

The temporal distribution and relative abundance of stable flies (*Stomoxys calcitrans*) (Diptera: Muscidae) in a feedlot near Heidelberg, Gauteng, South Africa

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ABSTRACT

The stable fly, *Stomoxys calcitrans* (L.) is a haematophagous fly that feeds primarily on the forelegs of cattle stimulating a range of avoidance behaviours in confinement situations such as feedlots. From literature it is apparent that stable flies associated with feedlots have a significant impact on cattle especially with regard to economic parameters such as a decline in feed intake and thus a lower average daily gain resulting in less meat production due to irritation caused by painful bites. The abundance of the stable flies was studied in a large commercial feedlot near Heidelberg from October 2012 to September 2013. Two tsetse fly traps, namely the NZI and the Vavoua fly traps, were used and evaluated in determining the seasonal abundance of the stable flies. The tsetse traps proved to be most effective for sampling stable flies compared to other designs. The NZI and Vavoua tsetse type trap were compared and although there were no significant difference the NZI trap proved to be more reliable and user friendly for this study. Stable flies were more abundant from late December with a peak in numbers late in January through February and became less abundant from early March. Minimum to no fly abundance occurred in the winter months from May to June 2013. The data indicated a strong edge effect for the stable flies, the flies were more abundant in pens and corridors that were surrounded by vegetation, manure run off and holding ponds. The numbers collected in traps were correlated with stable fly counts on the cattle to be used in calculating a future threshold in chemical control. Preliminary observations on the influence of temperature, wind speed and rainfall were also made. This research will form part of a larger project to determine an integrated fly management program for the feedlot.

Key words: *Stomoxys calcitrans*, stable flies, feedlots, meat production, seasonal abundance, South Africa

UITTREKSEL

Die stalvlieg, *Stomoxys calcitrans* (L.), is 'n bloedvoedende vlieg wat hoofsaaklik op die voorbene van beeste voed. Dit stimuleer verskeie vorme van vermydingsgedrag in die beeste, veral in voerkrale. Volgens die literatuur is dit duidelik dat stalvlieë in voerkrale 'n beduidende impak op beeste het, veral met betrekking tot die ekonomiese parameters soos 'n daling in voedselinname en dus 'n laer gemiddelde daaglikse gewigstoename. Dit lei tot laer vleisproduksie as gevolg van irritasie wat deur die vlieë se pynlike byt veroorsaak word. Die volopheid en verspreiding van die stalvlieë is vanaf Oktober 2012 tot September 2013 gemeet in 'n groot kommersiële voerkraal naby Heidelberg, Gauteng. Twee tsetsevliegvalle, naamlik die NZI- en die Vavoua-valle, is gebruik en geëvalueer in die bepaling van die seisoenale voorkoms van die stalvlieg. Die tsetsevalle is baie doeltreffend vir die vang van stalvlieë in vergelyking met ander modelle. Die Vavoua- en NZI-val is met mekaar vergelyk. Hoewel daar geen betekenisvolle verskille was nie, was die NZI-val as die mees betroubaar en verbruikersvriendelik van die twee beskou vir hierdie studie. Stalvlieë was meer aktief vanaf laat Desember met 'n hoogtepunt in getalle laat in Januarie tot in die middel van Februarie. Die getalle van die vlieë het gedaal vanaf Maart. Daar was baie min tot geen vlieë in die wintermaande van Mei tot Junie 2013 nie. 'n Sterk korrelasie is gevind tussen die vliegaktiwiteit en kanteffekte. Die vlieë was meer volop in krale wat deur plantegroei, misafvoer en misdamme omring is. Die stalvliegvangste is met stalvliegtellings op die beeste se voorpote gekorreleer sodat die drempelwaarde bereken kon word om chemiese beheer dienooreenkomstig aan te pas. Voorlopige kommentaar oor die invloed van temperatuur en reënval word ook gelewer. Hierdie navorsing sal deel vorm van 'n groter projek om 'n geïntegreerde vliegbeheerprogram vir die voerkraal te bepaal.

Sleutelwoorde: *Stomoxys calcitrans*, stalvlieë, voerkrale, vleisproduksie, seisoenale voorkoms, Suid Afrika.

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CHAPTER 1

INTRODUCTION

1.1 BACKGROUND INFORMATION

1.1.1 The importance of nuisance and biting flies

Contemporary studies have determined that biting flies are disruptive to cattle and have negative impacts on the welfare of livestock (Dougherty *et al.*, 1995). *Stomoxys calcitrans* (stable flies) (Diptera: Muscidae) are particularly associated with livestock facilities globally and are considered to be the most important pest of confined cattle (Morgan *et al.*, 1983; Kunz *et al.*, 1991; Thomas, 1993; Holdsworth *et al.*, 2006; Muenwon *et al.*, 2010). Both female and male stable flies are blood feeding and can act as mechanical and biological vectors of disease (Holdsworth *et al.*, 2006). They are the intermediate host of *Setaria cervi* (Nematode) and are also excellent mechanical vectors of blood dwelling pathogens including *Trypanosoma evansi* in a number of hosts (Holdsworth *et al.*, 2006) as well as Lumpy Skin Disease Virus and *Anaplasma* spp, both important pathogens for feedlot cattle in South Africa (Coetzer, 2004). *Stomoxys calcitrans* has a significant economic impact on feedlot cattle in a number of ways, in an attempt to avoid the painful bites, cattle will stomp their feet, throw their heads, twitch their skin, swish their tails and bunch together (Berry *et al.*, 1983; Wieman *et al.*, 1992; Dougherty *et al.*, 1993; 1994; Mullens *et al.*, 2006). The energy spent on this avoidance behaviour of cattle has an impact on their weight gain (Campbell *et al.*, 1977; 1987). Feed efficiency can be depressed by high levels of stable flies, however, cattle become desensitized to high numbers of stable fly bites after which weightloss is not affected to the same extent (Campbell *et al.*, 1987). Changes in vital signs, behaviour and nitrogen balance (retention) is likely to contribute to reduced rates of weight gain observed in cattle owing to stable fly infestation (Schwinghammer *et al.*, 1986). It is estimated that stable flies can cause more than \$1 billion in losses in the United States because of their irritating impact on confined cattle (Kneeland *et al.*, 2012; Taylor *et al.*, 2012).

Several estimates of the economic impacts of stable flies on feedlot cattle have been published since 1992 (Wieman *et al.*, 1992; Campbell *et al.*, 1993; Catangui *et al.*, 1993; 1995; 1997). Catangui *et al.* (1997) introduced an economic injury level for stable flies in feedlots. Factors included as variables in the economic injury level calculation are the market value of beef and the cost of controlling stable flies. Refinements yet to be included in future calculation are cattle breed, age, nutrition and efficacy of the stable fly management used (sanitation, insecticides, biological control) (Catangui *et al.*, 1997).

Other diptera species commonly associated with livestock are biting flies such as *Haematobia* spp., tabanids (horseflies) such as *Hybomitra sonomensis* and *Tabanus punctifer* can significantly reduce daily weight gain in yearling heifers and feedlot cattle (Foil & Hogsette, 1994; Jones & Anthony, 1964). Tabanids have been described as the vectors of more than 35 pathogenic agents of livestock, including equine infectious anaemia viruses (Foil & Hogsette, 1994).

Nuisance flies are listed as *Musca* spp., *Fannia* spp. and *Muscina* spp. (Holdsworth *et al.*, 2006). The nuisance fly most commonly associated with intensive animal facilities is *Musca domestica* and preferentially congregates in areas containing animals and feed (Meyer & Peterson, 1983; Miller *et al.*, 1993). Cattle will respond to increases of *M. domestica* abundance with more frequent defensive movements while flies prefer moist locations on animals resulting in mainly tail swishes, ear flicks and head tosses (Urech *et al.*, 2012). Movement of *Musca* species between faeces and food makes them ideal vectors of human and animal pathogens; for instance, *Musca autumnalis*, the face fly, transmits the nematode *Thelazia* spp. to the eyes of cattle (Holdsworth *et al.*, 2006).

1.1.2 Integrated fly management

The best way to address biting fly control is an integrated approach (Urech *et al.*, 2011). Good management and sanitation should always be the first and most important component of an integrated approach to fly control in feedlots (Morgan *et al.*, 1983; Kunz *et al.*, 1991; Thomas & Skoda, 1993; Thomas *et al.*, 1996). Relying on insecticides alone could prove to be expensive as proved by a study conducted on the control of houseflies and stable flies on dairy farms (Lazarus *et al.*, 1989). An integrated fly management program incorporates not only the use of chemicals and pesticides but also sanitation, manure management, animal management, facility design and biological control (Urech *et al.*, 2011). The advantages of an integrated fly management program are the reduced detrimental impact on the environment and natural enemies (predators and parasites), reduced impact on neighbours and recreational outdoor activity (Newson, 1977), reduction in production costs due to reduced insecticide usage, lower risk of insecticide resistance in target species and improved cattle welfare (Cilek & Greene, 1994; Marçon *et al.*, 1997; Memmi, 2010). The need for and success of integrated pest management components are determined by effective monitoring fly populations (Urech *et al.*, 2004; 2011).

At Karan Beef, a commercial feedlot in South Africa, chemical control of stable flies is currently based on presumed population densities. For the development of an integrated fly management program for a feedlot in South Africa a monitoring system should be in place. Studies on the impact of stable flies on feedlot cattle in South Africa is limited. The only work

found on stable flies in South Africa was conducted by Sutherland (1973), on the suitability of various types of dung as a larval breeding media for *Stomoxys calcitrans*, the effects of temperature on the adults, eggs and pupae (Sutherland, 1979) and the temperature preferences of the motile stages of *S. calcitrans* (Sutherland, 1980). Research on an integrated fly management program is therefore needed in South Africa.

1.1.3 Seasonal abundance of stable flies

Stable flies can complete their life cycle in approximately three weeks during the summer months (Campbell *et al.*, 1987; 2001; Johnson, 2011). As temperatures increase toward spring and summer the larvae migrate to the soil surface to pupate, after which the adult fly emerges (Campbell *et al.*, 1987; 2001; Johnson, 2011). The female stable fly lays up to 80 eggs at a time in organic material which serves as food for the developing larvae that develop through three larval instars. The adult stable fly can live up to six weeks (Campbell *et al.*, 1987; 2001; Johnson, 2011). The sub-adult population is usually also much bigger than the adult population. The duration of the adult stages are much longer than that of the sub-adult stages (up to 120 days) (Holdsworth *et al.*, 2006).

Stable flies have a seasonal occurrence with as many as three peaks in summer, these bimodal or trimodal peaks in abundance have been documented in the Midwest of the USA (Hall *et al.*, 1983; Black & Krasfur, 1985; Scholl, 1986). These peaks in abundance are presumably influenced by seasonal humidity and temperature conditions (Mullens & Meyer, 1987). The weekly variations in stable fly abundance in a study conducted in Alberta, Canada, by Lysyk (1993) showed four abundance peaks which were attributed to the emergence of an initial overwintering generation, followed by an additional three generations. Cruz-Vazquez *et al.* (2004) found two population abundance peaks in a semi-arid area in Mexico. Semakula *et al.* (1989) found that temperature affects the activity threshold of stable flies and that adult fly activity ceases completely at temperatures above 40 °C and below 11 °C. According to Urech *et al.* (2012), stable fly populations were higher during autumn and spring than in summer. Summer temperatures might have been too high in Queensland, Australia, as temperatures increased to higher than 30 °C and stable flies' maximum fecundity was at 25 °C (Lysyk, 1998). Stable flies were more active between 10:00 and 16:00 with very little activity between dusk and dawn (Catangui, 1997; Campbell *et al.*, 2001; Anthony, 2005). In Southeastern Nebraska, stable fly activity peaked at 14:00. During this time of the day the highest average stable fly counts per cattle forelegs occurred and the highest average number of flies were present in traps. The time of day can influence adult stable fly population estimates significantly (Thomas *et al.*, 1989).

Stable flies do not go into diapause (Greene *et al.*, 1989) nor do they have a freeze tolerance in their life cycle (Beerwinkle *et al.*, 1978; Jones & Kunz, 1997). Stable flies overwinter in their third larval stage. This stage can be prolonged up to 120 days under unfavourable conditions (Scholl, *et al.*, 1981; Berry *et al.*, 1978). Fresh corn silage and hay feeding sites are preferred over wintering sites for immature stable flies (Taylor & Berkebile, 2011; Scholl *et al.*, 1981). Some studies show that adult stable flies are active at low levels throughout winter (Berkebile *et al.*, 1994).

Degree days is a measurement of heat units over time, also known as “growing degree days” (GDD) to differentiate this value from “heating degree days” or “cooling degree days,” which are used to estimate energy demand. Accumulated degree days, or ADD, is a measurement of thermal units required for growth and development of an arthropod, based on 24 hour periods of time. Every insect requires a specific amount of heat accumulated to reach a certain life stage such as adult flight (Herms, 2004). Because arthropods are ectothermic, their development is influenced by the surrounding temperature. Every species requires a specific optimal temperature range for development to occur. When the temperature becomes too extreme, development will slow down and stop (Murray, 2008; Kowalsick & Clark, 2006; Herms, 2004).

In a study by Taylor & Berkebile (2011) the earliest stable fly emergence from hay feeding sites for cattle occurred at 235DD and decline in productivity at 900DD. Immature survival was highest at 20-25 °C and survival decreased below 15 °C and above 35 °C. Developmental time decreased from 71DD at 15 °C to 13DD at 30 °C. The developmental threshold for *S. calcitrans* is 12.2 °C. The weekly rate of change in stable fly populations was influenced by temperature and accumulated degree days above 10 °C (Taylor & Berkebile, 2011; Beresford & Sutcliffe, 2012; Krafur *et al.*, 1994; Lysyk, 1993). Correlations were found between stable fly emergence levels from hay feeding sites of cattle and temperature and rainfall (Berkebile *et al.*, 1994). The previous year plays an important role in the development of stable flies because of their biological interactions with temperature and moisture. However, the reduction in a stable fly population at certain times of the year could probably also be caused by endogenous factors, most likely the changes in the physical characteristics and bacterial communities in the breeding medium as a result of decomposition (Taylor & Berkebile, 2011).

1.1.4 Larval development sites

It is important to determine where stable flies breed in feedlots in order to control these flies in all their life stages. Sanitation and good drainage is the single most important method of controlling stable flies (Clymer, 1992). Stable fly larvae need a suitable medium (mixture of manure with decaying materials or silage is a favourable medium) and favourable

environmental conditions such as moisture availability to complete their life cycle (Dawit *et al.*, 2012). Sutherland (1978) reported that dung from swine, horses and cattle were suitable breeding media while larvae failed to survive in pure chicken dung. The most productive larval development sites spilled feed that accounted for 53.1% of the pupae collected in the feedlot and occurred constantly throughout the season (Meyer & Peterson, 1983). Other larval development sites were haylage, fresh and old corn silage, soiled straw bedding and manure along fence lines. Livestock facilities are associated with the late seasonal developmental sites of larvae (Taylor *et al.*, 2007).

1.1.5 Monitoring stable fly populations

For the development of an integrated fly management program, the above mentioned information (seasonal abundance, breeding and resting sites, sanitation practices, etc.) must be gathered by monitoring the stable fly populations using the most reliable and sustainable monitoring system. The most commonly used systems to monitor stable flies are with traps and fly counts on forelegs of cattle (Mullens & Meyer, 1987). Monitoring adult stable fly populations on cattle consists of counting the number of flies feeding on the outside of one foreleg and the inside of the other foreleg for a selected period of time, usually one minute (McNeal & Campbell, 1981; Berry & Campbell, 1985; Gerry *et al.*, 2007). Estimates of fly population levels using leg counts should not be done before 09:00 to 10:00 (Thomas *et al.*, 1989). The economic impacts are related to the number of stable flies on cattle/foreleg/minute (McNeal & Campbell, 1981). The lower foreleg is considered the preferred feeding site of stable flies (Hogsette *et al.*, 1987; Mullens *et al.*, 2006). More stable flies are found on the forelegs of cattle than on the rest of the body. Berry *et al.* (1983) observed that the ratio of flies/foreleg to the rest of the animal is 2.8:1. Foreleg counts are also more convenient and less time consuming than whole body counts (Campbell & Hermanussen, 1971). McNeal & Campbell (1981) used an economic threshold of five stable flies per foreleg per minute, formulated from research conducted on calves (Campbell *et al.*, 1977) and on dairy cattle (Bruce and Decker, 1958). The latest economic threshold for feedlot cattle is a mean of five flies per foreleg (Campbell *et al.*, 1987; 2001). Mullens & Meyer (1987) and Thomas *et al.* (1989) found a strong seasonal correlation between trap collections and leg counts. Urech *et al.* 2012 found a positive correlation between trap collections and leg stomps. These observations show the effectiveness of structured animal observations in estimating fly populations (Warnes & Finlayson, 1987; Mullens *et al.*, 2006) and that it can be used as an early warning system to apply insecticides according to a determined threshold.

The Alsynite fiber glass trap is most commonly used to monitor *S. calcitrans* (Broce, 1988; Mullen & Meyer, 1987; Scholl *et al.*, 1985; Scholl, 1986). The Alsynite trap needs a sticky adhesive and can lead to the samples being damaged which complicates identification thereof.

Another negative aspect of using the Alsynite trap is that environmental conditions can significantly influence trap performance, i.e. a higher number of flies landed on the Alsynite trap side protected from wind (Gersabeck & Merritt, 1983). Guo *et al.* (1998) found that two-thirds of the stable flies collected on the sticky traps were males and the effectiveness of controlling later generations were doubtful, whereas the NZI traps captured older flies (Taylor & Berkebile, 2006). The NZI trap and the Vavoua traps were developed in West-Africa and were widely used as an effective method to collect tsetse flies and other haematophagous flies (Mihok *et al.*, 1995; Mihok, 2002). The NZI tsetse type trap collected similar numbers to the Canopy trap, Vavoua trap and Alsynite cylinder traps, but with differences in relative performance among species or locations (Mihok *et al.*, 2006).

The proximity of the traps to the feedlot has a significant impact on the fly collections. In a monitoring design, the closer the traps are placed to the flies' breeding and feeding location, the more successful and efficient the experimental design will be (Holloway & Phelps, 1991; Abba *et al.*, 2011). According to Guo *et al.* (1998) attention should be focused on where the traps are placed as it is significantly influenced by factors such as vegetation, host availability and immature development sites. Traps as far as 2 km from confined livestock facilities will still catch stable flies in small numbers (Guo *et al.*, 1998). However, this will not give an accurate representation of the stable fly population compared to traps set closer to the animals (Urech *et al.*, 2012; Taylor *et al.*, 2007). Stable flies prefer shaded resting sites (Buschman & Patterson, 1981). Stable fly traps could be attractive sites for resting as some of the traps would provide shade and shelter where they can digest their blood meal (Berry & Campbell, 1985; Berry *et al.*, 1986).

1.2 RESEARCH QUESTION

What are the diversity and abundance, and the temporal and spatial distribution of stable flies in a feedlot in Gauteng Province, South Africa?

1.3 OBJECTIVES

- To develop a sustainable sampling method for *S. calcitrans*.
- To determine the seasonal and daily abundance (temporal distribution) of *S. calcitrans*.
- To determine *S. calcitrans* distribution within the feedlot (spatial distribution).
- To determine *S. calcitrans* density on cattle during the season.
- To correlate trap collections, bunching and fly counts on forelegs of cattle.
- To integrate all the above information into fly management recommendations.

CHAPTER 2

MATERIAL AND METHODS

2.1 STUDY SITE AND DESCRIPTION

Karan Beef, the feedlot selected for this study, (Fig. 2.1) situated on the Vaaldam Road (R549), Heidelberg, Gauteng Province (26° 36' 27" S, 28° 19' 13") (Fig. 2.2) in South Africa is the largest feedlot of its kind in Africa and can accommodate approximately 120 000 head of cattle.



Figure 2.1: Aerial map of the holding ponds (A), neighbouring game reserve, biofiltration wetlands (B), the feedlot (C) and the dung heaps (D).

The feedlot is built on a slope to promote drainage of dung and rain into drains at the lower end of the pens. There are manure holding ponds (A) in the feedlot and biofiltration wetlands (B) in a game reserve and pans associated with the Suikerbosrant River neighbouring the feedlot (C) (Fig.2.1). The dung heaps are approximately 5 km from the feedlot (D) (Fig. 2.1). The feedlot has corridors that are alphabetically numbered A from the bottom of the feedlot to VX at the upper side of the feedlot. Every corridor contains pens of approximately 30 x 70 m. There are 1 000 production pens (Fig. 2.3) each containing 120 to 130 cattle. There are 7 hospitals, one per section, each with dedicated recovery pens in a few hospital corridors per section where cattle that are showing clinical disease are treated appropriately and observed intensely.

Heidelberg (Fig. 2.2) has a mild climate. Summer months, October to March, average a minimum temperature of 17 degrees and a maximum of approximately 28. Winter months, July

and August, have the coldest weather. Winter days can reach 25 degrees but generally the winter months averages a low of 5 degrees and a maximum of 19 degrees. Hail is usually experienced during these thundershowers and snow is almost never found within this area (SouthAfrica.com, 2014)

Locality Map of Study area in South Africa

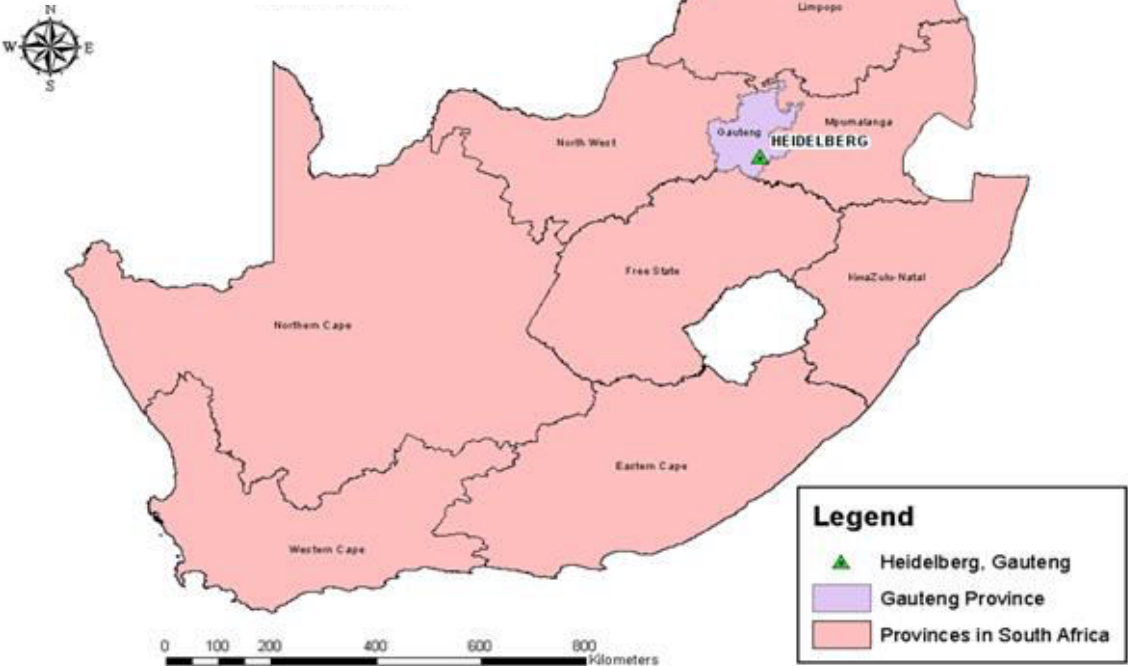


Figure 2.2: Locality of the feedlot used near Heidelberg, Gauteng, South Africa.



Figure 2.3: One of 30x70 m pens containing 120 cattle each. This is number R.

2.1.1 Insecticide treatments

The K-line pens were treated along with the rest of the feedlot as follows:

Insect growth regulator: Dimilin® (diflubenzuron) was applied underneath pen cables and edges of manure ponds (1-2 metres wide) about every 2-3 weeks when insect activity increased. Initial treatment was applied after the first rains in spring to curb the first generation. This was repeated as deemed necessary by subjective evaluation and observation conducted by feedlot management.

Residual surface spray: Deltamethrin 70 g/kg + PBO (Piperonyl Butoxide Synergist) WP, 350 g/kg; This was applied to all exterior walls of troughs and buildings.

Bait/attractant: Fly Bait (10 g/kg Methomyl [Carbamate]) + 1 g/kg Z - 9 Tricosene pheromone attractant. Granules were manually spread on the upper edges of feeding troughs and also in front of the troughs, walls of homes and on buildings. Many applications were applied reactively according to perceptions of fly number increases.

Dip/Spray: Cypermethrin 20%, 200 g / L; *Ad hoc* applied with a fogger, blowing a cloud of dip onto cattle. Direction was determined by wind direction. For most of the summer this was applied to the edges of pens, instead of onto the entire feedlot. A few times applications were made at the homes, blowing a dip cloud against flies that hide in the trees by day.

Concentration of control was on all the edges while middle pens were often skipped. This applied to all the chemicals used.

The H-line was not treated with any chemicals during the study period.

2.2 SAMPLING OF FLY POPULATIONS

2.2.1 Traps

An initial survey was conducted to determine a sustainable sampling method for stable flies. The traps evaluated were blue (Fig. 2.4.a) and green (Fig. 2.4.b) Chinese fly traps® purchased from *The no fly-zone CC*® and Redtops® (Fig. 2.5) distributed by Efekto® and the NZI trap® (Fig. 2.6).



Figure 2.4: A blue (a) and green (b) Chinese trap®.



Figure. 2.5: Redtop® fly trap.

The Vavoua® (Fig. 2.6) and NZI® (Fig. 2.7) tsetse type traps purchased from Vestergaard Frandsen (EA) (Ltd) (Disease Control Textiles) were used to sample the stable flies.



Figure 2.6: The Vavoua trap®.



Figure 2.7: The NZI trap®.



Figure 2.8: Modified 2-litre soda bottle for collecting the flies at the top of the trap.

The collection points at the end of the net at the top of the Vavoua, NZI and H-traps were modified to facilitate the fast and easy collection of the flies without damaging the samples. A two-litre soda bottle was attached to the top of the trap (Fig. 2.8).



Figure 2.9: The H-trap® (Tsetse.org, 2012)

The NZI®, Vavoua® and H-trap® tsetse type traps are especially effective for collecting haematophagous flies such as tabanids and *Stomoxys* spp. (Mihok, 2002; Mihok *et al.*, 1995). The design and colour of the traps resulted in optimal trap performance. Traps were made from appropriate fabrics in the colours produced by either copper phthalocyanine (phthalogen blue),

or its sulphonated forms (turquoise) (Mihok *et al.*, 2006). The Vavoua® trap is directed at 360° and is therefore multidirectional. The NZI® trap is directed in one direction although the colour of the material of the trap will still attract flies from the surroundings.

The H-trap® (Fig. 2.9) is a directional trap from the same company as the NZI® and Vavoua® traps from (Vestergaard Frandsen (EA) Ltd) (Disease Control Textiles). The H-traps will catch flies from two opposing sides.

2.2.2 Fly identifications

Fly trap collections were mounted over time and categorised into morphospecies. These samples were sent to Dr Ashley-Kirk Spriggs, a fly specialist at the National Museum, Bloemfontein, for identification.

2.3 SEASONAL ABUNDANCE AND SPATIAL DISTRIBUTION

2.3.1 Experimental design

A comparison was made to determine the efficiency of the NZI® and the Vavoua trap® types for future monitoring applications. Three replications of one NZI® and one Vavoua trap® were each placed in two lines (Fig. 2.10). Each replication of one NZI® trap and one Vavoua® trap in the K-line was placed 100 m apart. The NZI trap and the Vavoua trap in the K-line was placed 20 m apart. Each replication of one NZI trap and one Vavoua traps in the H-line was placed 20 m apart. The NZI trap and the Vavoua trap in the H-line was placed 4 m apart.

The traps were replicated per site to ensure representative monitoring of fly abundance. The one row was selected at the edge of the feedlot neighbouring an area covered by weeds and subjected to manure run-off. This H-line row of pens had a history of high levels of irritation observed during the previous years (Fig. 2.10). The second row selected was situated in the middle of the feedlot, the K-line (Fig. 2.10) with a history of relative low levels of observed irritation. The H-line and the K-line were approximately 1 km apart. All the traps were weekly sampled for haematophagous flies October 2012 to September 2013. The samples were collected at 10:00 every week to ensure the sampling was done consistently from week to week. The samples collected were sorted into morphospecies, and stable flies were identified and counted. Although these traps can be used to control stable fly populations in smaller feedlots, the use of these traps was not considered to result in destructive sampling because of the vastness of the feedlot used in this study.



Figure 2.10: An aerial map showing two experimental lines (H-line and K-line) and the position of s (V)-Vavoua® and (N)-NZI® trap selected to monitor the flies for the study in the feedlot near Heidelberg.

2.3.2 Daily abundance

The daily abundance of stable flies was determined by sampling the flies collected in the traps every hour. Hourly sampling started at 09:30 and last until 18:30 to 19:30. The bottles were emptied every hour on the hour. This was replicated on three days: 20 February 2013, 21 February 2013 and 5 March 2013.

2.4 FLY SURVEY ON CATTLE

2.4.1 Experimental design

Stomoxys calcitrans were counted on the forelegs of four cattle in each of the 12 pens in the H-line and in 12 pens selected at random in the K-line. A total of 48 cattle (4 per pen for 12 pens) for the K-line and H-line were counted per line per sampling day. The number of flies was counted per foreleg per minute on 14 days throughout the season. Monitoring adult stable fly populations on cattle consisted of counting the number of flies feeding on the outside of one lower foreleg and the inside of the other lower foreleg per minute (McNeal & Campbell, 1981;

Catangui *et al.*, 1997; Gerry *et al.*, 2007). The flies were counted from below the flank of the cattles' leg to the hoof (McNeal & Campbell, 1981; Eicher *et al.*, 2001).

Binoculars were used to count the number flies sitting on the lower forelegs of cattle (Fig. 2.11) and the number of foot stomps per minute were also counted (Catangui *et al.*, 1993; 1995). The counts were recorded on spreadsheets indicating date, time, number of flies counted, eartag number, row and pen number, and additional climate information. Random cattle were selected as far as possible within 10 m from the fence, two from the northern fence and two from the southern fence to ensure that the data was standardised and a realistic representation of the distribution of the cattle in the pen was used (Catangui *et al.*, 1995).

Estimates of fly population levels using leg counts should be done at a time during the day when there is relatively high stable fly activity. Thomas *et al.* (1989) suggest that fly counts should not be done before 09:00 and that the most reliable mean foreleg fly counts occurred at 14:00 when stable fly activity peaked in South-Eastern Nebraska. During this study the flies were collected from the traps every week at approximately 10:00 and then the daily fly counts on cattle were done at approximately 11:00. The traps were then emptied again on the same day after the fly counts had been made at approximately 15:00 to determine the mean daily trap collections per trap to be correlated with the daily cattle counts.



Figure 2.11: Stable flies feeding on cattle forelegs in Nebraska (Boxler, 2013).

2.5 DUNG HEAP IMPACTS

2.5.1 Experimental design

Feedlot dung was collected weekly from the pens and dumped on a large heap (Fig. 2.12) approximately 5 km from the feedlot. To determine the impact of these dung heaps on stable fly numbers into the feedlot, three H-traps® (Fig. 2.9) were placed along a distance gradient from the dung heaps (Fig. 2.13) to the feedlot. Trap number H-1 was placed next to the dung heaps approximately 5 km from the feedlot; trap H-2 was placed approximately 2.5 km from the feedlot next to hay bales; and trap H-3 was placed next to hay bales approximately 1 km from the feedlot. The traps were emptied weekly and the collected were sorted into morphospecies.



Figure 2.12: Dung gathered from pens and dumped on the dung heaps.



Figure 2.13: Aerial map showing location of the feedlot, hay bales, dung heaps and the position of H-traps [H1 (1 km), H2 (2.5 km) and H3 (5 km)] from the feedlot.

2.6 STATISTICAL ANALYSIS

The statistical analysis was conducted with the assistance of Prof Suria Ellis of the Statistical Consultation Service of the North-West University, Potchefstroom campus. The statistical program used was Statistica 64 Version 12.

Repeated measures ANOVA were used to determine statistically significant differences in the following:

- between numbers collected in the NZI and Vavoua trap,
- between numbers collected in the H-line and K-line,
- between the differences in the number of flies on forelegs in the H-line and K-line for edge effect,

- between flies counted on cattle in different pens in the H-line to determine edge effect - the pens at the edges of the corridor were pens 1-2 and 11-12 (group 1) and pen 3-4 and 7-8 (Group 2). Pens in the middle of the corridor were 5-8 (group 3).

Regression and correlation statistics were used to determine:

- climatic effects on trap collections,
- climatic effects on flies counted on forelegs,
- the correlation between trap collections and flies on forelegs,
- the correlation between flies on forelegs and foot stomps.

2.7 CLIMATOLOGICAL DATA

Extensive climate data were made available by dr Dirk Verwoerd that he had obtained from the local weather recording equipment on Karan Beef Feedlot.

Temperature, wind speed, wind chill, wind direction and barometric pressure was recorded at 07:00, 12:00, 17:00.

The weekly mean temperatures, rainfall and wind speed were calculated to correlate with weekly collections.

The daily weather data were correlated with the daily activity of the flies as represented by the number of flies observed on cattle forelegs per minute.

CHAPTER 3

RESULTS AND DISCUSSION

3.1 DIVERSITY OF NUISANCE FLIES IN THE FEEDLOT

In order to develop the most suitable sampling method for *S. calcitrans* the most abundant fly species caught in a green and blue Chinese trap, Redtop trap and one NZI trap during a preliminary study were identified. These flies were superficially allocated to 9 morphospecies. These were:

Morphospecies 1: Tabanidae. A tabanid species was found belonging to the genus *Haematopota*. The tabanids are haematophagous horse flies and cause cattle irritation when occurring in sufficient numbers (Foil & Hogsette, 1994).

Morphospecies 3: Muscidae, *Stomoxys calcitrans* (Linnaeus, 1758). *Stomoxys*, or stable flies, are haematophagous and were most likely to be the species responsible for cattle irritation in the feedlot (Berry *et al.*, 1983; Wieman *et al.*, 1992; Dougherty *et al.*, 1993; 1994; Mullens *et al.*, 2006).

Morphospecies 4: Muscidae, genus *Musca*. These species are nuisance flies and cause irritation in the eye and head regions. However, they are not haematophagous with biting irritation but they may transmit eye diseases (Holdsworth *et al.*, 2006).

Morphospecies 5: Scatopsidae. The larvae of Scatopsidae breed in decaying organic material, including animal dung. The adults appear to feed on nectar from flowers and this family may be ruled out as potential agents of irritation to cattle or other livestock (Coffey, 1966).

Morphospecies 6: Bibionidae, genus *Plecia*. Bibionidae larvae are herbivorous and feed predominately on decaying plant material; adults also feed on flowers and may be ruled out as nuisance flies (D'arcy-Burt & Blackshaw, 1991).

Morphospecies 7: Chironomidae. Larvae are aquatic and adults are non-biting midges. Although adults may occur in vast numbers following mass emergence events, and can be a nuisance, they are unlikely to cause serious irritation to cattle (Armitage *et al.*, 1995).

Morphospecies 11: Muscidae, genus *Atherigona*. This genus is specialised shoot-flies of grasses and cereals (Poacea). The larvae bore into the growing stem of grasses and cereals causing a condition known as "deadheart" and are therefore important agricultural pests in Africa and Asia. The adults feed on various liquids, but are not haematophagous and may be ruled out as nuisance flies (Skidmore, 1985).

Morphospecies 19: Milichiidae. They have a very varied biology, but the many non-specialised species have larvae that develop in various kinds of decomposing organic matter. Adults are not

haematophagous and are unlikely to occur in sufficient numbers to be regarded as causing irritation (Ferrar, 1987).

Morphospecies 42: Sphaeroceridae. This family consists of the lesser dung flies, and larvae of all species in this family develop in animal waste and other decomposing organic matter. These flies often occur in large swarms around dung under pen cables and may cause irritation. However, they are not haematophagous and may be ruled out as major agents of cattle irritation (Buck, 1996).

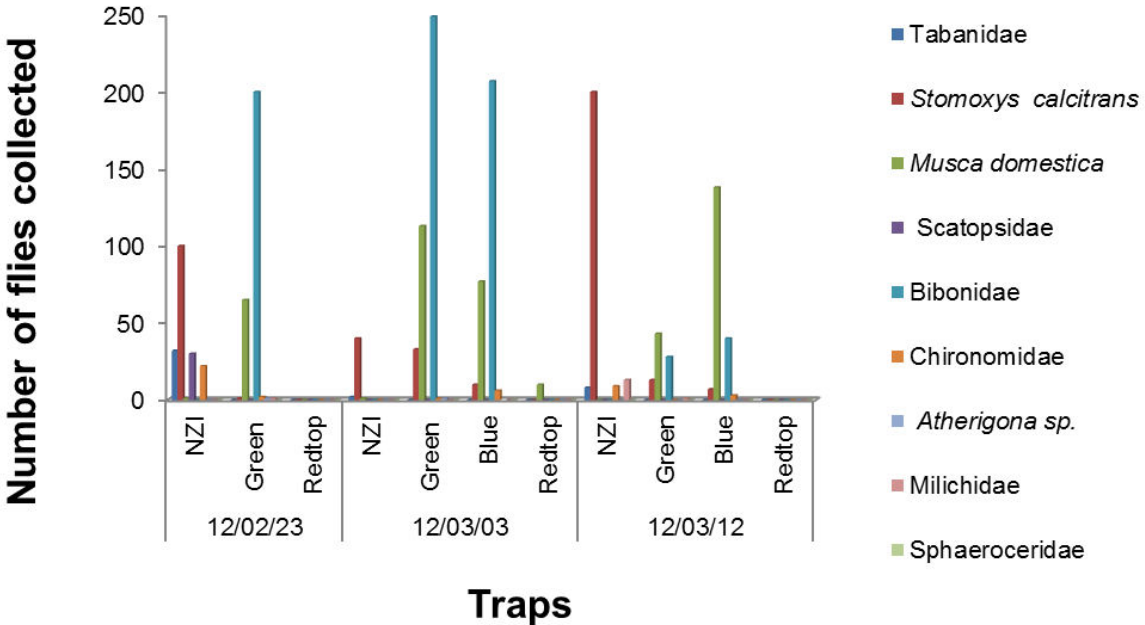


Figure 3.1: Diversity of flies collected in a NZI tsetse type trap, a green and blue Chinese trap and a redtop trap at Karan Beef on three days between 23 February 2012 and 12 March 2012.

The NZI trap showed to be the most effective trap for the collection of important biting flies (Appendix 1). An additional survey conducted using only the NZI trap from February 2012 to May 2012 and 23 collections were made, (Appendix 2) shows *S. calcitrans* to be the most abundant and important haematophagous fly species in the study area (Fig.3.2). The tabanids were the second most abundant haematophagous species. These results indicate that the NZI is an effective trap for collecting heamatophagous flies. *Stomoxys calcitrans* was the most important biting fly because of the high numbers collected and their proven irritation level due to their biting activity. These findings are supported in literature which indicates that *S. calcitrans* may have an economically important impact on feedlot cattle in our study (Morgan *et al.*, 1983; Kunz *et al.*, 1991; Thomas & Skoda, 1993; Holdsworth *et al.*, 2006; Muenworn *et al.*, 2010).

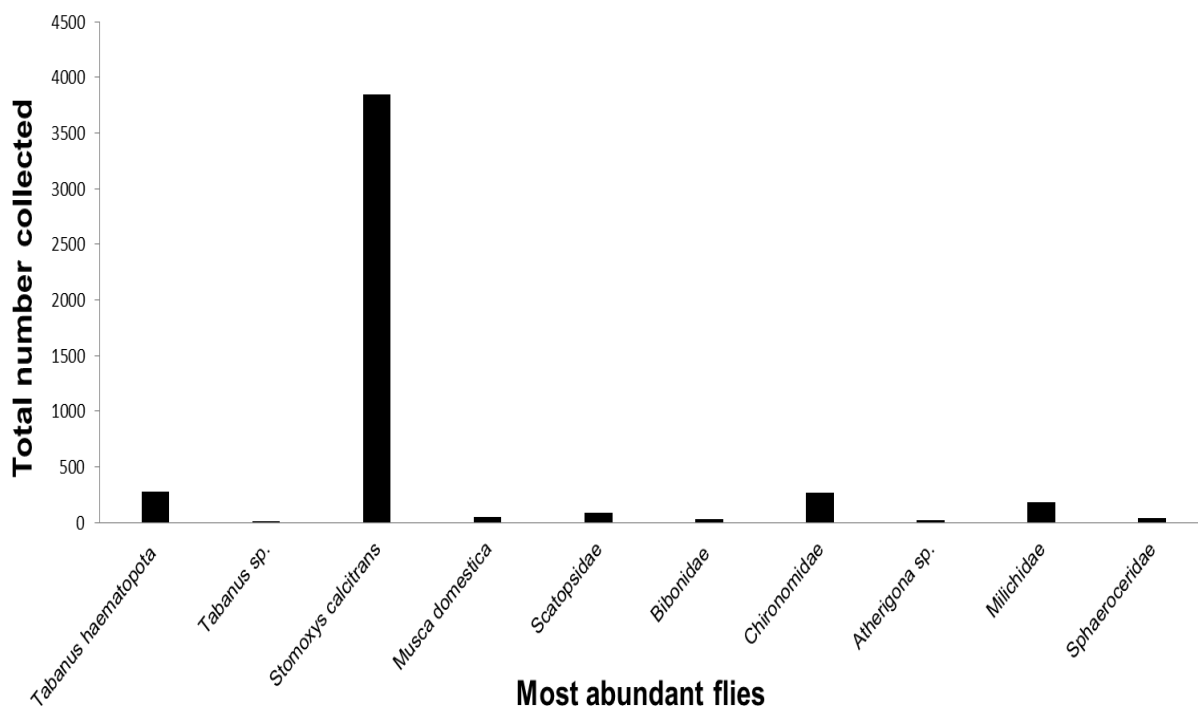


Figure 3.2: Total number of the most abundant flies collected in a NZI trap from February 2012 to May 2012.

The conclusion may be drawn that *S. calcitrans* (stable flies) is the most important biting fly in the feedlot with the greatest potential in causing irritation to the cattle. The NZI tsetse type traps were the most suitable trap type for collecting stable flies and to be used for further sampling (Mihok, 2002; Mihok *et al.*, 2006; Taylor & Berkebile, 2006).

3.2 SAMPLING METHODOLOGY OF HAEMATOPHAGOUS FLIES

The mean number of stable flies collected per week for the period 19 October 2012 to 14 June 2013, in 6 NZI and 6 Vavoua tsetse type traps were compared (Fig.3.3). The total number of flies collected by the NZI traps was 19 212 and the Vavoua traps 11 370 (Appendix 3). During one of the January peaks, on 18 January 2013, the NZI traps collected a mean of 275 stable flies per trap and the Vavoua traps 175. The NZI traps proved to be more efficient in collecting larger numbers of stable flies than the Vavoua traps. However, both traps were efficient in identifying stable fly population peaks and trends. A statistical analysis of the weekly number of flies collected between 19 October 2012 to 14 June 2013 done by repeated measures ANOVA, indicated that there was no significant statistical difference in the efficiency of the NZI and

Vavoua traps in collecting stable flies [F=1.0812, P=0.357]. The NZI traps proved to be more user friendly and reliable for monitoring purposes in this study and is found more efficient in other studies (Mihok, 2002; Mihok *et. al.*, 2006).

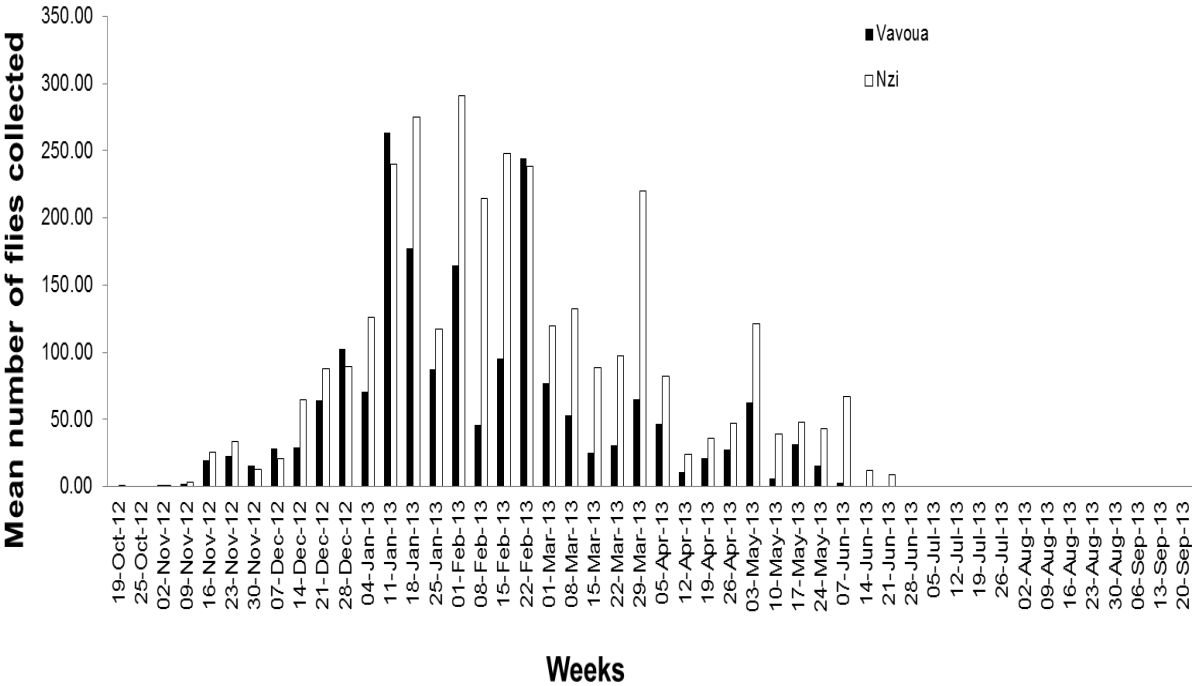


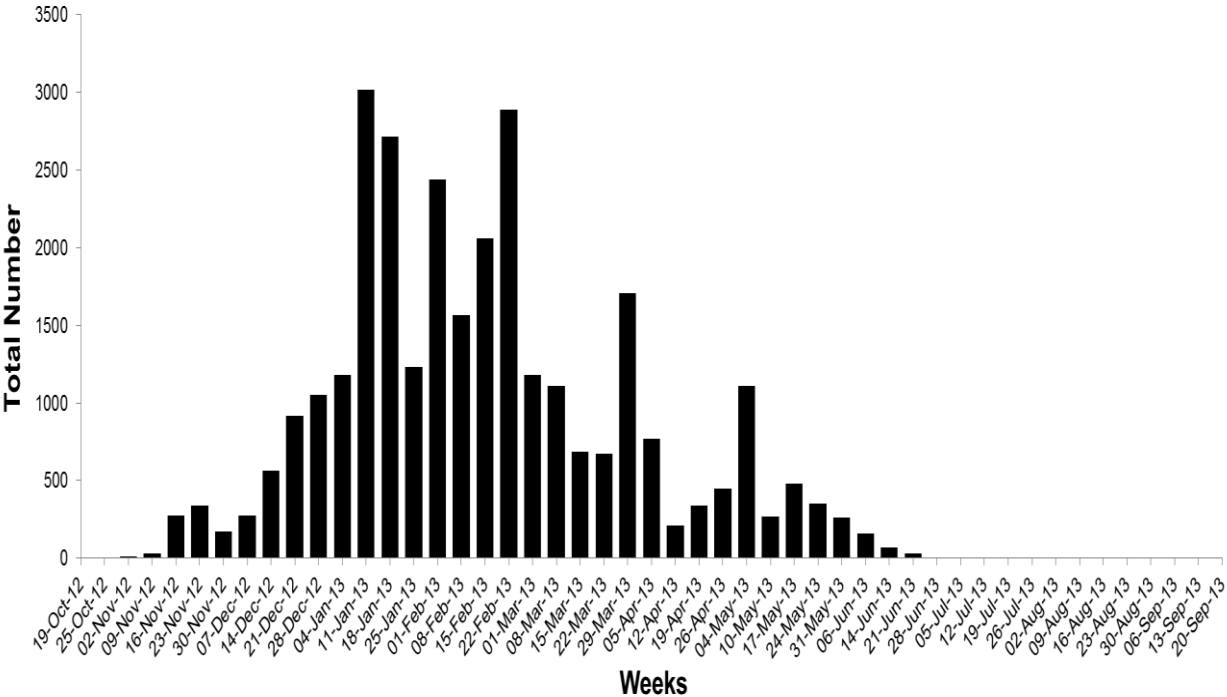
Figure 3.3: Mean number of *S. calcitrans* collected per week in 6 NZI and 6 Vavoua traps from 19 October 2012 to 13 September 2013. The dates given indicate the end dates for the weeks in which collections were made.

Although there was no statistical significant difference between the traps, the directional NZI trap collected more flies over the study period than the multidirectional Vavoua trap. This demonstrates that adult flies tend to enter the pens from outside. This indicates that adult flies probably rest in the neighbouring weeds along the edges of the pens (Cilek & Greene, 1994; Guo *et al.*, 1998).

This would not necessarily mean that pupae are found outside the pens and the pupation sites need to be investigated. This information has important fly management implications in directing chemical applications towards the weed surroundings during the fly resting periods.

3.3. SEASONAL ABUNDANCE OF *S. CALCITRANS*

The total number of flies collected over a period of 11 months in 12 traps (6xNZI and 6xVavoua traps) is given in Fig. 3.4. The total number of stable flies collected during that period was 30 582 (Appendix 3). Roughly five peaks were identified: the weeks ending 11 and 18 January 2013; 1 February 2013; 22 February 2013; 29 March 2013; and the last smaller peak was the end of the week of 4 May 2013. No stable flies were present in the traps from 21 June 2013 onwards.



occurs in the winter months when temperatures can drop to a low of -25 °C in Alberta, Canada (AlbertaCanada, 2014); the lowest temperature recorded on Karan Beef was -8 °C. The maximum temperatures in Alberta is much lower than that in Heidelberg. These very low temperatures may be the reason for Alberta having only four peaks as lower temperature result in lower developmental rates of the flies than at Heidelberg. The flies will be subjected to less degree days, probably resulting in the development of fewer generations (Lysyk, 1993; Krafur *et al.*, 1994; Taylor & Berkerbille, 2011; Beresford & Sutcliffe, 2012). The flies do not go into diapause but overwinter as a third larval instar when temperatures are low and development rates are very low until temperature increases occur (Beerwinkle *et al.*, 1978; Berry *et al.*, 1978; Scholl *et al.*, 1981; Greene, 1989; Jones & Kunz, 1997). This data may be useful for the development of control strategies. No control will be necessary during a large part of the year from May to October and peak numbers can indicate an alert period when it may be necessary to apply control measures.

3.4 FACTORS POSSIBLY INFLUENCING TEMPORAL ABUNDANCE

3.4.1 Temperature

Weather data was obtained from an onsite weather station since 2003 (Appendix 4). The possible influence of mean weekly temperature (and mean weekly maximum and minimum temperature) on the total number of *S. calcitrans* collected during the season is given in Fig. 3.5. The monthly distribution of mean weekly temperatures (Fig. 3.5.b) shows that the average temperatures for Heidelberg range from 16.6 °C in June to 26.3 °C in January. The monthly variation of minimum and maximum weekly temperatures as indicated Fig 3.5a. The region is coldest during July when the temperature drops to 0.2 °C during the night (Fig. 3.5.a). There was an increase in fly numbers as mean temperatures increased, which could be attributed to the first emergence of the fly population after winter. As temperatures started to increase from 2 November, the number of flies collected in the traps started to increase up to 23 November, at which point the mean maximum temperature reached a peak of approximately 33 °C. This high temperature could be a possible factor influencing the increase in fly numbers in the traps leading to the first peak in fly numbers. The following peaks in fly populations seem to be correlated with small temperature increases, possibly correlated with an increase in fly activity for the weeks ending 11 January 2013, and then 8 February 2013, 5 April 2013 and the week of 10 May 2013 respectively. The fly number peaks appear to occur farther apart as temperatures start to decrease indicating a decrease in development rate associated with a decrease in temperature (Lysyk, 1993; Taylor & Berkerbille, 2011). After the week ending 4 May, temperature decreased to a low of less than 5 °C after which the total number of flies collected

in the traps decreased significantly. No flies were present in the traps after the week ending on 14 June 2013. According to research conducted by Beresford and Sutcliffe (2012), the weekly rate of change of stable fly populations was influenced by temperature and accumulated degree days which means extremely low temperatures in winter and high temperatures in summer are probably unfavourable conditions for stable flies, which would increase the time the flies needed to develop (fewer degree days) and resulting in a decreasing population density (Mullens & Meyer, 1987; Gilles *et al.*, 2005).

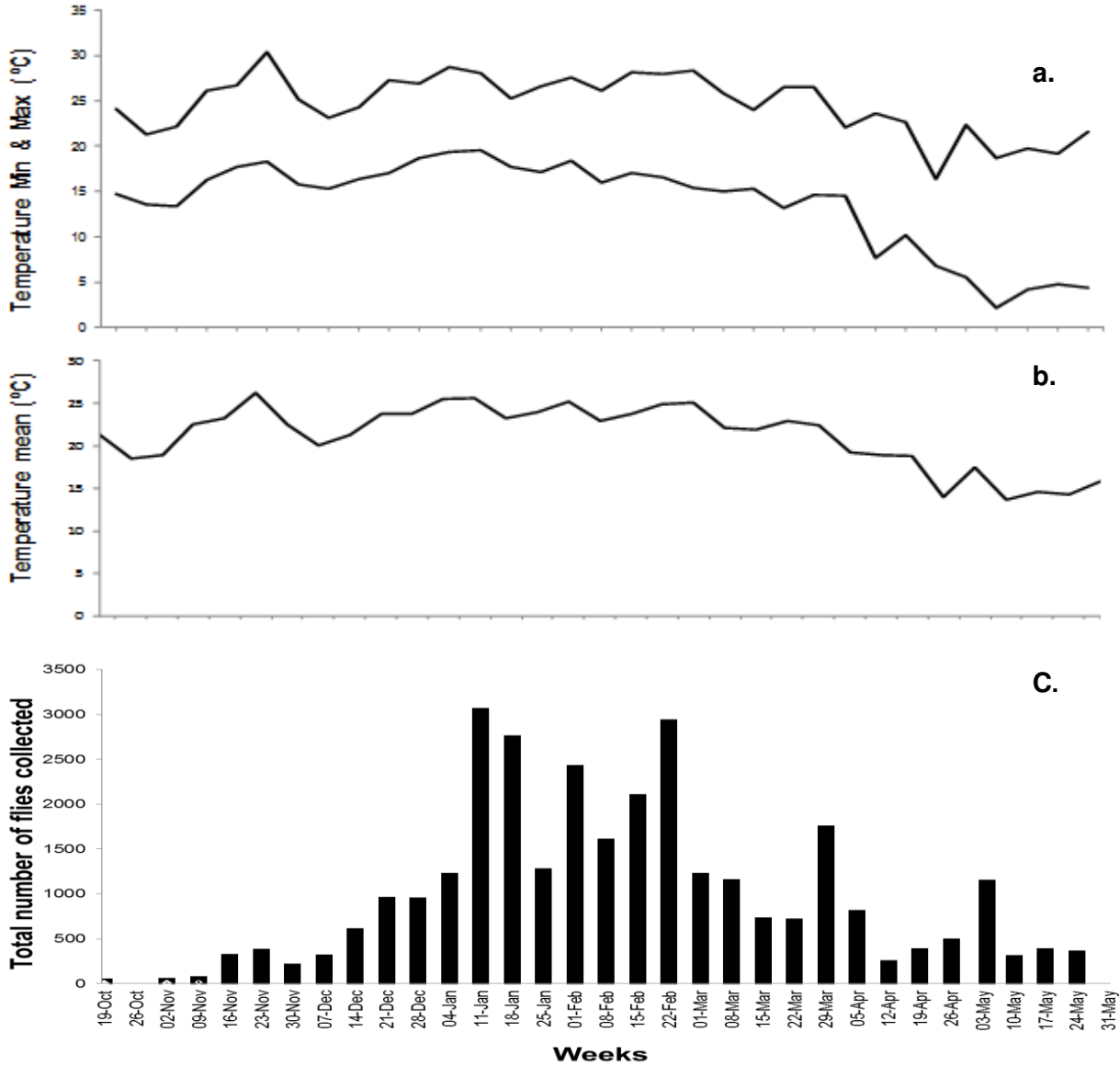


Figure 3.5: The relationships between mean weekly minimum and maximum temperature (a), mean weekly temperature (b) and total number of *S. calcitrans* collections per week (c) for the period 19 October 2012 to 31 May 2013. The dates given reflect the end dates of the weeks.

3.4.2 Other climatic factors

Other climatic factors that may have an influence on the number of flies collected in are wind speed and rainfall (Mullens & Meyer, 1987; Taylor *et al.*, 2007; Urech *et al.*, 2012; Taylor *et al.*, 2013). It is difficult to determine how meaningful these factors are in the field especially with regard to windspeed, since these factors are highly interactive and with other climatic factors. Effects of rainfall on the total number of flies collected are shown in Fig. 3.6. Heidelberg on average receives about 588 mm of rain per year, with most rainfall occurring during summer (SAexplorar, 2011). Fig. 3.6a shows the average rainfall values for Heidelberg per month. It receives the lowest rainfall (0 mm) in July and the highest (112 mm) in January (SAexplorar, 2011). Rainfall seems to have a significant effect and it is shown that after the first rainfall of the new season, fly numbers start building up (Mullens & Peterson, 2005). However, this increase in stable fly numbers also coincides with an increase in temperature, demonstrating the interaction between rainfall and temperature. The winter months are associated with no rainfall and zero fly collections. During the week ending on 7 December 2013 (Fig. 3.6) an average weekly rainfall of approximately 10 mm was recorded, that could be one of the factors contributing to the strong gradual increase in number of flies collected in the traps. Rainfall could be an important factor influencing collections, initiating the first emergence after a dry winter.

However, rainfall together with temperature could also play a regulating role in stable fly peak occurrences. In a study by Masmehatip *et al.*, (2006) on the daily activity of *S. calcitrans*, Vavoua traps were used in a dairy and a cattle farm in Nakhonpathom Province, Thailand, from July 2004 to June 2005. Over this period 80% of flies were captured during the rainy season from May to October and 20% during the dry season from November to April.

This corresponds with the results found in this study. In Heidelberg the rainy season is generally in January and February which also correlates with the highest numbers of flies collected.

Although farming activities (water usage in the feed lot) could have created breeding sites, it would not override the effect of rainfall because the water usage in the feedlot was a constant factor throughout the year.

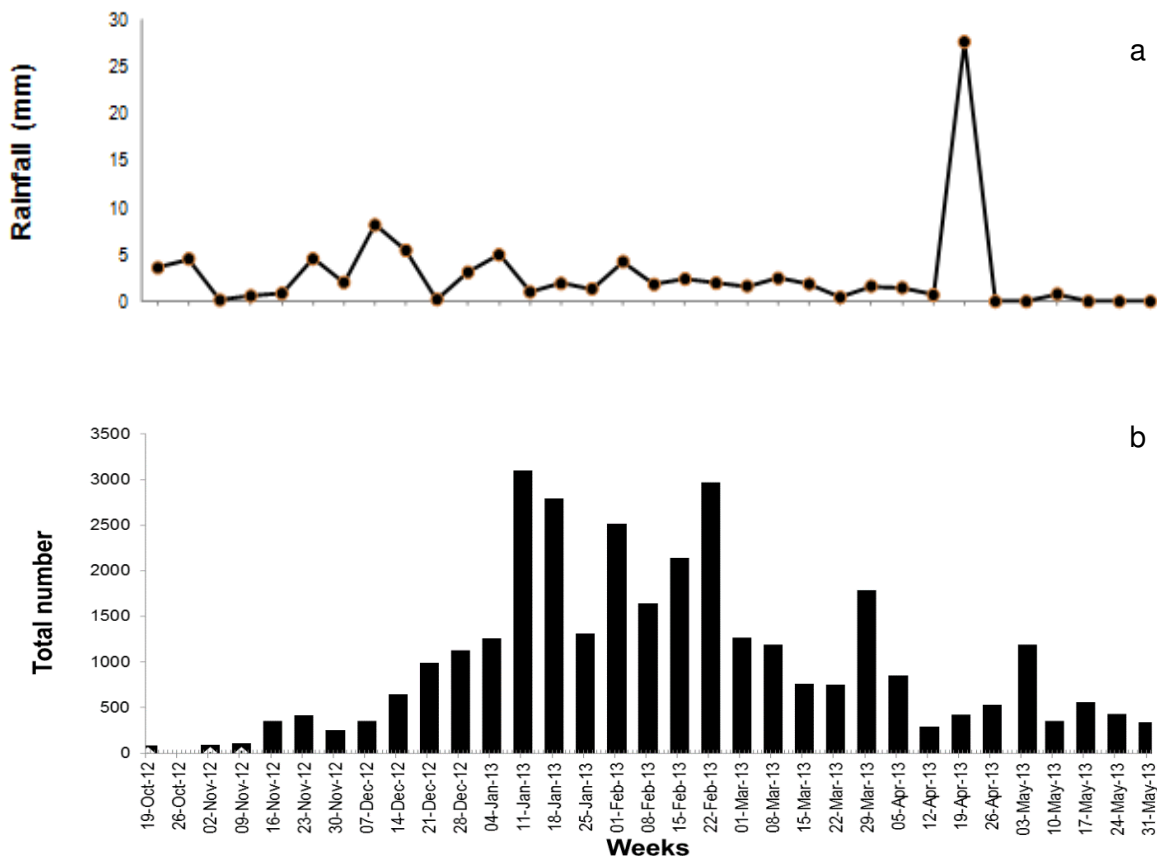


Figure 3.6: The relationships between rainfall (a) and total number of *S. calcitrans* collections (b) per week for the period 19 October 2012 to 31 May 2013. The dates given reflect the end dates of the weeks.

It is difficult to determine the impact of wind speed on the numbers collected in traps because the activity of the flies is more likely to be caused by daily temperatures (Mullens & Meyer, 1987). Further study is needed to determine the effect of wind on daily fly activity for use in the interpretation of trap collections and daily impacts on cattle irritation. The number of flies collected decreased significantly from the week ending on May, 3rd, 2013 and is probably the combined result of low temperatures, lack of rainfall and low relative humidity which could lead to rapid reductions in population size (Mullens & Meyer, 1987).

3.4.3 Daily distribution of stable flies

The daily distribution of stable flies was determined by removing and counting the stable flies collected in the traps every hour during the day. This was done on three days during the season, on 20 February 2013 (Table 5.1), 21 February 2013 (Table 5.2) and 05 March 2013 (Table 5.3). The total number collected per hour on these three days was determined (Fig. 3.7). Collections on 20 February and 5 March showed an early peak from 9:30-10:30, followed by a

decrease in activity. This decrease in activity was followed by an increase in numbers collected later in the afternoon extending over a longer period from 13:30 to 17:30 depending on daily condition. In South-Eastern Nebraska stable fly activity peaks were similar at 14:00 and stable flies were mostly active between 10:00 and 16:00 (Catangui *et al.*, 1997; Campbell *et al.*, 2001; Anthony, 2005). Low activity during the day may indicate periods that the flies need to rest and digest their blood meals during the day (Berry & Campbell, 1985; Berry *et al.*, 1986). There were no flies present in the traps after 19:30. This is also evident from a study done by Berry & Campbell, (1985) where stable flies' maximum feeding activity occurred in midday with very low activity between dusk and dawn. In a study by Masmataathip *et al.* (2006) in Thailand during July 2004 to June 2005 the activity pattern of *S. calcitrans* was diurnal with a peak between 08:00 to 10:00 and another less pronounced one in the afternoon. Although this literature support the findings in this study in general terms, the different patterns of daily distribution could be attributed to varying climatic conditions of Nebraska, Thailand and South Africa. This could imply that cattle have enough time during the night to compensate for the possible feeding losses during the day due to fly irritation. This data has important management implications for the determination of fly control threshold levels and the application of insecticides.

It is important to note that the traps could serve as resting sites for the flies. Stable flies prefer shaded resting sites which would make the traps an ideal resting site and it would ultimately increase the effectiveness of the traps (Buschman & Patterson, 1981). The significance of resting sites is that flies need a place to digest their blood meal. The flies ingest enough blood during a relatively short period of time to sustain them to the next day (Berry & Campbell, 1985). The control of weeds and vegetation surrounding the feedlot serving as resting sites could be an important stable fly management strategy in reducing fly numbers, or may be utilised as concentrated chemical control areas rather than the current more general application of pesticides.

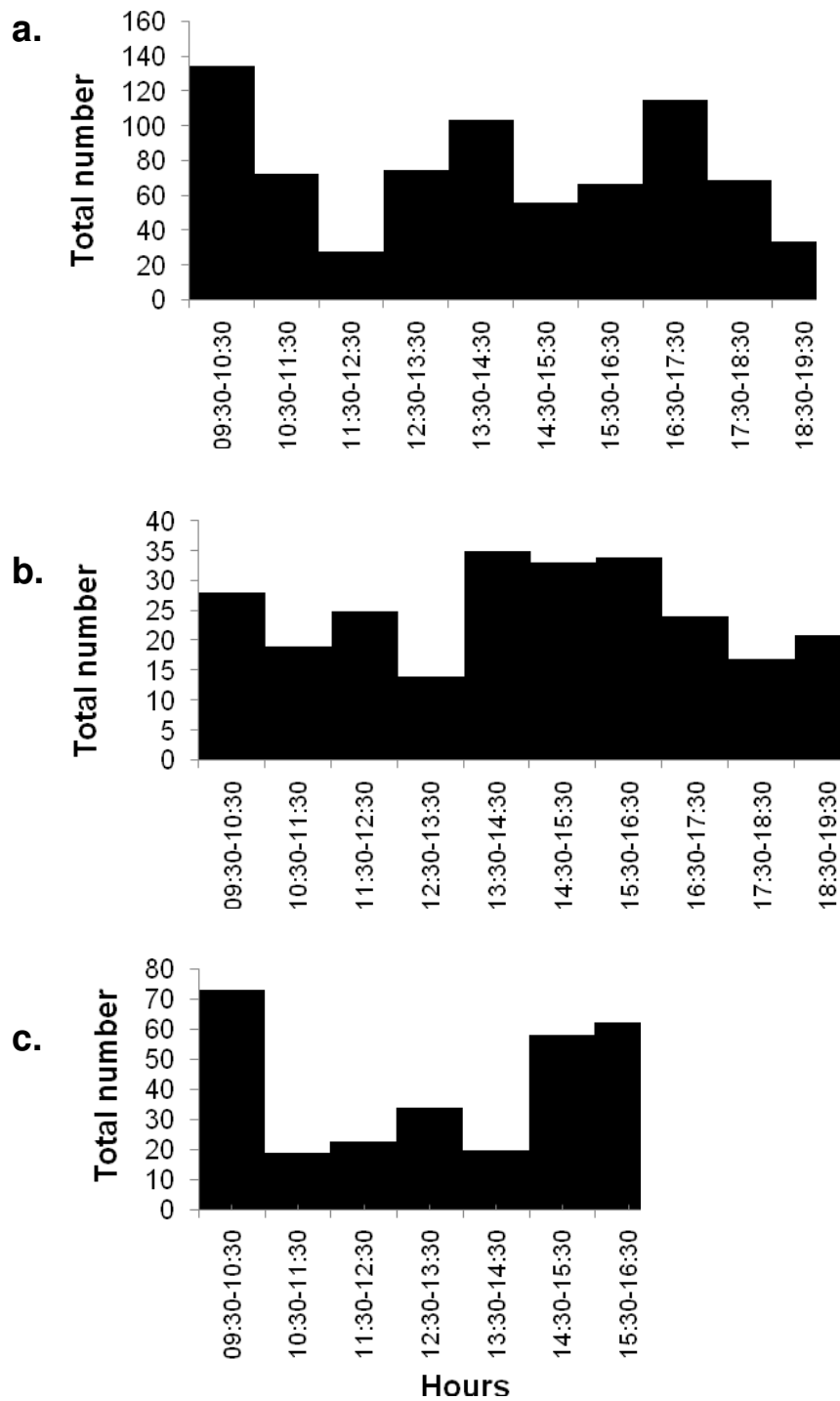


Figure 3.7: Total hourly collections of *S. calcitrans* in Vavoua and NZI traps in the H-line on days 20 February 2013 (a), 21 February 2013 (b) and 05 March 2013 (c).

3.5. STABLE FLY COUNTS ON CATTLE

3.5.1 Relationships between daily trap collections and *S. calcitrans* counted on cattle

Monitoring adult stable fly populations on cattle consists of counting the number of flies feeding on the outside of one lower foreleg and the inside of the other lower foreleg for a selected period of time, usually 1 minute (McNeal & Campbell, 1981; Berry & Campbell, 1985). The reason for counting the stable flies is that the lower foreleg is the preferred feeding site of stable flies (Hogsette *et al.*, 1987; Mullens *et al.*, 2006) and that literature exists on the relationship between stable flies per foreleg per minute and its economic impact (McNeal & Campbell, 1981; Wieman *et al.*, 1992; Campbell *et al.*, 1993; Catangui *et al.*, 1993; 1995; 1997). The relationship of the number of stable flies counted on the foreleg of cattle per minute (Appendix 9) and the total daily collections in three NZI traps and three Vavoua traps in the H-line (Table 6) were determined and are shown in Fig. 3.8. The highest number of stable flies counted on the cattle was on 25 January 2013. The highest number of flies collected in the traps was on 11 January 2013. The correlation between the daily stable fly collections and the number of flies on the forelegs of cattle was statistically significant at $P < 0.05$. The r value of 0.90 (Fig. 3.9) indicates a strong correlation between numbers collected per day in the traps and numbers counted on cattle forelegs. This implies that the numbers collected in the traps may be used as an indication of the level of fly activity on cattle on that same day. The management implications are that fly traps can be used either as a monitoring tool or as a control threshold indicator for insecticide application against stable flies (Mihok *et al.*, 1995; Mihok, 2002; Mihok *et al.*, 2006).

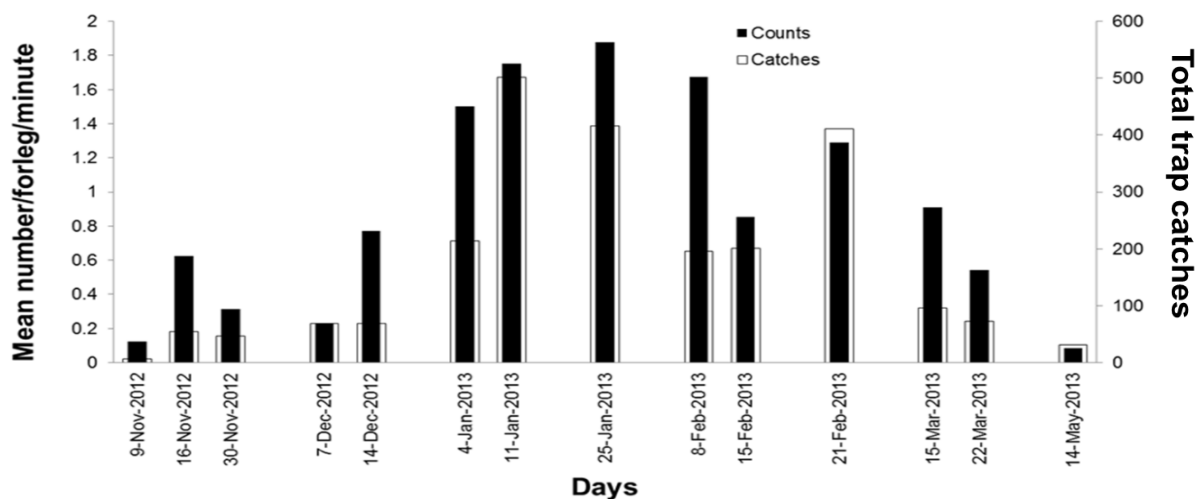


Figure 3.8: Total number of daily trap collections of *S. calcitrans* in 3 NZI and 3 Vavoua traps and mean *S. calcitrans* counted daily on cattle forelegs per minute in the H-line from 19 November 2012 to 14 May 2013.

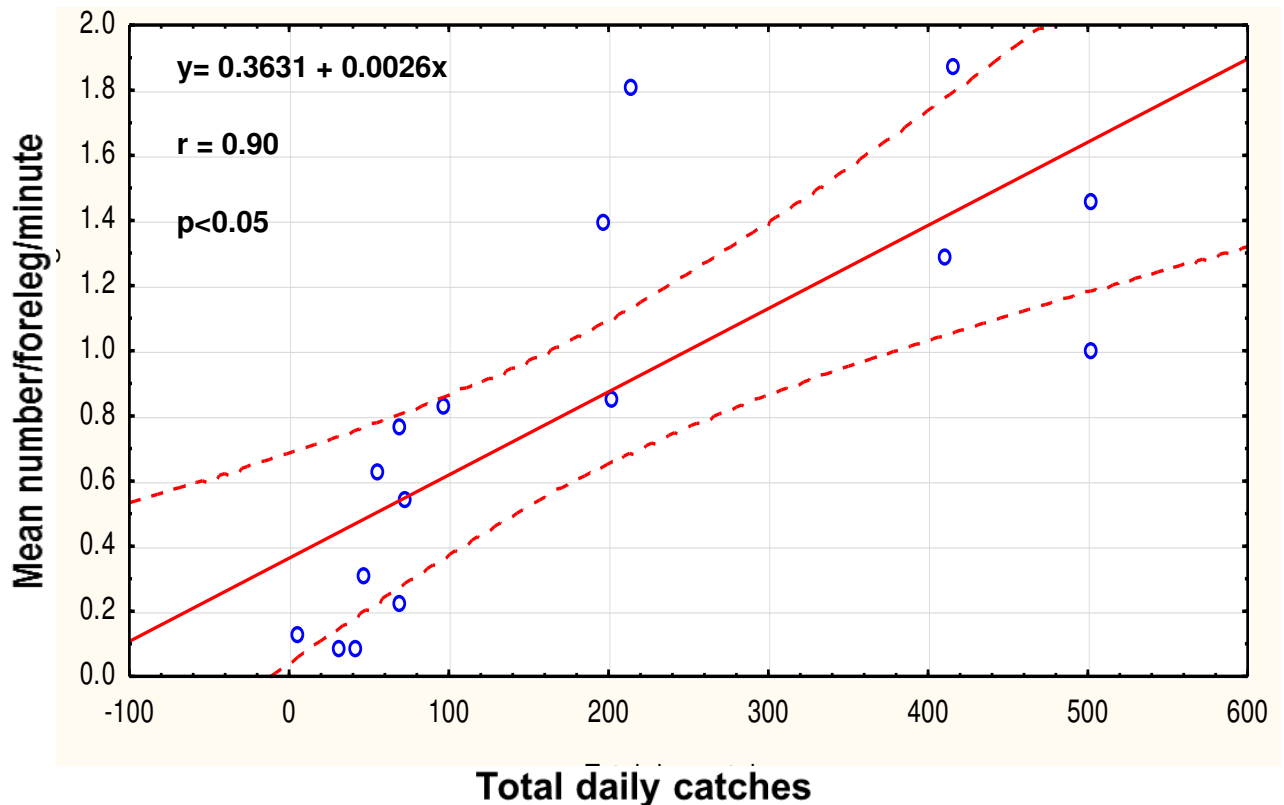


Figure 3.9: Regression between total daily stable fly collections in 3 NZI and 3 Vavoua traps in the H-line and mean number of flies counted per foreleg/min during daily surveys from 19 October 2012 to 31 May 2013.

The linear relationship between the numbers collected in the trap and the numbers counted on forelegs can be used to make predictions within the extent of the graph $y = 0.3631 + 0.0026x$ (Fig. 3.9) However, more research is needed to determine the economic threshold for larger stable fly populations for chemical fly control in South Africa.

3.5.2 Irritation indicators of stable flies on cattle

3.5.2.1 The relationship between cattle foot stomps and the number of stable flies counted on cattle forelegs per minute

It is evident from literature that stable flies have an irritating impact on cattle which may have a subsequent negative impact on meat production (Campbell *et al.*, 1977; 1987). *Stomoxys calcitrans* have a painful bite and can cause the cattle to display avoidance behaviour such as stomping their feet, throwing their heads, twitching their skin, swishing their tails and bunching together (Berry *et al.*, 1983; Wieman *et al.*, 1992; Dougherty *et al.*, 1993; 1994; Mullens *et al.*, 2006). This avoidance behaviour of cattle has an impact on their weight gain (Campbell *et al.*,

1977; 1978). Fig. 3.11 shows that the frequency of foot stomps (Appendix 10) correlates positively with the irritation caused by the number of stable flies counted on cattle forelegs (Appendix 9). This stable fly effect on leg stomping is confirmed in literature (Berry *et al.*, 1983; Wieman *et al.*, 1992; Dougherty *et al.*, 1993; 1994; Mullens *et al.* 2006).

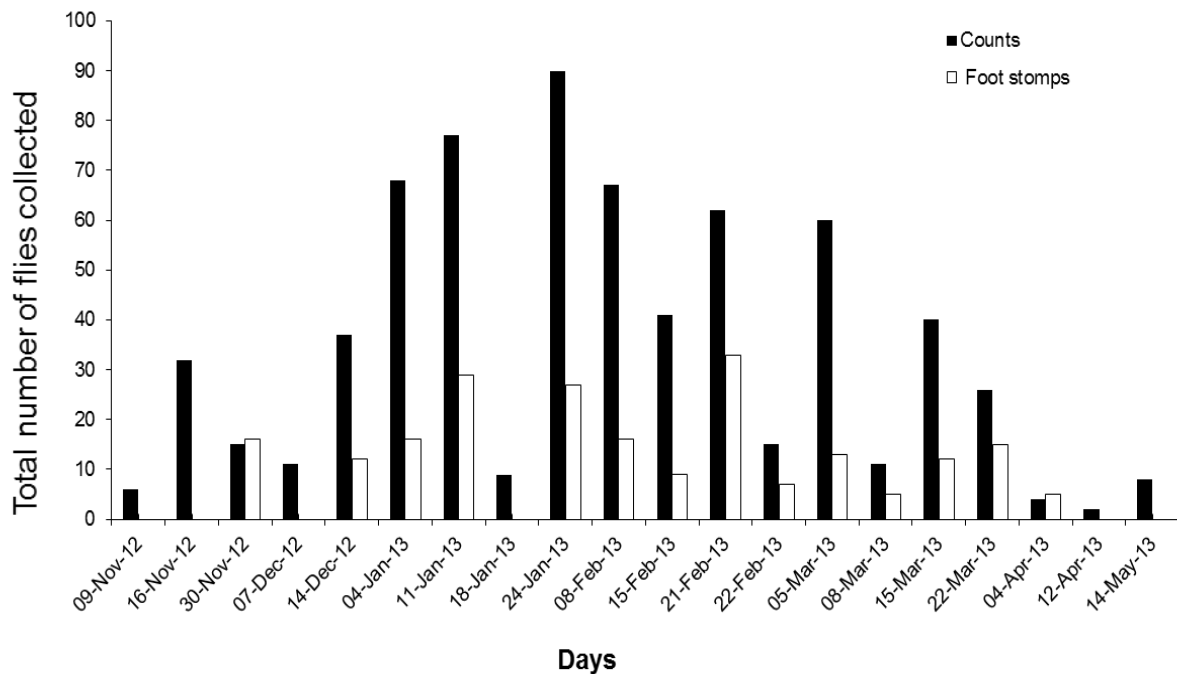


Figure 3.10: A comparison between the number of foot stomps/minute and the number of *S. calcitrans* counted/forelegs/minute on 48 cattle on selected days during the season in the high exposure H-line 19 October 2012 to 14 May 2013.

The regression analysis of the number of foot stomps per minute and the number of *S. calcitrans* counted per foreleg per minute, showed an r value=0.79 and $p < 0.05$ (Fig. 3.11). This strong correlation, is in agreement with literature (Urech *et al.*, 2012). However, the number of stable flies counted per foreleg never exceeded two flies per foreleg per minute. According to literature, the economic threshold for stable flies is five flies per foreleg per minute (Campbell *et al.*, 1987; 2001). The significant correlation found between stable fly activity and avoidance behaviour demonstrates that effective structured animal observations can be used to estimate the effect of fly populations on cattle weight gain and ultimately on economic losses (Warnes & Finlayson, 1987; Mullens *et al.*, 2006). Structured animal observations can be used as an early warning system to apply pesticides according when avoidance behaviour reaches a predetermined threshold to avoid economic losses.

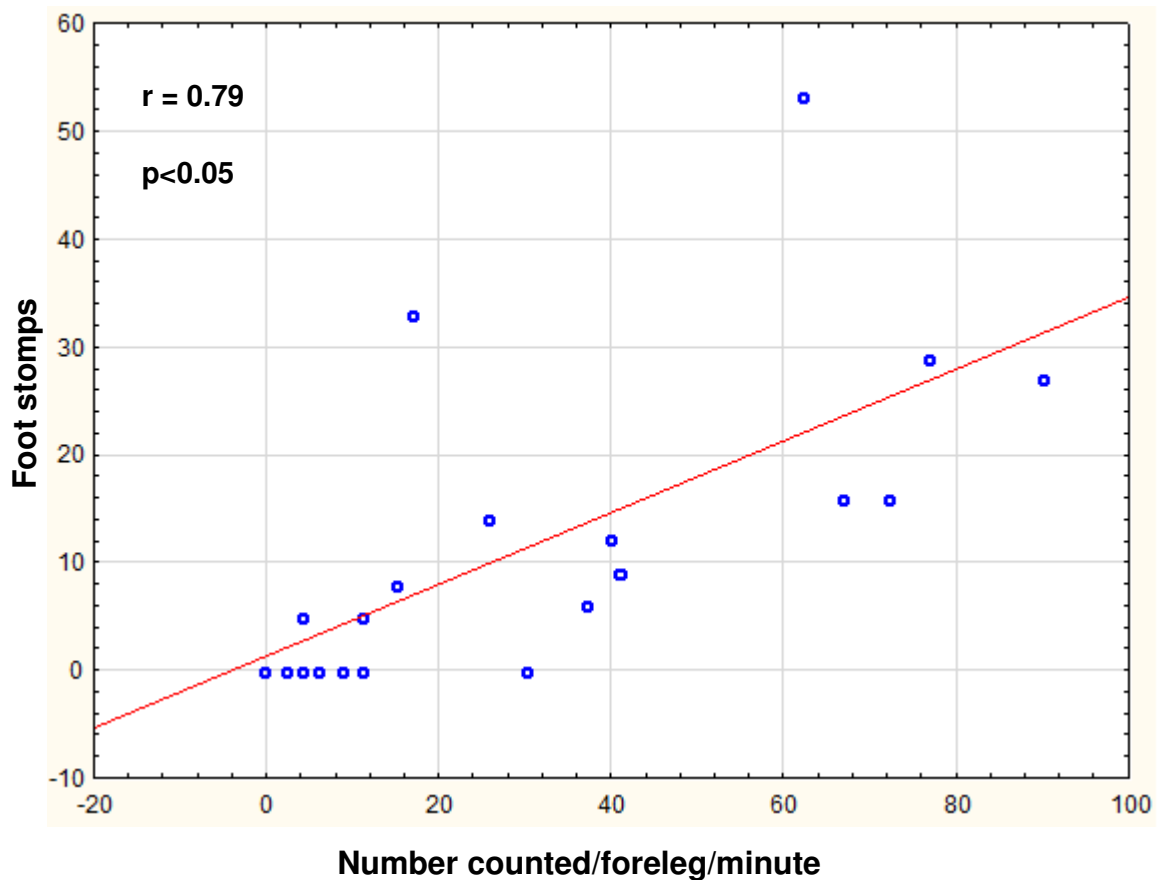


Figure 3.11: A scatter plot of the correlation between the total number of foot stomps per minute and the total number of *S. calcitrans* counted/forelegs/minute on 48 cattle on certain days during the season in the high exposure H-line 19 October 2012 to 31 May 2013.

3.6 FACTORS POSSIBLY INFLUENCING STABLE FLY FEEDING ACTIVITY ON CATTLE

The activity of flies is measured by their daily feeding patterns on the forelegs of cattle. The feeding patterns may be influenced by a number of factors such as radiation, relative humidity and wind speed (Berry & Campbell, 1985). A combination of high wind and high radiation can cause low relative humidity conditions which will decrease stable flies' feeding activity (Berry & Campbell, 1985). Temperature is the most important factor as it affects the flies' metabolic and other physiological rates although it could also cause behavioural responses (Berry & Campbell, 1985).

3.6.1 Temperature

The factors mentioned in 3.4.1 and 3.4.2, possibly influencing temporal abundance of stable flies collected in the fly traps was temperature and rainfall. It is also necessary to determine the influence of climatic factors on the daily feeding patterns of stable flies. In order to identify these effects, daily stable fly counts made on cattle forelegs during the season which were correlated with daily climatic factors (Appendix 8). The possible influence of mean daily as well as daily minimum and maximum temperatures on daily stable fly counts on cattle forelegs was determined to investigate the possible influence of temperature on daily stable fly activity patterns (Fig. 3.12). The minimum, maximum and mean daily temperatures were calculated for the days surveyed, using data obtained from a local on-site weather station.

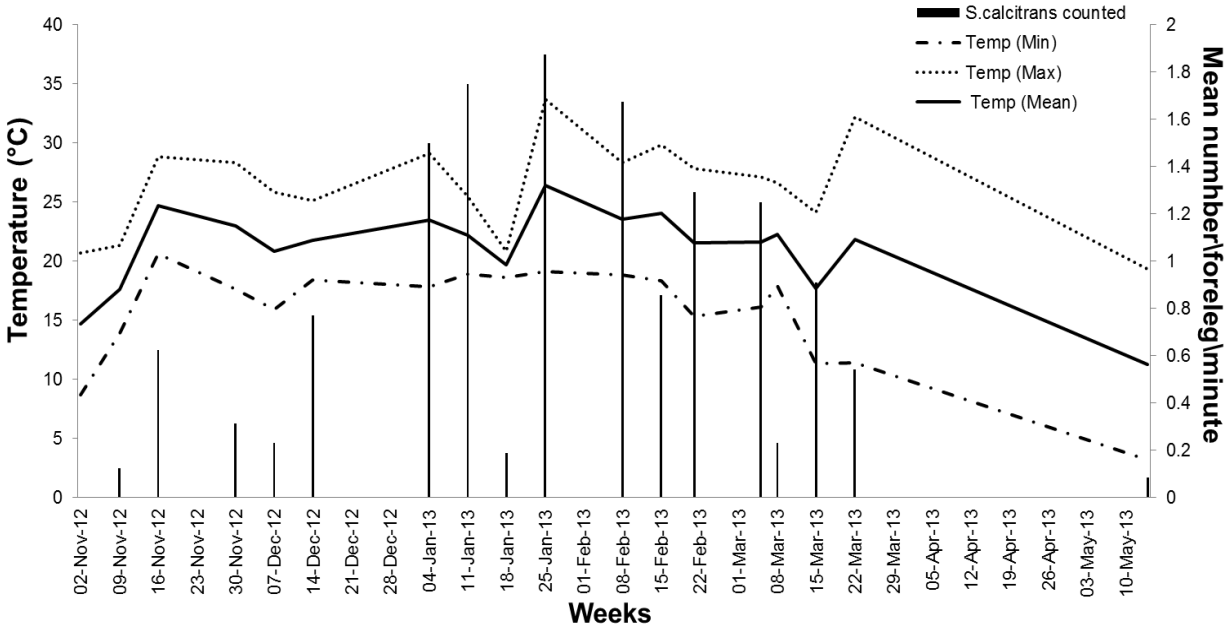


Figure 3.12: Relationship between mean daily, maximum and minimum temperatures and mean number of *S. calcitrans* counted/foreleg/minute from 02 October 2012 to 02 May 2013.

Figure 3.12 suggests that stable fly activity is influenced by temperature. This effect is quantified in Fig. 3.13. The relationship between mean daily temperature and mean number of stable flies per cattle fore leg per minute was calculated (Fig. 3.13). This indicated that there is a highly statistical significant positive correlation between the number of stable flies counted on cattle forelegs and mean daily temperature. Increasing temperature is therefore significantly positively correlated with feeding activity of stable flies.

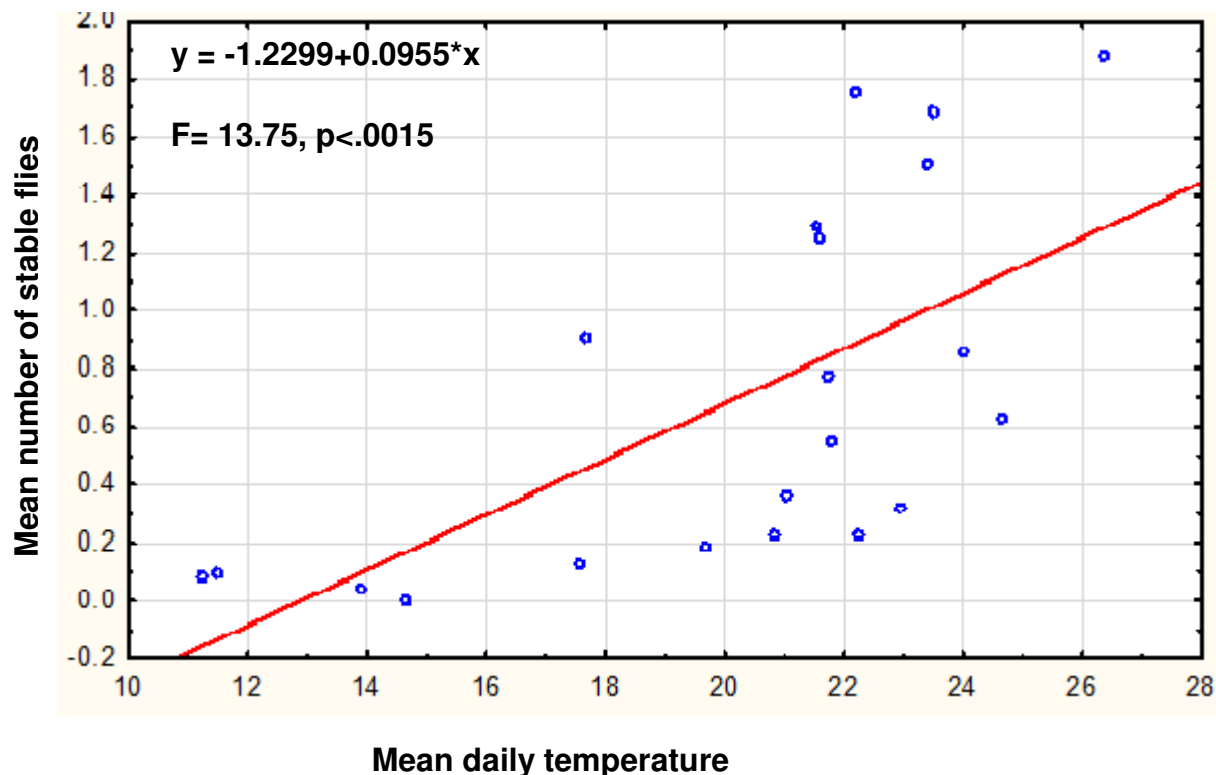


Figure 3.13: Regression analysis between mean daily temperature and mean number of *S. calcitrans* per foreleg/minute in the high exposure H-line from 19 October 2013 to 31 May 2013.

The management implication is that a combination of large trap collections and higher temperatures increases the risk of greater irritation to cattle. This could stimulate more bunching of cattle with the secondary effect of heat stress and possible dehydration (Wieman *et al.*, 1992). This effect should be taken into account when interpreting stable fly numbers on cattle. When chemicals are directly applied onto the cattle with mist blowers, the application should be done on days when the mean daily temperature is relatively high.

3.6.2 Wind speed

Wind speed is an important factor to consider when determining daily activity of stable flies. The impact of daily wind speed on the number of flies counted per minute per foreleg is shown in Fig.3.14. The highest number of flies was counted on 25 January 2013 while wind speed was very low.

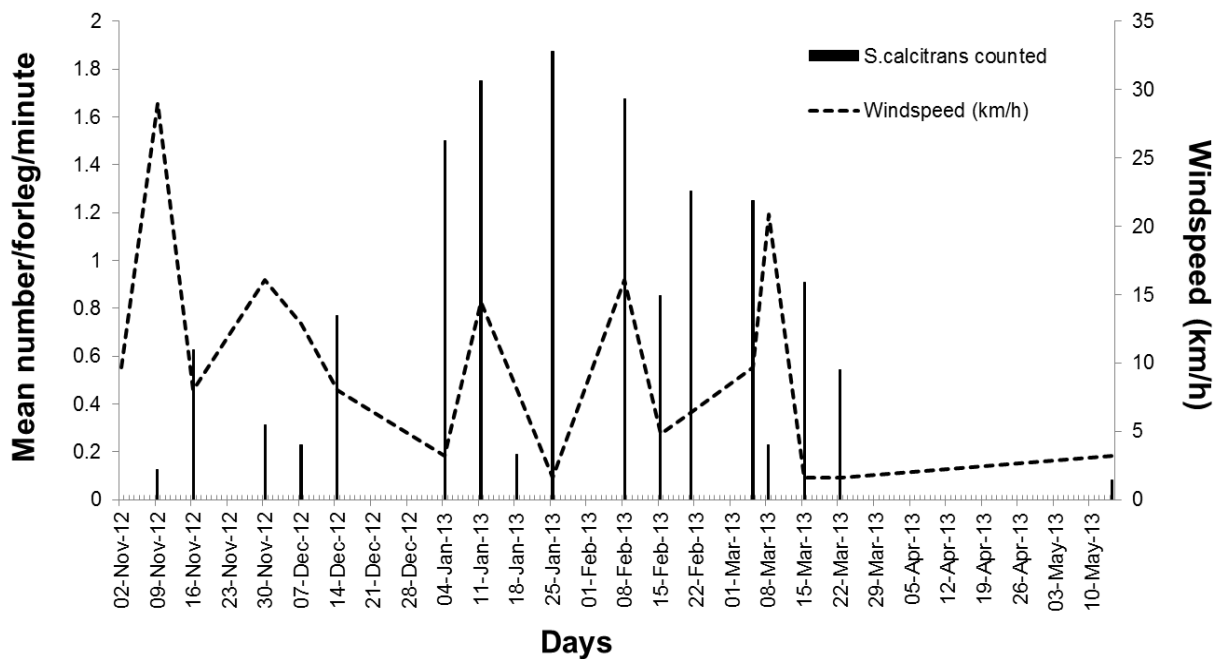


Figure 3.14: Relationship between daily wind speed and mean number of *S. calcitrans* counted/foreleg/minute on 48 cattle in the H-line during daily fly counts on days between 02 November 2012 to 02 May 2013.

Figure 3.14 suggests that wind speed has some effect on the number of stable flies on cattle. The linear correlation between daily wind speed and stable fly numbers on cattle forelegs is presented in Fig. 3.15. A relatively low negative correlation ($r=-0.29$) between wind speed and flies on forelegs was found. This was to be expected since the windmeter on site measures daily wind speed while the wind speed during the actual fly counting may vary considerably during the day. Furthermore, low or no fly numbers during the season is not necessarily a function of wind speed but could also be due to other factors. This resulted in a large variation in the data.

Further study is needed to determine the effect of wind on daily fly activity for use in the interpretation of daily impacts on cattle irritation. According to literature, stable flies are less active during strong wind as they seek shelter, such as vegetation and buildings, from the wind (Broce *et al.*, 1991). This information needs to be taken into consideration when interpreting stable fly counts on cattle as well as in trap collections. Although stable fly counting on cattle may be relatively low, this may be due to strong winds although the fly population may be very high.

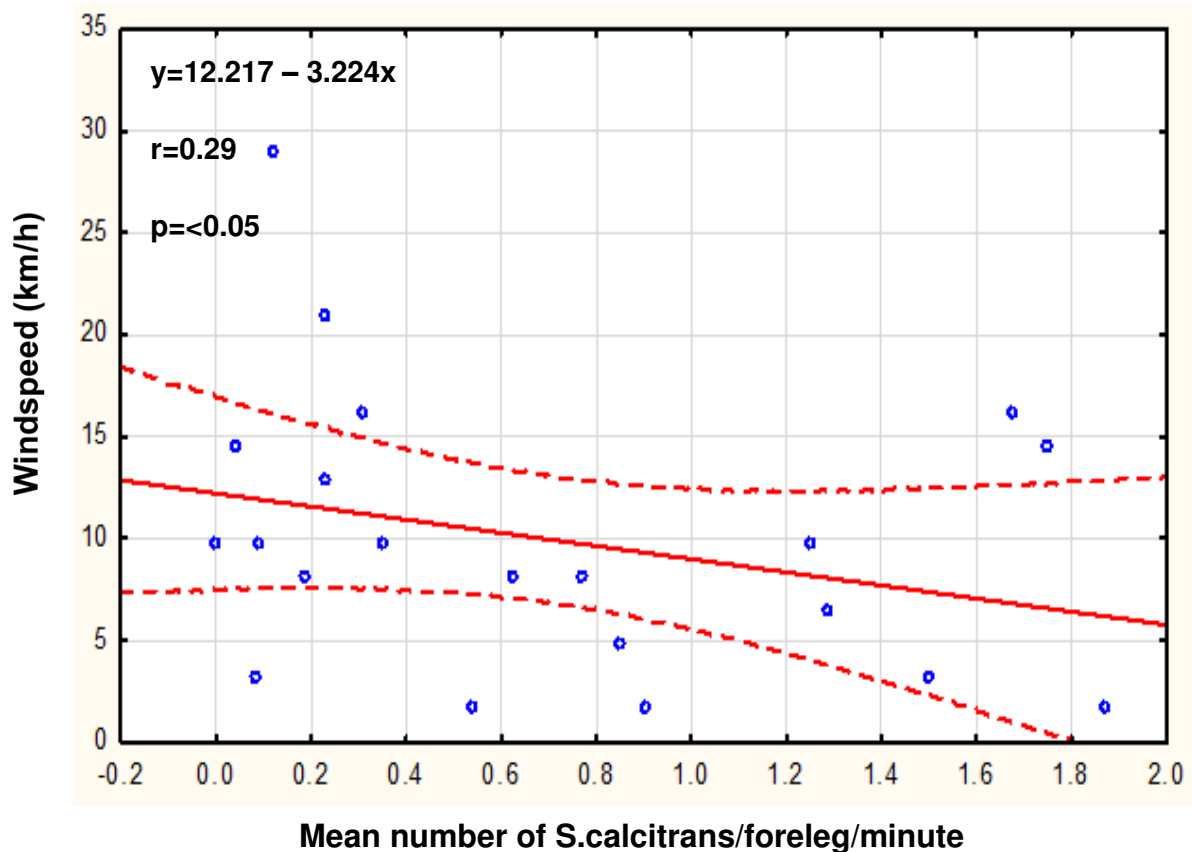


Figure 3.15: A regression between wind speed (km/h) and mean number of *S. calcitrans* counted/cattle/foreleg/minute during daily fly counts on certain days during the season from 02 November 2012 to 02 May 2013.

3.7 FACTORS INFLUENCING SPATIAL DISTRIBUTION OF STABLE FLIES IN THE FEEDLOT

3.7.1 Management effects, edge effects and resting sites

3.7.1.1 Comparison between the numbers of *S. calcitrans* collected in traps in the K-line and H-line

The H-line of pens is situated at the edge of the feedlot and surrounded by wetlands, manure run-off and vegetation and the pens were untreated with chemicals or pesticides. The K-line of traps is situated in the middle of the feedlot and pens were to some extent treated with pesticides (Fig 2.10). The total numbers of stable flies collected in the H-line and in the K-line during 19 October 2012 to 14 June 2013 are shown in Fig. 3.16. The number of stable flies collected in the unsprayed H-line traps at the edge of the feedlot is significantly higher than number collected in the sprayed K-line traps in the middle of the feedlot (Appendix 3). Fly population peaks show similar trends in both the H- and K-lines. Both lines showed a peak in

stable fly numbers in the week of 11-18 January 2013 and both lines showed an increase in numbers from 30 November 2012 with decreasing numbers from 7 June 2013 to zero numbers in winter.

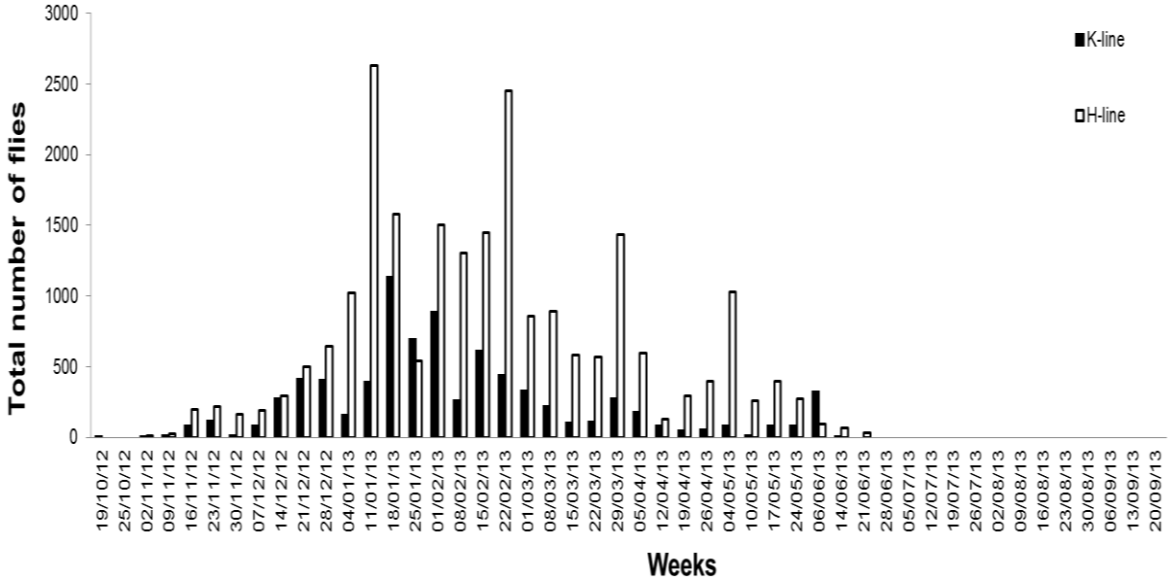


Figure 3.16: The total number of *S. calcitrans* collected per week per 3 NZI traps and 3 Vavoua traps in the H-line (high exposure) and K-line (low exposure) from 19 October 2012 to 14 June 2013.

To determine the statistical difference between the H- border line and K- middle line fly trap collections, repeated measures ANOVA was conducted (Fig. 3.17). The traps along the H borderline pens collected statistically highly significant more stable flies than the traps along the K middle line ($F=5.3545$, $P<0.001$). Factors such as high exposure to larval development sites, manure run-off, vegetation and the lack of insecticides applied, may be responsible for this significant difference (Fig. 3.17). The K-line in the middle of the feedlot was much less exposed to natural habitat and some chemicals were applied during the season as discussed in material and methods. The management implications is that chemical control should be concentrated on the edge pens to reduce the pressure from stable fly attacks on the edges of the feedlot.

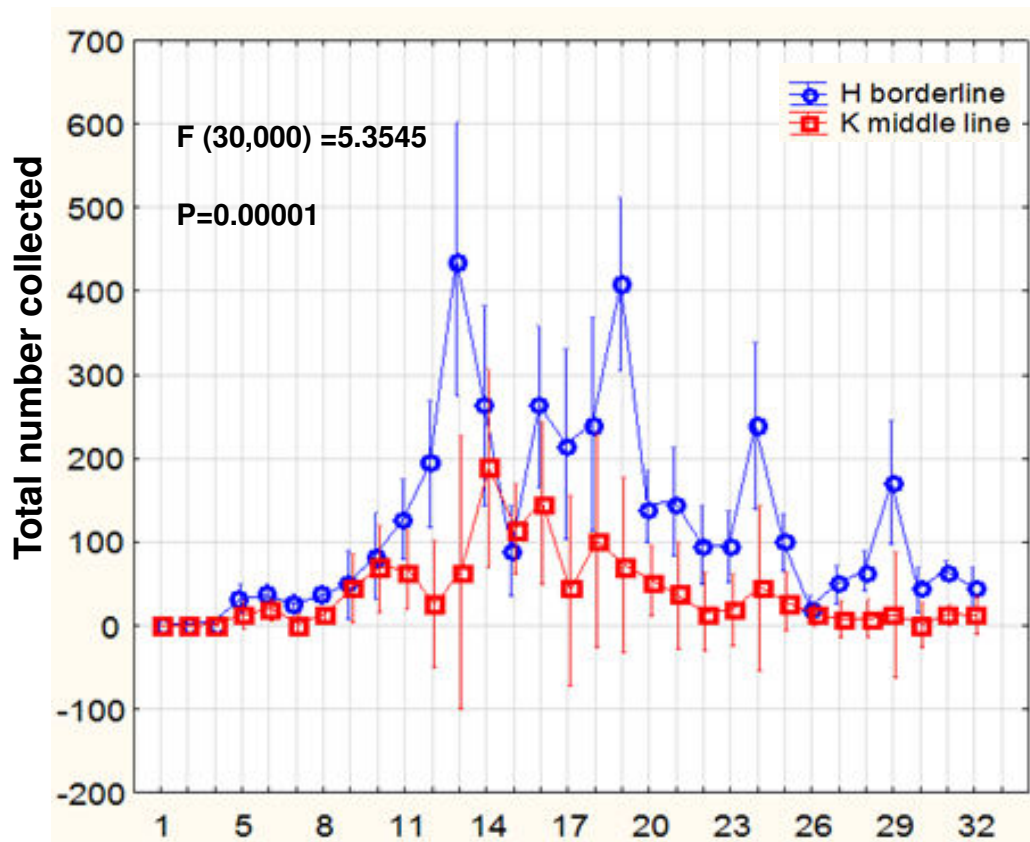


Figure 3.17: Repeated measures ANOVA between weekly *S. calcitrans* collected weekly between 19 October 2012 to 24 May 2013 in H- borderline line and K- middle line pens. Vertical bars denote 0.95 confidence intervals.

3.7.1.2 Comparison between *S. calcitrans* counted per cattle per minute in the K-line and H-line and statistical analysis.

The density of stable flies on cattle forelegs per minute in the H- borderline line and K- middle line during the season was determined (Appendix 8). It is clear that the density of flies on cattle was significantly higher in the H-line exposed to natural habitat and no-cover insecticide sprays on cattle (Fig. 3.18). The K-line had relatively low numbers of flies on the forelegs of cattle. The high number of stable flies' counted/cattle/foreleg/minute in the H-line could imply that there would be high levels of irritation and associated avoidance behaviour such as foot stomps. Urech *et al.* (2012) found a positive correlation between trap collections and foot stomps which implies that there is a high probability of higher avoidance behaviour in the H-line. The

management implication would be to treat the cattle in the pens bordering the surrounding veld with pesticides when stable fly populations reach unacceptable levels.

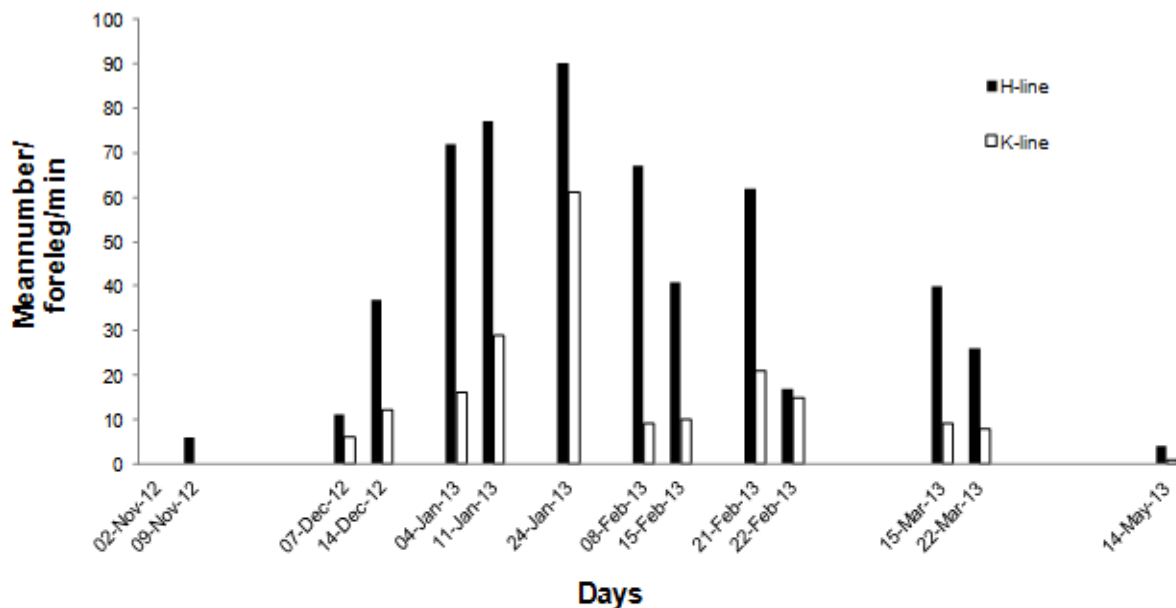


Figure 3.18: A comparison of the total daily number of *S. calcitrans* counted/foreleg/minute in the high exposure (H-line) and low exposure (K-line) lines.

3.7.1.3 Comparison between *S. calcitrans* count in the side paddocks and middle paddocks and statistical analysis.

The edge effect of the stable flies on the cattle was determined by comparing the number of flies' counted/foreleg/minute in the 12 pens in the unsprayed H-line, exposed to varying degrees of edge factors such as weed fields, manure run-offs and dirt roads (Fig. 2.10). Pen number 12 was subjected to manure run-off, vegetation and weeds and a dirt road. Pen number 1 was subjected to a large dirt road with more traffic and also manure run off.

The pens were divided into groups at the edge of the corridor and groups in the centre of the corridor (Appendix 9). The pens at the edges of the corridor were pens 1-2 and 11-12 (Group 1) and pen 3-4 and 7-8 (Group 2). Pens in the middle of the corridor were 5-8 (Group 3). The repeated measures ANOVA indicated a statistical significant effect ($F= 4.5023$) ($P=0.04414$) between stable flies on cattle and edge grouping (Appendix 11.1). In a Post Hoc test it was shown that that group 1 (pens 1,2,11 and 12 with the highest exposure to edge effects such as vegetation)(Appendix 11.2) had significantly more flies on cattle forelegs than in group 3 with the least edge exposure to surrounding vegetation). Groups 2 and 3 did not differ statistical significantly. This has important management implications. More chemicals can be applied to the pens in group 1 (1, 2 and 11, 12) than in the other pens, during the months with stable fly population peaks.

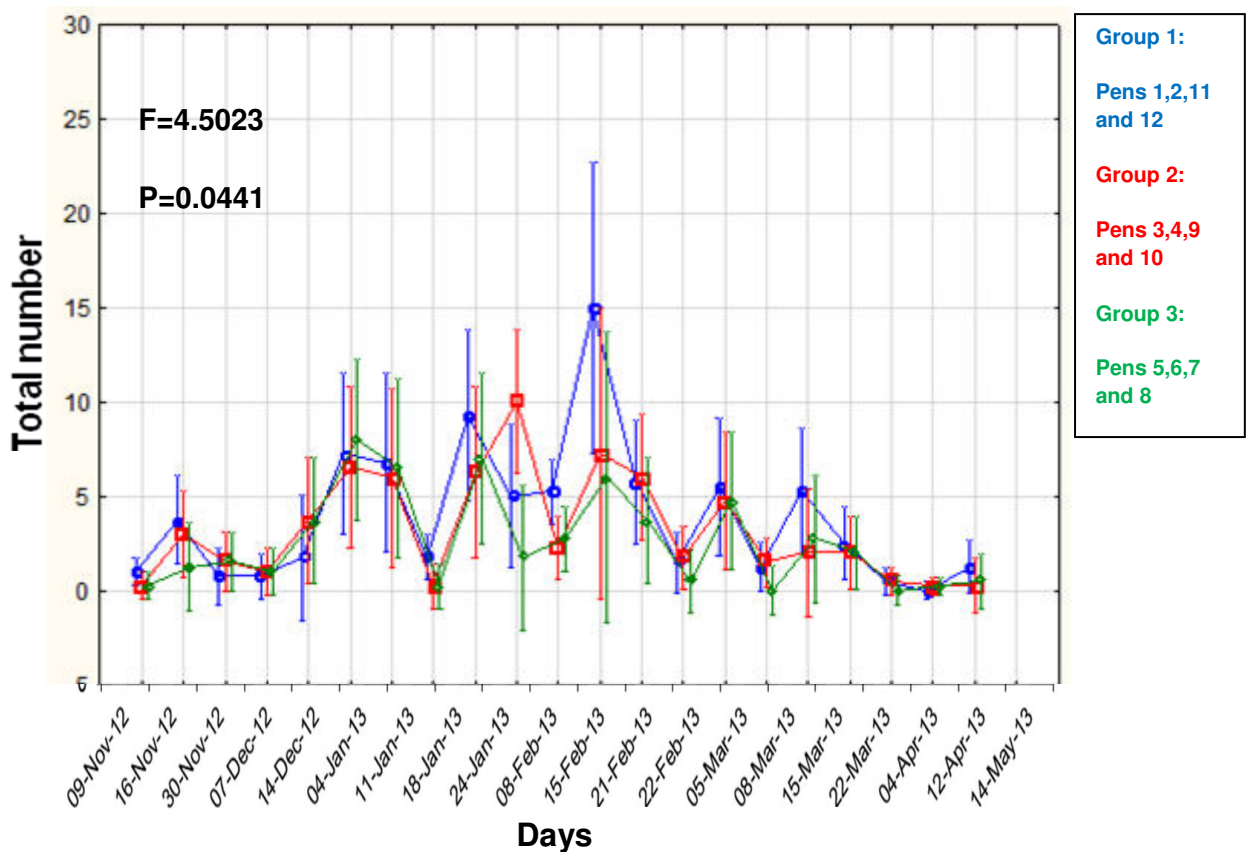


Figure 3.19: Repeated Measures Analysis of Variance for the total number of flies' counted/foreleg/minute/pen from 9 November 2012 to 14 May 2013 in the H-line.

3.7.1.4 Possible resting sites

The NZI traps are surrounded by vegetation that could serve as possible resting sites for stable flies after a feeding period during which digestion of blood meals occurs (Berry & Campbell, 1985) (Fig. 3.20). The vegetation surrounding this trap and the feedlot are possible reasons for large stable fly populations especially in the edge pens. The eradication or management of the surrounding vegetation should be part of a control strategy to reduce adult stable flies in the feedlot by reducing their resting sites (Urech *et al.*, 2011).



Figure 3.20: NZI trap surrounded by vegetation in the H-line – 19 February 2013.

The traps themselves could also be attractive sites for resting and digestion among many other possible sites (Berry *et al.*, 1986). The importance of the traps being possible resting sites is that it would increase the stable fly collections as the stable flies seek shelter within the traps.

3.8 DUNG HEAPS AND EFFECTS ON STABLE FLIES IN THE FEEDLOT

The dung in the pens are gathered into a pile in the middle of the pen and regularly removed from the pens. The dung is then dumped on large heaps approximately 5 km from the feedlot. The dung heaps were searched for fly pupae or other larval stages and very few pupae were found. It is therefore recommended that the dung heaps should be searched again in further studies. The total number of stable flies collected in H-traps along a gradient from the dung heap is shown in Fig. 3.21 (Appendix 7). Trap number H3, was placed 5 kilometres away from the feedlot and closest to the dung heaps. This trap collected the highest number of flies. Trap number H2 was placed 2.5 km away from the feedlot and collected lower numbers than H3. Trap number H1 was placed 1 km from the feedlot and collected a higher number of flies than trap H2. However, the numbers collected decreased drastically from the first week of collections (1 to 7 February 2013).

From 1 March 2013 the number of flies present in the H3 trap gradually decreased. There were no flies collected in any of the traps after 22 March 2014. The relatively low numbers of stable flies collected in the H-traps indicate that the dung heaps do not have a significant impact on the feedlot regarding an influx of flies from the dung heaps to the feedlot. This is supported by literature that states that stable flies do not prefer pure dung as a breeding medium (Dawit *et al.*, 2012; Sutherland, 1978).

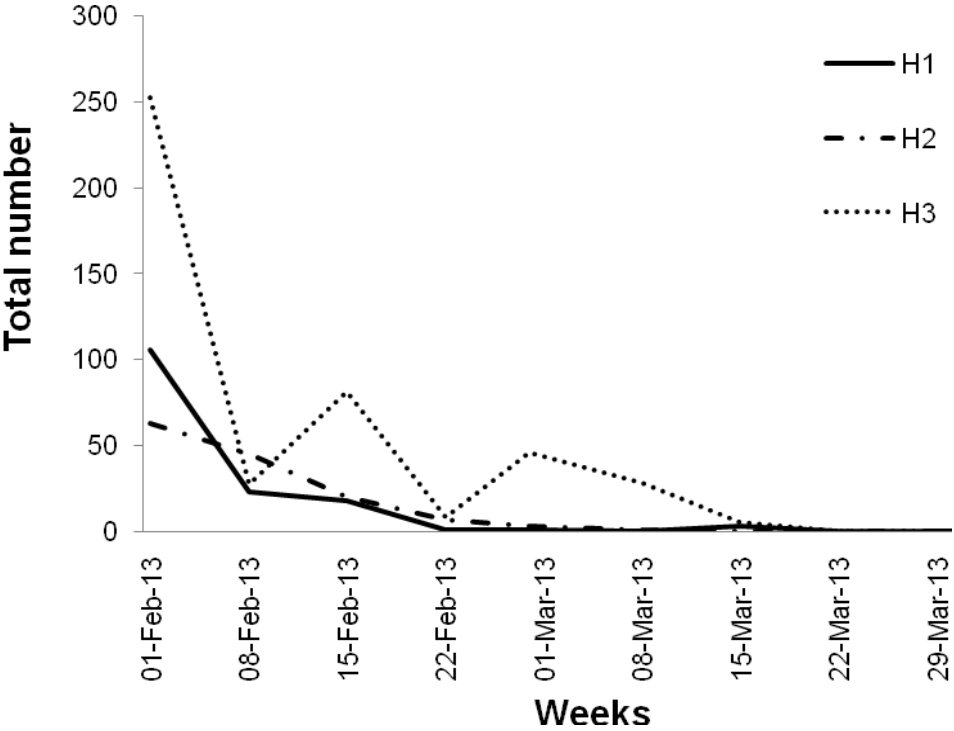


Figure 3.21: Influence of dung heaps on the total number of *S. calcitrans* collected in H-traps placed along a gradient from the dung heaps to the feedlot from 1 February 2013 to 4 April 2013.

CHAPTER 4

CONCLUSION

4.1 CONCLUSIONS AND RECOMMENDATIONS FOR EVERY OBJECTIVE

4.1.1 A sustainable sampling method for *S. calcitrans*

The NZI-tsetse type trap proves to be effective for the collecting of stable flies. This trap is durable and does not need fly bait to attract stable flies. These traps have been used in literature (Mihok, 2002; 2006) as a control method. The two tsetse type traps (NZI and Vavoua) were compared and although no significant difference in the number of flies collected in the traps was found, the NZI traps did seem to be more effective. NZI traps could be affectively used in monitoring stable fly populations in feedlots with relative ease.

4.1.2 The seasonal and daily abundance (temporal distribution) of *S. calcitrans*

Stable flies showed distinct seasonal distribution patterns. The highest numbers of flies collected were associated with summer months. Flies were present in the traps from 19 October to 20 September 2013 with five peaks occurring. Winter months were associated with very low to zero numbers of flies. Summer months should be closely monitored, especially from December to February for increased fly activity and associated irritation effects. However, economic threshold levels is needed to be adapted for feedlot specific conditions in the integrated management of nuisance fly management. There is not a visible pattern in the daily distribution of flies and more research is needed to identify daily distribution in fly activity to establish targeted timeframes for possible effective insecticide application. The preliminary recommendations from data obtained in this study is: applications of insecticides to reduce irritation during summer peaks, if threshold levels are exceeded, should be done early mornings from about 8:00 until 10:30, noon between 13:00 and 14:30 and late afternoon from 16:30 until 18:00. However, further studies are necessary to identify methods to move away from applying insecticides on cattle to breeding and resting sites of stable flies. Unnecessary and ineffective use of chemicals should be avoided which should enhance the natural mortality factors such as natural enemies. Insecticides should be used only according to economic threshold levels still to be determined as adult stable flies on cattle forelegs

4.1.3 *S. calcitrans* distribution within the feedlot (spatial distribution)

The results indicated a distinct spatial distribution of stable flies in the feedlot. A statistical significant edge effect was indicated, where more stable flies were found in the edge pens of the feedlot surrounded by vegetation, manure runoff and manure holding ponds than in the K-line in the middle pens of the feedlot. The weeds surrounding the edges need to be kept under control; it is a possible source of the fly infestation and irritation for cattle in the pens as it serves as resting places for adults (Urech *et al.*, 2011).

Some observations about the optimal feedlot design include that the drainage canals at the upper and lower end of the feedlot could be a possible larval development sites for flies and if treated with the necessary chemicals, could reduce the number of flies reaching adulthood. The use of growth regulators should be encouraged as is already the practice in the feedlot. The manure run off channels should be well managed to reduce vegetation growth which could be resting and larval development sites for the flies (Urech *et al.*, 2004; 2011). Many pupae were found in dung that accumulated underneath the fence lines. These pupae were found to be mainly houseflies. Management of dung under these cables by growth regulators and possible release of parasitoids would reduce nuisance fly populations. (Urech *et al.*, 2004; 2011). Manure heaps, approximately five kilometres from the feedlot, did not appear to have an effect on the feedlot in terms of stable flies. The impact should be continued to be monitored. The large number of hay bales and the hay in the pens should be managed properly to avoid spills. Hay spills in the pens together with dung and moisture becomes an ideal substrate for stable fly breeding (Meyer & Peterson, 1983).

4.1.4 *S. calcitrans* density on cattle during the season

Stable fly activity can also be monitored by animal observations by counting the number of flies on cattle forelegs/minute. According to literature if the number exceeds five flies/foreleg/minute there is a chance of a negative effect on the cattle's weight gain. (Campbell *et al.*, 1987; 2001). The maximum mean number of flies counted/foreleg/minute in this study was 1.8 and it seems as if the population density of stable flies is relatively low in comparison with literature. However, the adaption of of the equation used to develop a site specific economic threshold is needed. Irritation indicators asociated with stable fly densities corresponds with literature Urech *et al.* (2012) and the proven correlation between trap collections and stable flies on cattle can be used in management control decisions. The significant correlation found between stable fly activity and avoidance behaviour, can be used in effective structured animal observations to estimate the effect of fly populations on cattle behaviour (Warnes & Finlayson, 1987; Mullens *et*

al., 2006) and can be used as an early warning system to apply pesticides according to a determined economic threshold.

4.1.5 Correlation between trap collections, bunching and fly counts on cattle

There was a strong correlation between the number of flies collected in the traps and the flies counted on the cattle forelegs/minute. There was also a positive correlation between the number of flies counted/foreleg/minute and the number of foot stomps per minute. This is an indication that avoidance behaviours such as foot stomping and bunching are a good indication of the irritation caused by the flies and that these visible behaviours may be used to monitor the fly populations to apply chemicals to an observed threshold value (Berry *et al.*, 1983, Wieman *et al.*, 1992; Dougherty *et al.*, 1993; 1994; Mullens *et al.*, 2006).

4.2 FURTHER RESEARCH NEEDED

Further research needed in the development of an integrated fly management program:

- The adaptation of the equation used in the development of an economic threshold for stable fly control needs to be developed
- Finding stable fly larval development sites for controlling immature stages of the stable fly needs to be determined.
- Enhancing existing biological control factors and possible introduced biological control measures needs to be investigated to reduce the dependency on insecticides in fly control.
- The efficiency of baited fly nets needs to be investigated.
- The possible weight impact of stable fly irritation needs to be investigated.
- Calculations of the degree days of the stable flies can assist in predicting stable fly population peaks. Control methods to reduce the irritation impacts of the flies can be adjusted to these predictions.
- The possible impact of disease transmission by flies needs to be investigated.
- The integration of results obtained so far into an integrated fly management program and possible way to streamline stable fly monitoring, should be a priority.

4.3 FINAL CONCLUSION

This study addresses the research question in the following ways. The diversity and feedlot importance of the most abundant diptera species collected by tsetse type traps was determined. The seasonal abundance of stable flies were determined and could be used to predict stable fly infestations. The irritation impacts were determined by establishing the mean number of stable flies on cattle forelegs per minute as well as foot stomps. This information is important to determine an economic threshold. The spatial distribution of stable flies in the feedlot indicated a clear edge effect which has valuable implications integrated fly management .

REFERENCES

- Abba, J.O., Mwansat, G.S. & Goselle, O.N. 2011. The distribution, relative-abundance and the efficiency of traps of haematophagous flies at Assop Forest, plateau state, Nigeria. *Nigerian Annals of Natural Sciences* 11(1):13-21.
- AlbertaCanada. 2014. Climate and Geography.
<http://www.albertacanada.com/opportunity/choosing/province-climate-geography.aspx> Date of access: 4 April 2014.
- Anthony, C. 2005. Control of stable flies and house flies. Cooperative Extension Service Publ. 2045, College of Agriculture & Biological Sciences, South Dakota State University., USDA.
- Armitage, P.D., Cranston, P. & Pinder, L.C. 1995. The Chironomidae: Biology and Ecology of Non-Biting Midges. Springer, Nature - 572 pages. London: Chapman & Hall,
- Beerwinkle, K.R., Berry, I.L. & Kunz, S.E. 1978. Prediction models for mortality of immature stable flies caused by cold temperatures. *Environmental Entomology* 7:273-277.
- Beresford, D.V. & Sutcliffe, J.F. 2012. Field measurements of stable fly (Diptera: Muscidae) demography, fecundity, and survival based on daily trap collections at a beef farm in southern Ontario over a 5-yr period. *Medical Entomology* 49(6):1262-1269.
- Berkebile, D.R., Thomas, G.D. & Campbell, J.B. 1994. Overwintering of the stable fly (Diptera: Muscidae) in Southeastern Nebraska. *Journal of Economic Entomology* 87:1555-1563.
- Berry, I.L. & Campbell, J.B. 1985. Time and weather effects on daily feeding patterns of stable flies (Diptera: Muscidae). *Environmental Entomology* 14:336-342.
- Berry, I.L., Foerster, K.W. & Campbell, J.B. 1978. Overwintering behavior of stable flies in manure mounds. *Environmental Entomology* 7:67-72.
- Berry, I.L., Nelson, A.K. & Broce, A.B. 1986. Effect of weather on capture of stable flies (Diptera: Muscidae) by Alsynite fiber glass traps. *Environmental Entomology* 15:706-709.

- Berry, I.L., Stage, D.A. & Campbell, J.B. 1983. Populations and economic impacts of stable flies on cattle. *Transactions of the American Society of Agricultural Engineers* 26:873-877.
- Black, W.S. IV & Krasfur, E.S. 1985. Use of sticky traps to investigate seasonal trends in the spatial distribution of house flies and stable flies (Diptera: Muscidae). *Journal of Medical Entomology* 22:550-557.
- Boxler, D.J. 2013. Extension Educator West Central Research & Extension Center University of Nebraska–Lincoln, North Platte, NE.<http://beef.unl.edu/cattleproduction/controllingflies> Date of access: 6 February 2014.
- Broce, A.B. 1988. An improved Alsynite trap for stable flies, *Stomoxys calcitrans* L. (Diptera: Muscidae). *Journal of Medical Entomology* 25:406-409.
- Bruce, W.N. & Decker, G.C. 1958. The relationship of stable fly abundance to milk production to dairy cattle. *Journal of Economic Entomology* 51:269-174.
- Buck, M. 1996. Sphaeroceridae (Diptera) reared from various types of carrion and other decaying substrates in Southern Germany, including new faunistic data on some rarely collected species. *European Journal of Entomology* 94:137-152.
- Buschman, L.L. & Patterson, R.S. 1981. Assembly, mating, and thermo regulating behaviour of stable flies under field conditions. *Environmental Entomology* 10:16-21.
- Campbell, J.B., Berry, L., Boxler, D.J., Davis, R.L., Clanton, D.C. & Deutscher, G.H. 1987. Effects of stable flies (Diptera: Muscidae) on weight gain and feed efficiency of feedlot cattle. *Journal of Economic Entomology* 80:117-119.
- Campbell, J.B., Catangui, M.A., Thomas, G.D., Boxler, D.J. & Davis, R. 1993. Effects of stable flies (Diptera: Muscidae) and heat stress on weight gain and feed conversion of feeder cattle. *Journal of Agricultural Entomology* 10:155-161.
- Campbell, J.B. & Hermanussen, J.F. 1971. Efficacy of insecticides and methods of insecticidal applications for control of stable flies in Nebraska. *Journal of Economic Entomology* 64:1188-1190.

Campbell, J.B., Skoda, S.R., Berkebile, D.R., Boxler, D.J., Thomas, G.D., Adams, D.C. & Davis, R. 2001. Effects of stable flies (Diptera: Muscidae) on weight gains of grazing yearling cattle. *Journal of Economic Entomology* 94(3):780-783.

Campbell, J.B., White, R.G., Wright, J.E., Crookshank, R. & Clanton, D.C. 1977. Effects of stable flies on weight gains and feed efficiency of calves on growing or finishing rations. *Journal of Economic Entomology* 70(5):592–594.

Catangui, M.A., Campbell, J.B., Thomas, G.D. & Boxler, D.J. 1993. Average daily gains of Brahman-Crossbred and English x Exotic Feeder Heifers exposed to low, medium and high levels of stable flies (Diptera: Muscidae). *Journal of Economic Entomology* 86(4):1144-1150.

Catangui, M.A., Campbell, J.B., Thomas, G.D. & Boxler, D.J. 1995. Average daily gains of Brahman-crossbred and English X exotic feeder heifers during long-term exposure to stable flies (Diptera: Muscidae). *Journal of Economic Entomology* 88:1349-1352.

Catangui, M.A., Campbell, J.B., Thomas, G.D. & Boxler, D.J. 1997. Calculating economic injury levels for stable flies (Diptera: Muscidae) on feeder heifers. *Journal of Economic Entomology* 90(1):6-10.

Cilek, J.E. & Greene, G.L. 1994. Stable fly (Diptera: Muscidae) insecticide resistance in Kansas cattle feedlots. *Journal of Economic Entomology* 87(2):275-279.

Clymer, B.C. 1992. The stable fly as a pest in beef cattle feedlots, *In* G.D. Thomas and S.R. Skoda (eds.). *The stable fly: A pest of humans and domestic animals. Proceedings of the Entomological Society of America, Baltimore* 9-11.

Coetzer, J.A.W. 2004. Lumpy skin disease. *In: Infectious Diseases of Livestock, Second Edition.* Coetzer J.A.W. & Justin R.C., eds. Cape Town, South Africa: Oxford University Press, 1268–1276.

Coffey, M.D. 1966. Studies on the Association of Flies (Diptera) with Dung in South-eastern Washington. *Annals of the Entomological Society of America* 59(1):207-218.

Cruz-Vasquez, C., Mendoza, I.V., Parra, M.R. & Garzia-Vasquez, Z. 2004. Influence of temperature, humidity and rainfall on field population trend of *Stomoxys calcitrans* (Diptera: Muscidae) in a semiarid climate in Mexico. *Parasitología Latinoamericana* 59:99-103.

- D'arcy-Burt, S. & Blackshaw, R.P. 1991. Bibionids (Diptera: Bibionidae) in agricultural land: A review of damage, benefits, natural enemies and control. *Annals of Applied Biology*. 118(3):695–708.
- Dawit, L., Addis, M. & Gari, G. 2012. Distribution, seasonality and relative abundance of *Stomoxys* flies in selected districts of Central Ethiopia. *World Applied Sciences Journal* 19(7):998-1002.
- Dougherty, C.T., Knapp, F.W., Burrus, P.B., Willis, D.C., Cornelius, P.L & Bradley, N.W. 1993. Multiple releases of stable flies (*Stomoxys calcitrans*) and behaviour of grazing beef cattle. *Applied Animal Behavioural Sciences* 38(40):191-212.
- Dougherty, C.T., Knapp, F.W., Burrus, P.B., Willis, D.C. & Cornelius, P.L. 1994. Moderation of grazing behavior of beef cattle by stable flies (*Stomoxys calcitrans* L.). *Applied Animal Behavioural Sciences* 40:113-127.
- Dougherty, C.T., Knapp, F.W., Burrus, P.B., Willis, D.C. & Cornelius, P.L. 1995. Behavior of grazing cattle exposed to small populations of stable flies (*Stomoxys calcitrans* L.). *Applied Animal Behaviour Science* 42:231–248.
- Eicher, S.D., Morrow-Tesch, J.L., Albright, J.L. & Williams, R.E. 2001. Tail-docking alters fly numbers, fly-avoidance behaviours, and cleanliness, but not physiological measures. *Journal of Dairy Science* 84:1822-1828.
- Ferrar, P. 1987. A guide to the breeding habits and immature stages of Diptera Cyclorrhapha. *Entomonograph* 8:222-223.
- Foil, L.D. & Hogsette, J.A. 1994. Biology and control of Tabanids, stable flies and horn flies. In Ectoparasites of animals and control methods. *Scientific and Technical Review of the Office International des Epizooties (Paris)* 13:1125-1158.
- Gerry, A.C., Peterson N.G. & Mullens B.A. 2007. Predicting and controlling stable flies on California dairies. *University of California Division of Agriculture and Natural Resources*. Res. Publ. 8258.
- Gersabeck, E.F. & Merritt, R.W. 1983. Vertical and temporal aspects of Alsynite panel sampling for adult *Stomoxys calcitrans* (L.) (Diptera: Muscidae). *Florida Entomologist* 66:222-227.

Gilles, J., David, J.F. & Duvallet, G. 2005. Temperature effects on development and survival of two stable flies, *Stomoxys calcitrans* and *Stomoxys niger niger* (Diptera: muscidae), in La Réunion Island. *Journal of Medical Entomology* 42(3):260-265.

Greene, G.L., Hogsette, J.A. & Patterson, R.S. 1989. Parasites that attack stable fly and house fly (Diptera:Muscidae) puparia during the winter on dairies in northwestern Florida. *Journal of Economic Entomology* 82:412-418.

Guo, Y., Greene, G.L. & Butine, M.D. 1998. Population profile of stable flies (Diptera: Muscidae) collected on Alsynite traps in various feedlot habitats. *Journal of Economic Entomology* 91:159-164.

Hall, R.D., Thomas G.D., Berry, I.L., Fischer, F.J & Foehse, M.C. 1983. Relative abundance of stable flies, *Stomoxys calcitrans* (L.) (Diptera: Muscidae), at dairies, feedlots and pastures in Missouri. *Journal of Kansas Entomology Society* 56:223-228.

Harms, D.A. 2004. "Using Degree Days and Plant Phenology to Predict Pest Activity" in: IPM of Midwest Landscapes. MN Agriculture Experiment Station. 49-59.

Hogsette, J.A., Ruff, J.P. & Jones, C.J. 1987. Stable Fly Biology and Control in Northwest Florida. *Journal of Agricultural Entomology* 4:1-11.

Holdsworth, P.A., Vercruysse, J., Rehbein, S., Peter, R.J., De Bruin, C., Letonja, T. & Green, P. 2006. World Association for the Advancement of Veterinary Parasitology (W.A.A.V.P) guidelines for evaluating the efficacy of ectoparasites against biting and nuisance flies on ruminants. *Veterinary Parasitology* 136:3-13.

Holloway, M.T.P. & Phelps, R.J. 1991. The responses of *Stomoxys spp.* (Diptera: Muscidae) to traps and artificial host odours in the field. *Bulletin for Entomological Research* 80:51-55.

Johnson, G. 2011. Managing Stable Flies on Livestock and Around Livestock Facilities by Professor of Veterinary Entomology, Montana State University Bozeman File under: Agriculture and Natural Resources (Pest Management) New November 2011 300-1111SA.

Jones, C.M. & Anthony, D.W. 1964. The Tabanidae (Diptera) of Florida. Technical bulletin no. 1295 Agricultural Research Service United States Department of Agriculture In cooperation with Florida Agricultural Experiment Station Washington, DC.

Jones, S.R. & Kunz, S.E. 1997. Importance of supercooling points in the overwintering of the horn fly and stable fly (Diptera: Muscidae). *Journal of Medical Entomology* 34:426-429.

Kneeland, K.M., Skoda, S.R., Hogsette, J.A., Li, A.Y., Molina-Ochoa, J., Lohmeyer, K.H. & Foster, J.E. 2012. A Century and a Half of Research on the Stable Fly, *Stomoxys calcitrans* (L.) (Diptera: Muscidae), 1862-2011: An Annotated Bibliography. Department of Agriculture, Agricultural Research Service, ARS-173.

Kowalsick, T. & Clark, S. 2006. Using Growing Degree Days for Insect Pest Management. *Cornell Cooperative Extension*.

Krafsur, E.S., Moon, R.D & Lysyk, T.J. 1994. Adult Age And Reproductive Success In Stable Fly Populations (Diptera: Muscidae). *The Canadian Entomologist* 126(2):239-249.

Kunz, S.E., Murrell, K.D., Lambert, G., James L.F. & Terrill, C.E. 1991. Estimated losses of livestock to pests, pp. 69-98. In D. Pimentel (ed.), CRC handbook of pest management in agriculture, vol. I. CRC, Boca Raton, FL.

Lazarus, W.F., Rutz, D.A., Miller, R.W. & Brow, D.A. 1989. Costs of Existing and Recommended Manure Management Practices for House Fly and Stable Fly (Diptera: Muscidae) Control on Dairy Farms. *Journal of Economic Entomology* 82(4):1145-1151.

Lysyk, T.J. 1993. Seasonal abundance of stable flies and house flies (Diptera: Muscidae) in dairies in Alberta, Canada. *Journal of Medical Entomology* 30(5):888-895

Lysyk, T. J. 1998. Relationships between temperature and life-history param of *Stomoxys calcitrans* (Diptera: Muscidae). *Journal of Medical Entomology* 35:107-119.

Marçon, P.C.R.G., Thomas, G.D., Siegfried, B.D. & Campbell, J.B. 1997. Susceptibility of stable flies (Diptera: Muscidae) from Southeastern Nebraska beef cattle feedlots to selected insecticides and comparison of 3 bioassay techniques. *Journal of Economic Entomology* 90:293-298.

Masmeatathip, R., Gilles, J., Ketavan, C. & Duvallet, G. 2006. First survey of seasonal abundance and daily activity of *Stomoxys* spp. (Diptera: Muscidae) in Kamphaengsaen Campus, Nakornpathom Province. *Thailand Parasite* 13(3):245-250.

McNeal, C.D., JR. & Campbell, J.B. 1981. Insect pest management in Nebraska feedlots and dairies: A pilot integrated pest management project. Cooperative Extensive Service, Report 10, University of Nebraska, Lincoln.

Memmi, B.K. 2010. Mortality and knockdown effects of imidacloprid and methomyl in house fly (*Musca domestica* L., Diptera: Muscidae) populations. *Journal of Vector Ecology* 35:144-148.

Meowweather. 2014. Heidelberg, Gauteng, South Africa.

<http://www.meowweather.com/history/South%20Africa/na/-26.5/28.35/Heidelberg.html?units=c#>

Date of access: 4 April 2014.

Meyer, J.A. & Peterson, J.J. 1983. Characterization and seasonal distribution of larval development sites of stable flies and house flies (Diptera: Muscidae) on Eastern Nebraska feedlots and dairies. *Journal of Economic Entomology* 76:103-108.

Mihok, S. 2002. The development of a multipurpose trap (the NZI) for tsetse and other biting flies. *Bulletin of Entomological Research* 92(5):385-403.

Mihok, S., Carlson, D.A., Krafur, E.S. & Foil, L.D. 2006. Performance of the NZI and other traps for biting flies in North America. *Bulletin of Entomological Research* 96:387-397.

Mihok, S., Kang'ethe, E.K. & Kamau, G.K. 1995. Trials of traps and attractants for *Stomoxys* spp. (Diptera: Muscidae). *Journal of Medical Entomology* 32(3):283-9.

Miller, R.W., Pickens, L.G. & Potts, W.E. 1993. Comparison of traps and an integrated program to manage house flies and stable flies on dairy farms. *Journal of Agricultural Entomology* 10:189-196.

Morgan, C.E., Thomas, G.D. & Hall, R.D. 1983. Annotated bibliography of the stable fly, *Stomoxys calcitrans* (L.), including references on other species belonging to the genus *Stomoxys*. North Central Regional Res. Publication No. 291. University of Missouri Agricultural Experiment Station Research Bulletin 1049, Columbia.

Muenworn, V., Duvallat, G., Thainchum, K., Tuntakom, S., Tanasilchayakul, S., Prabaripai, A., Akkratanakul, P., Sukonthabhirom, S. & Chareonviriyaphap, T. 2010. Geographic distribution of Stomoxyine flies (Diptera: Muscidae) and diurnal activity of *Stomoxys calcitrans* in Thailand. *Journal of Medical Entomology* 47:791-797.

Mullens, B.A. & Meyer, J.A. 1987. Seasonal abundance of stable flies (Diptera: Muscidae) on California dairies. *Journal of Economic Entomology* 80(5):1039-1043.

Mullens, B.A., Lii, K.S., Mao, Y., Meyer, J.A., Peterson, N.G. & Szijj, C.E. 2006. Behavioral responses of dairy cattle to the stable fly, *Stomoxys calcitrans*, in an open field environment. *Medical and Veterinary Entomology* 20:122-137.

Mullens, B.A. & Peterson, N.G. 2005. Relationship between rainfall and stable fly (Diptera: Muscidae) abundance on California dairies. *Journal of Medical Entomology* 42(4):705-708.

Murray, S.M. 2008. Using Degree Days to Time Treatments for Insect Pests. Published by Utah State University Extension and Utah Plant Pest Diagnostic Laboratory.

Newson, H. D. 1977. Arthropod problems in recreation areas. *Annual Review of Entomology* 22:333-353.

SAexplorer. 2011. Heidelberg (GT) climate.

[http://www.saexplorer.co.za/south-africa/climate/heidelberg_\(gt\)_climate.asp](http://www.saexplorer.co.za/south-africa/climate/heidelberg_(gt)_climate.asp) Date of access: 28 March 2014.

Scholl, P.L. 1986. Field population studies of *Stomoxys calcitrans* (L.) in eastern Nebraska. *Southwestern Entomologist* 11:155-160.

Scholl, P.J., Petersen, J.J., Stage, D.A. & Meyer, J.A. 1981. Open silage as an overwintering site for immature stable flies in eastern Nebraska. *Southwestern Entomologist* 6:253-258.

Scholl, P.J., Lowry, S.R. & Rabe, G.G. 1985. Modified Williams sticky traps used to measure activity of adult stable flies, *Stomoxys calcitrans* (L.) in eastern Nebraska. *Southwest Entomologist* 14:881-890.

Schwinghammer, K.A., Knapp, F.W., Boling, J.A. & Schillo, K.K. 1986. Physiological and nutritional response of beef steers to infestations of the stable fly (Diptera: Muscidae). *Journal of Economic Entomology* 79(5):1294-1298.

Semakula, L.M., Taylor, R.A.J. & Pitts, C.W. 1989. Flight behavior of *Musca domestica* and *Stomoxys calcitrans* (Diptera: Muscidae) in a Kansas dairy barn. *Journal of Medical Entomology* 26:501-509.

Skidmore, P. 1985. The Biology of the Muscidae of the World. Springer. *Science* - 550 pages.

SouthAfrica.com. 2014. The climate of Gauteng province.

<http://www.southafrica.com/gauteng/climate/> Date of access: 14 September 2014.

Sutherland, B. 1973. The suitability of various types of dung and vegetable matter as larval breeding media for *Stomoxys calcitrans* L. (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research* 47:7-11.

Sutherland, B. 1978. The suitability of various types of dung and vegetable matter as larval breeding media for *Stomoxys calcitrans* L. (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research* 45:241-243.

Sutherland, B. 1979. The effects of temperature on the adults, eggs and pupae of *Stomoxys calcitrans* Linnaeus (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research* 46:223-227.

Sutherland, B. 1980. The temperature preferences of the motile stages of *Stomoxys calcitrans* Linnaeus (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research* 47:7-11.

Taylor, D.B. & Berkebile, D.R. 2006. Comparative efficiency of six stable fly (Diptera: Muscidae) traps. *Journal of Economic Entomology* 99:1415-1419.

Taylor, D.B. & Berkebile, D.R. 2011. Phenology of stable fly (Diptera: Muscidae) larvae in round bale hay feeding sites in Eastern Nebraska. *Environmental Entomology* 40:184-219.

Taylor, D.B., Berkebile, D.R. & Scholl, P.J. 2007. Stable fly population dynamics in Eastern Nebraska in relation to climatic variables. *Journal of Medical Entomology* 44:765-771.

Taylor, D.B., Friesen, K. & Zhu, J.J. 2013. Spatial-Temporal Dynamics of Stable Fly (Diptera: Muscidae) Trap Collections in Eastern Nebraska. *Environmental Entomology* 42(3):524-531.

Taylor, D.B., Moon, R.D. & Darrell, R.M. 2012. Economic impact of stable flies (Diptera: Muscidae) on dairy and beef cattle production. *Journal of Medical Entomology* 49(1):198-209.

Thomas, G.D. 1993. The influence of beef cattle feedlots on the urban fly problem, pp. 1-6. In G.D. Thomas and S.R. Skoda (eds.). Rural flies in the urban environment? Proc. of a

Symposium, 1989 annual meeting of the ESA; N. Cent. Reg. Publ. No. 335. Agric. Research Div., Institute of Agric. and Natural Resources, Univ. of Nebraska Rs. Bull. No. 317.

Thomas, G.D., Berry, I.L., Berkebile, D.R. & Skoda, S.R. 1989. Comparison of three sampling methods for estimating adult stable fly (Diptera: Muscidae) populations. *Environmental Entomology* 18:513-520.

Thomas, G.D. & Skoda, S.R. 1993. Rural flies in the urban environment. North Central Regional Publication, University of Nebraska, Lincoln, No. 335.

Thomas, G.D., Skoda, S.R., Berkebile, D.R. & Campbell, J.B. 1996. Scheduled sanitation to reduce stable fly (Diptera: Muscidae) populations in beef cattle feedlots. *Journal of Economic Entomology* 89:411-414.

Tsetse.org. 2012. H-trap. <http://www.tsetse.org/FAQ/htrap.htm> Date of access: 18 April 2014.

Urech, R., Bright, R.L., Green, E.P., Brown, G.W., Hogsette, J.A., Skerman, A.G., Elson-Harris, M.M. & Mayer, D.G. 2012. Temporal and spatial trends in adult nuisance fly populations at Australian cattle feedlots. *Australian Journal of Entomology* 51:81-96.

Urech, R., Green, E.P., Skerman, A.G., Elson-Harris, M.M., Hogsette, J.A., Bright, R.L. & Brown, G.W. 2004. Management of nuisance fly populations on cattle feedlots. Department of Primary Industries and Fisheries, Queensland.

Urech, R., Green, E.P., Skerman, A.G., Elson-Harris, M.M., Hogsette, J.A., Bright, R.L., Brown, G.W., O'Shea, V., Everingham, G. & Leemon, D. 2011. Integrated pest management for nuisance flies in cattle feedlots. Department of Employment, Economic Development and Innovation, Queensland.

Warnes, M.L. & Finlayson, L.H. 1987. Effects of host behaviour on host preference in *Stomoxys calcitrans*. *Medical and Veterinary Entomology* 1:53-57.

Wieman, G.A., Campbell, J.B., Deshazer, J.A. & Berry, I.L. 1992. Effects of stable flies (Diptera: Muscidae) and heat stress on weight gain and feed efficiency of feeder cattle. *Journal of Economic Entomology* 85(5):1835-184

Appendix 1: The diversity of most abundant flies collected in a NZI tsetse type trap, green and blue Chinese traps and Redtop traps at Karan Beef on three days from 23 February 2012 to 12 March 2012.

	12/02/23			12/03/03				12/03/12			
	NZI	Green	Redtop	NZI	Green	Blue	Redtop	NZI	Green	Blue	Redtop
Tabanidae	32	0	0	2	0	0	0	8	0	0	0
<i>Stomoxys calcitrans</i>	100	1	0	40	33	10	0	200	13	7	0
<i>Musca domestica</i>	1	65	0	1	113	77	10	0	43	138	0
Scatopsidae	30	0	0	0	0	0	0	0	0	0	0
Bibonidae	0	200	0	0	249	207	0	0	28	40	0
Chironomidae	22	2	0	0	1	6	0	9	0	3	0
<i>Atherigona sp.</i>	0	1	0	0	1	0	0	0	0	0	0
Milichidae	0	1	0	0	0	0	0	13	1	0	0
Sphaeroceridae	0	0	0	0	0	0	0	0	0	0	0

Appendix 2: The total number of the most abundant flies collected in a NZI trap from February 2012 to May 2012.

	20 - 27 Feb	27 Feb- 4 Mar	5-11 Mar	12-18 Mar	19-25 Mar	26 Mar- 1 Apr	2-6 Apr	7-13 Apr	14-20 Apr	5-11 May	19-25 May	26 -28 May	Total
<i>Tabanus haematopota</i>	180	31	40	10	13	7	0	0	0	0	0	0	281
<i>Stomoxys calcitrans</i>	1508	451	654	302	37	777	17	40	18	0	3	9	3816
<i>Musca domestica</i>	6	6	12	6	2	8	2	1	1	0	0	2	46
Scatopsidae	67	5	2	0	2	5	0	0	0	0	2	1	84
Bibonidae	18	11	3	0	0	2	0	0	0	0	0	0	34
Chironomidae	0	8	18	17	17	57	0	0	0	1	1	2	121
<i>Atherigona sp.</i>	4	0	0	0	9	7	2	0	0	0	1	0	23
Milichidae	0	4	20	17	23	18	0	3	0	2	7	6	100
Sphaeroceridae	11	8	4	0	2	0	0	0	0	0	0	0	25

Appendix 3: The total and mean number of *S. calcitrans* collected per week in 6 NZI and 6 Vavoua traps in the H-line and K-line from 19 October 2012 to 20 September 2013.

	K-line			H-line			Total	Mean	K-line			H-line			Total	Mean	Total	Total	Total
	V 1	V 2	V 3	V 1	V 2	V 3	Vavoua	Vavoua	N1	N2	N3	N1	N2	N3	NZI	NZI	K line	H line	for all the traps
19-Oct-12	0	0	0	0	0	0	0	0.00	0	0	0	1	0	0	1	0.17	0	1	1
25-Oct-12	0	0	0	0	0	0	0	0.00	0	0	0	0	0	0	0	0.00	0	0	0
02-Nov-12	2	1	0	2	1	0	6	1.00	3	1	1	0	0	0	5	0.83	8	3	11
09-Nov-12	0	1	0	6	3	1	11	1.83	0	5	7	4	0	3	19	3.17	13	17	30
16-Nov-12	7	3	7	53	31	19	120	20.00	1	59	32	39	5	20	156	26.00	109	167	276
23-Nov-12	21	17	0	41	34	23	136	22.67	25	48	49	33	3	43	201	33.50	160	177	337
30-Nov-12	1	0	2	6	26	59	94	15.67	32	27	10	4	1	3	77	12.83	72	99	171
07-Dec-12	25	10	18	38	49	29	169	28.17	40	no data	35	7	3	19	104	20.80	128	145	273
14-Dec-12	35	9	26	24	33	50	177	29.50	35	41	112	157	12	32	389	64.83	258	308	566
21-Dec-12	62	9	40	77	80	118	386	64.33	64	29	133	210	41	51	528	88.00	337	577	914
28-Dec-12	146	37	48	145	138	no data	514	102.80	144	17	201	72	29	73	536	89.33	593	457	1050
04-Jan-13	46	20	20	93	125	119	423	70.50	no data	318	367	20	41	11	757	126.17	771	409	1180
11-Jan-13	96	31	62	836	153	401	1579	263.17	628	360	254	113	5	79	1439	239.83	1431	1587	3018
18-Jan-13	40	109	134	120	366	296	1065	177.50	142	174	481	205	342	305	1649	274.83	1080	1634	2714
25-Jan-13	37	57	99	75	155	103	526	87.67	73	33	100	142	112	243	703	117.17	399	830	1229
01-Feb-13	35	90	97	45	280	440	987	164.50	211	301	274	246	153	268	1453	290.60	1008	1432	2440
08-Feb-13	17	11	40	102	58	49	277	46.17	252	425	421	51	72	66	1287	214.50	1166	398	1564

15-Feb-13	24	31	60	13	27	419	574	95.67	246	401	342	232	100	164	1485	247.50	1104	955	2059
22-Feb-13	70	90	92	378	304	530	1464	244.00	642	187	413	91	46	49	1428	238.00	1494	1398	2892
01-Mar-13	12	26	13	215	128	69	463	77.17	143	132	169	109	102	65	720	120.00	495	688	1183
08-Mar-13	37	10	18	103	59	91	318	53.00	315	190	136	113	13	26	793	132.17	706	405	1111
15-Mar-13	5	15	2	58	44	29	153	25.50	221	107	126	40	9	27	530	88.33	476	207	683
22-Mar-13	11	30	1	35	56	54	187	31.17	208	140	76	32	no data	32	488	97.60	466	209	675
29-Mar-13	34	36	10	77	135	97	389	64.83	404	367	357	65	40	88	1321	220.17	1208	502	1710
05-Apr-13	28	11	3	94	63	79	278	46.33	77	150	134	111	7	13	492	82.00	403	367	770
12-Apr-13	13	2	1	23	15	13	67	11.17	18	40	21	50	6	7	142	23.67	95	114	209
19-Apr-13	19	8	3	41	41	14	126	21.00	79	19	101	10	5	0	214	35.67	229	111	340
26-Apr-13	12	1	0	37	50	65	165	27.50	67	44	134	26	6	6	283	47.17	258	190	448
03-May-13	5	0	2	76	227	68	378	63.00	130	157	372	51	11	8	729	121.50	666	441	1107
10-May-13	0	1	1	21	5	7	35	5.83	81	40	106	6	0	2	235	39.17	229	41	270
17-May-13	14	4	1	68	34	70	191	31.83	83	68	72	27	28	9	287	47.83	242	236	478
24-May-13	15	3	3	30	24	19	94	15.67	51	112	39	38	6	12	258	43.00	223	129	352
07-Jun-13	1	1	1	1	14	0	18	3.00	50	3	28	127	97	96	401	66.83	84	335	419
14-Jun-13	0	0	0	0	0	0	0	0.00	10	30	27	5	0	0	72	12.00	67	5	72
21-Jun-13	0	0	0	0	0	0	0	0.00	0	0	0	15	9	6	30	8.57	0	30	30
28-Jun-13	0	0	0	0	0	0	0	0.00	0	0	0	0	0	0	0	0.00	0	0	0
05-Jul-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
12-Jul-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

19-Jul-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
26-Jul-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
02-Aug-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
09-Aug-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
16-Aug-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
23-Aug-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
30-Aug-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
06-Sep-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
13-Sep-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
20-Sep-13		0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total							11370							19212		15978	14604	30582	

Appendix 4: The total and mean number of flies collected per week in the H- and K-line, the total and mean number flies collected in only the H-line and climate data -mean weekly minimum and maximum temperature, mean weekly temperature from 19 October 2012 to 31 May 2013.

	Total <i>S.calcitrans</i> / 12 traps collections	Total traps H-line(6)	Mean H-line (6)	Minimum temperature	Wind Speed 07:00	Maximum temperature	Wind Speed 12:00	Rain mean per week	Rain farm meter mean per week	Mean temperature	Total rainfall	Total rainfall farm meter
19-Oct-12	1	0	0.00	14.71	5.29	24.09	10.56	3.59	4.29	21.31	25.14	30.00
25-Oct-12	0	0	0.00	13.60	6.90	21.30	14.50	4.53	5.14	18.54	31.74	36.00
02-Nov-12	11	8	1.33	13.40	10.56	22.13	12.87	0.14	0.00	18.97	1.00	0.00
09-Nov-12	30	22	3.67	16.30	9.19	26.10	15.41	0.59	0.71	22.54	4.10	5.00
16-Nov-12	276	195	32.50	17.73	8.27	26.71	10.34	0.86	1.14	23.26	6.00	8.00
23-Nov-12	337	220	36.67	18.29	8.74	30.39	14.27	4.54	5.57	26.25	31.80	39.00
30-Nov-12	171	160	26.67	15.83	3.69	25.20	9.87	2.03	2.71	22.54	14.20	19.00
07-Dec-12	273	191	38.20	15.34	5.74	23.10	15.41	8.15	9.43	20.08	57.07	66.00
14-Dec-12	566	295	49.17	16.33	2.76	24.24	8.96	5.44	7.00	21.30	38.08	49.00
21-Dec-12	914	501	83.50	17.06	7.60	27.27	7.13	0.22	0.14	23.72	1.51	1.00
28-Dec-12	906	645	129.00	18.69	3.91	26.93	5.96	3.12	4.71	23.74	21.82	33.00
04-Jan-13	1180	1022	204.40	19.36	3.90	28.71	7.13	4.97	5.71	25.51	34.80	40.00
11-Jan-13	3018	2632	438.67	19.59	4.36	28.09	7.81	1.01	1.57	25.60	7.09	11.00
18-Jan-13	2714	1579	263.17	17.69	5.30	25.29	9.64	1.92	3.00	23.20	13.43	21.00
25-Jan-13	1229	539	89.83	17.13	0.91	26.64	7.34	1.33	1.43	23.97	9.34	10.00
01-Feb-13	1456	1504	300.80	18.38	3.73	27.58	7.75	4.24	4.86	25.24	29.71	34.00
08-Feb-13	1564	1307	217.83	16.01	1.60	26.14	7.81	1.81	2.43	22.95	12.68	17.00
15-Feb-13	2059	1448	241.33	17.07	2.51	28.17	5.04	2.40	2.86	23.76	16.77	20.00
22-Feb-13	2892	2454	409.00	16.57	1.37	27.96	6.64	1.96	2.29	24.90	13.70	16.00
01-Mar-13	1183	856	142.67	15.44	3.44	28.36	8.30	1.63	2.00	25.13	0.00	0.00
08-Mar-13	1111	894	149.00	15.06	2.99	25.80	5.51	2.47	2.86	22.11	17.27	20.00
15-Mar-13	683	585	97.50	15.31	3.20	24.00	7.36	1.85	3.71	21.93	12.94	26.00
22-Mar-13	675	569	94.83	13.14	4.83	26.47	8.29	0.43	1.00	22.91	3.04	7.00
29-Mar-13	1710	1437	239.50	14.59	5.74	26.54	7.11	1.63	2.00	22.44	11.43	14.00

05-Apr-13	770	597	99.50	14.53	2.76	22.09	7.59	1.45	1.43	19.24	10.15	10.00
12-Apr-13	209	130	21.67	7.71	0.69	23.57	6.67	0.69	1.00	18.91	4.82	7.00
19-Apr-13	340	295	49.17	10.17	3.44	22.67	6.41	27.68	38.33	18.79	83.04	115.00
26-Apr-13	448	397	66.17	6.83	0.00	16.40	4.37	0.00	0.00	13.95	0.00	0.00
04-May-13	1107	1030	171.67	5.51	0.00	22.39	4.34	0.00	0.00	17.46	0.00	0.00
10-May-13	262	260	43.33	2.16	0.00	18.71	4.81	0.80	1.29	13.71	5.57	9.00
17-May-13	343	395	65.83	4.21	0.46	19.77	8.71	0.00	0.00	14.60	0.00	0.00
24-May-13	313	275	45.83	4.74	0.00	19.17	7.13	0.00	0.00	14.31	0.00	0.00
31-May-13	210	48	8.00	4.40	0.00	21.54	7.59	0.00	0.00	15.80	0.00	0.00

Appendix 5.1: The total hourly collections of *S. calcitrans* in Vavoua and NZI traps in the H-line on days 20 February 2013.

	HV3	HN3	HV2	HN2	HV1	HN1	Total
09:30-10:30	17	45	22	4	12	35	135
10:30-11:30	3	18	9	8	14	21	73
11:30-12:30	2	8	3	4	4	7	28
12:30-13:30	5	6	12	7	14	31	75
13:30-14:30	12	16	9	9	23	35	104
14:30-15:30	6	19	8	3	9	11	56
15:30-16:30	9	19	5	2	11	21	67
16:30-17:30	4	17	7	11	25	51	115
17:30-18:30	17	16	12	0	20	4	69
18:30-19:30	15	5	4	0	5	5	34
Total	90	169	91	48	137	221	756

Appendix 5.2: The total hourly collections of *S. calcitrans* in Vavoua and NZI traps in the H-line on 21 February 2013.

	HV3	HN3	HV2	HN2	HV1	HN1	Total
09:30-10:30	10	24	12	1	10	16	73
10:30-11:30	2	0	7	3	2	5	19
11:30-12:30	0	4	4	0	3	12	23
12:30-13:30	5	1	9	0	2	17	34
13:30-14:30	5	1	9	1	3	1	20
14:30-15:30	9	0	4	17	8	20	58
15:30-16:30	9	4	4	7	20	18	62
Total	40	34	49	29	48	89	289

Appendix 5.3: The total hourly collections of *S. calcitrans* in Vavoua and NZI traps in the H-line on 05 March 2013.

	HV3	HN3	HV2	HN2	HV1	HN1	Total
09:30-10:30	0	9	9	4	2	4	28
10:30-11:30	1	5	2	3	4	4	19
11:30-12:30	3	4	6	4	4	4	25
12:30-13:30	2	1	1	5	0	5	14
13:30-14:30	1	5	6	6	8	9	35
14:30-15:30	3	12	8	2	3	5	33
15:30-16:30	3	6	5	7	1	12	34
16:30-17:30	0	5	2	4	1	12	24
17:30-18:30	1	8	1	3	1	3	17
18:30-19:30	1	5	1	0	3	11	21
Total	15	60	41	38	27	69	250

Appendix 6: Daily *S. calcitrans* collections in the H-line in 3 NZI and 3 Vavoua traps

	Vavoua 1	Vavoua 2	Vavoua 3	NZI 1	NZI 2	NZI 3	Total
09-Nov-12	3	2	1	0	0	0	6
16-Nov-12	12	19	13	2	3	6	55
30-Nov-12	7	18	8	0	10	4	47
07-Dec-12	8	8	16	4	17	16	69
14-Dec-12	8	11	5	7	3	35	69
21-Dec-12	77	80	118	64	29	133	501
04-Jan-13	29	47	39	3	42	54	214
11-Jan-13	50	151	83	46	74	97	501
25-Jan-13	79	20	20	0	7	20	146
08-Feb-13	13	27	38	42	32	44	196
15-Feb-13	35	54	26	10	9	67	201
21-Feb-13	71	55	58	140	27	60	411
15-Mar-13	19	13	20	21	18	5	96
22-Mar-13	0	10	9	18	27	9	73
04-Apr-13	7	4	4	11	4	12	42
14-May-13	6	1	4	6	1	13	31

Appendix 7: The total number of *S. calcitrans* collected in 3 H-traps along the dung heaps approximately 5 km from the feedlot

	H1	H2	H3
01-Feb-13	106	63	252
08-Feb-13	23	45	27
15-Feb-13	18	20	81
22-Feb-13	1	7	8
28-Feb-13	1	3	46
08-Mar-13	0	1	28
15-Mar-13	3	0	5
22-Mar-13	0	0	0
29-Mar-13	0	0	0
04-Apr-13	0	0	0

Appendix 8: The total number of daily trap collections of *S. calcitrans* in 3 NZI and 3 Vavoua traps in the H-line, mean *S. calcitrans* counted daily on cattle forelegs per minute in the H-line and KI-line and daily climate data from 19 November 2012 to 14 May 2013.

	Total H-line counts/foreleg /min	Total K-line counts/foreleg /min	Mean counts/foreleg /min/48 cattle in H-line	Total daily collections in 6 traps in H-line	Wind Speed	Temp (Min)	Temp (Max)	Temp (Mean)
9-Nov-12	6	0	0.13	6	29	13.9	21.3	17.6
16-Nov-12	32	0	0.67	55	8	20.6	28.8	24.7
30-Nov-12	15	0	0.31	47	16.1	17.6	28.3	22.95
7-Dec-12	11	6	0.23	69	12.9	15.9	25.8	20.85
14-Dec-12	37	12	0.77	69	8	18.4	25.1	21.75
4-Jan-13	67	16	1.5	214	3.2	17.8	29.1	23.45
11-Jan-13	77	29	1.60	501	14.5	18.9	25.5	22.2
25-Jan-13	90	61	1.88	416	1.6	19.1	33.7	26.4
8-Feb-13	67	9	1.40	196	16.1	18.8	28.3	23.55
15-Feb-13	41	10	0.85	201	4.8	18.3	29.8	24.05
21-Feb-13	62	21	1.29	411	6.4	15.3	27.8	21.55
15-Mar-13	40	9	0.83	96	1.6	11.3	24.1	17.7
22-Mar-13	26	8	0.54	73	1.6	11.4	32.2	21.8
14-May-13	8	1	0.17	31	3.2	3.2	19.3	11.25

Appendix 9: The total and mean number of *S.calcitrans* flies counted/foreleg/minute in the H-line from 9 November 2012 to 14 May 2013.

	Group 1		Group 2		Group 3				Group 2		Group 1		Total <i>S.calcitrans</i> counted/foreleg	Mean counts/foreleg/min
	Pen 1	Pen 2	Pen 3	Pen 4	Pen 5	Pen 6	Pen 7	Pen 8	Pen 9	Pen 10	Pen 11	Pen 12		
09-Nov-12	2	1	1	0	0	0	0	1	0	0	0	1	6	0,13
16-Nov-12	1	4	5	3	2	1	0	2	2	2	2	8	32	0,67
30-Nov-12	0	2	2	3	0	1	4	1	1	0	0	1	15	0,31
07-Dec-12	0	1	0	0	0	0	3	1	2	2	1	1	11	0,23
14-Dec-12	1	0	2	2	0	3	5	7	2	9	1	5	37	0,77
04-Jan-13	2	10	4	10	2	7	3	11	7	4	9	8	67	1,5
11-Jan-13	11	5	5	11	4	5	10	7	1	7	11	0	77	1,60
18-Jan-13	0	1	0	1	0	0	1	0	0	0	4	2	9	0,19
25-Jan-13	7	4	2	7	9	1	11	7	8	8	12	14	90	1,88
08-Feb-13	9	4	8	16	6	1	0	0	10	6	3	4	67	1,40
15-Feb-13	7	6	3	1	1	4	4	2	4	1	4	4	41	0,85
20-Feb-13	17	14	2	2	4	9	9	2	1	24	15	14	113	2,35
21-Feb-13	8	7	10	8	3	8	2	2	3	3	3	5	62	1,29
22-Feb-13	0	4	1	3	0	0	1	1	0	3	0	2	15	0,31
05-Mar-13	4	4	3	4	8	8	2	1	3	9	10	4	60	1,25
08-Mar-13	2	1	4	0	0	0	0	0	1	1	0	2	11	0,23
15-Mar-13	6	0	2	1	2	0	7	2	3	2	10	5	40	0,83
22-Mar-13	2	1	0	0	1	4	1	2	4	4	4	3	26	0,54
04-Apr-13	1	0	0	0	0	0	0	0	0	2	0	1	4	0,08
12-Apr-13	0	0	0	0	0	0	1	0	1	0	0	0	2	0,04
14-May-13	0	1	1	0	0	0	0	2	0	0	0	4	8	0,17

Appendix 10: The total and mean number of foot stomps counted on the cattle/minute for 48 cattle in the H-line 9 November 2012 to 14 May 2013.

	Group 1		Group 2		Group 3				Group 2		Group 1		Total Foot stomps	Mean foot stomps/48 cattle
	Pen 1	Pen 2	Pen 3	Pen 4	Pen 5	Pen 6	Pen 7	Pen 8	Pen 9	Pen 10	Pen 11	Pen 12		
09-Nov-12	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00
16-Nov-12	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00
30-Nov-12	0	4	0	3	0	0	0	0	0	0	1	8	16	0,33
07-Dec-12	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00
14-Dec-12	0	0	1	0	1	1	0	0	3	0	0	6	12	0,25
04-Jan-13	1	3	0	1	0	0	3	4	1	0	2	1	16	0,33
11-Jan-13	3	4	3	5	2	4	2	2	0	1	3	0	29	0,60
18-Jan-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00
25-Jan-13	0	1	2	2	4	1	1	3	2	0	1	10	27	0,56
08-Feb-13	1	3	0	5	0	2	0	0	2	2	0	1	16	0,33
15-Feb-13	1	0	0	0	0	1	0	0	1	1	0	5	9	0,19
20-Feb-13	4	1	0	4	0	2	5	4	4	20	8	1	53	1,10
21-Feb-13	3	6	3	4	4	2	2	1	2	0	1	5	33	0,69
22-Feb-13	0	0	0	1	0	0	1	0	1	2	0	2	7	0,15
05-Mar-13	0	0	1	2	3	1	0	0	1	0	5	0	13	0,27
08-Mar-13	0	0	4	0	0	0	0	0	0	1	0	0	5	0,10
15-Mar-13	3	0	0	1	1	1	3	0	1	0	2	0	12	0,25
22-Mar-13	1	0	0	1	2	1	0	4	2	2	1	1	15	0,31
04-Apr-13	2	0	0	0	0	0	0	0	0	1	0	2	5	0,10
12-Apr-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00
14-May-13	0	0	0	0	0	0	0	0	0	0	0	0	0	0,00

Appendix 11.1: Repeated Measures Analysis of Variance for the total number of flies counted/foreleg/minute/pen in 3 groups from 9 November 2012 to 14 May 2013 in the H-line.

Sigma-restricted parameterization. Effective hypothesis decomposition					
Effect	SS	Degr. of Freedom	MS	F	P
Intercept	2622.893	1	2622.893	333.2057	0.000000
Group	70.881	2	35.440	4.5023	0.044144
Error	70.845	9	7.872		
R1	1839.04	20	91.951	11.9425	0.000000
R1*Group	385.452	40	9.636	1.2516	0.163797

Group; LS Means						
Current effect F(2,9) = 4.5023, p=0.04414						
Effective hypothesis decomposition						
Cell No.	Group	DV_1 Mean	DV_1 Std. Err.	DV_1 -95.00%	DV_1 +95.00%	N
1	1	3.892857	0.306122	3.200361	4.585353	4
2	2	3.190476	0.306122	2.497980	3.882972	4
3	3	2.595238	0.306122	1.902742	3.287734	4

Appendix 11.2: Unequal N HSD; variable DV _1 Approximate Probabilities for Post Hoc Tests

Error: Between Ms= 7.8717, df = 9.0000				
Cell no.	Group	3.8929 {1}	3.1905 {2}	2.5952 {3}
1	1		0.285925	0.036423
2	2	0.285925		0.393009
3	3	0.036423	0.393009	