	24.4±0.4	28.7±0.7	25.1±0.5	27.4±0.5
	Nov.	22.8±0.2	25.8±0.6	23.7±0.4	25.6±0.6
	Dec.	23.7±0.6	27.4±0.8	22.7±0.3	27.1±0.7
1988	Jan.	22.2±0.3	26.3±0.8	23.2±0.4	25.2±0.4
	Feb.	21.0±0.3	25.3±0.3	23.9±0.4	27.6±0.3
	Mar.	23.4±0.6	27.7±1.3	24.2±0.5	26.7±0.6
	Apr.	23.2±0.3	26.6±1.2	23.6±0.5	25.7±0.7
	May	22.8±0.2	26.3±0.7	24.0±0.4	26.5±0.7
	June	21.4±0.2	25.4±0.5	22.5±0.3	24.8±0.8
	July	19.1±0.5	24.2±0.7	20.5±0.5	23.8±0.8
	Aug.	21.6±0.2	25.0±1.0	21.6±0.3	23.9±0.7
	Sept.	21.1±0.4	26.4±0.9	21.6±0.5	25.0±0.9
	Oct.	23.5±0.2	27.6±0.8	23.5±0.4	26.3±0.9

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