

The impact of *Stomoxys calcitrans* populations on cattle in a feedlot near Heidelberg, Gauteng, South Africa

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PREFACE

The research discussed in this dissertation was conducted in the Unit for Environmental Sciences and Management, North-West University, Potchefstroom Campus, Potchefstroom, South Africa.

The research conducted and presented in this dissertation represents original work undertaken by the author and has not been previously submitted for degree purposes to any university. Where use was made of the work of other researchers, it is duly acknowledged in the text. The reference style used in this thesis is according to the specifications given by the NWU Harvard Referencing Guide.

SUMMARY

The stable fly (Diptera: Muscidae), *Stomoxys calcitrans*, is a widespread economically important pest of livestock at confined production facilities such as dairies and feedlots. Stable flies are haematophagous insects that frequently feed on the forelegs of cattle. Stable flies can cause significant production losses and are of severe animal health and welfare concerns. The present study evaluated the impact of stable fly populations on cattle. In order to achieve this aim, the following were investigated: (1) the temporal and spatial distribution of stable flies; (2) stable fly density on cattle in sprayed and unsprayed pens as sampled with traps and counted on cattle forelegs; (3) impact of stable flies on the feed intake of cattle; (4) impact of stable flies on the weight gain performance of cattle. Knowledge gathered during this study was used as recommendations for an integrated fly management programme. The seasonal abundance of stable flies was monitored from 24 October 2013 to 3 December 2014 with Nzi tsetse type traps. The diurnal and seasonal distribution of stable flies was investigated. Stable fly populations in vegetation have been observed to follow peak feeding periods on cattle. A fairly good correlation between stable flies collected from traps and the number of stable flies counted on cattle forelegs, confirmed the use of trap collection rates in accurately predicting the degree of stable fly feeding and irritation on cattle. Feed intake were related to the various levels of stable fly pressures and feed management practices. Statistically significant differences observed, were identified as having a little practical impact on meat production, specifically for the Karan Beef environment. This indicates little need of routine chemical control. Continuous monitoring of stable fly populations remains necessary to identify abnormal seasonal increases in fly populations. The results of this study have important implications for the development of an integrated fly management program.

Key words: *Stomoxys calcitrans*, stable fly, temporal distribution, integrated fly management, impact on meat production.

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LIST OF ABBREVIATIONS

ADG	average daily gain
ADGC	average daily gain cold
ADGL	average daily gain live
ANOVA	univariate two-way analysis of variance
CCW	cold carcass weight
DMI	dry matter intake
DW	dead weight
pb	proboscis
HDC	high fly density control (untreated)
HDT	high fly density treated
HSD	honest significant difference
IGR	insect growth regulator
IPM	integrated pest management
LDT	low fly density treated
SPSS	Statistical Package for Social Sciences
UV	ultraviolet
WCW	warm carcass weight

CHAPTER 1

INTRODUCTION

1.1 BACKGROUND INFORMATION ON *STOMOXYS CALCITRANS*

1.1.1. Systematics and distribution of stable flies

The species *Stomoxys calcitrans*, also known as the stable fly, dog fly (Johnson, 2011; Mavoungou *et al.*, 2012; Talley, 2008) or biting house fly (Bishop, 1913, 1927; Dawit *et al.*, 2012; Talley, 2008), is one of 18 recognised species from the genus *Stomoxys* (Diptera: Muscidae) (Baldacchino *et al.*, 2013; Dsouli *et al.*, 2011; Zumpt, 1973). It belongs to the tribe *Stomoxiinae*, a subfamily of Muscidae (Dsouli *et al.*, 2011; Zumpt, 1973). The Muscidae family is a large monophyletic group that contains about 4 500 well-defined species divided into 180 genera (Dsouli *et al.*, 2011). Within the subfamily *Stomoxiinae*, *Stomoxys calcitrans* is the only species with a worldwide distribution (Baldacchino *et al.*, 2013; Dsouli-Aymes *et al.*, 2011; Hall *et al.*, 1983; Muenworn *et al.*, 2010a; Thomas *et al.*, 1989; Urech *et al.*, 2012; Zumpt, 1973) whereas the other seventeen species are exclusively found in tropical regions (Dsouli *et al.*, 2011; Zumpt, 1973). Twelve are located on the African continent, four on the Asian continent, and one on both the African and Asian continents (Dsouli-Aymes *et al.*, 2011; Zumpt, 1973). According to literature, New World stable flies have a Palearctic origin (Brues, 1913; Kneeland, 2011; Marquez *et al.*, 2007). It is believed that the *Stomoxys* genus was introduced into the New World during colonization (Kneeland, 2011; Marquez *et al.*, 2007) and human migration (Kneeland, 2011; Marquez *et al.*, 2007). Dsouli-Aymes *et al.* (2011) suggested stable flies from the Palearctic, Nearctic, Neotropical and Oceanic region originated from the Afrotropical region. Presently, as humans and livestock spread across the continent due to economical and recreational activities, stable fly populations continuously grow across New World regions (Kneeland, 2011). Stable flies are recognized as one of the most important pests of livestock in many parts of the world (Baldacchino *et al.*, 2013; Campbell *et al.*, 1987; Dawit *et al.*, 2012; Kunz *et al.*, 1991; Morgan *et al.*, 1983; Thomas, 1993; Zhu *et al.*, 2012; Zumpt, 1973). They are a great nuisance to livestock, especially cattle, and have a deleterious impact on their welfare (Dougherty *et al.*, 1995). These species are of economical, medical and veterinary importance (Bruce & Decker, 1958; Campbell *et al.*, 1977, Campbell *et al.*, 2001; Miller *et al.*, 1973; Wieman *et al.*, 1992).

1.1.2. General morphology of the stable fly

The adult stable fly is 4 to 7 mm in body length (Gerry *et al.*, 2007; Masmehatathip *et al.*, 2006b), about the same size as the common housefly, *Musca domestica* (Gerry *et al.*, 2007; Howell *et al.*, 1978; Johnson, 2011; Masmehatathip *et al.*, 2006b). The adult stable fly has a

grey abdomen and thorax marked with a set of dark-grey patterns (Masmearthip *et al.*, 2006b) (Fig. 1.1). The abdomen has a distinct checkered pattern, one median spot and two lateral round spots, on the dorsal side of the second and third segment (Howell *et al.*, 1978; Masmearthip *et al.*, 2006b; Talley, 2008; Tam, 2003; Tangtrakulwanich, 2012) (Fig. 1.1). The thorax has four dorsal longitudinal stripes of which the two outermost stripes are shorter (Howell *et al.*, 1978; Masmearthip *et al.*, 2006b; Tam, 2003). Stable flies are easily distinguished from other Muscidae species through their proboscis (Bishop, 1913; Masmearthip *et al.*, 2006b) (Fig. 1.2). The stable fly has a bayonet-like proboscis instead of a sucking, sponge-like proboscis such as the house fly (Bishop, 1913; Tangtrakulwanich, 2012). The proboscis is a long, thin, piercing organ that protrudes forward from under the head (Bishop, 1913) (Fig. 1.2). The base is equipped with sclerotized teeth adapted for cutting, tearing and piercing (Stephens & Newstead, 1907). The stable fly can also be distinguished from other *Stomoxys* species by their moderately bent fourth wing vein (Dodge, 1953; Foil & Hogsette, 1994; Masmearthip *et al.*, 2006b) and maxillary palps which are shorter than the proboscis (Bishop, 1913; Howell *et al.*, 1978). Other characteristics used to differentiate between *Stomoxys* flies include male genitalia and external morphological characteristics such as the dorsal abdominal pattern (Masmearthip *et al.*, 2006b).

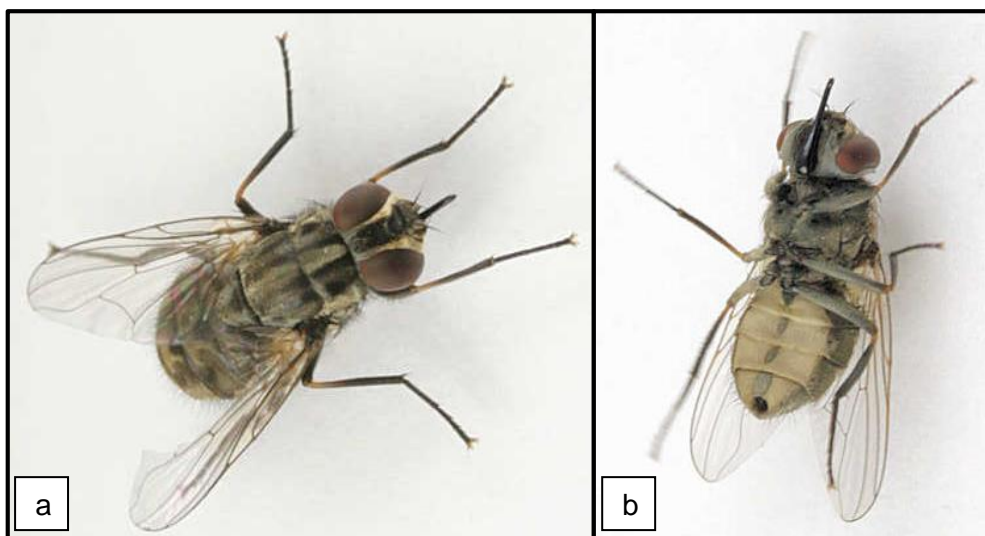


Figure 1.1: Dorsal (a), and ventral (b) view of an adult stable fly (*Stomoxys calcitrans*) (Schaefer, 2015).

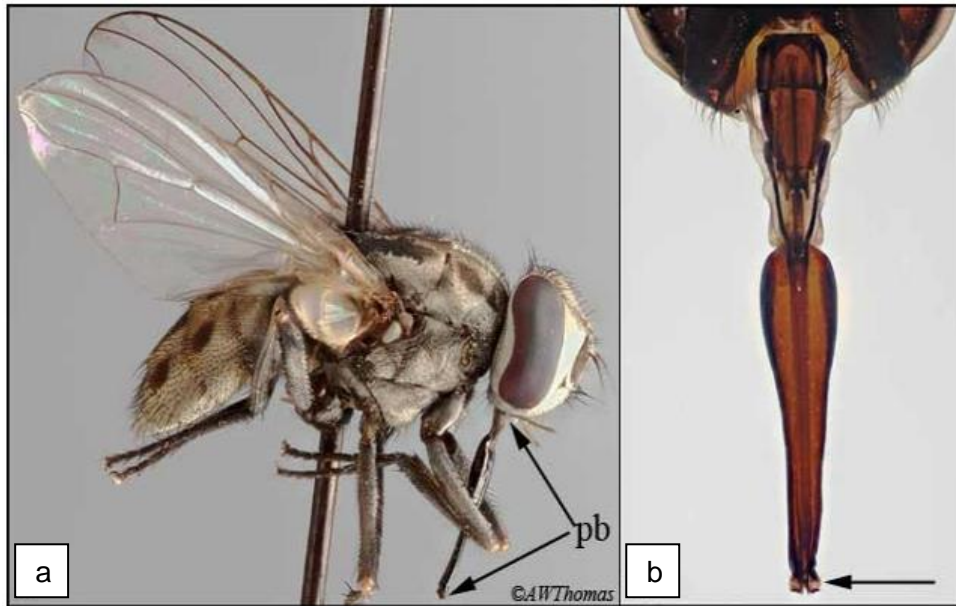


Figure 1.2: Stable fly (*Stomoxys calcitrans*) showing proboscis (pb) (a), and front view of proboscis indicating the position of the sclerotized teeth (b) (Thomas, 2012).

Stable fly eggs are about 1 mm long (Bishop, 1913; Newstead, 1906; Tangtrakulwanich, 2012; Wall & Shearer, 1997), slightly curved on one side, and straight with a deep and broad longitudinal groove on the other (Newstead, 1906). Stable fly eggs are white when first laid but change to a creamy white colour as time passes (Ajidagba, 1979; Newstead, 1906).

The stable fly larva is characterized by a creamy-white colour (Newstead 1906; Russel *et al.*, 2013) and black sub-cutaneous mouth-hook (Newstead, 1906). The stable fly has three instar larvae. The first instar larva has round posterior spiracles discs with straight slits, whereas the second and third instar larvae have triangular discs with two and three sinuous slits, respectively (Friesen *et al.*, 2015). The newly hatched larva is translucent and characterized by mouth-parts not yet fully developed (Newstead, 1906). The older larva is slightly less transparent compared to the younger one (Newstead, 1906), and characterized by a pale yellow to nearly white colour (Bishop, 1927). The larvae of both housefly and stable fly are cylindrical (Bishop, 1913), rounded posteriorly and tapered anteriorly (Newstead, 1906). The stable fly pupa is about 4 to 7 mm long (Bishop, 1913) with three S-shaped yellow slits (Bishop, 1913) and eleven visible segments which are slightly wider anteriorly than posteriorly (Newstead, 1906). The exteriors are lightly sclerotized and a reddish-to-dark brown colour (Tangtrakulwanich, 2012).

1.1.3. Feeding habits of the stable fly

Stable flies, both male and female, are obligatory, haematophagous insects (Bishop, 1927; Holdsworth *et al.*, 2006; Thomas *et al.*, 1989; Zumpt, 1973) that feed on wild and domestic animals (Dawit *et al.*, 2012; Muenworn *et al.*, 2010a; Wall & Shearer, 1997), birds, reptiles (Mitzmain, 1913), amphibians and even humans in the absence of a preferred host (Baldacchino *et al.*, 2013; Dawit *et al.*, 2012). Large populations of adult stable flies are problematic because of their painful bites (Kaufman & Weeks, 2012; Müller *et al.*, 2012) and aggressive, persistent feeding behaviour (Baldacchino *et al.*, 2013; Bishop, 1913; Muenworn *et al.*, 2010a; Newson, 1977; Schofield & Torr, 2002). Stable flies prefer to feed on the lower extremities of cattle (Berry *et al.*, 1983; Pitzer *et al.*, 2011; Urech *et al.*, 2012) (Fig. 1.3a) and horses (Pitzer *et al.*, 2011; Russel *et al.*, 2013) (Fig. 1.3b), the ankles of humans (Pitzer *et al.*, 2011; Russel *et al.*, 2013), and ears of dogs (Baldacchino *et al.*, 2013; Pitzer *et al.*, 2011; Russel *et al.*, 2013).

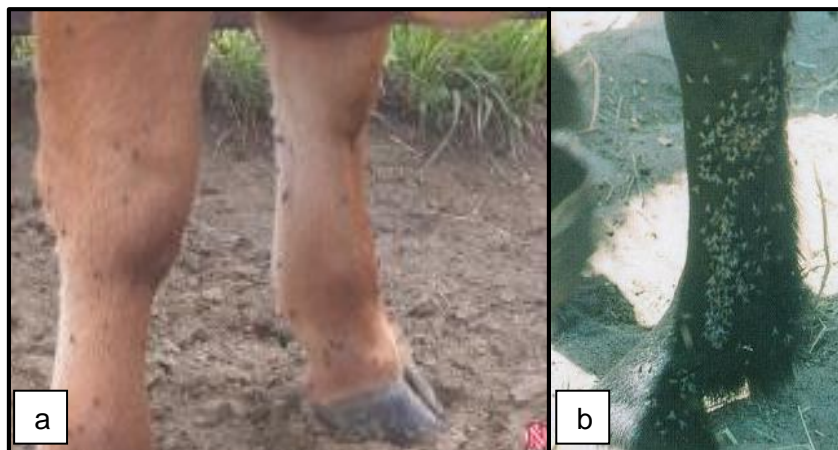


Figure 1.3: Stable flies (*Stomoxys calcitrans*) feeding on the front legs of cattle (Boxler, 2013) (a), and horses (b) (Western Australia, 2015).

Stable flies take between one (Berry & Campbell, 1985; LaBrecque *et al.*, 1975) and two (Charlwood & Lopes, 1980; Hafez & Gamal-Eddin, 1959; Kunz & Monty, 1976) bloodmeals a day, depending on the climate, for a period of about 2 to 5 minutes (Bishop, 1927). During each bloodmeal, they pierce and tear the host's skin with their long, bayonet-type proboscis until a pool of blood forms at the surface of the skin (Gerry *et al.*, 2007). Stable flies require blood for successful mating (Müller *et al.*, 2012), reproduction (Friesen *et al.*, 2015; Hogsette *et al.*, 1987) and ovarian development (Friesen *et al.*, 2015; Müller *et al.*, 2012; Phasuk *et al.*, 2013). However, according to various research studies, some *Stomoxys* species feed on the nectar of fruits and flowers (Hogsette *et al.*, 1987; Müller *et al.*, 2012; Phasuk *et al.*, 2013; Showler & Osbrink, 2015). In Mali, stable flies have been reported to be most attracted to a single fruit (*Piliostigma reticulatum*) and three flower species (*Acacia albida*, *Ziziphus*

mauritiana and *Acacia macrostachya*) (Müller *et al.*, 2012). Nectar does not facilitate reproduction (Showler & Osbrink, 2015), but has been proven to serve as a great source of energy during long-distance flight activity when stable flies are searching for bloodmeals (Hogsette *et al.*, 1987; Phasuk *et al.*, 2013). A bimodal blood-feeding (Hogsette *et al.*, 1987; Kunz & Monty, 1976; Mihok & Clausen, 1996) and an unimodal sugar-feeding pattern were reported in Mali (Hogsette *et al.*, 1987). In a more recent study by Müller *et al.* (2012), *Stomoxys* flies were recorded to feed on sugar as frequently as on blood.

1.1.4. Lifecycle of the stable fly

The stable fly lifecycle consists of four distinct stages typical to the Muscoid family: egg, larva (maggot), pupa and adult (Bishop, 1927; Ross *et al.*, 1982). Stable fly larvae develop best at temperatures between 15°C and 30°C (Gilles *et al.*, 2005) and have poor survival rates at temperatures above 30°C (Gerry *et al.*, 2007). The female stable fly begins depositing eggs after about three bloodmeals (Bishop, 1927). Mitzmain (1913) has shown that female stable flies can deposit between 632 and 820 eggs during 20 ovipositions in a single lifetime, whereas Bishop (1913) recorded a total of 273 eggs during three ovipositions. Although eggs are deposited in irregular masses (Bishop, 1927), they are generally found in batches of 25 to 50 (Wall & Shearer, 1997). The female often carefully separates and scatters the eggs throughout the breeding medium, using her proboscis and legs (Newstead, 1906; Russel *et al.*, 2013). After the eggs have been laid, in about one to three days (Bishop, 1927), saprophagous larvae (maggots) hatch from the eggs (Russel *et al.*, 2013) by splitting the anterior end of the groove (Bishop, 1927; Newstead, 1906), and immediately start feeding on the surrounding decaying, organic material (Bishop, 1927; Johnson, 2011). Within 10 to 11 days, the larvae rapidly develop through three instars, each being larger than the previous one (Bishop, 1927). After feeding, when the larvae become full-grown, they move to drier, cooler areas to start pupation (Bishop, 1927). At first, the larvae start shortening by contracting their anterior segments into a barrel-shaped pupa (Bishop, 1927; Newstead, 1906). Afterwards the skin (integument) hardens to form a protective case (puparium) surrounding the developing pupa (Bishop, 1927; Newstead, 1906). The soft yellowish puparium hardens and changes into a reddish brown colour as it ages (Bishop, 1927; Newstead, 1906). The whole pupation process is complete within about two hours (Newstead, 1906). The entire transformation from maggot into adult fly occurs within the puparium (Bishop, 1927; Newstead, 1906) and takes about 6 to 20 days (Bishop, 1927). Before emergence: the puparium darkens; the nymph pushes the skin of the final molting into the posterior end of the puparium and then emerges from the anterior end of the puparium (Newstead, 1906). After emergence, the fly tries to escape from its environment by pushing its way out using its ptilinum (Newstead, 1906). Afterwards, the fly waits until its

integument and wings harden, positions its proboscis, and flies (Newstead, 1906; Shipley, 1915).

1.1.5. Stable fly larval developmental sites

Knowledge concerning the source of stable fly development is important in formulating fly-control methods in feedlots (Berkebile *et al.*, 1994). An effective method for stable fly control is elimination of developmental sites (Mcpheron & Broce, 1996). Stable flies are able to develop in a wide range of physiochemical environments; a favourable larval medium is characterised by a suitable pH, temperature, moisture content (Berkebile *et al.*, 1994; Broce & Haas, 1999; Rasmussen & Campbell, 1981) and organic content (Rasmussen & Campbell, 1981). In addition to these factors, a suitable pupariation site is associated with other environmental factors such as light and osmolality (Mcpheron & Broce, 1996). Meyer and Peterson (1983) identified sixteen different types of developmental sites for stable flies and house flies. The most frequent and consistent breeding sites of stable flies at feedlots and dairies were provided by spilled feed, stored manure and various forms of silage (hay, corn and oats) (Meyer & Peterson, 1983). Other larval developmental sites at feedlots included protected areas where manure, spilled feed and bedding material accumulate (Skode *et al.*, 1919), such as under fencelines (Meyer & Peterson, 1983), or in drainage ditches, potholes, (Meyer & Peterson, 1983; McNeal & Campbell, 1981) and empty lots (Meyer & Peterson, 1983). Stable flies avoid development in undisturbed cowpats and fresh cattle manure, and prefer manure older than two weeks (Bishop, 1913; Broce & Haas, 1999; Broce *et al.*, 2005). High summer temperatures dry the surface of manure and create a crust that seals moisture and maintains a long-term habitat suitable for larval development (Showler & Osbrink, 2015). Stable fly larvae will bury deep into manure to feed and avoid desiccation (Showler & Osbrink, 2015). In addition to being found in cattle manure, immature stable flies have been found in other manure such as that of swine (Meyer & Peterson, 1983) and horse (Jeanbourquin & Guerin, 2007).

The method of overwintering is important in the development of methods for stable fly control at feedlots (Berkebile *et al.*, 1994). Stable flies have no diapause (Greene *et al.*, 1989; Taylor *et al.*, 2011), or freeze-tolerant stage (Beerwinkle *et al.*, 1978; Jones and Kunz, 1997; Taylor *et al.*, 2011). Berkebile *et al.* (1994) found that stable flies do not overwinter as diapausing adults, but rather as developing larvae. This was confirmed by the absence of hypertrophied fat bodies in adult stable flies and ovaries in stage two development. Stable flies have been reported to overwinter as slowly developing larvae when found below the frost line (Berry *et al.*, 1978; Campbell *et al.*, 1987; Scholl *et al.*, 1981). The larvae have an estimated threshold temperature of 12.3°C (Larson & Thomsen, 1940) and can survive short

periods of subfreezing temperatures (Beerwinkle *et al.*, 1978). Additionally, the third instar larvae stage can extend for up to 120 days in unfavourable conditions (Berry *et al.*, 1978; Scholl *et al.*, 1981). Stable fly overwintering sites may differ from warmer months, due to the heat produced during fermentation (Berkebile *et al.*, 1994). Ideal breeding mediums should not freeze for long periods or generate too much heat (Berkebile *et al.*, 1994). In the winter, immature stable flies are generally found in heat-generating mediums (Taylor *et al.*, 2011) such as peanut litter or manure mixed with grain or hay, straw stacks or open silage (Berkebile *et al.*, 1994). In south-eastern Nebraska, a small number of third instar larvae were found in silage during the winter (Berkebile *et al.*, 1994). Silage has a temperature gradient suitable for overwinter pupae (Berkebile *et al.*, 1994). During the winter, the top layer freezes while temperatures increase in the layers below (Berkebile *et al.*, 1994). Other overwinter sites may include cattle-confinement buildings (Matthysse, 1945; Somme, 1961), or pig manure (Mellor, 1919). Piled manure and grass clippings have a temperature gradient that extend development and allow developing stable flies to survive freezing temperatures (Berkebile *et al.*, 1994; Berry *et al.*, 1978). Large, round hay bales fed to cattle during the winter might be the primary source of stable fly populations in early summer (Broce *et al.*, 2005; Taylor *et al.*, 2011). In Missouri, numerous developing stable fly larvae have been found on the edge of round hay bales stored in the field (Hall *et al.*, 1982). An estimated 28 000 larvae per m² have been found in wasted hay at dairies in north-western Florida (Broce *et al.*, 2005). However, as the summer progresses, it seems as if stable fly production from overwintering sites decline, presumably due to higher temperatures and lower rainfall, which makes these sites unfavourable for developing stable fly larvae (Broce *et al.*, 2005). Adoption of this practice made stable flies an important pest of pastured and grazing cattle (Broce *et al.*, 2005; Taylor *et al.*, 2011).

1.2. MONITORING OF STABLE FLY POPULATIONS

1.2.1. Stable fly trapping

Development of an integrated pest management (IPM) approach requires knowledge of the stable fly's ecology, biology (Masmehatip *et al.*, 2006a; Morgan *et al.*, 1983) and seasonal and spatial distribution in and around the intensive production facility (Urech *et al.*, 2004). Thus, fly populations needs be monitored to keep track of fluctuations and to determine the effectiveness of control strategies (Urech *et al.*, 2004). It is important to use a reliable and sustainable method to monitor stable fly populations at intensive cattle production facilities (Mullens & Meyer, 1987). In various studies, several types of methods have been used to survey, estimate and monitor stable fly abundance at feedlot facilities (Baldacchino *et al.*, 2013; Evert, 2014; Hall *et al.*, 1983; Urech *et al.*, 2012; Mullens & Meyer, 1987). Some of

these include animal-baited traps (Hall *et al.*, 1983; Harley, 1965; Thomas *et al.*, 1989; Williams *et al.*, 1977), sticky traps (Broce, 1988; Williams, 1973), sweep-nets (Masmeatathip *et al.*, 2006b), Malaise traps (Hall *et al.*, 1983), tsetse type traps such as Vavoua traps (Mihok, 2002; Mihok *et al.*, 1995a; Muenworn *et al.*, 2010a; Muenworn *et al.*, 2010b) and Nzi traps (Evert, 2014; Mihok, 2002; Mihok *et al.*, 1995a). The latter is the most commonly used method to monitor stable fly activity (Mullens & Meyer, 1987). The Nzi and Vavoua traps are simple, safe, economical traps developed in West Africa (Mihok, 2002; Mihok *et al.* 1995a). These traps are designed to trap tsetse flies and other haematophagous flies (Mihok, 2002; Mihok *et al.* 1995a). In an initial survey conducted by Evert (2014), a sustainable and effective method for monitoring stable flies was determined. The traps evaluated were the Nzi® (Fig. 2.3a), Vavoua® and H-trap® tsetse type traps (Evert, 2014). The Nzi tsetse type trap proved to be effective in collecting stable flies and other haematophagous flies (Evert, 2014; Mihok, 2002; Mihok *et al.*, 1995a).

Other selective traps include Alsynite (Broce, 1988; Mullens & Meyer, 1987; Scholl, 1986; Scholl *et al.*, 1985; Thomas *et al.*, 1989; Urech *et al.*, 2012) or Coroplast (Corflute) traps (Urech *et al.*, 2012). The Alsynite trap is commonly used in the USA for the purpose of research, while the NZI trap is commonly used in South Africa. The NZI trap is especially used for the purpose of identification. The Alsynite fiber glass trap is a highly efficient, cost effective, portable trap (Hogsette & Ruff, 1990). It is a cylindrical trap consisting of alsynite fiber glass and adhesive coated sleeves (Ose & Hogsette, 2014). Stable flies are attracted to the ultraviolet (UV) light reflected from the panels and are caught on the sticky sheets (Urech *et al.*, 2004). The traps have some limitations as the sticky adhesive coatings may damage the samples and make identification difficult. There is also the risk of environmental conditions influencing trap performance (Gersabeck & Merritt, 1983).

1.2.2. Stable fly sweep-netting

Sweep-netting is often used to monitor adult stable fly populations. Several literature studies proposed that stable flies seek shaded areas - such as fences, walls or vegetation - to rest and to digest their bloodmeal (Berry & Campbell, 1985). Sweep-netting may be used to collect adult stable fly populations from grass, brush and weeds. In contrast to various trap types, stable fly sweep-netting does not depend on the attraction of the insects to the trap. It is a method commonly used for monitoring insects (Avancini & Silveira, 2000; Soto *et al.*, 2014; Szalanski *et al.*, 1996), especially in agricultural arthropod surveys (Spafford & Lortie, 2013).

1.2.3. Stable fly density on cattle

Fly counts are done by visually counting the number of stable flies resting and feeding on the front legs of cattle (Berry & Campbell, 1985; McNeal & Campbell, 1981; Mullens & Meyer, 1987; Thomas *et al.*, 1989). This is a convenient and time-saving method, compared to whole body counts (Campbell & Hermanussen, 1971). The number of adult flies feeding on the outside of one foreleg, and the inside of the other foreleg, is counted per minute (Berry & Campbell, 1985; Gerry *et al.*, 2007; McNeal & Campbell, 1981). Estimating stable fly abundance through leg counts should be done on a sunny day, after 09:00 to 10:00 (Thomas *et al.*, 1989). The economic impacts of stable flies on feedlot cattle are then related to the numbers of flies feeding on cattle front legs per minute (McNeal & Campbell, 1981). Several studies used 5 flies per front leg per minute as an economic injury threshold for stable flies (Gerry *et al.*, 2007; McNeal & Campbell, 1981). This estimation is based on research conducted on dairy cattle (Bruce & Decker, 1985) and calves (Campbell *et al.*, 1977).

1.3. INTEGRATED PEST MANAGEMENT (IPM) APPROACH

A significant amount of research has been done on methods of stable fly control. Stable fly control is more effective when a combination of control strategies is implemented, thus a more integrated approach to stable fly control has been recommended for intensive production facilities (Urech *et al.*, 2004). An integrated pest management (IPM) approach should provide the intensive production facility with an effective, sustainable and economical method of stable fly control (Urech *et al.*, 2004). In general, an IPM system consists of three types of control measures, including cultural (mechanical/ physical), biological and chemical (Urech *et al.*, 2004).

1.3.1. Cultural control

Cultural control is the most practical, economical (Kaufman & Weeks, 2012) and cost-effective (Gerry *et al.*, 2007) approach for managing stable fly populations. It is directed towards reducing and eliminating stable fly developmental sites at production facilities (Baldacchino *et al.*, 2013; Kaufman & Weeks, 2012). In an IPM approach for nuisance flies on cattle feedlots, Urech *et al.* (2004) recommended reduction of stable fly developmental sites by managing manure, spilled feed, silage and carcasses. Additionally, feedlot maintenance was also mentioned for the control of stable flies (Urech *et al.*, 2004). Stable fly resting sites should be reduced or removed by maintaining vegetation, particularly weeds and grass surrounding pens, drains and sedimentation ponds (Urech *et al.*, 2004). However,

if cultural control is not sufficient to control stable flies and the numbers exceed the economic injury threshold for stable flies on cattle, it is recommended to use cultural control in combination with another control strategy.

1.3.2. Biological control

Biological control plays an important role in reducing stable fly populations. Entomopathogenic fungi (such as *Beauveria bassiana* and *Metarhizium anisopliae*), predatory mites and parasitic wasps (from the family Pteromalidae (Hymenoptera) (Baldacchino *et al.*, 2013)) have been the most commonly used tool to control nuisance flies in cattle feedlot facilities. Since biological control is temporary, costly and does not achieve instant results, it is recommended to be used in combination with other control strategies (Urech *et al.*, 2004).

1.3.3. Chemical control

Insecticide application runs the risk of environmental pollution, has various health and safety issues, leads to insecticide resistance (Urech *et al.*, 2012) and destroys natural enemies (Gerry *et al.*, 2007). Therefore, chemical control should not be used as the primary method of stable fly control, but rather in combination with cultural and biological controls. Insecticides should be used in moderation to preserve biological control agents (Urech *et al.*, 2004). Larvicides, fly baits and insect growth regulators (IGR) such as cyromazine are recommended over adulticides, because they do not affect beneficial insects. However, if adulticide application is unavoidable, knockdown insecticides are recommended because the effect is short lived and might allow stable fly populations to recover quickly (Urech *et al.*, 2004).

The feedlot design should facilitate fly control. It should promote effective cleaning and removal of fly developmental sites at intensive feedlot facilities (Urech *et al.*, 2004).

1.4. AIM AND OBJECTIVES

The main aim of the present study was to determine the impacts of *Stomoxys calcitrans* populations on cattle in a feedlot near Heidelberg, Gauteng, South Africa.

Specific objectives were:

- To determine the temporal and spatial distribution of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae) in a feedlot near Heidelberg, Gauteng, South Africa.
- To determine the stable fly density on cattle in sprayed and unsprayed pens, as sampled with traps and counted on cattle forelegs.
- To determine the impact of stable flies on the feed intake of feedlot cattle
- To determine the impact of stable flies on the weight-gain performance of feedlot cattle.
- To identify integrated stable fly management options based on knowledge gathered during the study.

1.5. OUTLINE OF DISSERTATION

A brief description of each chapter is given below to provide the context of the present study:

Chapter 1 contains a description of the literature review of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae), along with the aim and objectives.

Chapter 2 provides a description of the site, general sampling methods and statistical methods used. Material and methods, specifically dealing with the results in chapter 3 and 4, are given in these relevant chapters.

Chapter 3 contains a description of the temporal and spatial distribution of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae), at a feedlot near Heidelberg, Gauteng, South Africa. Chapter 3 also contains material and methods, along with the results and discussions of the stated objectives.

Chapter 4 provides a description of the impact of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae), on the feed consumption and weight-gain performance of feedlot cattle near Heidelberg, Gauteng South Africa. Chapter 4 also contains material and methods, along with the results and discussions of the stated objectives.

Chapter 5 provides a discussion of the conclusions of all the stated objectives, and stable fly management recommendations based on the results of this study.

CHAPTER 2

GENERAL MATERIAL AND METHODS

2.1. STUDY SITE AND DESCRIPTION

This study was conducted at Karan Beef, a cattle feedlot located near Heidelberg, Gauteng, South Africa (26° 36' 27" S, 28° 19' 13" E) (Fig. 2.1). It is situated outside Heidelberg on the Vaal dam Road (R594), about 50 km south-east of Johannesburg.



Figure 2.1: An aerial map of Karan Beef, showing the feedlot (A), holding ponds (B), neighbouring game reserve, biofiltration wetlands (C as indicated by the arrow), and dung heaps (D) (Evert, 2014).

The feedlot is situated on a slope, to facilitate drainage of manure and run-off water into holding dams. On the property there are also manure-holding ponds, pastured cattle sites and a game reserve with a wetland eco-development (Karan Beef, 2015). Manure heaps are located about 5 km from the feedlot (Evert, 2014). The feedlot is surrounded by grasslands and perennial plants. Karan Beef extends over 2 330 hectares and can accommodate over 120 000 head of cattle - which has recently been extended to 150 000 head of cattle, making it the largest feedlot in Africa (Karan Beef, 2015). The feedlot consists of several rows of concrete pens, arranged with feed bunks on the one side and drainage channels on the other (Fig. 2.2). The pens are alphabetically numbered, starting from A at the lower side of the feedlot, to VX at the upper side of the feedlot (Evert, 2014). Feedlot pens contain the

same number of calves per surface area (Fig. 2.2 & Table 4.1). Seven hospital areas are operational alone for each section (Evert, 2014). Cattle showing clinical symptoms of diseases are immediately moved to recovery pens for observation and treatment (Evert, 2014).



Figure 2.2: Feedlot pens containing 96 cattle (Table 4.1). This is row number VX.

The facility also owns the largest, most modern feed mill in the world, occupying an area of 15 000 square meters (Karan Beef, 2015). The feed mill is capable of producing up to 15 000 tons of mixed feeds on a daily basis. Finished cattle are transported to the harvest facility, the most modern facility in Africa capable of processing 2 000 head of cattle on a daily basis (Karan Beef, 2015). As soon as the cattle are slaughtered, they are moved to the deboning plant and meat processing facility and then through to the client. Currently, Karan Beef supplies beef to Hong Kong, Indian Ocean islands, Middle East and African countries (Karan Beef, 2015).

The Gauteng province, located on the interior plateau of South Africa (Dyson, 2009), has a mild climate with two distinct different seasons: a wet summer season and dry winter season (SouthAfrica.com, 2014). Gauteng has a consistent climate that is warm and wind free in the summer, and cold at night in the winter (SouthAfrica.com, 2014). Summers are from October to March, while winters are from July to August (SouthAfrica.com, 2014). Average midday temperatures at Heidelberg range from 16.6°C in June to 26.3°C in January (SAexplorer, 2014). The coldest weather is in July, below an average of 0.2°C during the night (SAexplorer, 2014). Heidelberg receives about 588mm rain per year (SAexplorer, 2014), with most rainfall occurring during the summer (SAexplorer, 2014). It receives the highest

rainfall (average of 112mm per month) in January and the lowest (average of 0mm per month) in July (SAexplorer, 2014). Hail is usually expected during thunderstorms, and snow almost never (SouthAfrica.com, 2014).

2.2. SAMPLING OF STABLE FLY POPULATIONS

2.2.1. Trapping

In this study, the collection points located at the end of the net on top of the Vavoua, Nzi and H-traps were modified to improve removal of captured flies (Fig. 2.3b). A two-litre plastic container with a lid was attached to the end of the trap (Evert, 2014). This modification was designed for easy and fast removal of collected flies without damage to the samples (Evert, 2014) (Fig. 2.3b). Evert (2014) recommended the modified Nzi tsetse type trap for collecting stable flies in future studies, since this trap has been used as a method of stable fly control in several research studies (Evert, 2014; Mihok, 2002; Mihok *et al.*, 2006). The trap does not require fly bait to attract stable flies, it is easy to deploy and durable in stormy weather conditions (Evert, 2014). Flies were collected dry, and stored in ethanol. No predation were observed in plastic containers.

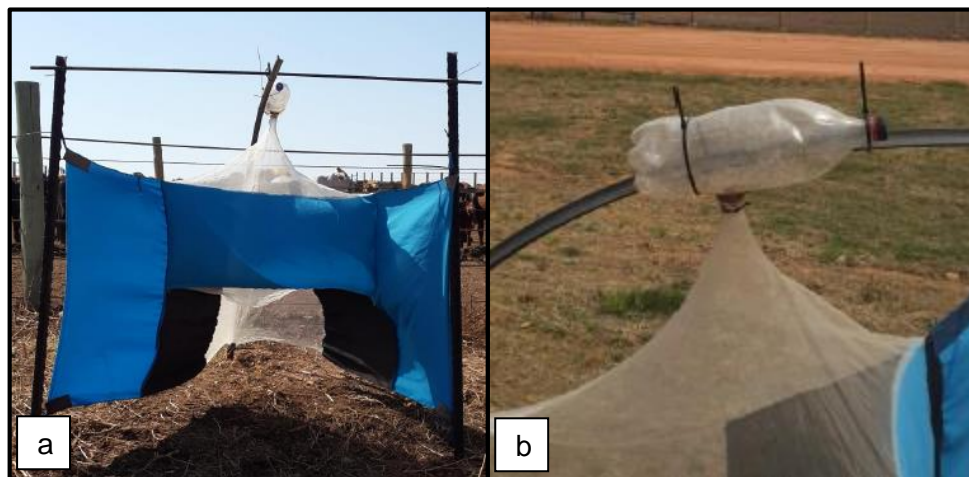


Figure 2.3: The Nzi® tsetse type trap (a), and modified two-litre plastic container with lid for collecting stable flies at the top of the trap (b) (Evert, 2014).

The design and colour of the traps imitate the shape of an animal, resulting in optimal trap performance (Evert, 2014; Horvath, 2004; Mihok, 2002). The traps were made from fabrics, white insect netting, and cloth with either a copper phthalocyanine (phthalogen blue), or its sulphonated forms (turquoise colour) (Mihok *et al.*, 2006). These selected colours proved to be very effective in attracting stable flies and other biting flies (Dawit *et al.*, 2012; Mihok *et al.*, 2006). The traps have a triangular layout, phthalogen blue front, and colour-fast black back and lower-front entrance (Mihok, 2002). The top of each trap has a tetrahedron white

netting (Mihok, 2002) extending into a collection bottle. The Nzi tsetse type trap is a one-directional passive trap that attracts flies from the surroundings (Evert, 2014). It is most effective when directed towards surrounding grassland away from obstructing vegetation. The Nzi® tsetse type traps used in this study were purchased from Vestergaard Frandsen (EA) (Ltd) (Disease Control Textiles).

2.2.2. Sweep-netting

Sweepings were done using a standard-size net with a 381 mm hoop and 1.5 m handle. During each sweeping, the net was repeatedly swept back and forth through the vegetation in a 180° sweep. The net was kept a few centimeters below the top of the vegetation during sweeping. Afterwards, the net was flipped over to prevent flies from escaping. Three sweeps of a 180° sweep of the net represented one independent sweep-net sample. Each sweep-net sample was an estimate of the hourly number of stable flies resting in the vegetation surrounding the feedlot. Collected stable flies were removed from traps and nets, separately stored in bottles containing 70% alcohol, and brought back to the laboratory for identification and counting. The number of stable flies collected were recorded by capture date, hour and location.

2.3. COUNTING STABLE FLIES ON CATTLE

Stable flies feeding per minute on cattle forelegs were viewed through binoculars and counted. The number of stable flies feeding on the inside of the one foreleg and the outside of the other foreleg were counted when viewed from one direction (Berry *et al.*, 1983; Catangui *et al.*, 1997; Gerry *et al.*, 2007; McNeal & Campbell, 1981; Mullens & Meyer, 1987). Only stable flies feeding on cattle forelegs between the flank and hoof were counted (McNeal & Campbell, 1981). The mean number of stable flies feeding per foreleg per minute (Berry *et al.*, 1983) was calculated for every counting event and treatment. Stable fly leg counts were done weekly on sunny-days, generally Wednesdays.

Leg counts were done on 4 randomly selected animals per pen. Cattle that were counted were approximately within 10 m of the fence-line and feed bunk. Two random cattle from the feed bunk and two from the opposite fence-line were selected for foreleg counts, to ensure standardised and representative data (Catangui *et al.*, 1995; Evert, 2014). Cattle from 16 pens were counted over the course of the study, 6 pens in the VX-line and 10 randomly selected pens from the H-line. A total of 64 cattle (4 cattle per pen for 16 pens) from both the VX-pens and H-pens were counted per sampling day. Leg count data from each day were recorded on spreadsheets indicating day, time, number of stable flies, eartag number, breed, colour and pen number.

Regardless of the type of method used, findings should be used in the development of an effective IPM programme adapted to the specific conditions unique to each type of livestock production facility (Baldacchino *et al.*, 2013).

2.4. STATISTICAL ANALYSES

2.4.1. Temporal and spatial distribution of stable flies

Stable fly data was entered into Excel spreadsheets for statistical analysis. Statistical analysis was done in Statistica (STATISTICA 12) (StatSoft Inc., 2014). Where appropriate, analysis of variance (ANOVA) was used. Regression and correlation analyses were used to determine: (1) the effects of climatic factors on trap collections and the number of flies counted on cattle forelegs per minute; and (2) the correlation between stable fly collections and stable flies on cattle forelegs.

Data of stable fly occurrence on different breeds of cattle of various colours were analysed using Durbin-Watson statistics, to determine if there was an autocorrelation in stable fly foreleg counts over time. Indicating time dependency as shown by Durbin-Watson statistics, fly data from all 20 weeks were analysed per week to search for the occurrence of a stable fly preferences pattern over time. An analysis of the fly counts on cattle breeds and fly counts on cattle of different coat colours during 20 weeks was done separately. The total stable fly count data was examined for significant differences by using parametric and non-parametric statistical analyses. An Unequal N HSD (honest significant difference) test, a modification of the Tukey HSD test, Kruskal-Wallis test, and ANOVA statistics were performed on the number of stable flies counted on the forelegs of cattle of different breeds. ANOVA ($P < 0.05$) was used to differentiate between the mean numbers of stable flies counted on the forelegs of cattle of different coat colours.

Since it was difficult to determine a stable fly preferences pattern and show significant effects, ANOVA ($P < 0.05$) was performed on the fly counts on cattle breeds and fly counts on cattle of different coat colours across the whole survey period.

2.4.2. Impact of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae) on the feed consumption and weight gain performance of feedlot cattle

Multiple regression analyses were performed in Statistica (STATISTICA 12) (StatSoft Inc., 2014) with Bonferroni-correction to determine the impacts of various levels of stable fly pressures on the average daily dry-matter intake of confined steers and on the average daily gains of finishing steers. Multiple regression analyses using Statistical Package for the Social Sciences (SPSS) (IBM SPSS Statistics for Windows, Version 22.0, 2014) were performed by applying the Bonferroni-correction (or adjustment) to the p -values to avoid making a Type I error (Armstrong, 2014; Davis, 2013). Simply put, the uncorrected p -value was multiplied by the number of comparisons done. Excel spreadsheets were used to examine the relationship between the average daily dry-matter intake, various stable fly pressures and climatic conditions during different seasons.

Subsequently, the effects of stable flies feeding on the average daily gain light (ADGL) weight and average daily gain cold (ADGC) weight of light and heavy weight cattle were analysed using ANOVA and Cohen's d (effect size) tests. A practical significance test was performed on the weight gain data because, other than ANOVA, it is independent of sample size (Ellis & Steyn, 2003). Additionally, the practical significant test indicates whether an effect is large enough to have some practical implication. It indicates the strength of an observed effect between variables. The statistical significant test shows the probability of a relationship between variables. It is important to know that the statistical significant test may not necessarily guarantee practical significance, but to be practically significant the data must be statistically significant.

Practical significance is measured through effect sizes. It is based on standardised mean differences between two groups (Ellis & Steyn, 2003; Steyn, 1999; Steyn, 2000). These effect sizes are described through the use of Cohen's d effect sizes (Ellis & Steyn, 2003; Lakens, 2013). The effect sizes in the present study were interpreted using the following benchmarks suggested by Cohen (1988): small effect ($d = 0.2$), medium effect ($d = 0.5$) and, large effect ($d = 0.8$). Data with an effect size of $d \geq 0.8$ was considered practically significant because it is an indication of a difference with a large effect (Ellis & Steyn, 2003). These values are guidelines for the interpretations of effects sizes, and are generally used in practice (Lakens, 2013).

2.5. INSECTICIDE TREATMENT

In a preliminary study based on the temporal and spatial distribution of stable flies at Karan Beef, Evert (2014) proposed, from the data collected, that a more carefully planned programme with fewer insecticide applications should be implemented at Karan Beef. The number of insecticide types has been reduced to Dimilin® (diflubenzuron), an IGR. Previously applied insecticides were Deltamethrin (residual surface spray), fly baits (bait/attractant), and Cypermethrin (dip/spray) (Evert, 2014).

The cattle feedlot was under regular fly management. The feedlot was treated for the control of stable flies, except for VX-pens which were kept untreated. Information regarding insecticide usage at Karan beef was limited. Dimilin® was applied to manure underneath pen cables. Dimilin® was applied after the first rain in the spring, at an early stage of population development. If necessary, Dimilin® applications were repeated when subjective observations by the feedlot management indicated increased fly activity.

Dimilin® belongs to the benzoyl ureas group of insecticides. It is an IGR which inhibits synthesis of chitin in the larvae's cuticle and prevents insect larvae from moulting, resulting in the death of the insects. A wide variety of formulations exist, each formulated to perform a specific larvicide function. Dimilin® SC-48, SC-15, G-4 and GR-2 are formulated to be applied to developmental sites. For best results, Dimilin® is applied to the upper layer (10-15 cm) of the developmental medium. Dimilin® is used before large fly populations becomes established, and repeated after 2 to 3 weeks (Chemtura, 2015). Recommended rates for fly control in g/10 m² of surface area are summarised in Table 2.1.

Table 2.1: Recommended rates for various Dimilin® formulations for fly control in g/10 m² of surface area. Higher rates are used for first application and lower rates are used every 2 weeks in a control programme (Chemtura, 2015).

	Dimilin® formulations			
	SC - 48	SC - 15	G - 4	GR - 2
Dry and wet surfaces	10 - 20	35 – 70	-	-
Wet surfaces only	10 - 20	35 – 70	125 - 250	250 - 500

2.6. FLY IDENTIFICATION

Collected flies, from both traps and sweep-nets, were mounted and categorised into morphospecies. These fly samples were then sent for identification to Dr Ashley-Kirk Spriggs, a fly specialist at the National Museum, Bloemfontein (Evert, 2014).

CHAPTER 3

TEMPORAL AND SPATIAL DISTRIBUTION OF STABLE FLIES, *STOMOXYS CALCITRANS* (DIPTERA: MUSCIDAE)

3.1. INTRODUCTION

3.1.1. Seasonal abundance of stable flies

Stable flies have a seasonal occurrence pattern with unimodal (Mullens & Meyer, 1987), bimodal (Doud *et al.*, 2012; Jacquiet *et al.*, 2014; Taylor *et al.*, 2007; Taylor *et al.*, 2013) or trimodal peaks in abundance (Lysyk, 1998). These bimodal peaks in abundance were recorded in cattle feedlots in Australia (Urech *et al.*, 2012), semi-arid areas in Mexico (Cruz-Vázquez *et al.*, 2004) as well as in eastern Nebraska (Taylor *et al.*, 2013) and Missouri in the United States (Hall *et al.*, 1983). The seasonal abundance of stable fly populations has been widely studied and has been found to be associated with climatic factors (Rodriguez-Batista *et al.*, 2005). The main climatic factors determining seasonal and year-to-year stable fly abundance were reported by several studies to be temperature (Cruz-Vázquez *et al.*, 2004; Mullens & Meyer, 1987), humidity (Cruz-Vázquez *et al.*, 2004; Mullens & Meyer, 1987) and rainfall (Cruz-Vázquez *et al.*, 2004; Dawit *et al.*, 2012; Taylor *et al.*, 2007).

Temperature affects the rate of change in stable fly populations and is associated with stable fly emergence from overwintering sites (Lysyk, 1993). Severe low temperatures can decrease the development rates of developing immatures, change adult emergence times, or kill adult flies and alter the generation structure (Lysyk, 1993). Adult stable flies feed more readily during spring, summer and autumn than during winter, and cease feeding at temperatures lower than 15°C (Bailey & Meifert, 1973). Stable fly fecundity is highest at 25°C, while fewer eggs are produced at 35°C (Lysyk, 1998). In Australian cattle feedlots, seasonal effects were the main factor determining adult fly populations (Urech *et al.*, 2012). Stable fly abundance was observed to be the lowest during winter months, and highest during autumn and spring (Urech *et al.*, 2012). Lower stable fly density, compared to autumn and spring, was recorded during summer months (Urech *et al.*, 2012). Stable flies are active throughout the year in regions with less severe winter temperatures (Mullens & Meyer, 1987). Stable flies have been reported to be present throughout the year, with minor fluctuations in seasonal trends in central and southern California (Mullens & Meyer, 1987).

In the western province of Canada, stable flies on dairy cattle were active from May to October and showed four population peaks in late August, mid-September and early October (Lysyk, 1993). Low stable fly population numbers observed during winter months (Urech *et*

al., 2012), are most likely a consequence of (1) low rainfall during winter months and high temperatures during summer months, resulting in desiccation of developmental sites (Masmeatathip *et al.*, 2006a), or (2) insecticide application (Masmeatathip *et al.*, 2006a), and/or (3) interspecific competition for developmental sites (Masmeatathip *et al.*, 2006a). Also related with an increase or decrease in stable fly populations are endogenous factors (Taylor & Berkebile, 2011). Decomposition may change the physical characteristics and bacterial communities present in stable fly developmental sites (Taylor & Berkebile, 2011). These bacterial communities are important and necessary for stable fly larvae to complete development (Taylor & Berkebile, 2011). Stable fly abundance was highest during rainy seasons in Thailand (Keawrayup *et al.*, 2012; Masmeatathip *et al.*, 2006a), Pedro Leopoldo and Minas Gerais, Brazil (Rodriguez-Batista *et al.*, 2005) and southern Kaduna, Nigeria (Cruz-Vázquez *et al.*, 2004).

3.1.2. Diurnal activity of stable flies

Stable flies have a diurnal activity pattern, while other muscoid flies such as *Stomoxys sitchensis* and *Stomoxys indica* have a crepuscular pattern (Masmeatathip *et al.*, 2010a). The time of day significantly affects estimates of stable fly populations (Thomas *et al.*, 1989). Stable flies have been recorded to be most active during the day between 10:00 and 16:00, with decreasing activity between dusk and dawn (Campbell *et al.*, 2001; Catangui, 1997). In south-eastern Nebraska, the highest stable fly activity observed at feedlots through front-leg counts on cattle and alsynite fiber glass trap collections was at 14:00 (Thomas *et al.*, 1989).

Unimodal (Berry & Campbell, 1985; Keawrayup *et al.*, 2012) and bimodal (Charlwood & Lopes, 1980; Keawrayup *et al.*, 2012; Kunz & Monty, 1976; Lysyk, 1998; Masmeatathip *et al.*, 2006a; Mitzmain, 1913; Phasuk *et al.*, 2013) feeding patterns have been recorded for adult stable flies. Berry and Campbell (1985) suggested that stable flies with a unimodal, diurnal pattern feed enough to survive until the following bloodmeal at the same time, whereas the bimodal feeding pattern observed in stable flies might be the result of three phenomena: (1) stable flies taking a single bloodmeal during midday depress feeding rates until a second bloodmeal takes place; (2) continuous high temperatures during night and day causes some stable flies to engorge twice a day; and (3) cool weather conditions limit some stable flies from feeding, thereby resulting in different feeding patterns in the following days.

In a study by Berry and Campbell (1985) on the effects of weather on the daily feeding pattern of stable flies, feeding rates were strongly correlated with weather conditions. Temperatures were identified as the major factor influencing stable fly feeding activity (Berry & Campbell, 1985). In winter, stable flies are known to bask in sunny areas (Showler &

Osbrink, 2015) on vertical surfaces such as walls, and on hay bales (Gerry *et al.*, 2007), trees, barns and pole sheds (Showler & Osbrink, 2015). Stable flies were found on the sunny side of animals when temperatures were below 30°C, and on the shaded parts during higher temperatures (Showler & Osbrink, 2015). Stable fly feeding was the highest at temperatures between 24°C and 33.2°C (Berry & Campbell, 1985), and lowest at temperatures below 15°C (Berry & Campbell, 1985; Showler & Osbrink, 2015). Stable flies rest in shaded areas sheltered from extreme heat when their internal temperatures reach 31°C to 34°C (Berry & Campbell, 1985; Buschman & Patterson, 1981; Patterson *et al.*, 1981). In addition to temperature, other factors such as relative humidity, solar radiation and wind have also been identified as factors influencing stable fly feeding activity (Berry & Campbell, 1985). Campbell and Berry (1985) observed that a combination of humidity, radiation and wind increased moisture loss and decreased feeding rates in stable flies (Berry & Campbell, 1985). Depending on the type of feedlot, solar radiation may also increase the internal temperature of stable flies above the ambient temperature (Berry & Campbell, 1985; Mameathip *et al.*, 2006a). Therefore, stable flies present in open feedlots, fully exposed to the sun, rest between feeding periods in shaded areas between bunching cattle. (Berry & Campbell, 1985). At Lake Superior, the direction of the wind (rather than the velocity) had a greater effect on the biting activity of stable flies at beaches (Berry & Campbell, 1985). The wind carries stable flies for great distances onto the beach, approximately 10 to 15 miles inland (Hogsette *et al.*, 1987; Kaufman & Weeks, 2012).

The objective formulated in Chapter 1:

To determine the temporal and spatial distribution of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae) in a feedlot near Heidelberg, Gauteng, South Africa is the aim in Chapter 3.

Specific objectives:

- To determine the seasonal and daily abundance (temporal distribution) of stable flies.
- To determine the stable fly density on cattle during the season.
- To correlate foreleg counts on cattle with trap collections.
- To determine the diversity of immature stable fly stages around feedlot cattle, and the degree of pupal parasitism.
- To determine the influence of cattle breeds and colour on stable fly feeding preference.

3.2. MATERIAL AND METHODS

3.2.1. Seasonal abundance of stable flies

Fly data obtained from a similar study conducted at Karan Beef by Evert (2014) was used as representative data of stable fly abundance from October 2012 to September 2013. The data from Evert (2014) was included in this study to provide a more comprehensive dataset, strengthen previous findings (Evert, 2014) and validate results.

In the present study, adult stable flies were monitored by collecting flies using six Nzi tsetse type traps. A grid of three Nzi tsetse type traps was maintained at two selected sites within the feedlot. Three Nzi tsetse type traps were placed at both the VX-line and H-line (Fig. 3.1). The VX-line is located at the upper side of the feedlot, neighbouring an area covered by weeds and various other vegetation (Fig. 3.1). The H-line (Fig. 3.1), also adjacent to an area of vegetation, was selected due to its history of a high level of irritation observed during the previous years (Evert, 2014). The traps were placed in the open, a few metres from the cattle pens. There was no shading by trees or large vegetation.

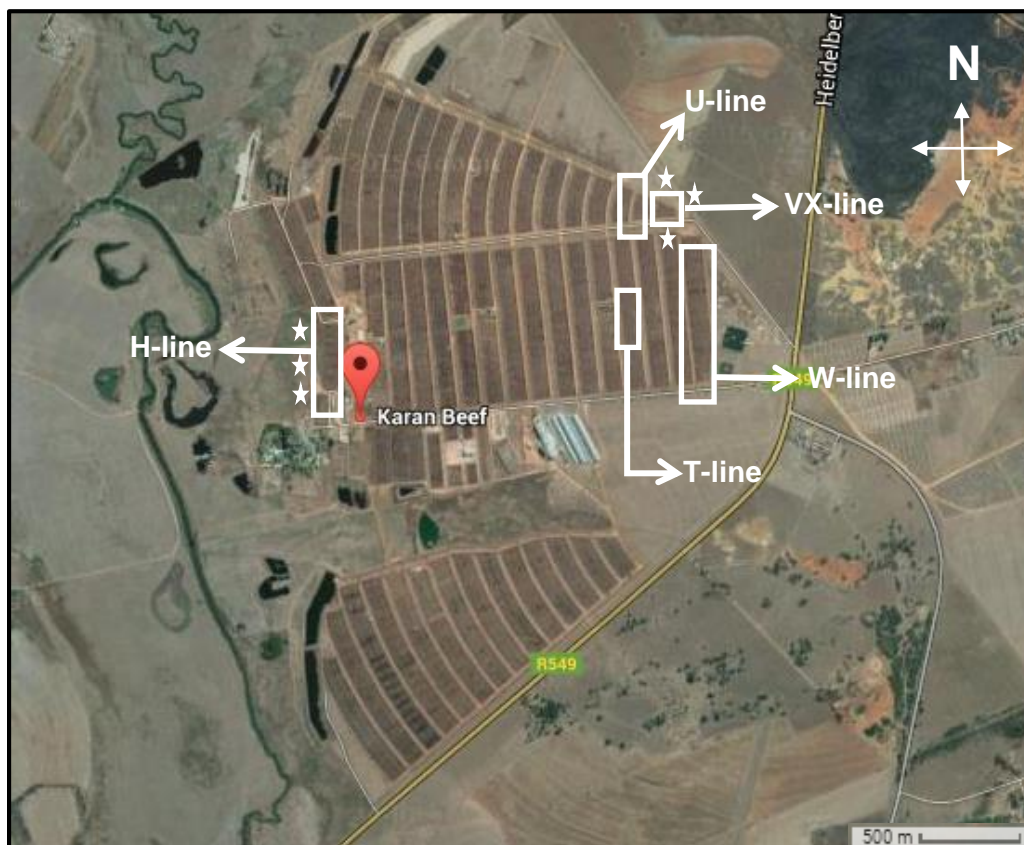


Figure 3.1: An aerial map showing a portion of Karan Beef feedlot, five experimental lines (U-, VX-, W-, T-, and H-line), and the position of the Nzi tsetse type traps and sweeping sites (as indicated by white stars) (Google Maps, 2015).

Stable flies were collected at each site over a seven-day period from May 2015 to December 2015. The traps were serviced weekly throughout the seasons, at 10:00, to ensure consistent sampling from week to week (Evert, 2014). However, during the winter season when fly catches were reduced to zero, sampling was stopped and the traps were removed for repairs. The weekly fly collections were brought back to the laboratory and stored in 70% alcohol until they were identified and counted. The number of stable flies collected were recorded by collection date, time and location.

3.2.2. Diurnal abundance of stable flies

3.2.2.1. Trap and sweep-net collections

Evert (2014) suggested that stable flies rest between bloodmeals in nearby vegetation surrounding Karan Beef feedlot. In this study, the diurnal activity of the adult stable flies at Karan Beef feedlot was determined by collecting flies in 6 Nzi tsetse type traps and sweep-nets (Fig. 3.1). Trap collections and sweep-net catches from weeds and vegetation (see Chapter 2) were done every hour, from sunset to sunrise during the day on 20 February 2014 - from 07:30 to 17:30 at the VX-line and H-line (three Nzi tsetse type traps per site).

3.2.3. Stable fly density on cattle

The stable fly density on cattle was monitored by counting the number of stable flies feeding per minute on the lower extremities and chest floor of cattle (see Section 3.2.3). Stable flies on the forelegs of four cattle per pen were counted. In addition to weekly stable fly leg counts, weekly stable fly trap collections were done. The stable fly density on and around cattle was monitored weekly through trap collections and leg counts, respectively. Stable flies were collected every week from traps at approximately 10:00 while leg counts on cattle forelegs were done at approximately 11:00. Leg counts were done when stable fly activity was expected to be the highest (Evert, 2014). Traps were emptied at the beginning of leg counts (11:00) and re-emptied at 15:00, at the end of each day's stable fly leg counts. This data was used to determine the correlation between the mean daily trap counts and the mean number of stable flies counted per forelegs per minute. This was done to determine the efficiency of the Nzi tsetse type trap as a monitoring tool or control threshold indicator for predicting the stable fly density on cattle - and for the timing of insecticide application (Evert, 2014; Mihok, 2002; Mihok *et al.*, 2006; Mihok *et al.*, 1995a).

3.2.4. Sampling, extraction and care taking of stable fly pupae

The distribution of stable fly developmental sites was determined by removing surface manure (top 10 cm) and vegetative material from four possible stable fly developmental sites at Karan Beef feedlot. Samples were taken from hospital pens, manure heaps, and manure piles underneath border cables of pens at the W-line and VX-line (Fig. 3.1). Vegetative material, consisting of moist manure and hay used as bedding material during the winter, was removed from hospital pens. Manure samples from large manure heaps located approximately 5 km from the feedlot were removed (Evert, 2014). These heaps represented manure from feedlot pens cleaned on a weekly basis and dumped on the large heaps outside the feedlot (Evert, 2014). All samples were done in triplicate (three samples per site). Samples were stored in plastic bags, and immediately sorted for puparia on arrival at the laboratory.

Stable fly developmental media were collected and weighed, after which pupae were removed. The methods most commonly used for removing muscoid fly pupae from stable fly developmental media are the flotation, forced air and autoseparation method (Hogsette, 1992). The flotation method has been used in several studies to collect fly pupae (Berkebile *et al.*, 2009; Fisher & Bergman, 1986; Hogsette, 1992; Jones & Weinzierl, 1997; Ratcliffe *et al.*, 2002). The flotation method entails a seven-step procedure in which samples are firstly placed in a large bucket and flooded with water while thoroughly stirring by hand. After the samples were allowed to stand, the liquid portion was transferred to a second bucket, while the solid portion was flooded again. The solid portion was flooded three times, or until no pupae were recovered. This procedure allowed pupae to float to the surface. All the liquid portions were transferred through a sieve, after which the retained material was transferred to a large white pan where pupae were manually removed with soft forceps and dried on paper towels. The solid portion was examined for puparia that did not float (Utt & Hall, 1992). Puparia were counted, and incubated in labelled petri-dishes at room temperature until adult emergence. The bottom of the petri-dishes were lined with cotton wadding, and kept moist by regularly misting with water every 2 to 3 days. Upon examination, puparia were examined for exit holes that may indicate parasitoid emergence. Emerged flies and parasitoids were removed, counted, identified and stored in 70% ethanol. Puparia from which no adult insects emerged were dissected. Data was recorded on spreadsheets indicating the total number of puparia collected, including the total number of parasitized, emerged, and non-emerged puparia. Emerged flies were categorised as stable flies, house flies and parasitoids.

3.2.5. Influence of cattle breeds and colour on stable fly preference

The influences of cattle breeds and cattle colour on possible stable fly preference were determined by counting the number of stable flies feeding per minute on the forelegs of cattle of different breeds and colours. African beef breeds of cattle selected for the study were Brahman breeds from the *Bos indicus* group; Nguni, Drakensberger and Bonsmara breeds from the *B. taurus* x *B. indicus* crossbreed (sanga) group and European breeds (Charolais, Limousin) from the *Bos taurus* group. Cattle were classified as white, grey, brown, black, beige and mixed-coat coloured animals, based on the predominant colour of the animal's coat. Fly count data was recorded in spreadsheets indicating day, time, number of stable flies, breed, colour, eartag number and pen number.

3.2.6. Climatological data

Stable fly activity in response to climatological factors is important since it could affect stable fly monitoring. Evert (2014) evaluated the possible influence of climatic factors on the daily activity of stable flies using weather data obtained from a local on-site weather station at Karan Beef (Evert, 2014). From these findings it appears as if high rainfall early in the year correlates with the increasing number of stable flies collected weekly in traps (Evert, 2014). A statistically significant positive correlation between mean daily temperature and stable flies collected weekly in traps was recorded (Evert, 2014). On the other hand, a relatively low negative correlation was reported between wind speed and the number of stable flies counted on cattle forelegs. The influence of wind speed on the number of stable flies collected weekly in traps was more difficult to determine and interpret since the on-site weather station recorded daily windspeed measurements and not the actual wind speed at the time of stable fly collection (Evert, 2014). Drawing meaningful conclusions regarding the influence of these climatic factors on the number of stable flies collected weekly in traps were difficult due to the interactivity between the climatic factors and stable fly responses (Evert, 2014).

As per recommendation from a preliminary study by Evert (2014), more direct weather measurements were taken in the field. Temperature and wind speed measurements were taken at the feedlot about 1 m from the pens; at the same time stable fly counts on cattle forelegs were done to interpret the more direct climate impacts on the daily flight activity of stable flies. On-site wind speed measurements were taken with a portable mechanical anemometer while temperature measurements were taken with a HI 9052 portable microprocessor-based thermometer (HANNA© Instruments Product).

3.3. RESULTS AND DISCUSSION

3.3.1. Seasonal abundance of stable flies

The seasonal abundance of stable flies was determined at Karan Beef feedlot with the objective to correlate stable fly numbers with possible meat production impacts (see Chapter 4). Stable fly data collected per week for the period 19 October 2012 to 20 September 2013 at the same feedlot were extracted from Evert (2014). The mean number of stable flies collected weekly with 6 Nzi tsetse type traps from 19 October 2012 to 20 September 2013 (Evert, 2014) and 24 October 2013 to 3 December 2014 is presented in Fig. 3.2 and Fig. 3.3, respectively.

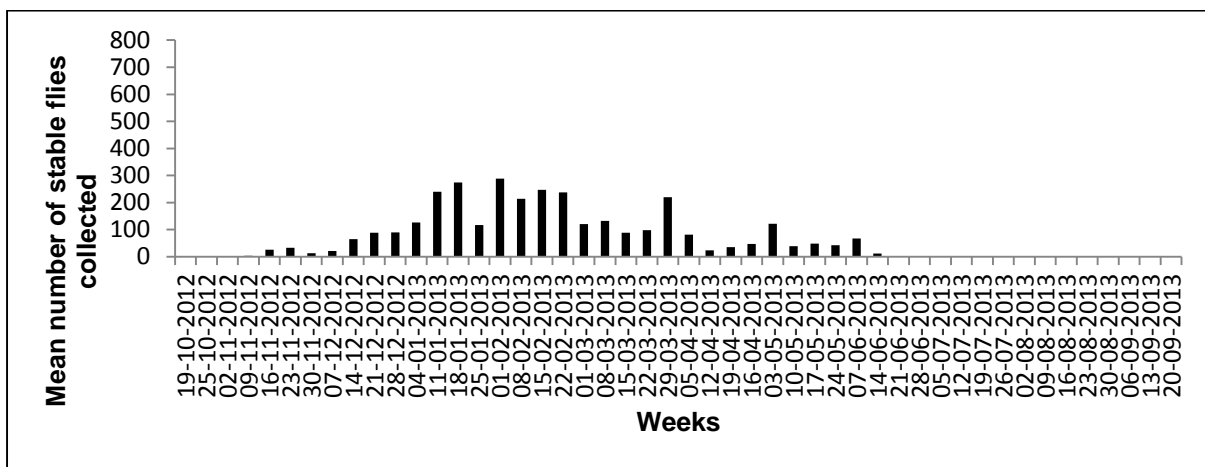


Figure 3.2: Mean number of stable flies collected weekly with 6 Nzi tsetse type traps from 19 October 2012 to 20 September 2013 (Evert, 2014). The given dates indicate periods during which no or few stable flies were collected.

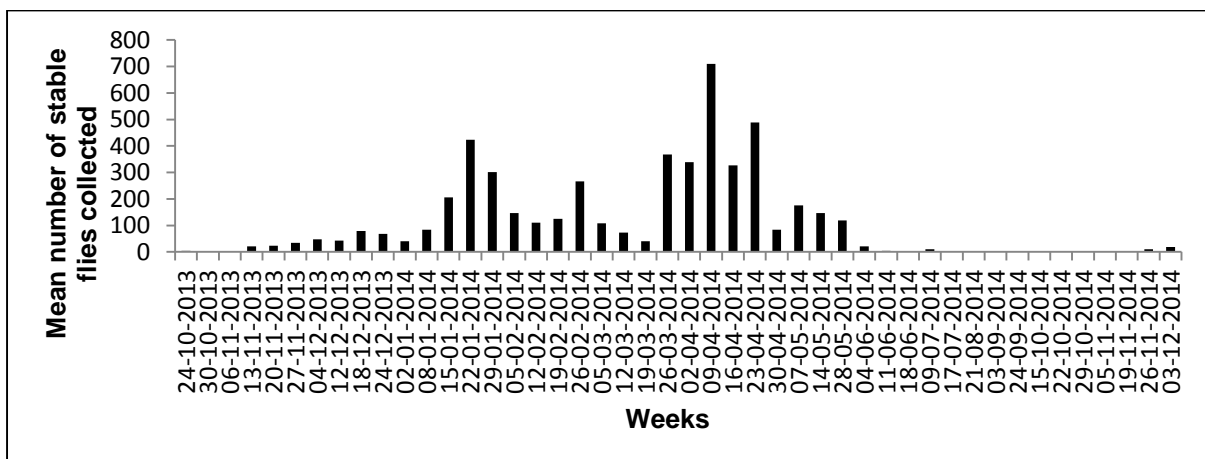


Figure 3.3: Mean number of stable flies collected weekly with 6 Nzi tsetse type traps from 24 October 2013 to 3 December 2014. The given dates indicate the periods during which no or few stable flies were collected.

The total number of stable flies collected during the 11 months for the 2012-2013 survey was 19 204 (Evert, 2014). This study collected a total of 30 478 adult stable flies over a period of 15 months for the 2013-2014 survey. It is clear from Fig. 3.2 and Fig. 3.3 that stable fly numbers were much higher during the 2013-2014 survey than during the 2012-2013 survey.

Roughly five stable fly population peaks were identified during the 2012-2013 and 2013-2014 survey at Karan Beef feedlot. In both surveys after the cold winter months in the following year, stable fly numbers slowly build-up to the first stable fly population peak in January. The number of stable fly population peaks observed during both survey years was relatively similar, with the exception of the size of stable fly population peaks which differed between years. Stable fly population peaks during the 2013-2014 survey, with the exception of the second peak, were much higher than peaks during the 2012-2013 survey. This may be explained by annual differences in environmental factors such as temperature, rainfall and humidity. The absence of stable flies during the winter may be explained by severe frost. The same type of seasonal abundance distribution with the exception of an additional stable fly population peak in Heidelberg was also noted at dairies in Alberta, Canada (Evert, 2014). Climatic differences between areas, such as extremely low winter temperatures in Alberta (-25°C) - much lower compared to those recorded in Heidelberg (-8°C) - may explain the four stable fly population peaks observed in Alberta (Evert, 2014). At lower temperatures, stable flies may have lower developing rates and less degree days (measurement of heat units over time) which may result in fewer generations (Beresford & Sutcliffe, 2012; Evert, 2014; Krafur *et al.*, 1994; Lysyk, 1993; Taylor & Berkebile, 2011).

These findings may prove to be very useful in the development of an IPM programme. High stable fly population peak periods provide significant management directions for the control of stable flies, and serve as an indication of when to expect to apply control measures. Possible control strategies might not be necessary before January (Fig. 3.2 and Fig. 3.3). No fly control is necessary during a large part of the year, especially from May to December (Fig. 3.2 and Fig. 3.3).

3.3.2. Diurnal distribution of stable flies

The total number of stable flies collected hourly on 20 February 2014 at the H-line, using 3 Nzi tsetse type traps and sweep-nets, is summarised in Fig. 3.4.

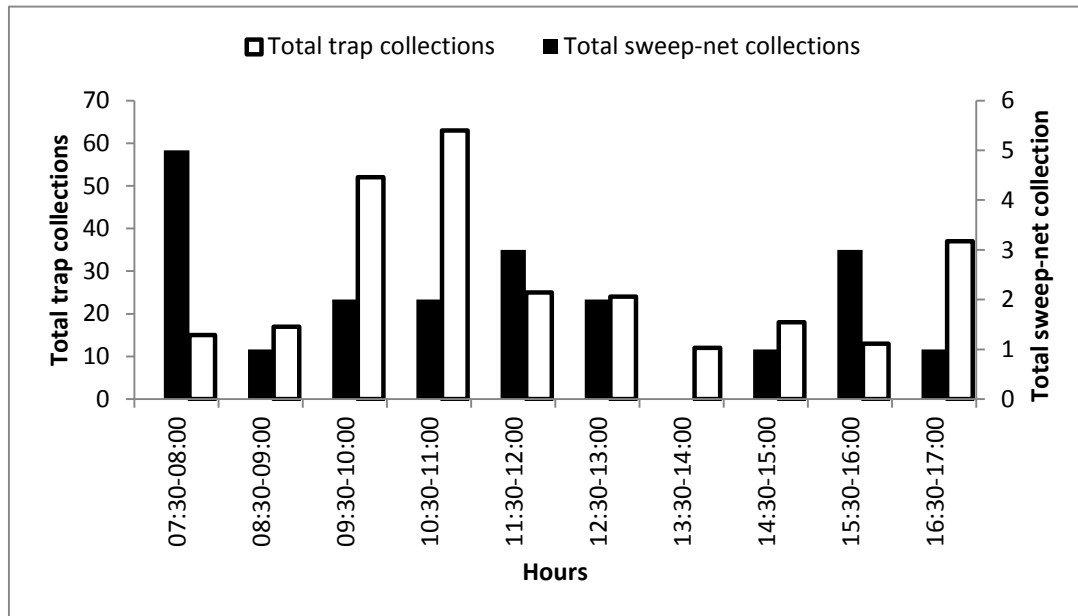


Figure 3.4: Relationship between the total numbers of stable flies collected using 3 Nzi tsetse type traps and sweep-netting in vegetation every hour from 07:30 to 17:00 at the **H-line** on 20 February 2014.

Stable fly trap collections on 20 February 2014 at the H-line showed a bimodal stable fly activity pattern, with an early peak from 09:30 to 11:00 and another between 16:30 and 17:00 (Fig. 3.4). Stable flies were present in all three traps throughout the day. The diurnal activity pattern as observed in these findings is similar to those reported in Thailand, with a bimodal stable fly activity pattern between 08:00 and 10:00 and from 16:00 (Masmeatathip *et al.*, 2006). High temperatures may force stable flies to take a second bloodmeal, resulting in a bimodal feeding pattern (Berry & Campbell, 1985). (see Section 3.1.2, Chapter 3).

High stable fly sweep-net collections early in the morning correspond with low trap collection. Whereas an increase in stable fly traps collections between 08:30 and 11:00 correspond with a decrease in sweep-net collections (Fig. 3.4). A slow increase in sweep-net collection until 12:00 corresponds again with a decrease in trap collections (Fig. 3.4). A second peak in sweep-net collections between 15:30 and 16:00 corresponds with a decrease in trap collections. This is an illustration of the diurnal activity pattern of stable flies feeding and resting at Karan Beef feedlot.

The total number of stable flies collected hourly on 20 February 2014 at the VX-line, using 3 Nzi tsetse type traps and sweep-nets, is summarised in Fig. 3.5.

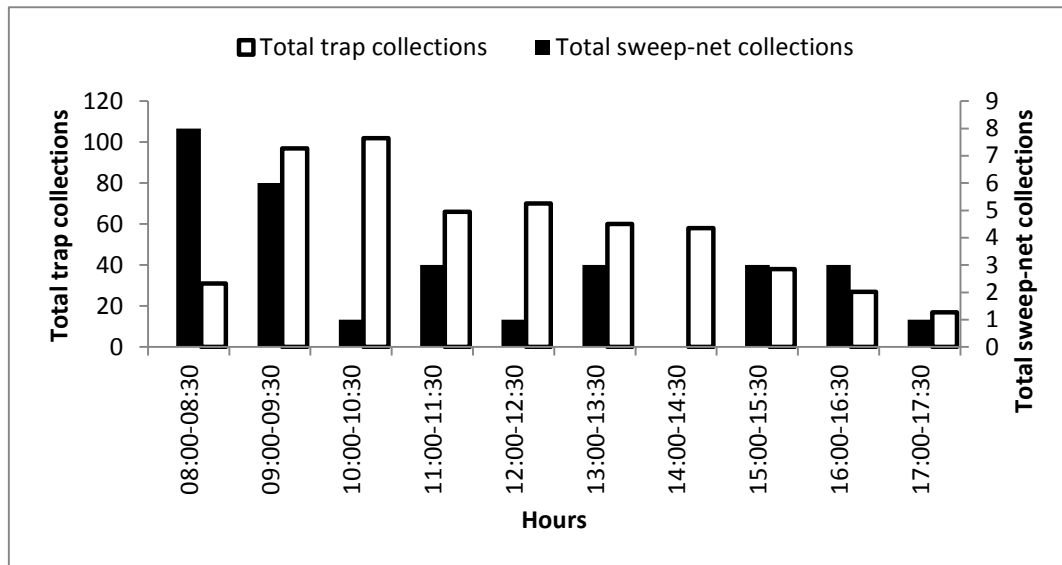


Figure 3.5: Relationship between the total number of stable flies collected using 3 Nzi tsetse type traps and sweep-netting in vegetation every hour from 08:00 to 17:30 at the **VX-line** on the 20 February 2014.

In contrast with the bimodal stable fly activity pattern observed at the H-line, it appears as if stable fly trap collections on 20 February 2014 at the VX-line have a less bimodal stable fly activity pattern, with a major peak occurring between 09:00 and 10:30. However, the sweep-net peak from 15:00 to 16:00 is still present. Stable flies were present in all three traps throughout the day. The diurnal activity pattern observed in these findings is similar to one reported in Nebraska (Berry & Campbell, 1985). Berry and Campbell (1985) noted an unimodal stable fly activity pattern with a peak at midday. Stable flies take enough blood to survive until the next engorgement, resulting in an unimodal feeding pattern (Berry & Campbell, 1985).

High stable fly sweep-net collections early in the morning correspond with low trap collection. An increase in stable fly trap collections between 09:00 and 10:30 correspond with a decrease in sweep-net collections. Low stable fly activity recorded during the day, especially after the peak feeding period, might be due to stable flies resting in nearby weeds and vegetation. Stable flies are also known to rest on vertical surfaces such as walls, hay bales (Gerry *et al.*, 2007), trees, barns and pole sheds (Showler & Osbrink, 2015) between feeding periods to digest ingested blood (Berry & Campbell, 1985). This explanation is supported by the inverse relationship between trap collections and sweep-net collections. These results have important implications for the development of an integrated fly management

programme at Karan Beef feedlot. Weeds and vegetation surrounding the feedlot should be kept short, since it serves as a resting site for stable flies between blood meals. In addition to controlling weeds and vegetation, insecticide application should be concentrated on these resting sites rather than on the entire feedlot. (see Section 1.1.5).

3.3.3. Relationship between daily trap collections and cattle foreleg counts

The relationship between the mean number of stable flies counted per foreleg per minute and the mean number of trap collections using 3 Nzi tsetse type traps are summarised in Fig. 3.6.

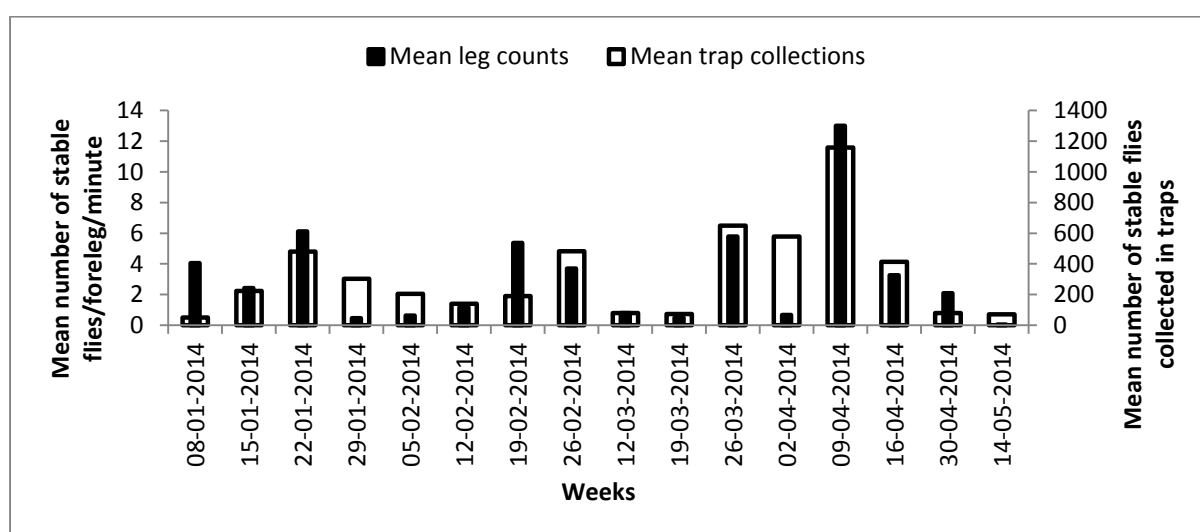


Figure 3.6: Relationship between the mean number of stable flies counted daily per foreleg per minute and the mean number of stable flies collected with 3 Nzi tsetse type traps at the VX-line from 8 January 2014 to 14 May 2014.

Stable fly leg counts fluctuated considerably between daily observations, whereas trap counts were more consistent. Roughly five stable fly population peaks were identified from the mean number of stable flies counted per foreleg per minute (Fig. 3.6). Stable fly numbers were low early March 2014 and May 2014, whereas higher numbers of stable flies were observed on cattle forelegs during January 2014, late March 2014 and April 2014 (Fig. 3.6). The reason trap collections are more consistent (compared to leg counts) may be because stable fly leg counts were more susceptible to weather conditions and other variables at that time (Mullens *et al.*, 1995).

A regression analysis between the mean numbers of stable flies collected with 3 Nzi tsetse type traps and the mean number of stable flies counted on cattle forelegs per minute at the VX-line from 8 January 2014 to 14 May 2014 is summarised in Fig. 3.7.

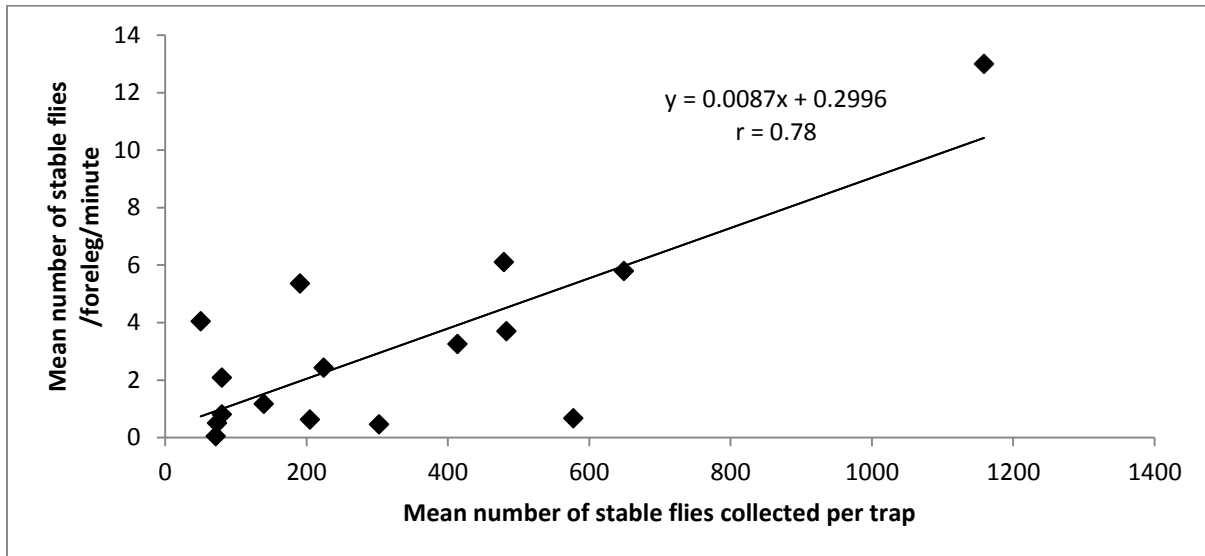


Figure 3.7: Regression analysis between the mean number of stable flies counted weekly per foreleg per minute and the mean number of stable flies collected weekly per Nzi tsetse type trap at the VX-line from 8 January 2014 to 14 May 2014.

Regression analyses between the mean number of stable flies counted weekly per foreleg per minute and the mean number of stable flies collected weekly per Nzi tsetse type trap, delivered a positive linear regression (Fig. 3.7). The linear relationship between the mean number of stable flies collected per trap and the mean number of stable flies counted per minute on cattle forelegs can be used as a model to make predictions within the extent of the regression $y = 0.0087x + 0.2996$ (Fig. 3.7). These findings show that the numbers of stable flies collected in the traps are a fairly good indication of the degree of stable fly activity on cattle during that selected period. Thus it is possible to use stable fly catch rates as predictions for the degree of stable fly irritation on cattle. Evert (2014) reported, from findings in a preliminary study at the same feedlot, that stable fly trap collections could also be used to indicate the level of stable fly activity on cattle in a feedlot on the same day. A strong seasonal correlation between trap collections and foreleg counts was recorded from various studies (Mullens & Meyer, 1987; Thomas *et al.*, 1989). From these observations, it appeared that animal counts were effective in estimating fly populations (Mullens *et al.*, 2006; Warnes & Finlayson, 1987) and could be used as an indication for insecticide application (Evert, 2014). However, further research on the economic threshold for larger stable fly populations for chemical control in South Africa is still needed (Evert, 2014).

3.3.4. Possible influence of climatic factors on the diurnal activity of stable flies

The possible influence of climate factors (temperature and rainfall) on the diurnal activity of stable flies was analysed by correlating temperature and mean daily wind speed with the mean number of stable flies counted weekly on cattle forelegs per minute. Weather data obtained from on-site measurements was used (see Section 3.2.6). The possible influence of temperature and wind speed on the mean number of stable flies counted weekly on cattle forelegs per minute is summarised in Fig. 3.8.

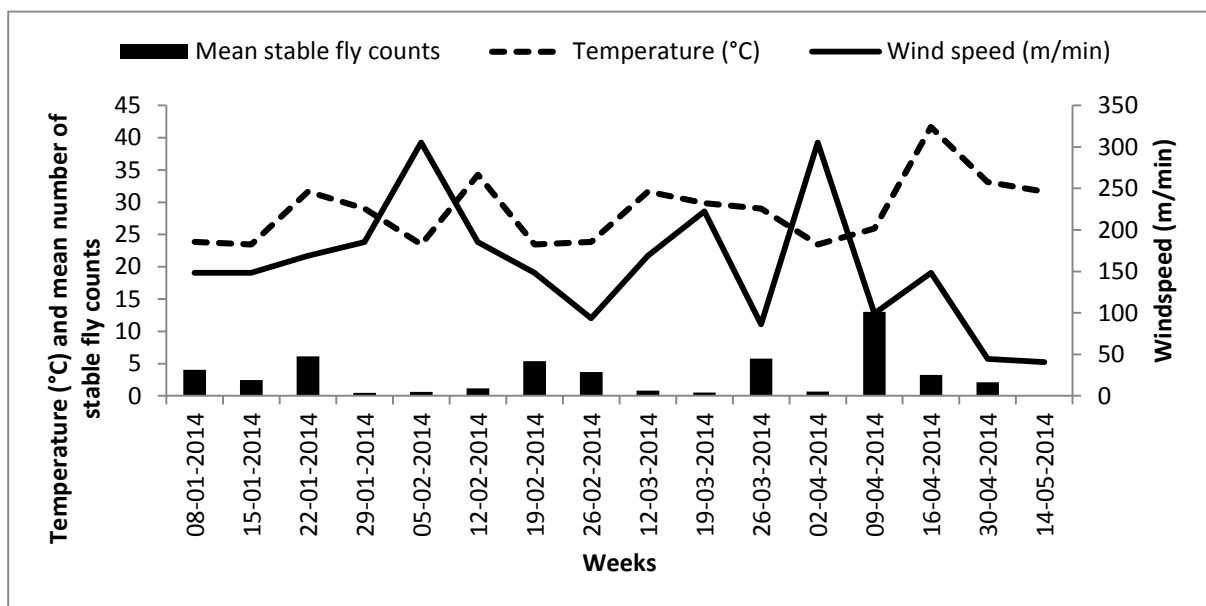


Figure 3.8: Relationship between temperature (°C), wind speed (m/min) and mean number of stable flies counted weekly on cattle forelegs per minute from 8 January 2014 to 14 May 2014.

The possible influence of temperature on the diurnal activity of stable flies: It appears as if temperature had some effect on the mean number of stable flies counted weekly on cattle forelegs per minute (Fig. 3.8). Temperature is the most important factor influencing stable fly feeding activity, since it affects metabolic and other physiological rates which in turn affect behavioural responses of stable flies (Berry & Campbell, 1985). Temperature influences the rates of stable fly development and populations (Lysyk, 1993), as reflected by the increase in leg counts (Fig. 3.8). A similar observation was also noted by Evert (2014), between weekly stable fly trap collections and mean temperature measurement from a local on-site weather station.

The relationship between temperature and mean number of stable flies counted weekly on cattle forelegs per minute was quantified in a regression analysis summarised in Fig. 3.9.

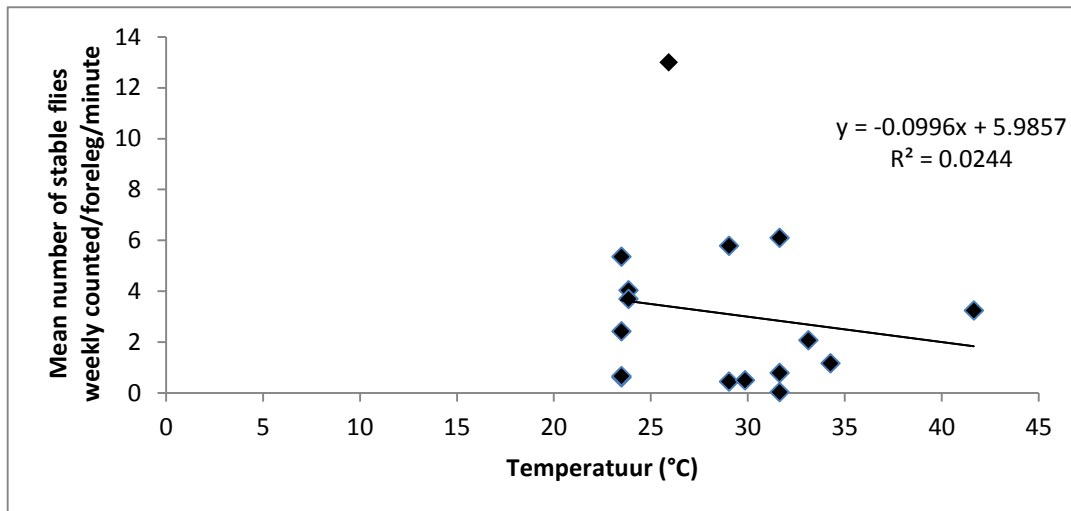


Figure 3.9: Regression analyses showing the relationship between temperature (°C) and mean number of stable flies weekly counted on cattle forelegs per minute from 8 January 2014 to 14 May 2014.

Regression analyses between temperature and mean number of stable flies counted weekly on cattle forelegs presented a negative linear regression (Fig. 3.9). The linear relationship between temperature and mean number of stable flies counted weekly on cattle forelegs per minute is very weak ($r=0.17$), and not suitable for predictions (Fig. 3.9).

The possible influence of wind speed on the diurnal activity of stable flies: It appears as if wind speed had some effect on the mean number of stable flies counted weekly on cattle forelegs per minute (Fig. 3.8). In addition to temperature, wind speed is an important factor to take into account when determining the diurnal activity pattern of stable flies. During strong winds, stable flies have been reported to shelter in vegetation and buildings (Broce *et al.*, 1991) and on cattle. Thus the high number of stable flies observed on cattle forelegs on 2 April 2015 might be due to stable flies sheltering from strong winds. Stable flies resting between bunching cattle might increase heat stress in cattle and cause significant weight gain reductions (Wieman *et al.*, 1992). The relationship between wind speed and mean number of stable flies counted weekly on cattle forelegs per minute was quantified in a regression analysis summarised in Fig. 3.10.

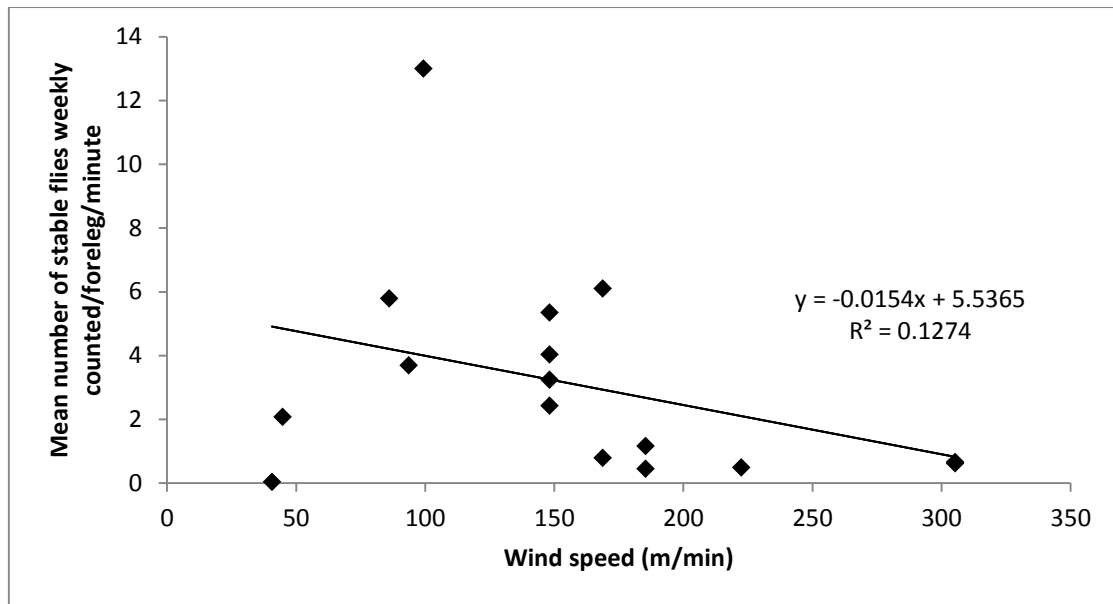


Figure 3.10: Regression analyses showing the relationship between wind speed (m/min) and mean number of stable flies weekly counted on cattle forelegs per minute from 8 January 2014 to 14 May 2014.

Regression analyses between wind speed and mean number of stable flies counted weekly on cattle forelegs presents a negative linear regression (Fig. 3.10). Although the linear relationship between wind speed and mean number of stable flies counted weekly on cattle forelegs per minute is relatively weak ($r=0.36$)(Fig. 3.10).

3.3.5. Diversity of developmental sites and degree of parasitism of stable fly pupae

The diversity of stable fly developmental sites at Karan Beef feedlot was determined (see Section 2.1). The total number of stable fly pupae collected per gram breeding media per site from 19 March 2014 to 17 July 2014 is presented in Fig. 3.11.

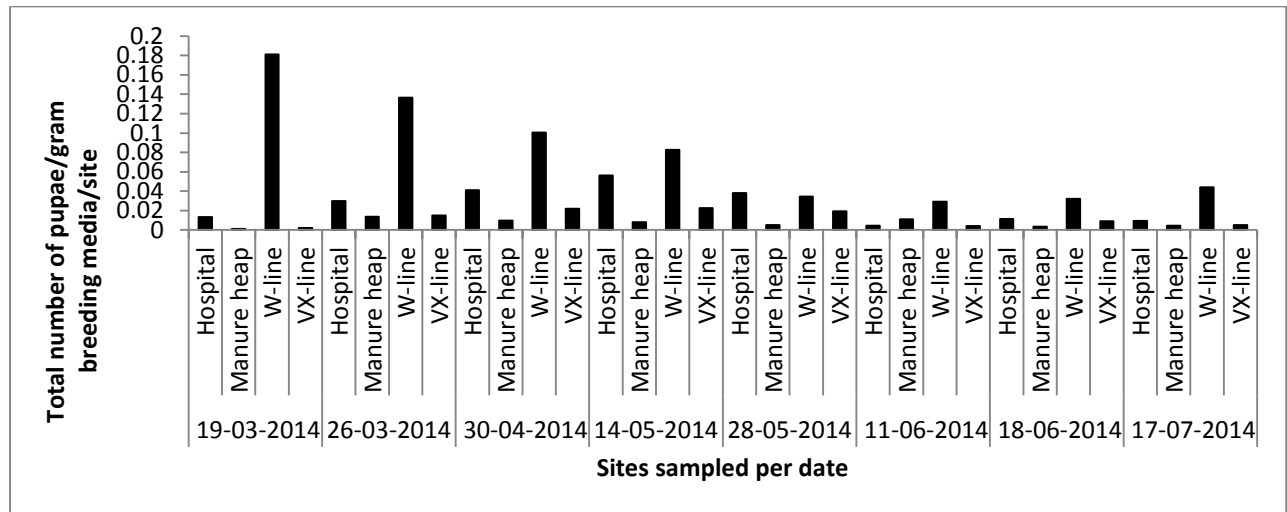


Figure 3.11: Total numbers of stable fly pupae/gram breeding media/site collected from four potential developmental sites from 19 March 2014 to 17 July 2014 at Karan Beef feedlot.

The highest number of pupae/gram breeding media was collected from the W-line. Manure build-up under fence lines at the W-line might have allowed continuous stable fly breeding during the season, with relatively high moisture content under the fence cables. Although treated for the control of stable flies, it could be that manure piles were impenetrable by insecticides and pupation was not affected by insecticide and IGR application. In addition to the W-line, a relatively high number of pupae/gram breeding media was also collected from the hospital pens. Piled manure and piled hay might provide a temperature gradient suitable for developing stable flies, especially overwintering stable fly immature stages (see Chapter 1). Although not treated for the control of stable flies, routine removal of developmental sites might explain low numbers of pupae/gram breeding media collected at the VX-line. Stable fly immature stages at both the VX-line and manure heaps might have been more susceptible to desiccation, due to high temperatures and low moisture content, thus explaining low numbers of pupae/gram breeding media/site. Piled manure, piled haylage and similar habitats at Karan Beef feedlot need to be further examined as potential developmental sites for stable fly immature stages during the winter.

The relative abundance (%) of emerged stable fly and house fly puparia, and parasitoids recovered from fly puparia collected from possible stable fly developmental sites is summarised in Fig. 3.12.

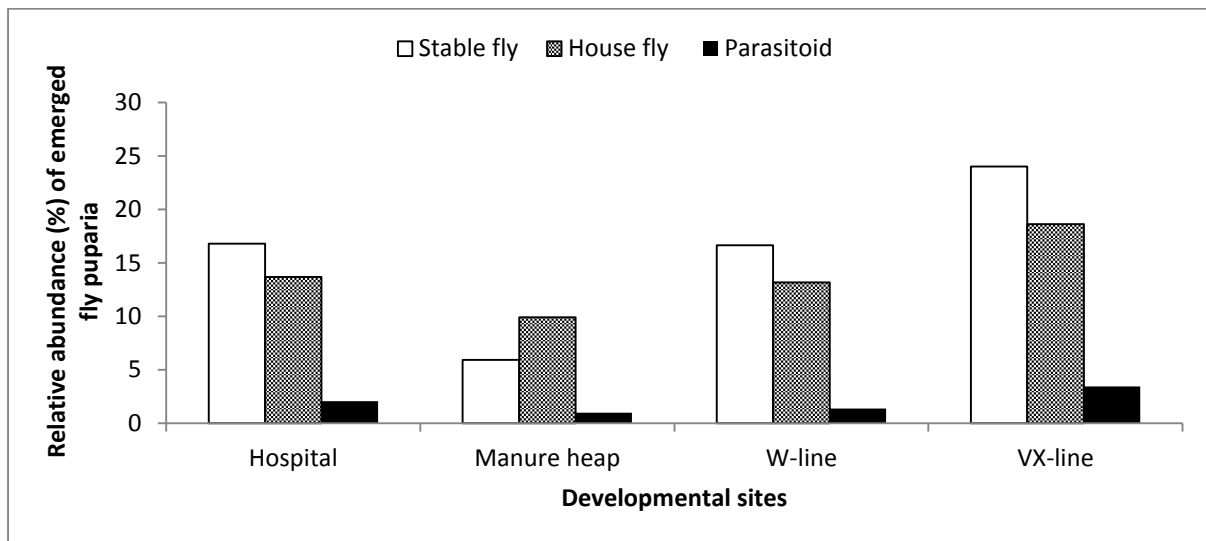


Figure 3.12: Relative abundance (%) of emerged stable fly and house fly puparia, and parasitoids recovered from fly puparia collected from possible stable fly developmental sites, from 19 March 2014 to 17 July 2014 at Karan Beef feedlot.

Overall parasitism of the total number of fly puparia collected per developmental site was relatively low. However, percentage parasitism of the total number of fly puparia collected at the VX-line was higher compared to parasitism at the W-line. In addition to higher parasitism of fly puparia, relatively more fly puparia emerged at the VX-line than the other three developmental sites, most likely due to no stable fly control at the VX-line. The effect of Dimilin® on the emergence of fly puparia is visible at the hospital pens and W-line. Parasitoids as a potential biological control agent of stable flies and house flies need to be further examined.

3.3.6. The influence of cattle breeds and colour on stable fly preference

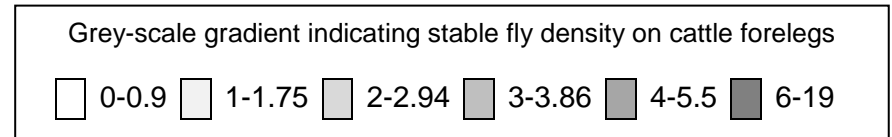
Stable fly abundance was monitored weekly through leg counts on cattle, to determine the influence of cattle breeds and cattle colour on stable fly preference. The possible influence of cattle breed on stable fly preference per week is summarised in Table 3.1.

Table 3.1: Stable flies (*S. calcitrans*) counted per week on the forelegs of individual cattle breeds from 8 January 2014 to 25 March 2015.

	Breeds																		ANOVA P value	Kruskal-Wallis P value
	Charolais			Brahman			Bonsmara			Nguni			Drakensberger			Limousin				
	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD		
Week 1	2	3.50	2.12	16	3.63	3.07	33	4.67	3.01	8	2.13	2.10	0	-	-	0	-	-	0.1598	1.000
Week 2	1	2	0	10	5.10	3.84	7	3.29	1.38	1	3	0	2	2.50	2.12	0	-	-	0.6180	1.000
Week 3	2	0.50	0.71	23	3.78	2.43	26	4.27	3.57	7	3.86	3.02	3	7.33	11.02	8	4.38	2.72	0.4444	0.4622
Week 4	1	19 ^A	0	19	3.26 ^B	3.43	29	2.72 ^B	2.23	8	4 ^B	3.21	6	2.17 ^B	1.47	6	3.17 ^B	0.75	0.0001*	0.4074
Week 5	2	0	0	20	2.25 ^C	1.77	36	0.67 ^D	0.89	4	0.75	1.50	5	1	1.22	4	0.50	1	0.0010*	0.0117*
Week 6	4	1	1.15	20	2	2.10	27	1.22	1.39	5	0.6	0.54	10	1.4	1.34	1	2	0	0.4549	0.7094
Week 7	0	-	-	8	4.5	2	9	5.22	3.38	1	0	0	4	4	2.82	1	5	0	0.5287	1.000
Week 8	2	5.5	0.70	17	2.94	3.76	32	4.03	3.21	8	4.37	5.09	4	3	2.30	3	1	1.73	0.5897	0.3144
Week 9	1	0	0	11	1.27	2.05	45	0.84	1.13	8	0.25	0.46	8	0.5	0.53	1	0	0	0.4671	0.5366
Week 10	7	2.57	5.94	26	2	5.49	26	1.12	1.80	14	1	1.18	11	1.36	2.11	0	-	-	0.8134	1.000
Week 11	10	11.4 ^F	5.80	22	2.18 ^E	4.08	31	7.32	5.76	11	7.18	5.93	10	10.9 ^F	8.96	1	2	0	0.0004*	0.0001*
Week 12	4	0.75	0.96	14	1.07	1.64	44	0.90	2.01	6	0	0	13	0.31	0.63	0	-	-	0.5651	1.000
Week 13	3	0	0	19	0.58	1.35	28	0.82	1.44	14	0.43	0.94	8	0.88	2.47	0	-	-	0.8195	1.000
Week 14	1	0	0	5	0	0	8	1.5	1.41	3	2.33	0.58	6	3.17	2.93	1	5	0	0.0572	0.0478*
Week 15	0	-	-	5	0.4	0.05	7	0.29	0.49	7	0.29	0.76	4	1	0.82	1	2	0	0.0937	1.000
Week 16	0	-	-	5	0.6	0.55	10	2.4	2.27	4	3.25	2.06	4	2.5	2.52	1	6	0	0.1494	1.000
Week 17	0	-	-	6	1.5	2.26	11	0	0	2	0	0	4	0	0	1	0	0	0.1544	1.000
Week 18	0	-	-	1	3	0	13	2.92	2.10	5	1.4	0.89	3	1.67	1.15	0	-	-	0.3848	1.000
Week 19	0	-	-	5	1	1.41	10	1.2	1.62	3	2.33	2.52	5	1.2	2.68	0	-	-	0.8040	1.000
Week 20	0	-	-	3	0.33	0.58	12	1.42	2.07	4	1.75	1.5	3	0.67	0.58	0	-	-	0.6670	1.000

Per breed: *n* = total number of cattle per breed per week, \bar{x} = mean number of stable flies counted per foreleg, SD = standard deviation. Per week: Means followed by the same letters do not differ significantly between breeds. Means followed by different letters differ significantly (Unequal N HSD test, $p < 0.05$) between breeds.

*Statistically significant difference at $p < 0.05$.



ANOVA was used to analyse stable fly leg count data. In addition to ANOVA, the Kruskal-Wallis test was performed because of unequal numbers of breeds and very low numbers of some breeds counted during some weeks. The data was analysed per week due to time dependency. Several breeds of cattle seemed to be more attractive to feeding stable fly populations. However, the relative mean number of stable flies counted on breeds fluctuated significantly between weeks and no significant pattern in stable fly preference for the selected cattle breeds could be established. Leg counts differed significantly between breeds in week 4, 5 and 11 (ANOVA; $p < 0.05$). Kruskal-Wallis showed statistically significant differences in leg count data in week 5, 11 and 14 ($p < 0.05$).

The cattle breed preferred by feeding stable flies varied extensively between weekly observations. Charolais breeds elicited the highest feeding response from stable flies, significantly higher than Brahman, Bonsmara, Nguni, Drakensberger and Limousin breeds in week 4. In week 11, stable fly leg counts on Charolais breeds were significantly higher than Brahman and Drakensberger breeds. However, this high count was a once-off phenomenon and seems like an outlier. It seems as if cattle breed is not the only factor influencing stable fly preference. Stable fly counts on Brahman breeds were significantly higher than Bonsmara breeds in week 5, but significantly lower than Charolais and Drakensberger breeds in week 11. Little conformation in literature regarding the influence of cattle breed and colour on stable fly preference were available. However, other flies such as horn fly populations were reported to be higher on both purebred Angus and Charolais breeds compared to purebred Brahmans (Catangui *et al.*, 1993). These studies also reported that animals with a higher percentage of Brahman blood, regardless of coat colour, are less attractive to horn flies than Angus and Charolais (Catangui *et al.*, 1993). These findings are in contrast to the study by Catangui *et al.* (1995) in which both Brahman-crossbreeds heifers and European breeds were similarly susceptible to stable fly feeding.

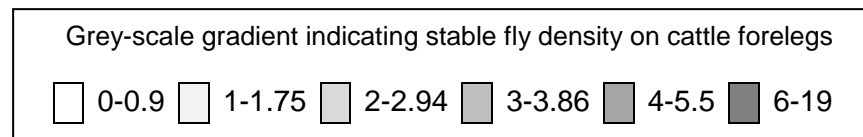
The possible influence of cattle coat colour on stable fly preference per week is summarised in Table 3.2.

Table 3.2: Stable flies counted per week on the forelegs of cattle of different coat colours from 8 January 2014 to 25 March 2015.

	Colours																		ANOVA P value	Kruskal-Wallis P value
	White			Grey			Brown			Mix			Black			Beige				
	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD		
Week 1	2	3.50	2.12	14	3.71	3.07	35	4.57	3.03	8	2.12	2.1	0	-	-	0	-	-	0.1989	1.000
Week 2	1	2	0	8	5.63	3.85	9	3.22	1.92	1	3	0	2	2.5	2.12	0	-	-	0.4145	1.000
Week 3	2	0.5	0.71	14	3.57	2.68	33	4.09	3.25	7	3.86	3.02	5	7	8.87	8	4.38	2.72	0.3356	0.4071
Week 4	1	19 ^A	0	13	1.69 ^B	1.80	34	3.35 ^B	2.94	8	4 ^B	3.21	7	2.57 ^B	1.72	6	3.17 ^B	0.75	0.0001*	0.1200
Week 5	2	0	0	15	1.87	1.81	40	0.95	1.26	4	0.75	1.5	6	1.33	1.37	4	0.5	1	0.2100	0.2377
Week 6	4	1	1.15	10	1.9	2.08	36	1.36	1.59	5	0.6	0.55	11	1.73	1.68	1	2	0	0.7022	0.7543
Week 7	0	-	-	3	2.33	0.58	9	5.22	3.38	1	0	0	9	5	2.12	1	5	0	0.2565	1.000
Week 8	2	5.5	0.71	10	1.1	1.20	34	3.94	3.16	8	4.38	5.10	9	5.11	4.31	3	1	1.73	0.0773	0.0382*
Week 9	1	0	0	5	2.4 ^C	2.61	49	0.82	1.11	8	0.25 ^D	0.46	10	0.4	0.52	1	0	0	0.0267*	0.0858
Week 10	7	2.57	5.94	9	4.67	8.96	34	1.06	1.65	14	1	1.18	20	0.9	1.68	0	-	-	0.0713	1.000
Week 11	10	11.4	5.80	13	3.31	5.02	36	6.44	5.78	11	7.18	5.93	14	7.86	9.04	1	2	0	0.0812	0.0373*
Week 12	4	0.75	0.96	10	0.6	0.84	46	1.07	2.10	6	0	0	15	0.27	0.59	0	-	-	0.3921	1.000
Week 13	3	0	0	11	0.64	1.57	34	0.71	1.34	14	0.43	0.94	10	1	2.31	0	-	-	0.8187	1.000
Week 14	1	0	0	3	0	0	10	1.2	1.40	3	2.33	0.58	6	3.17	2.92	1	5	0	0.0892	0.0906
Week 15	0	-	-	2	0	0	9	0.44	0.53	7	0.29	0.76	5	0.8	0.84	1	2	0	0.1362	1.000
Week 16	1	0	0	2	0.5	0.70	10	2.4	2.27	4	3.25	2.06	6	2	2.10	1	6	0	0.3042	0.1922
Week 17	0	-	-	3	0.33	0.58	13	0.54	1.66	2	0	0	5	0.2	0.45	1	0	0	0.9708	1.000
Week 18	0	-	-	1	3	0	13	2.92	2.10	5	1.4	0.89	3	1.67	1.15	0	-	-	0.3848	1.000
Week 19	0	-	-	5	0.75	1.5	11	1.27	1.56	3	2.33	2.52	5	1.2	2.68	0	-	-	0.7608	1.000
Week 20	0	-	-	2	0	0	13	1.38	1.98	4	1.75	1.5	3	0.67	0.58	0	-	-	0.6287	1.000

Per coat colour: *n* = total number of cattle per coat colour per week, \bar{x} = mean number of stable flies counted per foreleg, SD = standard deviation. Per week: Means followed by the same letters do not differ significantly between coat colours. Means followed by different letters differ significantly (Unequal N HSD test, $p < 0.05$) between coat colours.

* Statistically significant difference at $p < 0.05$.



ANOVA was used to analyse stable fly leg count data. In addition to ANOVA, a Kruskal-Wallis test was performed due to unequal numbers of cattle per colour and very low numbers of some colours counted during some weeks. This data was analysed per week due to time dependency. Several cattle of various coat colours seemed to be attractive to feeding stable fly populations. Cattle of both dark and light coloured coats were continuously attacked by stable flies. However, the relative mean number of stable flies counted on cattle of various coat colours fluctuated significantly between weeks, and no significant pattern in stable fly preference for the selected coat colour could be established. Leg counts differed significantly in week 4 and 9 (ANOVA; $p < 0.05$). Kruskal-Wallis showed statistical significant differences in leg count data in week 8 and 11 ($p < 0.05$). Unequal number of cattle per coat colour might explain significant differences

The coat colour preferred by feeding stable flies varied extensively between weekly observations. White-coloured coats elicited the highest feeding response from stable flies, significantly higher than grey, brown, mix, black and beige coloured cattle in week 4. In week 9, stable fly leg counts on cattle with a grey coloured coat were significantly higher than cattle with a mixed coloured coat. It seems as if coat colour is not the only factor influencing stable fly preference. Franks *et al.* (1964) suggested that the colour of the breed might not be the only factor that determines the activeness, especially to horn flies.

The possible influence of cattle breed on stable fly (*Stomoxys calcitrans*) preference per season is summarised in Table 3.3.

Table 3.3: Mean number of stable flies counted per week on the foreleg of individual cattle breeds from 8 January 2014 to 25 March 2015.

Breeds																			
Charolais			Brahman			Bonsmara			Nguni			Drakensberger			Limousin			ANOVA P value	Kruskal-Wallis P value
<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD		
40	4.48 ^A	6.26	255	2.32	3.24	444	3.40	3.21	123	2.15 ^B	3.29	113	2.41	4.46	29	2.79	2.34	0.0117*	0.1932

Per breed: *n* = total number of cattle per breed, \bar{x} = mean number of stable flies counted per foreleg, SD = standard deviation. Means followed by different letters differ significantly (Unequal N HSD test; $p < 0.05$) between breeds.

* Statistically significant difference at $p < 0.05$

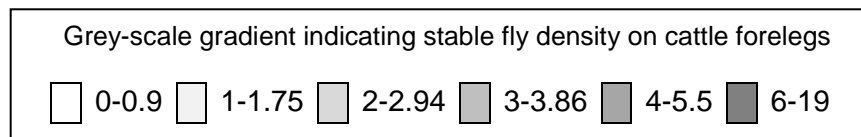
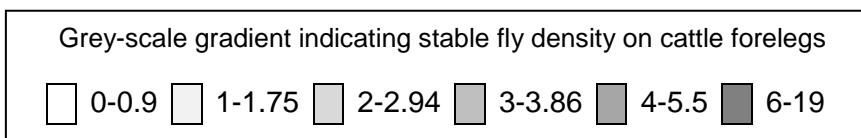


Table 3.4: Mean number of Stable flies counted on the foreleg of cattle of different coat colours from 8 January 2014 to 25 March 2015.

Colours																			
White			Grey			Brown			Mix			Black			Beige			ANOVA P value	Kruskal-Wallis P value
<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD	<i>n</i>	\bar{x}	SD		
41	4.37 ^A	6.22	152	2.32	3.47	508	2.37	3.14	123	2.15 ^B	3.29	151	2.45	4.20	29	2.79	2.34	0.0182*	0.2326

Per coat colour: *n* = total number of cattle per colour; \bar{x} = mean number of stable flies counted per foreleg, SD = standard deviation. Means followed by different letters differ significantly (Unequal N HSD test; $p < 0.05$) between coat colours.

* Statistically significant difference at $p < 0.05$



Since no significant pattern in stable fly preference for selected cattle breeds between weeks was established, time dependency was ignored. ANOVA and Kruskal-Wallis analyses were performed on stable fly leg count data to determine the possible influence of cattle breed on stable fly preference. Several breeds of cattle seemed to be attractive to feeding stable fly populations. The relative mean number of stable flies counted on cattle forelegs fluctuated significantly between breeds ($p = 0.0117$). Stable fly leg counts on Charolais breeds were significantly higher than Nguni breeds.

The possible influence of cattle coat colour on stable fly preference per season is summarised in Table 3.4. Since no significant pattern in stable fly preference for selected cattle coat colours between weeks was established, time dependency was ignored. ANOVA and Kruskal-Wallis was performed on stable fly leg count data to determine the possible influence of cattle breed on stable fly preference. Several cattle of various coat colours seemed to be attractive to feeding stable fly populations. The relative mean number of stable flies counted on cattle forelegs differed significantly between cattle of various coat colours ($p = 0.0182$). Stable fly leg counts on breeds with a white-coloured coat were statistically significant higher than breeds with a mixed-colour coat.

CHAPTER 4

IMPACT OF STABLE FLIES, *STOMOXYS CALCITRANS* (DIPTERA: MUSCIDAE) ON THE FEED CONSUMPTION AND WEIGHT GAIN PERFORMANCE OF FEEDLOT CATTLE

4.1. INTRODUCTION

Stable flies are a major nuisance in and around confined cattle feeding operations such as dairies (Bruce & Decker, 1958; Meyer & Petersen, 1983), feedlots (Campbell *et al.*, 1987, 2001; Hall *et al.*, 1983; Meyer & Peterson, 1983), pastures (Broce *et al.*, 2005; Campbell *et al.*, 2001; Hall *et al.*, 1982) and rangelands (Campbell *et al.*, 2001; Hall *et al.*, 1982; Taylor & Berkebile, 2011; Taylor *et al.*, 2013). Evert (2014) determined the seasonal abundance and distribution of nuisance fly species at Karan Beef. The most commonly collected adult flies at Karan Beef were *Haematopota* (Tabanidae), *Stomoxys calcitrans* (Muscidae), *Musca* (Muscidae), Scatopsidae, *Plecia* (Bibionidae), Chironomidae, *Atherigona* (Muscidae), Milichiidae and Sphaeroceridae (Evert, 2014). Stable flies were identified as a major species of concern on feedlot cattle at Karan Beef, due to their high numbers and proven levels of irritation (Evert, 2014). Therefore, this study investigates stable fly populations and their impact on feedlot cattle at Karan Beef.

High stable fly densities can cause significant production losses in dairy and beef industries (Meyer & Petersen, 1983; Taylor *et al.*, 2012) and severe animal health and welfare concerns (Baldacchino *et al.*, 2013; Schwinghammer *et al.*, 1986; Urech *et al.*, 2012). When attacked by stable flies, cattle move to areas with lower fly activity such as windy hilltops or water (Campbell *et al.*, 1977; Johnson, 2011; Mullens *et al.*, 2006; Schole *et al.*, 2011). Cattle aggregate in a tight bunch in an attempt to avoid attacks (Campbell *et al.*, 1993; Johnson, 2011; Mullens *et al.*, 2006; Schofield & Torr, 2002; Schole *et al.*, 2011; Taylor *et al.*, 2012; Wieman *et al.*, 1992). Each animal will attempt to move deeper into the centre of the bunch to reduce the intensity of the biting activity (Berry *et al.*, 1983, Johnson, 2011). Furthermore, in an attempt to dislodge stable flies, cattle will exhibit fly repelling behaviours such as head throwing, foot stomping, skin twitching and tail flicking (Broce *et al.*, 2005; Campbell *et al.*, 1993; Dougherty *et al.*, 1993; Johnson, 2011; Mullens *et al.*, 2006; Schole *et al.*, 2011; Taylor *et al.*, 2012). The severe biting activity, together with the attempts to fight them off, result in metabolic costs (Jonsson & Mayer, 1999; Schole *et al.*, 2011; Taylor *et al.*, 2012), irritation, stress (Schwinghammer *et al.*, 1986), exsanguinations (Baldacchino *et al.*, 2013; Bishop, 1927; Hall *et al.*, 1982; Hall *et al.*, 1983), fatigue (Baldacchino *et al.*, 2013), transmission of pathogenic organisms (Baldacchino *et al.*, 2013; Hall *et al.*, 1982; Hall *et al.*, 1983; Holdsworth *et al.*, 2006; Sumba *et al.*, 1998), a decrease in feeding time (Dougherty *et*

al., 1993; Hall *et al.*, 1982) and feed intake (Dougherty *et al.*, 1993; Meyer & Peterson, 1983). Furthermore, heat generated as a consequence of bunching and defensive behaviour may induce a thermostatic satiety-regulating mechanism in cattle which inhibits feeding (Dougherty *et al.*, 1993; Dougherty *et al.*, 1995; Wieman *et al.*, 1992). As a result, cattle under these pressures become more susceptible to secondary infections (Hall *et al.*, 1982) and show signs of reduced weight gains (Campbell *et al.*, 1977; Campbell *et al.*, 2001; Catangui *et al.*, 1997) and milk production (Bruce & Decker, 1958; Hall *et al.*, 1982; Miller *et al.*, 1973).

It is important to know in what way animals respond to environmental challenges in order to develop improved strategies to minimize or eliminate potential production losses during extreme weather conditions (Hahn, 1999). Changes in the thermal environment of cattle as a result of weather events may have adverse impacts on cattle performance (Belasco *et al.* 2015; Bolsen & Pollard, 2004; Hahn, 1985). Extreme weather conditions induce a variety of physiological responses (Christopherson & Kennedy, 1983) which can reduce weight gain and feed efficiency in cattle (Belasco *et al.*, 2015; Hoelscher, 2001). Severe weather-related responses in cattle can significantly increase animal mortality, which could result in production losses (Belasco *et al.*, 2015; Mader, 2003). Cattle experiencing extremely cold weather conditions, especially when combined with increasing wind speed, snow or precipitation, (Hahn, 1985) increase their daily feed intake to compensate for heat loss (Belasco *et al.*, 2015; Nisa *et al.*, 1999). These animals experience higher energy requirements for maintaining body reserves and body temperature (Vining, 1990). The opposite occurs in cattle experiencing severely warm weather conditions. Increasing temperatures, coupled with avoidance behaviours (bunching behaviour, for example) can induce or promote heat stress (Campbell *et al.*, 1993; Taylor *et al.*, 2012). As a result, cattle under these pressures start expending extra energy to reduce the heat load in order to maintain a normal body temperature (Beede *et al.*, 1983; Campbell *et al.*, 1993). In an attempt to dissipate the heat load, cattle will exhibit heat loss mechanisms such as panting, breathing and sweating (Beede *et al.*, 1983; Brown-Brandl *et al.*, 2006; Campbell *et al.*, 1993; Mader *et al.*, 2006). However, if the heat stress period continues, the animals will reduce their daily feed intake (Hahn, 1985; Hahn, 1999; Nisa *et al.*, 1999; Wieman *et al.*, 1992) to balance the amount of heat dissipated with the metabolic heat produced during digestion (Campbell *et al.*, 1993). In addition to the responses experienced by cattle under heat stress, cattle mortality rates increase (Hahn, 1985). Cattle under continuous heat stress periods will require more time to reach market weight, resulting in higher operating costs (Wieman *et al.*, 1992).

The most important effect of stable fly feeding on cattle is reduced production (Taylor *et al.*, 2012). Taylor *et al.* (2012) analyzed published studies and developed a statistical model for describing the effect of stable fly feeding on cattle performance. In the United States, production losses were estimated to be 139 kg milk by dairy cows, 6 kg body weight for pre-weanling calves, 26 kg body weight for pastured stockers, and 9 kg bodyweight for feeder cattle. Using 2005 to 2009 averages prices, the national losses of each industry sector were estimated to be \$226 million for cattle on feed, \$360 million for dairy cattle, \$358 million for cow-calf herds and \$1.268 million for pastured cattle (Taylor *et al.*, 2012). The total impact of stable fly feeding on cattle production in the United States was estimated to be \$2.211 million per year (Taylor *et al.*, 2012). Presently, several estimates on the economic impact of stable flies on cattle have been published (Campbell *et al.*, 1993; Catangui *et al.*, 1992; Catangui *et al.*, 1993, 1995, 1997; Grisi *et al.*, 2014; Taylor *et al.*, 2012; Wieman *et al.*, 1992). However, the economic impact of stable flies in South Africa has never been researched. Thus, studies to quantify the effect of stable fly feeding on cattle performance in South Africa are necessary.

Campbell *et al.* (1987) recorded significant weight gain reductions and decreased feed efficiency in feedlot cattle exposed to an average of 2.58, 5.21 and 7.07 stable flies per leg. Considering, weight gain, feed efficiency, and cost of control with insecticides, the economic threshold for stable flies is less than two stable flies per foreleg per heifer (Campbell *et al.*, 1987). However, according to Catangui *et al.* (1997), stable fly numbers should not exceed 7 stable flies per foreleg per animal per minute, to avoid economic losses. Campbell *et al.* (2001) studied the impact of stable fly feeding on grazing cattle and reported that an average of 2.79 stable flies per leg caused an average weight gain reduction of 0.2 kg/day/steer. An average of 0.85 stable flies/front leg were reported on treated calves, while an average of 3.64 stable flies/front leg were observed on control calves (Campbell *et al.*, 2001). However, these values cannot be extrapolated directly to this study since climatic factors such as humidity and temperature (Cruz-Vázquez *et al.*, 2004) could influence stable fly population densities which could in turn influence cattle production. In addition to stable fly populations, cattle production could be influenced by a wide range of other factors including climatic factors and other flies such as house flies, horn flies and horse flies (see Introduction - Chapter 4).

The objective formulated in Chapter 1:

To determine the impact of stable flies (Diptera: Muscidae) on the feed consumption and weight gain performance of feedlot cattle near Heidelberg, Gauteng, South Africa is the aim in Chapter 4.

The specific objectives were:

- To determine the impact of various levels of stable fly pressures on the average daily dry matter intakes of confined steers.
- To evaluate the relationship between the average daily dry matter intakes of confined steers, various levels of stable fly pressures, and climatic conditions.
- To determine the impact of various levels of stable fly pressure on the average daily gains of finishing steers.

4.2. MATERIAL AND METHODS

4.2.1. The overall experimental design and feedlot pen layout during a 113-day finishing trial

Refer to Chapter 2 for a comprehensive description of Karan Beef feedlot. The outline of the 113-day finishing trial, conducted from 3 December 2013 to 8 April 2014 at Karan beef feedlot is summarized in Fig. 4.1.

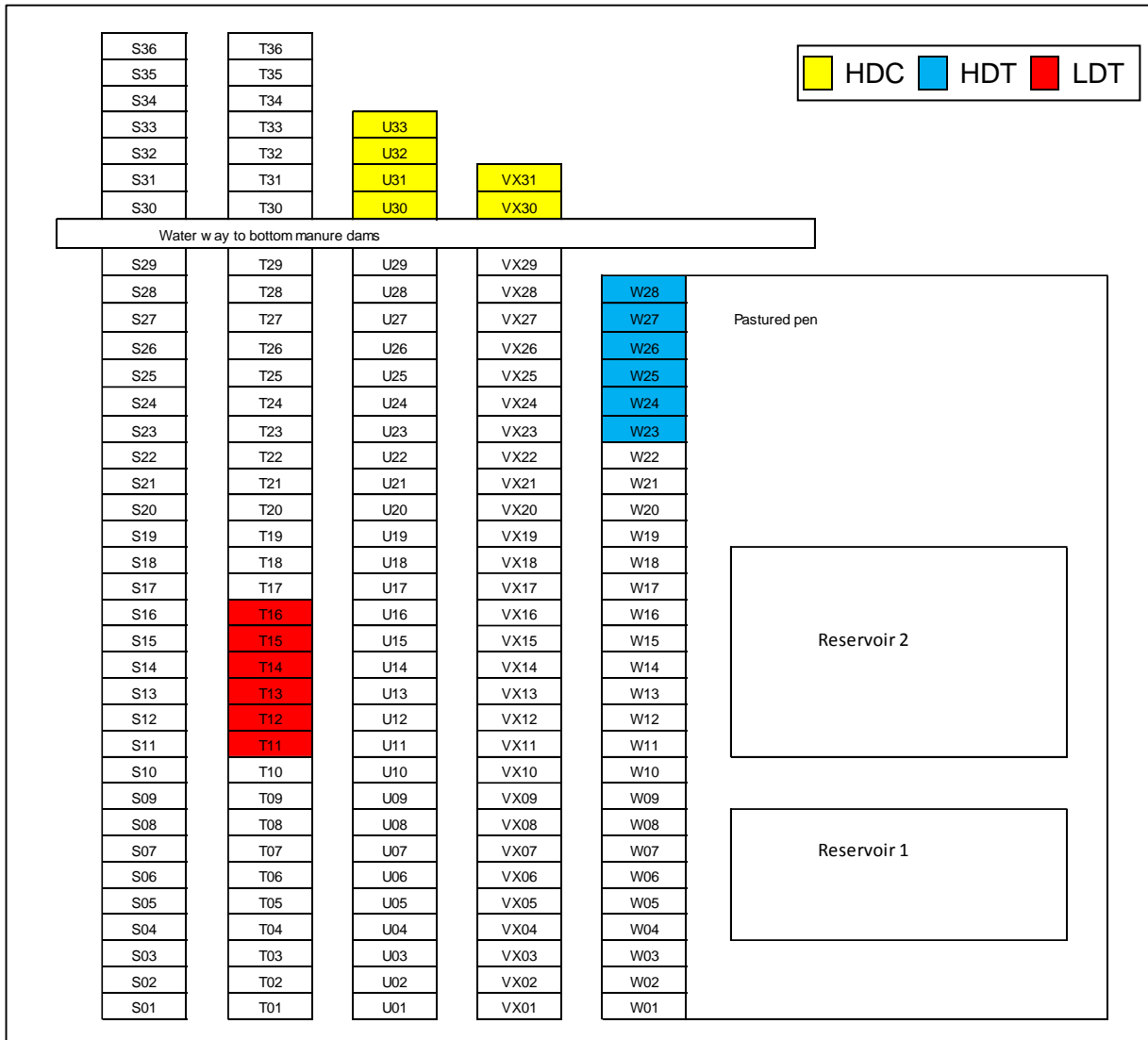


Figure 4.1: The overall feedlot pen layout showing the 113-day finishing trial, conducted from 3 December 2013 to 8 April 2014 at Karan Beef feedlot. Treatments: HDC = high fly density control (untreated); HDT = high fly density treated; LDT = low fly density treated.

The 113-day finishing trial consisted of three treatments as outlined in Fig. 4.1 and Table 4.1. Treatments were located at different locations within the feedlot, depending on stable fly density and absence or presence of stable fly control. The high fly density control (HDC)

treatment consisted of six pens (U30, U31, U32, U33, VX30 and VX31), historically experiencing high stable fly density with no chemical control (Evert, 2014). These pens were located at the upper corner of the feedlot, neighbouring an area covered by weeds and a variety of other vegetation offering stable fly refuge after feeding. Stable fly densities in this treatment were expected to be very high since no stable fly control measures were applied during the course of the trial. This treatment was used as the control treatment.

The high fly density treated (HDT) treatment consisted of six pens (W23, W24, W25, W26, W27 and W28), historically experiencing relatively high stable fly density (Evert, 2014) and routinely controlled with insecticides. These pens were located at the upper side of the feedlot neighbouring the pastured pen and other vegetation. This treatment was selected due to its history of a high level of irritation observed during the previous years (Evert, 2014). Stable fly densities in this treatment were expected to be lower than the HDC treatment due to application of stable fly control measures at a site with a known history of high levels of stable fly irritation. Stable fly populations were controlled by regularly applying Dimilin, an insect growth regulator (diflubenzuren), and pyrethrin contact sprays through mist blowers. (see Chapter 2).

The low fly density treated (LDT) treatment consisted of six pens (T11, T12, T13, T14, T15 and T16), experiencing low stable fly density and routine chemical control applications, similar to the HDT treatment. These pens were located in the middle of the feedlot surrounded by other pens (Fig. 4.1). The pens were connected side-by-side. The pens had concrete floors and were equipped with feed bunks and automatic waterers. The pens were cleaned on a regular basis to prevent manure buildup. These pens were selected due to their history of low stable fly counts and low irritation levels observed during previous studies (Evert, 2014).

4.2.2. Stable fly density on cattle

The stable fly population feeding on steers in the HDC treatment and HDT treatment were regularly monitored through foreleg counts (see Section 2.3). Stable fly density data from the LDT treatment was extrapolated from a similar preliminary study by Evert (2014) at Karan Beef feedlot. The average numbers of stable flies counted weekly per minute on cattle front legs were calculated for each treatment. These counts were expressed as a mean number of stable flies per minute per front leg per animal (see Section 3.2.3).

4.2.3. Allocation of calves into feedlot experimental pens

A total number of 2 204 multiple breed crossbred cattle from varying locations was used in the trial. These breeds included Nguni, Hereford, Limousine, Charolais, Drakensberger, Bonsmara and Brahman cattle. Upon arrival, the calves were randomly selected and assigned to the three treatments to ensure that each treatment received calves of different backgrounds and breeds. Two initial weight groups were stratified in each treatment; initial light weight calves ranging between 150-210 kg and initial heavy weight calves ranging between 211-250 kg. The two weight groups within each treatment (six pens per treatment) were contained in separate pens (three replicate pens per weight group). Some pens were somewhat smaller than the others, but each pen received the same number of calves per surface area (Table 4.1).

Table 4.1: Experimental layout showing the 113-day finishing trial and three treatments conducted from 3 December 2013 to 8 April 2014 at Karan Beef feedlot. Each pen received the same number of calves per surface area. Each treatment was divided into two initial weight groups, and randomly replicated three times into three pens.

Treatment	Light weight steers (150-210 kg)			Heavy weight steers (211-250 kg)		
HDC	VX30	U32	U30	VX31	U33	U31
<i>n</i>	96	113	113	96	113	113
HDT	W27	W25	W23	W28	W26	W24
<i>n</i>	130	130	130	130	130	130
LDT	T15	T13	T11	T16	T14	T12
<i>n</i>	130	130	130	130	130	130

Treatments: HDC = high fly density control (untreated); HDT = high fly density treated; LDT = low fly density treated. For each treatment: *n* = total number of calves per pen.

Apart from sick or injured animals which were permanently removed from the pens, cattle once allocated to pens stayed intact as a group for the duration of the study. Cattle were fed a daily standardized amount of a predetermined diet from a scale-equipped feed truck for the duration of the study (Fig. 4.2). The animals' daily feeding requirements within each treatment were monitored and evaluated.



Figure 4.2: Scale-equipped feed truck delivering a standardized amount of predetermined diet.

4.2.4. Average daily gain (ADG; kg/steer/day)

Definitions of key terminology:

Average daily gain cold (ADGC; kg/steer/day) weight: Average amount of cold weight gain per steer per day. Average daily gain cold (ADGC) weight was calculated according to Equation 4.7.

Average daily gain live (ADGL; kg/steer/day) weight: Average amount of live weight gain per steer per day. Average daily gain live (ADGL) weight was calculated according to Equation 4.2.

Dead weight (DW): Cattle weight after slaughtering and bleeding. The weight received from Karan Beef feedlot.

Dressing percentage: Dressing percentage is an estimated 57%. This value is based on past weight experience from Karan Beef feedlot.

Final cold carcass weight (CCW_f): Calculated using WCW_f and an estimated 1.80% shrinkage per total body weight. The shrinkage value used in this study was based on past weight experience from Karan Beef feedlot. Final cold carcass weight (CCW_f) was calculated according to Equation 4.6.

Final live weight (W_f): Cattle were weighed after slaughtering, after weight was lost from bleeding. Therefore the final live weight was calculated using DW and an estimated 1.48%

blood volume loss per total body weight. The final live weight (W_f) was calculated according to Equation 4.1.

Final warm carcass weight (WCW_f): Calculated using W_f and an estimated dressing percentage. The final warm carcass weight (WCW_f) was calculated according to Equation 4.5.

Initial cold carcass weight (CCW_i): Calculated using WCW_i and an estimated 1.80% shrinkage per total body weight. The shrinkage value used in this study was based on past weight experience from Karan Beef feedlot. Initial cold carcass weight (CCW_i) was calculated according to Equation 4.4.

Initial live weight (W_i): Initial weight of steers at the beginning of the 113-day finishing trial.

Initial warm carcass weight (WCW_i): Calculated using W_i and an estimated dressing percentage. The initial warm carcass weight (WCW_i) was calculated according to Equation 4.3.

The final live weight (W_f) was calculated according

Equation 4.1:

$$W_f = DW + \text{Blood percentage lost}$$

Where:

W_f = final live weight

DW = dead weight

Blood percentage lost = estimated 1.48% blood volume lost per total body weight.

The average daily gain live (ADGL) weight was calculated according

Equation 4.2:

$$ADGL = \frac{(W_f - W_i)}{d}$$

Where:

ADGL = average daily gain live weight

W_f = final live weight

W_i = initial live weight

d = number of days on feed (113-day finishing trial)

The initial warm carcass weight (WCW_i) was calculated according

Equation 4.3:

$$WCW_i = W_i \times \text{Dressing percentage}$$

Where:

WCW_i = initial warm carcass weight

W_i = initial live weight

Dressing percentage = estimated 57%

The initial cold carcass weight (CCW_i) was calculated according

Equation 4.4:

$$CCW_i = WCW_i - \text{Shrinkage percentage}$$

Where:

CCW_i = initial cold carcass weight

WCW_i = initial warm carcass weight

Shrinkage percentage = estimated 1.80%.

The final warm carcass weight (WCW_f) was calculated according

Equation 4.5:

$$WCW_f = W_f \times \text{Dressing percentage}$$

Where:

WCW_f = final warm carcass weight

W_f = final live weight

Dressing percentage = estimated 57%

The final cold carcass weight (CCW_f) was calculated according

Equation 4.6:

$$CCW_f = WCW_f - \text{Shrinkage percentage}$$

Where:

CCW_f = final cold carcass weight

WCW_f = final warm carcass weight

Shrinkage percentage = estimated 1.80%.

The average daily gain cold (ADGC) weight was calculated according

Equation 4.7:

$$ADGC = \frac{(CCW_f - CCW_i)}{d}$$

Where:

ADGC = average daily gain cold weight

CCW_f = final cold carcass weight

CCW_i = initial cold carcass weight

d = number of days on feed (113-day finishing trial)

Data available for each group of steers per pen per treatment included initial live weight and dead weight. Information concerning background or history of steers was limited. The two steps for calculating the ADGL weight are the following: (1) Calculate final live weight according to Equation 4.1. (2) Calculate the ADGL weight according to Equation 4.2.

The five steps for calculating the ADGC weight are the following: (1) Calculate initial warm carcass weight according to Equation 4.3. (2) Calculate initial cold carcass weight according to Equation 4.4. (3) Calculate final warm carcass weight according to Equation 4.5. (4) Calculate final cold carcass weight according to Equation 4.6. (5) Calculate ADGC weight according to Equation 4.7. Before data analysis, data was checked for false values and outliers, which were removed.

4.3. RESULTS AND DISCUSSION

4.3.1. Impacts of various levels of stable fly pressures on average DMI of confined feedlot steers

4.3.1.1. Various levels of stable fly pressures at the HDC, HDT and LDT treatments

Mean number of stable flies counted on cattle front legs at the HDC, HDT and LDT treatment is summarised in Table 4.2. Estimated fly counts from the LDT treatment was extrapolated from a study by Evert (2014), at Karan Beef feedlot.

Table 4.2: Mean number of stable flies (*Stomoxys calcitrans*) weekly counted per minute on cattle front legs at the HDC and HDT treatment, from 8 January 2014 to 12 February 2014, and from 26 February 2014 to 9 April 2014 as well as estimated fly counts based on 2012-2013 survey (Evert, 2014).

	Treatments		
	HDC	HDT	LDT*
<i>n</i>	259	432	574
\bar{x}	3.44	2.78	0.37

Treatments: HDC = high fly density control treatment (untreated), HDT = high fly density treated treatment, LDT = low stable fly density treated treatment. For each treatment: *n* = total number of steers; \bar{x} = mean number of stable flies weekly counted per minute on cattle front legs [(total stable flies weekly counted per minute on cattle front legs)/(total number of steers)].

* Estimated fly counts based on 2012-2013 survey at Karan Beef feedlot (Evert, 2014).

Mean stable fly leg counts were higher in the HDC treatment than the HDT treatment. This is no surprise; higher stable fly population densities were expected in the HDC treatment because no stable fly control measures were applied at this treatment during the course of the trial. Stable fly leg counts at the HDT treatment were expected to be lower compared to the HDC treatment, because of stable fly control. Low stable fly densities (compared to the HDC and HDT treatment) were recorded at the LDT treatment in previous years (Evert, 2014) and were added as treatment with expected estimate of low fly density as well as stable fly control.

Overseas studies on economic injury levels (stable flies per foreleg per minute) for stable flies determined an average of 7 stable flies per foreleg per animal per minute as the economic injury level for stable flies (Catangui *et al.*, 1997). Meaning the feedlot industry could suffer economic losses when stable fly numbers are allowed to reach or exceed this number (Catangui *et al.*, 1997).

4.3.1.2. The impact of various levels of stable fly pressures on average DMI of heavy weight steers from the HDC, HDT and LDT treatments

The impact of various levels of stable fly pressures on the average daily DMI of heavy weight steers is shown in Fig. 4.3.

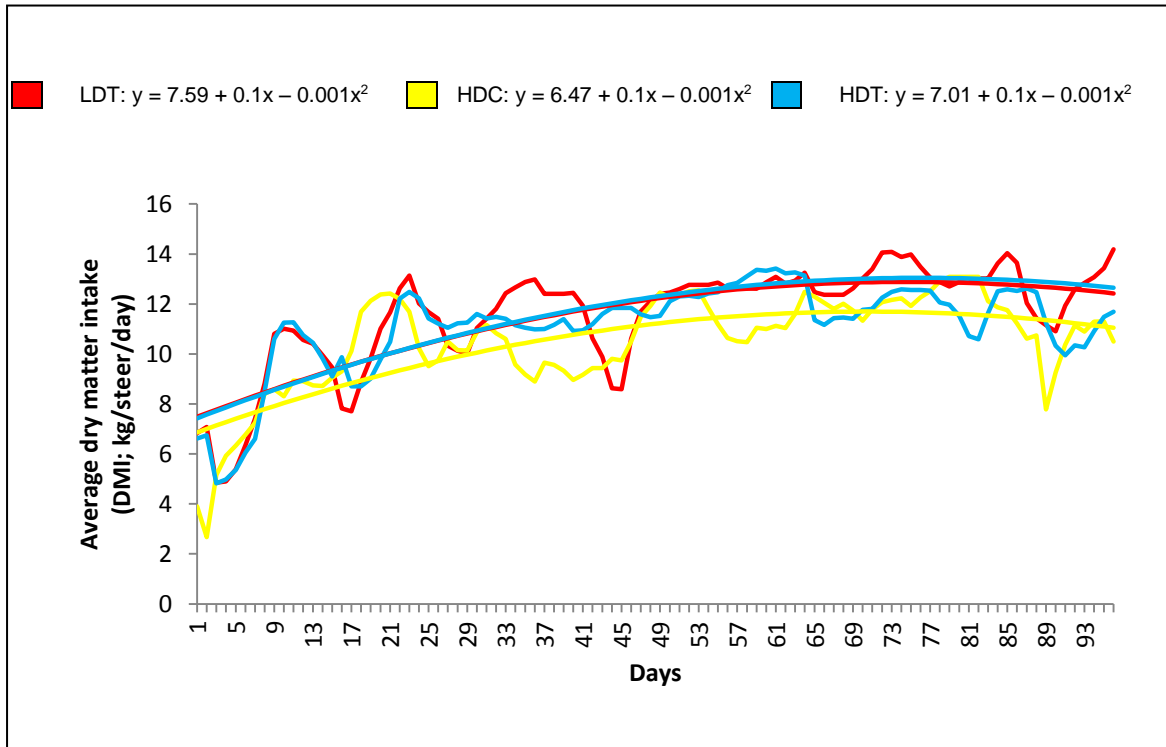


Figure 4.3: The average daily dry matter intake (DMI; kg/steer/day) of steers with mean initial weights ranging between 211 and 250 kg showing the impact of different stable fly population treatments. Fitted quadratic regression lines are shown. Treatments: LDT = low fly density treated; HDC = high fly density control (untreated); HDT = high fly density treated.

Quadratic regression lines were fitted statistically significantly to the average daily DMI data of heavy weight steers using SPSS. The overall average daily DMI gradually increased over time, then flattened steadily before decreasing slowly for all three treatments. Multiple regression analyses between the average daily DMI of heavy weight steers from both the HDC and LDT treatment presented a significant regression equation ($f(3, 222) = 120.72$, $p < 0.0001$) with an R^2 of 0.62. The model predicted that the average daily DMI per treatment was equal to $7.59 + 0.1(\text{time}) - 0.001(\text{time})^2 - 1.12(\text{treatment})$, where treatments were coded as 0 = LDT and 1 = HDC. A multiple regression analysis showed that the intercept of the HDC treatment was statistically significant from the LDT treatment ($p < 0.0003$; Bonferroni-correction p-value). Multiple regression analyses with Bonferroni-correction

showed that the average daily DMI from the HDC treatment was statistically significantly lower than the LDT treatment ($p < 0.0003$; Bonferroni-correction p -value).

Multiple regression analyses between the average daily DMI of heavy weight steers from both the HDC and HDT treatment presented a statistical significant regression equation ($f(3, 222) = 103.09, p < 0.0001$) with an R^2 of 0.58. The model predicted that the average daily DMI per treatment was equal to $6.72 + 0.1(\text{time}) - 0.001(\text{time})^2 + 0.55(\text{treatment})$, where treatments were coded as 0 = HDC and 1 = HDT. A multiple regression analysis showed that the intercept of the HDC treatment was statistically significant from the HDT treatment ($p < 0.0003$; Bonferroni-correction p -value). Multiple regression analysis with Bonferroni-correction showed that the HDT treatment was statistically significant higher than the HDC treatment ($p < 0.003$; Bonferroni-correction p -value).

Multiple regression analysis between the average daily DMI of heavy weight steers from both the LDT and HDT treatment presented a statistical significant regression equation ($f(3, 222) = 129.52, p < 0.0001$) with an R^2 of 0.64. The model predicted that the average daily DMI per treatment was equal to $7.02 + 0.1(\text{time}) - 0.001(\text{time})^2 + 0.57(\text{treatment})$, where treatments were coded as 0 = HDT and 1 = LDT. A multiple regression analysis showed that the intercept of the LDT treatment was statistically significantly different from the HDT treatment ($p < 0.0003$; Bonferroni-correction p -value). Multiple regression analysis with Bonferroni-correction showed that the LDT treatment was statistically significant higher than the HDT treatment ($p < 0.003$; Bonferroni-correction p -value).

4.3.1.3. The impact of various levels of stable fly pressures on average DMI of light weight steers at the HDC, HDT and LDT treatments

The impact of various levels of stable fly pressures on the average daily DMI of light weight steers is shown in Fig. 4.4.

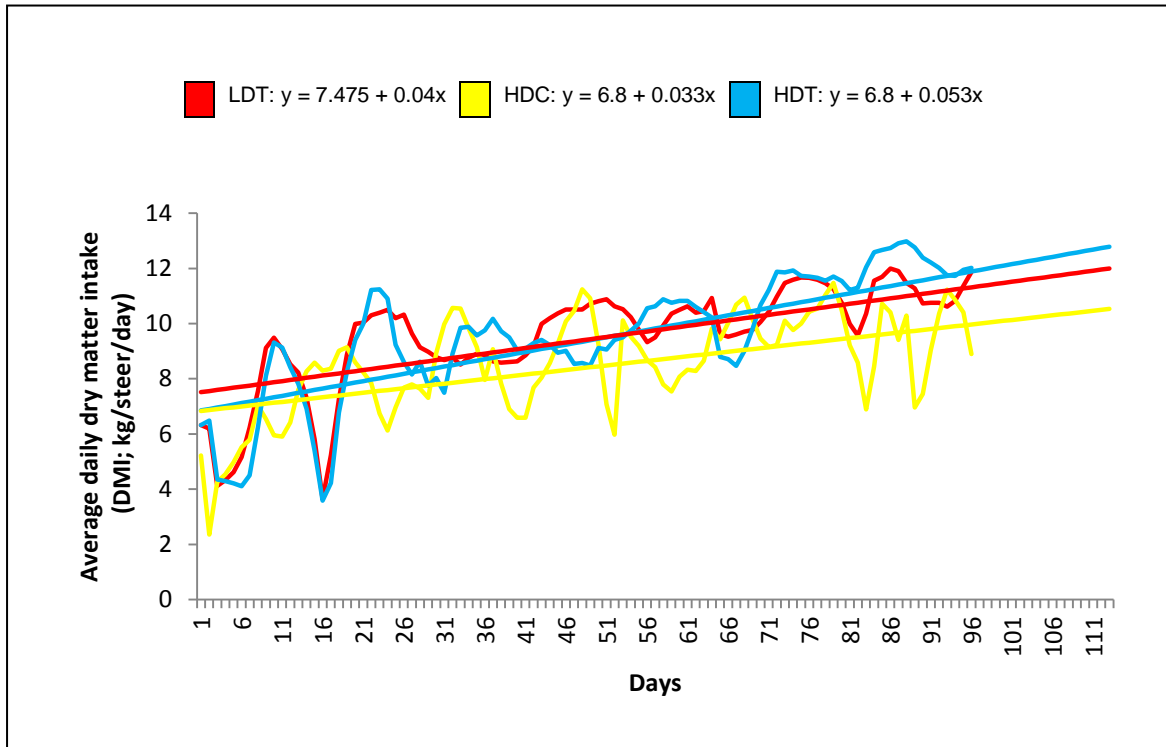


Figure 4.4: Linear regression lines showing the impacts of various levels of stable fly pressures on the average dry matter intake (DMI; kg/steer/day) of steers with mean initial weights ranging between 150 and 210 kg. DMI = dry matter intake. Treatments: LDT = low fly density treated; HDC = high fly density control (untreated); HDT = high fly density treated.

Linear regression lines were fitted statistical significantly to the average daily DMI data of light weight steers using SPSS. The overall average daily DMI of the light weight steers gradually increased over time. Examination of the relationship between the average DMI of light weight steers from the HDC and LDT treatment presented a statistical significant linear regression equation ($f(3, 248) = 84.71, p < 0.0001$) with an R^2 of 0.51. The model predicted that the average daily DMI per treatment was equal to $7.703 + 0.034(\text{time}) - 0.799(\text{treatment})$, where treatments were coded as 0 = HDC and 1 = LDT. A multiple regression analysis showed that the intercept of the HDC treatment was statistically significantly different from the LDT treatment ($p < 0.042$; Bonferroni-correction p-value). Multiple regression analysis with Bonferroni-correction showed that the average daily DMI of the

HDC treatment was statistically significantly lower than the LDT treatment ($p < 0.0003$; Bonferroni-correction p -value). The LDT treatment regression slope was parallel but higher than that of the HDC treatment, showing a higher rate of DMI for the LDT treatment.

Multiple regression analysis between the average daily DMI of light weight steers from the HDC and HDT treatment presented a statistically significant linear regression equation ($f(3, 248) = 125.42, p < 0.0001$) with an R^2 of 0.60. The model predicted that the average daily DMI per treatment was equal to $6.904 + 0.031(\text{time}) - 0.019(\text{treatment})$, where treatments were coded as 0 = HDC and 1 = HDT. A multiple regression analysis showed that the intercept of the HDC treatment was not statistically significantly different from the HDT treatment ($p < 2.61$; Bonferroni-correction p -value). Multiple regression analysis with Bonferroni-correction also showed that the average daily DMI of the HDC treatment was statistically significant lower than the HDT treatment ($p < 0.0003$; Bonferroni-correction p -value).

Multiple regression analysis between the average daily DMI of light weight steers from the LDT and HDT treatments presented a statistically significant linear regression equation ($f(3, 248) = 126.49, p < 0.0001$) with an R^2 of 0.60. The model predicted that the average daily DMI per treatment was equal to $6.96 + 0.04(\text{time}) - 0.74(\text{treatment})$, where treatments were coded as 0 = HDT and 1 = LDT. A multiple regression analysis showed that the intercept of the LDT treatment was not statistically significant from the HDT treatment ($p < 0.06$; Bonferroni-correction p -value). Multiple regression analysis with Bonferroni-correction also showed that the average daily DMI of the LDT treatment was statistically significant lower than the HDT treatment ($p < 0.0003$; Bonferroni-correction p -value).

Variation in average daily DMI between treatments might be explained by environmental factors and/or different levels of stable fly pressures between treatments. The overall stable fly activity was higher at the HDC treatment than at the HDT treatment (see Table 4.2). Another possible factor explaining differences in average daily DMI between the HDC and HDT treatments could have been the type of feed management practice. The type of feed management practice from the HDC treatment might have been somewhat different from the rest of the treatments, since different feed bunk managers had been responsible for the HDC and HDT treatment. Slightly different management practices between managers might have been a possible confounder, explaining unexpected average daily DMI differences observed among all three treatments.

DeHaan *et al.* (1995) reported a DMI pattern consisting of three distinct segments: an adaptation, a plateau, and a declining phase. The DMI pattern observed by DeHaan *et al.*

(1995) is similar to the DMI pattern for the heavy weight steers from the three treatments, although the light weight steers still show a linear increase in the later phase without declining. It appears as if the average daily DMI in heavy weight steers gradually increased in a linear fashion for the first few days (adaptation phase), then reached a plateau (plateau phase), before steadily declining as steers reach slaughtering weights (declining phase). The large fluctuations in the average daily DMI of both the light and heavy weight steers among all three of the treatments during the first few days of the trial is probably associated with the adaptation phase. It is believed that steers were adapting to the new environment, pen mates and high-concentrated finishing diet (DeHaan *et al.*, 1995). It appears as if the overall trend of the average daily DMI of light weight steers from the three treatments increased for a longer period of time compared to the overall average daily DMI trend of the heavy weight steers from all three treatments. DeHaan *et al.* (1995) also noted that the DMI curve for light weight steers increased for a longer period of time and that the DMI patterns for the calves and yearling cattle differ throughout a feeding period. This might be due to physiological age or initial weight (DeHaan *et al.*, 1995). Additionally, irregular fluctuation within the average daily DMI of the light and heavy weight steers from the HDT, HDC and LDT treatments might also have been associated with seasonal climatological patterns.

4.3.2. The possible impacts of various climatic conditions on average daily DMI of steers

The possible impacts of daily rainfall (mm) and mean daily temperature (°C) on the average daily DMI of heavy and light weight steers from the HDC, HDT and LDT treatments is demonstrated in Fig. 4.5 and Fig. 4.6, respectively.

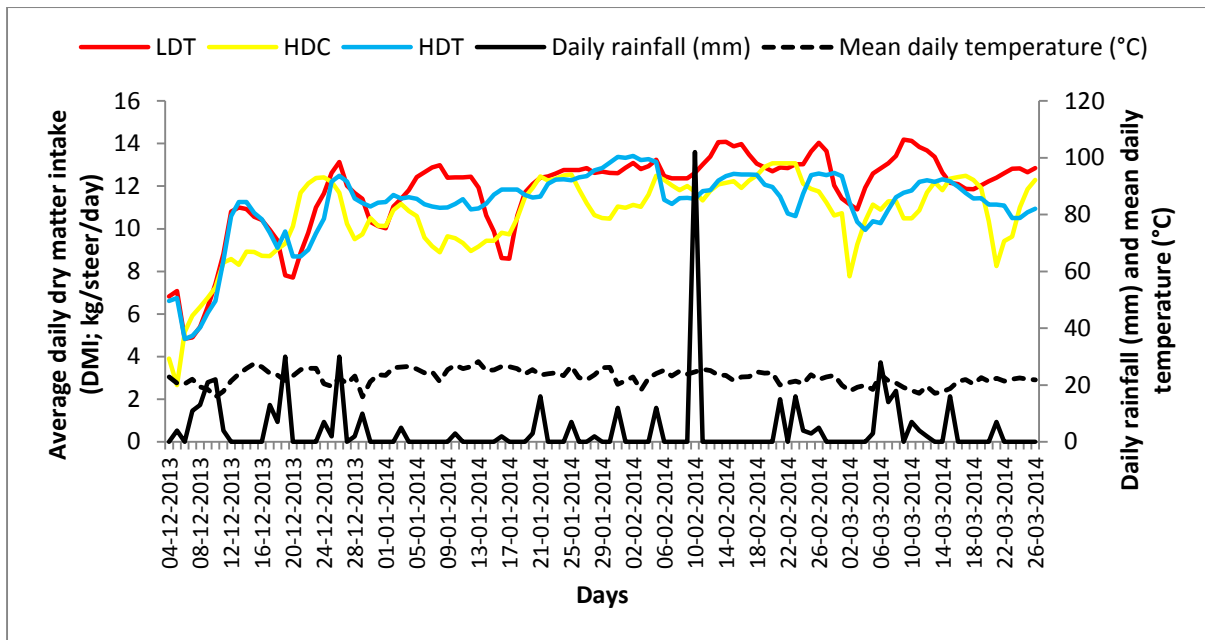


Figure 4.5: The possible influence of daily rainfall (mm) and mean daily temperature (°C) on the average daily dry matter intake (DMI; kg/steer/day) of heavy weight (211-250 kg) steers from the HDC, HDT and LDT treatments. DMI = dry matter intake, Treatments: HDC = high fly density control (untreated); HDT = high fly density treated; LDT = low fly density treated.

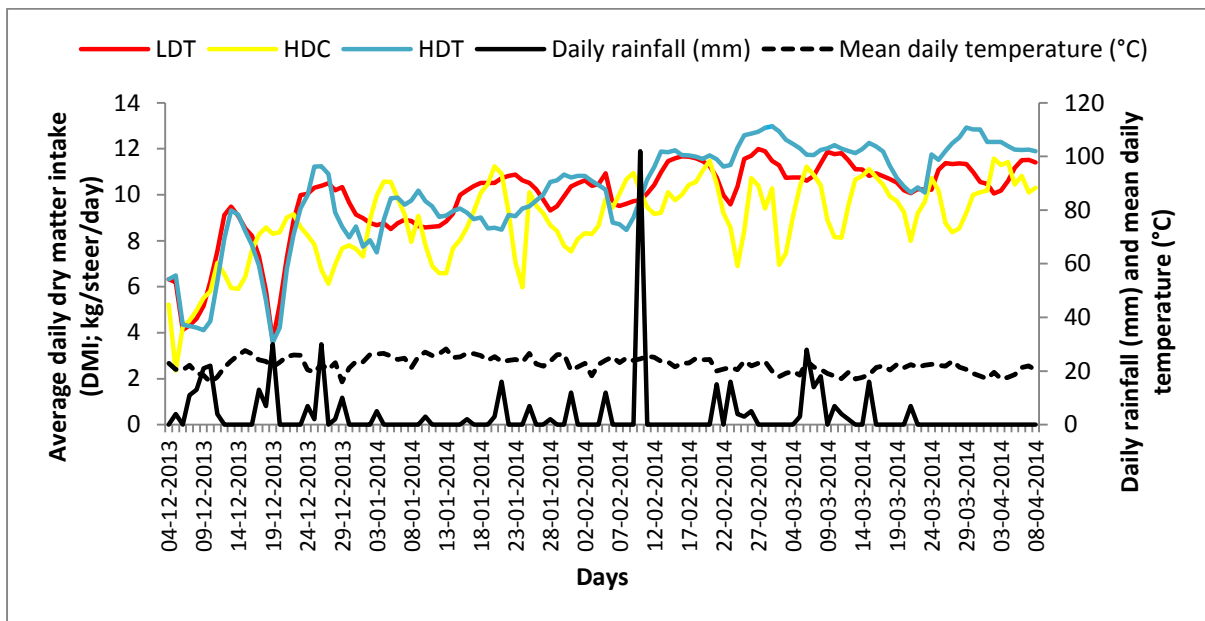


Figure 4.6: The possible influence of daily rainfall (mm) and mean daily temperature (°C) on the average daily dry matter intake (DMI; kg/steer/day) of light weight (150-210 kg) steers from the HDC, HDT and LDT treatments. DMI = dry matter intake, Treatments: HDC = high fly density control (untreated); HDT = high fly density treated; LDT = low fly density treated.

The lowest mean daily temperature (15.86°C) recorded at Karan Beef was on 10 December 2013 and the highest mean daily temperature (28.23°C) on 13 January 2014. The highest

rainfall received was 102 mm. It appears as if the average daily DMI of heavy and light weight steers from the three treatments might have been affected by rainfall and/or temperature. Daily rainfall periods seem to result in short-term decreases in average daily DMI in both heavy weight and light weight groups (Fig. 4.5 and Fig. 4.6). However, rainfall peaks at the beginning of the trial seemed to correspond with increases in average daily DMI. It seemed as if steers were able to cope or adapt to gradual changes in the environment, as explained by increase and decrease rates of average daily DMI. The interactions between various levels of stable fly pressures, weather conditions and management factors (e.g. breed selection, feed adaption (Young, 1993), and environmental modifications (Mitlöhner *et al.*, 2001; Young, 1993)) are highly interrelated and not clearly discernible.

Further research on the responses of steers to stable flies and weather conditions is needed to understand the relationship between the average daily DMI, various levels of stable fly pressures and weather conditions.

4.3.3. The impact of various levels of stable fly pressures on ADGL and ADGC weight of steers

4.3.3.1. Process of identifying outliers in the ADGL weights of heavy and light weight steers

The impact of various levels of stable fly pressures, as provided in the three treatments - on ADGL and ADGC weights of light and heavy weight steers - were investigated from 4 December 2013 to 8 April 2014 (see Section 4.2.4). Multiple outliers in data sets have the risk of increasing error variance and reducing the power variance of statistical tests (Osborne & Overbay, 2004). Outliers non-randomly distributed may decrease normality and violate assumptions of sphericity and multivariate normality, hereby altering the odds of making both Type I and Type II errors. Additionally, outliers may also influence estimates of substitutive interest. Since outliers far outside the norm of variables may exhibit a negative impact on statistical analysis tests, data was screened for outliers before analysis. Data was subjected to a normal probability plot to analyse data for normality, ensure a normal distribution, and identify outliers far from the mass of data. In the normal probability plot, the ADGL weight data of steers was sorted from smallest to largest and fitted to a straight normal line. Subsequently, the distributions of the ADGL weight values relative to the normal straight line were examined.

A normal probability plot of ADGL weights of heavy weight steers is presented in Fig. 4.7.

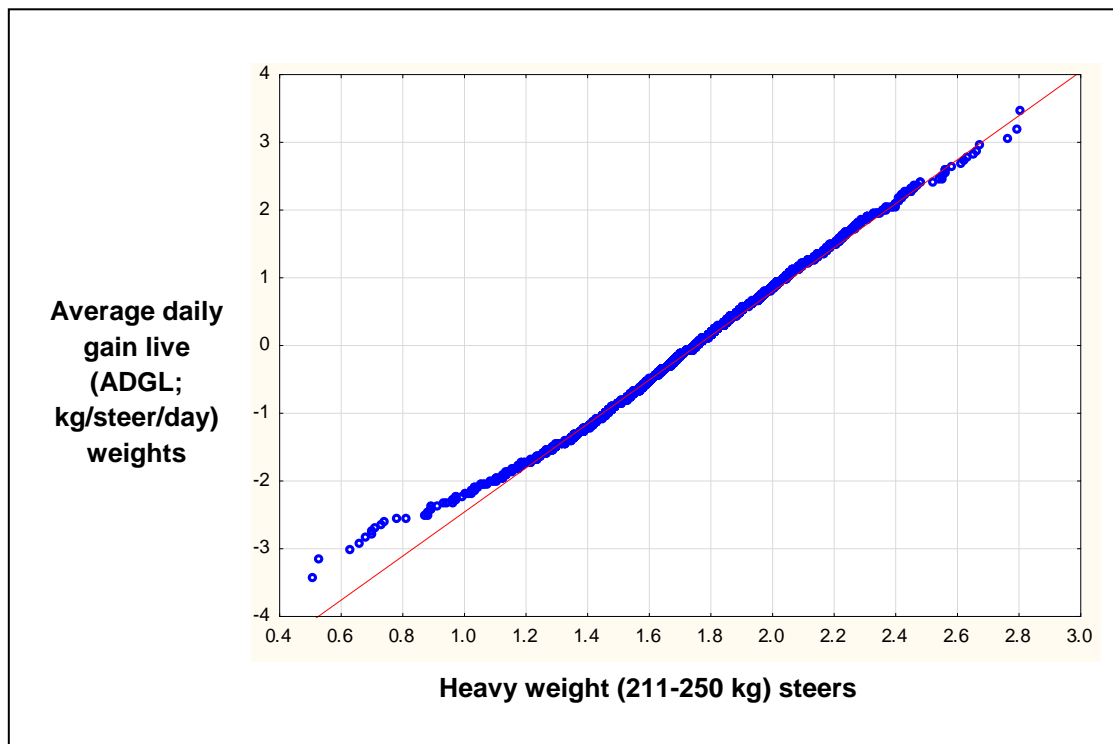


Figure 4.7: Normal probability plot of the average daily gain live (ADGL; kg/steer/day) weights of heavy weight (211-250 kg) steers ($n = 803$ steers).

In the normal probability plot (Fig. 4.7), a relatively linear pattern was found in the centre of the data. However, ADGL weight values of heavy weight steers appeared to deviate at both ends of the normal straight line, thus indicating some non-normality in the data set. Since the normal distribution of the ADGL weight values of heavy weight steers did not provide an adequate fit to the data set, an appropriate distribution was identified using box-and-whisker plot analyses in SPSS (Fig. 4.9).

A normal probability plot for the average daily gain live (ADGL; kg/steer/day) weights of light weight (150-210) is presented in Fig. 4.8.

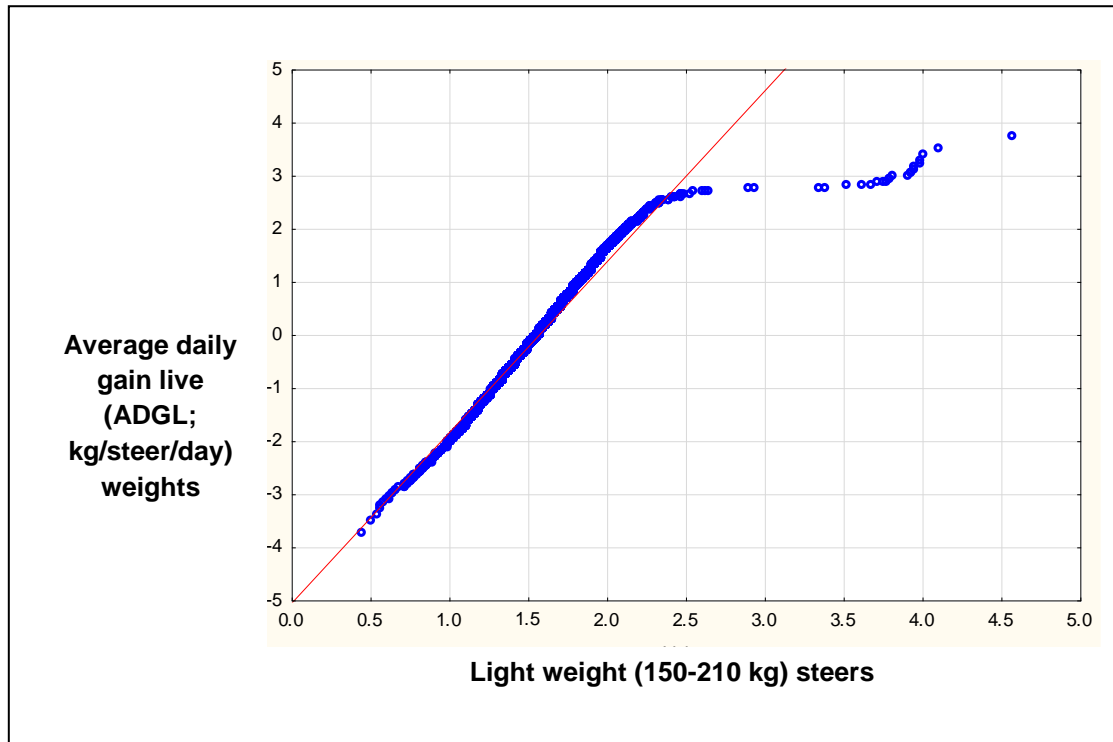


Figure 4.8: Normal probability plot of the average daily gain live (ADGL; kg/steer/day) weights of light weight (150-210 kg) steers ($n = 751$ steers).

In the normal probability plot, the values follow the relatively linear pattern most of the time initially. However, at upper end of the normal straight line, ADGL weight values showed a marked deviation from the normal straight line, indicating non-normality in the data set. Since the normal distribution of the ADGL weight values of light weight steers did not provide an adequate fit to the data set, an appropriate distribution was identified using box-and-whisker plot analysis in SPSS (see Fig. 4.10).

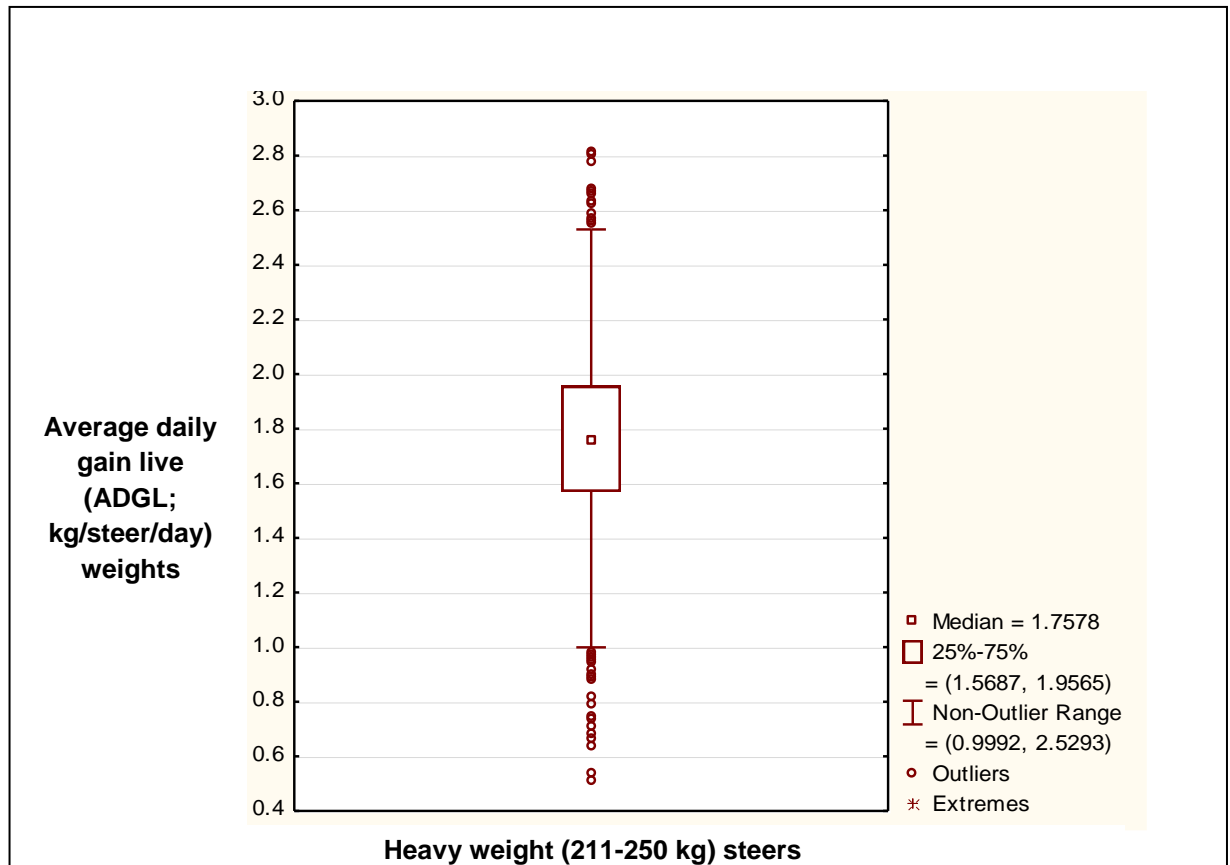


Figure 4.9: Box-and-whisker plot showing the average daily gain live (ADGL; kg/steer/day) weights of heavy weight (211-250 kg) steers ($n = 803$). The figure shows the median ADGL weights of light weight steers (small box within), 25% to 75% quartiles (larger box), non-outlier range (whiskers), outliers (dots) and extremes (stars).

The minimum and maximum ADGL weight gain values of heavy weight steers were 0.51 kg and 2.81 kg, respectively. As calculated using the box-and-whisker plot analysis in SPSS, ADGL weight values of heavy weight steers that were equal or higher than 2.54 were identified as outliers and removed at the upper end of the box-and-whisker plot (Fig. 4.9). These values were also identified as unrealistic from feedlot experience.

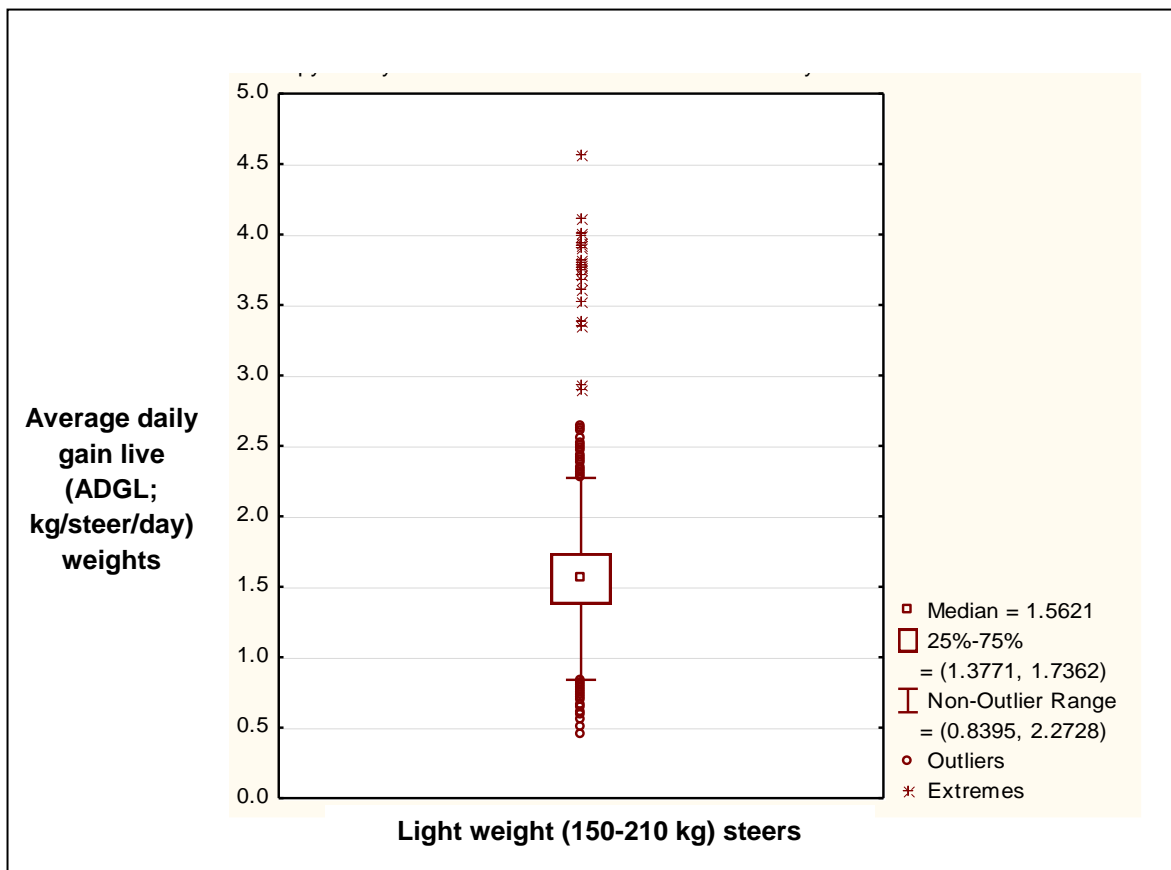


Figure 4.10: Box-and-whisker plot showing the average daily gain live (ADGL; kg/steer/day) weights of light weight (150-120 kg) steers ($n = 751$ steers). The figure shows the median ADGL weights of heavy weight steers (small box within), 25% to 75% quartiles (larger box), non-outlier range (whiskers), outliers (dots) and extremes (stars).

The minimum and maximum ADGL weight gain values of light weight steers were 0.45 kg and 4.56 kg, respectively. As calculated using the box-and-whisker plot analysis in SPSS, ADGL weight values of light weight steers that were equal or higher than 2.27 were identified as outliers and removed at the upper end of the box-and-whisker plot (Fig 4.10). These values were also identified as unrealistic from feedlot experience

Hawkins (1980) described an outlier as an observation that “deviates so much from other observations as to arouse suspicion that it was generated by a different mechanism”. Selected steers were removed because they were not representative of the whole population. Outlier values of the ADGL weight data which were very different to the rest of the population could have been due to a number of reasons, including human error.

4.3.3.2. The impact of various levels of stable fly pressures on ADGL (average daily gain live) and ADGC (averaged daily gain cold) weight of finishing steers

The ADGL and ADGC weight gains were expressed as the average kg/steer/day. The impact of various levels of stable fly pressures on the ADGL weights of light and heavy weight steers is presented in Fig. 4.11 and Table 4.3.

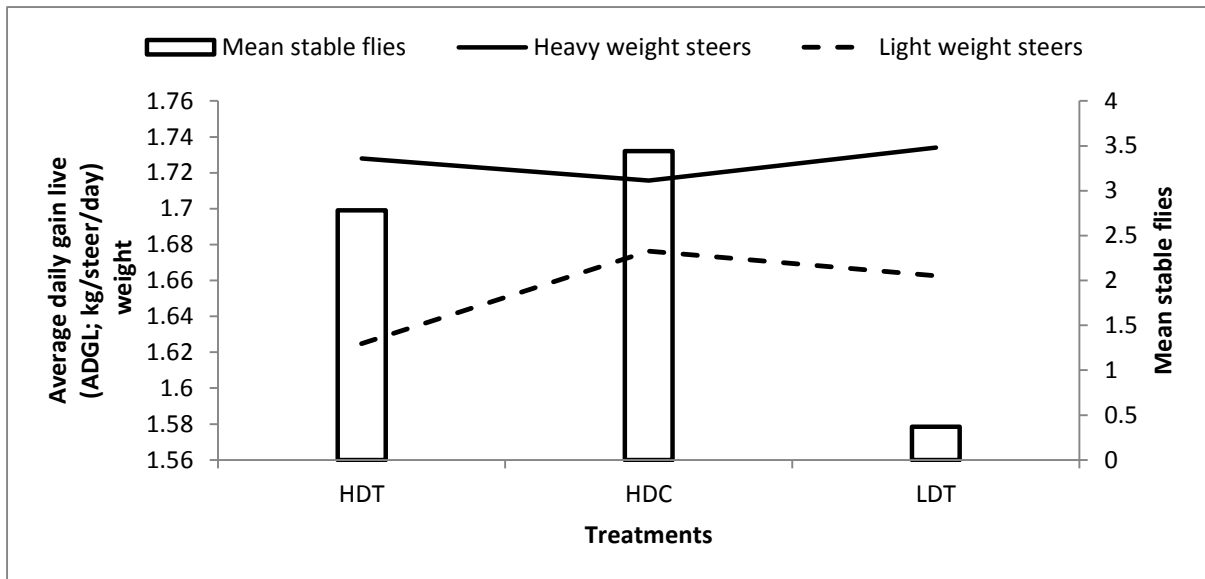


Figure 4.11: The impacts of various levels of stable fly pressures on the average daily gain live (ADGL; kg/steer/day) weight of light weight (150-210 kg) and heavy weight (211-250 kg) steers. ADGL = average daily gain live, Treatments: HDT = high fly density treated; HDC = high fly density control (untreated); LDT = low fly density treated, Mean stable flies = [(total stable flies counted weekly per minute on cattle front legs)/(total number of steers)].

Table 4.3: Mean average daily gain live (ADGL) weight of heavy weight (211-250 kg) and light weight (150-210 kg) steers from 4 December 2013 to 8 April 2014.

Average daily gain live (ADGL; kg/steer/day) weight								
	HDT		HDC		LDT		ANOVA	
	<i>n</i>	\bar{x}	<i>n</i>	\bar{x}	<i>n</i>	\bar{x}	<i>P value</i>	MSE*
Light weight steers (150-210 kg)	261	1.62	207	1.68	283	1.66	0.127	0.17
Heavy weight steers (211-250 kg)	291	1.73	209	1.72	303	1.73	0.803	0.02

ADGL = average daily gain live, Treatments: HDT = high fly density treated, HDC = high fly density control (untreated), LDT = low fly density treated. For each treatment: *n* = total number of steers, \bar{x} = mean number of stable flies counted weekly per minute on cattle front legs.

*Mean square error

Univariate tests of significance showed no statistically significant differences in the mean ADGL weights of light weight and heavy steers from the HDT, HDC and LDT treatments. Additionally, the Cohen's effect size test showed no practical significance between treatments. Non-responsiveness in the ADGL weights from the light and heavy weight steers at any of the three treatments may be an indication of adaptation to stable fly feeding or a degree of tolerance in steers to higher levels of stable fly populations. Catangui *et al.* (1993) suggested that cattle exposed to higher densities of stable flies are becoming conditioned, with lower response to irritation and decreased effect on weight gain.

The impact of various levels of stable fly pressures on ADGC weights of light and heavy weight steers is presented in Fig. 4.12 and Table 4.4.

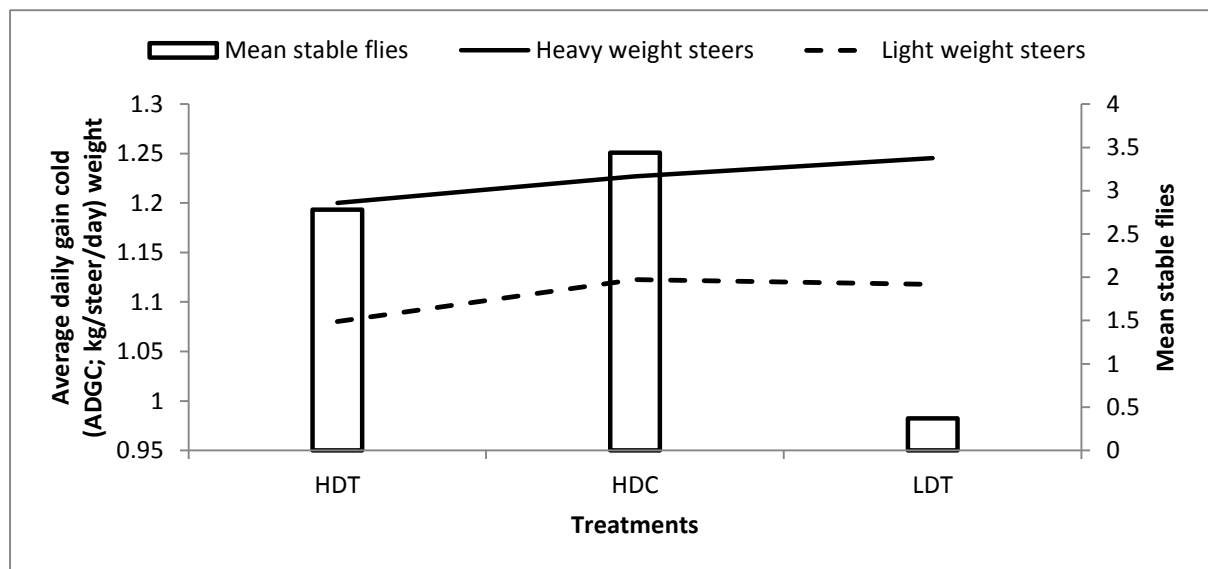


Figure 4.12: The impacts of various levels of stable fly pressures on the average daily gain cold (ADGC) weight of light weight 150-210 kg) and heavy weight (211-250 kg) steers. ADGC = average daily gain cold, Treatments: HDT = high fly density treated; HDC = high fly density control (untreated); LDT = low fly density treated. Mean stable flies = [(total stable flies weekly counted per minute on cattle front legs)/(total number of steers)].

Table 4.4: Mean average daily gain cold (ADGC; kg/steer/day) weight of heavy weight (211-250 kg) and light weight (150-210 kg) steers from 4 December 2013 to 8 April 2014, assessed by analyses of variance (ANOVA).

Average daily gain cold (ADGC; kg/steer/day) weight											
	HDT		HDC		LDT		ANOVA		Cohen's <i>d</i> (effect size) ^{***}		
	<i>n</i>	\bar{x}	<i>n</i>	\bar{x}	<i>n</i>	\bar{x}	<i>P value</i>	MSE ^{**}	HDT/HDC	HDT/LDT	HDC/LDT
Light weight steers (150-210 kg)	261	1.08	207	1.12	283	1.12	0.0137*	0.1346	0.23	0.20	0.03
Heavy weight steers (211-250 kg)	291	1.20	209	1.23	303	1.25	0.0126*	0.1556	0.15	0.25	0.10

ADGC = average daily gain cold, Treatments: HDT = high fly density treated, HDC = high fly density control (untreated), LDT = low fly density treated. For each treatment: *n* = total number of steers, \bar{x} = mean number of stable flies counted weekly per minute on cattle front legs [(total stable flies weekly counted per minute on cattle front legs)/(total number of steers)].

* Statistically significant difference at $p < 0.05$.

** Mean square error

*** Cohen's *d* (effect size) values depict the comparison of the ADGC weight of both the light and heavy weight steers between treatments. Cohen's *d* \approx 0.2, low effect; 0.5, moderate effect; and \geq 0.8, large effect.

Univariate test of significance indicates statistically significant differences in the mean ADGC weights of light weight and heavy weight steers from the HDT, HDC and LDT treatments. The Cohen's effect size value showed a moderate practically significant difference in the mean ADGC weights of the light weight steers, when the HDT treatment is compared with the HDC ($d = 0.23$) and LDT (0.20) treatment. Furthermore, the Cohen's effect size value showed a moderate practically significant difference in the mean ADGC weights of the heavy weight steers, between the HDT and LDT treatment ($d = 0.25$). Further research is needed to determine whether various levels of stable fly pressures, management differences, or other factors such as environmental factors, are associated with differences in the mean ADGC weights of steers.

Since stable flies disrupt feeding of cattle during the day, it is believed that cattle may compensate for feeding interference by feeding at night - and possibly very early in the morning, when stable flies are not active. Similar behaviour was noted in a study by Marlow and Pogacnik (1986), who suggested that late resting levels during darkness in riparian areas are associated with low insect pest activity. This behaviour also appears in cattle compensating for reduced grazing time in hot weather conditions (Kendall *et al.*, 2006). Other factors, such as time of sunrise and sunset, have been reported to influence the pattern of feeding behaviour of cattle (Stricklin, 1986). Further knowledge and research on factors influencing stable fly densities and weight gain performance of feedlot cattle may assist in the development of a stable fly management programme.

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1. Temporal and spatial distribution of stable flies, *Stomoxys calcitrans* (Diptera: Muscidae)

A distinct stable fly seasonal distribution pattern, with five stable fly peaks, was observed during this study, confirming an earlier study (Evert, 2014). High stable fly population numbers were associated with summer months. There was a slow build-up in stable fly numbers after winter. Distinct diurnal stable fly patterns, with bimodal peaks, were observed. Stable fly peak periods in vegetation (sweep-net catches), have been identified alternating with peak periods of stable fly counts on cattle

5.2. Stable fly density on cattle in sprayed and unsprayed pens as sampled with traps and counted on cattle forelegs

There was a positive correlation between the mean number of stable flies counted on cattle front legs per minute and the mean number of stable flies collected in traps. This indicates that the number of stable flies collected in the traps is a fairly good estimate of the degree of stable fly activity on cattle for the period monitored. Therefore, stable fly collection rates from Nzi tsetse type traps can be used as accurate predictors of the degree of stable fly feeding and irritation on cattle.

5.3. Impact of stable flies on the feed intake of feedlot cattle

It was concluded that the various levels of stable fly populations have an adverse effect on the average daily dry matter intake (DMI) of confined heavy and light weight steers. Variations in the average daily dry matter intake (DMI) data of steers from the three treatments are related to various levels of stable fly pressures and feed management practices.

5.4. Impact of stable flies on the weight gain performance of feedlot cattle

The interaction between various levels of stable fly pressures and the average daily gain (ADG) weights of light and heavy weight steers is complex. No statistically significant differences were found in the average daily gain live (ADGL) weights of light and heavy weight steers. Statistically significant differences were observed in the average daily gain cold (ADGC) weights of light and heavy weight steers. However, these differences were of little practical impact on meat production, with little need for routine chemical control. Steers

seem to develop a degree of tolerance for, or adaptation to, stable fly feeding and might be compensating for feeding interferences during the day by feeding at night and possibly very early in the morning when stable flies are not active. These findings are an indication of the advantage of focused integrated stable fly control at feedlots, especially with the application of insect growth regulators, to keep stable fly populations at a low level. Continuous stable fly monitoring is necessary to identify abnormal seasonal increases in fly populations.

It is concluded that there is an edge effect and seasonal impact on *S. calcitrans* populations. Stable flies were more abundant at pens surrounded by vegetation, manure run-off and holding ponds. However, stable fly density is probably below the economic threshold levels at the Karan Beef site. The seasonal build-up of flies after winter is slow and control measures are probably not necessary before January, whereas no control is necessary during a large part of the year, especially from May to December. Chemical application should be applied, based on continuous monitoring of stable fly populations, to prevent the building up of larger populations. These results have important implications for the development of an integrated fly management programme and insecticide use in the feedlot. Chemical control, when necessary, should be aimed at major resting sites, such as weeds and vegetation surrounding feedlot pens. Insecticide applications should be done early in the mornings, from around 07:30 and up to 10:30, and later in the evening from around 15:30 onwards. Insecticides should not be used as the principal method of stable fly control, but rather in combination with cultural and biological control methods. The preservation of natural mortality factors should be optimized and investigated. The use of insect growth regulators should be optimized and used in combination with cultural and biological control. Other methods for reducing stable fly population numbers at Karan Beef feedlot could be to reduce or remove stable fly resting sites by managing vegetation, weeds and grass surrounding pens, drains and sedimentation ponds to reduce refuge opportunities for stable flies.

5.2. FUTURE RESEARCH

- Cattle bunching behaviour and aggregation as a possible index of fly densities on cattle needs to be investigated.
- Photo imaging as a possible tool for quantifying bunching behaviour needs to be investigated.
- An economic threshold equation specific to Karan Beef needs to be developed.
- Possible impacts of stable flies as vectors of diseases needs to be investigated.
- Existing biological control agents and biological control measures need to be investigated to reduce the dependency on chemicals for stable fly control.
- Information gathered needs to be integrated into a fly management system.

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