Locust and grasshopper outbreaks in Zululand sugarcane, Kwazulu-Natal, South Africa

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ABSTRACT

The aim of the research presented in this dissertation was to investigate localised acridid outbreaks causing damage to sugarcane in Empangeni, Kwazulu-Natal, South Africa. Reports of outbreaks with increasing severity have occurred for the last six or seven years, which prompted this research. In general, little is known about acridid outbreaks in sugarcane due to their sporadic nature, therefore this project set out to identify morphologically and molecularly, all species associated with Empangeni sugarcane and to determine the ecology and population dynamics of the most important species, to provide baseline data for the development of an integrated pest management (IPM) plan against these crop pests.

A year-long population survey was conducted on a number of affected farms and adjacent grassland sites to determine the life cycles, population density and composition of this acridid complex on these farms. Surveys showed that this complex comprised five species: *Nomadacris septemfasciata, Petamella prosternalis, Ornithacris cyanea, Cyrtacanthacris aeruginosa* and *Cataloipus zuluensis*. Observed damage was recorded throughout the year in order to correlate with species densities. It was found that damage was closely associated with two species, namely *Petamella prosternalis* and *Nomadacris septemfasciat*a, therefore these two species are of particular concern due to their higher densities and large body size resulting in them being the biggest threat to Empangeni sugarcane. All five species are univoltine but two different life cycle strategies were discovered, 3 species exhibit a winter egg diapause while 2 species exhibit a winter adult reproductive diapause, an important finding considering the knowledge intensive method of control which has been proposed. Population surveys revealed a large difference in species composition among farms (sugarcane sites) and among grassland sites. *Nomadacris septemfasciata* and *Petamella prosternalis* showed a significant preference for sugarcane while species such as *Cyrtacanthacris aeruginosa, Zonocerus elegans* and *Orthocta* sp. seemed to prefer grassland sites.

Feeding potential trials were completed on the two most economically important species, *Nomadacris septemfasciata* and *Petamella prosternalis*. The results showed that *Petamella prosternalis* has a significantly higher feeding potential (% leaf damage) compared to *Nomadacris septemfasciata* even though in terms of dry weight and length, it is a smaller specimen. Feeding data were used to determine the voracity of these two species under laboratory conditions. *Petamella prosternalis* eats roughly 1.83 grams of fresh sugarcane per day while *Nomadacris septemfasciata* eats approximately 1.16 grams per day. These results were then compared to observed field damage data in order to gauge their accuracy and applicability within a field setting, which showed that although sugarcane field damage is significantly correlated with the population fluctuations of both these species, it is more closely associated with *Petamella prosternalis* resulting in a correlation coefficient of 0.429143 while *Nomadacris septemfasciata* had a correlation coefficient of 0.250.

Phase polyphenism in the red locust *Nomadacris septemfasciata* was investigated using three methods including traditional morphometrics (Elytra/Femur (E/F) ratio), hopper colouration and eye stripe data. The

E/F ratio indicated that the majority of the Empangeni population sampled is in the transiens and gregarious phase with a mean E/F ratio of 2.0063 and that they seem to be becoming more gregarious over time, with populations in 2012 having a mean E/F ratio of 1.9973 and in 2013 a mean E/F ratio of 2.01315. Hopper colouration also indicates that populations are generally showing gregarious tendencies with the majority of hoppers exhibiting gregarious type colouration. Eye stripe data showed that the majority of adult specimens observed had seven eye stripes, an indication of gregarious individuals. Geometric morphometrics, a relatively new, software based technique which has not been used in phase polyphenism studies before was used as a means to accurately measure variations among populations of *N. septemfasciata* by measuring the variation in forewing shape according to allocated landmarks. The resulting shape variation were compared to traditional morphometrics in an attempt to relate the two techniques so that geometric morphometrics can possibly be used as a tool to study phase polyphenism in locusts in the future. The results indicate that similarities in terms of location and gender exist between the two methods, however the exact same individuals should be used for both methods which will improve the accuracy of comparisons.

This study, for the first time identified the acridid complex causing damage to Empangeni sugarcane and provided a broad summary of the potential impact the complex has on the crop as well as what may be causing them to occur in this area such as optimal habitat conditions and sugarcane as a preferable host plant being two factors which are likely to be having an influence. The findings of this study provide the baseline data needed in order to develop a more integrated and sustainable approach to controlling these acridids pests. Understanding the biology of the species enables practitioners to make more effective management decisions which is clearly needed as the current techniques being used have not solved the ‘grasshopper problem’.

Key words: Grasshoppers, locusts, sugarcane, Empangeni, population surveys, feeding potential, phase polyphenism, morphometrics, strategic control, IPM.

OPSOMMING

Die doel van die navorsing wat in hierdie verhandeling was om ondersoek in te gelokaliseerde acridid uitbrekings veroorsaak skade aan suikerriet in Empangeni, KwaZulu- Natal, Suid-Afrika. Verslae van die uitbreek met toenemende erns plaasgevind het vir die laaste 6-7 jaar , wat gelei het tot hierdie navorsing. In die algemeen, is min bekend oor acridid uitbreek in suikerriet as gevolg van hul sporadiese aard , daarom is hierdie projek uiteengesit morfologies en molekulêre identifiseer , al die spesies wat verband hou met Empangeni suikerriet en die ekologie en bevolkingsdinamika van die belangrikste spesies te bepaal , te voorsien basislyn data vir die ontwikkeling van 'n geïntegreerde plaagbestuur (GPB) plan teen hierdie oes peste.

'N jaar lank bevolking opname is uitgevoer op 'n aantal plase wat geraak is en aangrensende grasveld plekke om die lewensiklus te bepaal , bevolkingsdigtheid en samestelling van hierdie acridid kompleks op die plase . Opnames het getoon dat hierdie komplekse bestaan uit vyf spesies: *Nomadacris septemfasciata , Petamella prosternalis , Ornithacris Cyanea , Cyrtacanthacris aeruginosa* en *Cataloipus zuluensis* Waargeneem skade is aangeteken deur die jaar ten einde te ooreenstem met spesie digtheid . Daar is gevind dat die skade was nou verbind met twee spesies , naamlik *P. prosternalis* en *N. septemfasciata* , dus is die twee spesies van besondere belang as gevolg van hul hoër digtheid en 'n groot liggaam grootte lei daartoe dat hulle die grootste bedreiging vir die Empangeni suikerriet. Al vyf spesies is univoltine maar twee verskillende lewensiklus strategieë ontdek , 3 spesies toon 'n winter eier diapouse terwyl 2 spesies toon 'n winter volwasse reproduktiewe diapouse , 'n belangrike bevinding oorweging van die kennis intensiewe metode van beheer wat is voorgestel . Bevolking opnames openbaar 'n groot verskil in spesies samestelling onder plase (suikerriet webwerwe) en onder grasveld webwerwe. *Nomadacris septemfasciata* en *Petamella prosternalis* het 'n beduidende voorkeur vir suikerriet, terwyl spesies soos *Cyrtacanthacris aeruginosa* , *Zonocerus elegans* en *Orthocta* SP . gelyk grasveld webwerwe te verkies .

Voeding potensiaal proewe is op die twee mees ekonomies belangrike spesies voltooi , *Nomadacris septemfasciata* en *Petamella prosternalis* Die resultate het getoon dat Petamella prosternalis het 'n aansienlik hoër voeding potensiaal (% blaarskade) in vergelyking met *Nomadacris septemfasciata* selfs al in terme van droë gewig en lengte , is dit is 'n kleiner model . Voeding data is gebruik om die gulzig van hierdie twee spesies onder laboratorium toestande te bepaal. *Petamella prosternalis* eet ongeveer 1.83 gram vars suikerriet per dag, terwyl *Nomadacris septemfasciata* eet ongeveer 1,16 gram per dag. Hierdie resultate is dan in vergelyking met waargeneem veld skade data om die akkuraatheid en toepaslikheid in 'n veld omgewing, wat getoon het dat hoewel suikerriet veld skade beduidend gekorreleer met die bevolking skommelinge van beide hierdie spesies, is nouer dit verband hou met Petamella prosternalis te meet lei tot 'n korrelasie koëffisiënt van 0.429143 terwyl *Nomadacris septemfasciata* het 'n korrelasie koëffisiënt van 0.250408

Fase polyphenism in die rooisprinkaan *Nomadacris septemfasciata* is ondersoek met behulp van drie metodes, insluitend tradisionele Morphometricsveld (Elytra / Femur (E / F) verhouding), hopper kleur en oog streep data. Die Elytra te Femur verhouding (E / F -verhouding) het aangedui dat die meerderheid van die bevolking is Empangeni in die transiens en gesellige fase met 'n gemiddelde E / F verhouding van 2,0063 en dat dit lyk asof hulle meer kuddedier met verloop van tyd , met bevolkings in 2012 met 'n gemiddelde E / F verhouding van 1,9973 en in 2013 'n gemiddelde E / F verhouding van 2,01315 . Hopper kleur dui ook aan dat die bevolking in die algemeen toon kuddedier neigings Thye meerderheid van hoppers uitstal gesellige tipe kleur. Oog streep het getoon dat die meerderheid van die volwasse eksemplare waargeneem het sewe oog strepe: 'n aanduiding van die gesellige individue. Geometriese Morphometricsveld , 'n relatief nuwe , sagteware gebaseer tegniek wat nie gebruik is in fase polyphenism studies voor was gebruik as 'n middel tot verskille tussen bevolkings van die rooisprinkaan akkuraat te meet deur die variasie meet in voorvlerk vorm volgens toegeken landmerke Die gevolglike vorm variasie is in vergelyking met die tradisionele Morphometricsveld in 'n poging om die twee tegnieke in verband te bring , sodat meetkundige Morphometricsveld kan moontlik as 'n instrument fase polyphenism om te studeer in sprinkane in die toekoms gebruik kan word. Die resultate dui daarop dat die ooreenkomste in terme van ligging en geslag bestaan tussen die twee metodes is egter presies dieselfde individue moet gebruik word vir beide metodes wat die akkuraatheid van vergelykings verbeter.

Hierdie studie , wat vir die eerste keer geïdentifiseer die acridid kompleks om skade aan Empangeni suikerriet en verskaf 'n breë opsomming van die potensiële impak van die kompleks het op die gewas sowel as wat kan veroorsaak word om hulle te kom in hierdie gebied soos optimale habitat toestande en suikerriet as 'n beter gasheer plant. Die bevindinge van hierdie studie verskaf die basislyn data wat nodig is om 'n meer geïntegreerde en volhoubare benadering tot die beheer van hulle as die begrip van die biologie van die spesie in staat stel om praktisyns meer effektiewe bestuur besluite wat duidelik nodig is as 'n die huidige tegnieke gebruik moet maak om te ontwikkel nie opgelos die " sprinkaan probleem".

Sleutel woorde: Locusts, sprinkane, suikerriet, Empangeni, bevolking opnames, voeding potensiaal, fase polyphenism Morphometricsveld, strategiese beheer, IPM.

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Chapter 1: General Introduction

1.1 Introduction

The sugarcane industry in South Africa is the 11th largest in the world in terms of production (FAO 2011), with gross income from all producers being R6.12 Billion for the 2011/2012 season (DAFF 2012). Like many commercially grown crops however, a variety of insect pests attack different parts of the plant. Insect species feeding on sugarcane are characteristically of limited geographical distribution, very few are cosmopolitan species (Williams et al. 1969). This phenomenon means that cane growing areas of the world are subject to a large diversity of insect pests which are often unique to a limited geographical area. Williams et al. (1969) lists five insect orders as major crop pests which include: Hemiptera (sap suckers), Coleoptera and Blattodea (root feeders), Lepidoptera (stalk borers) and Orthoptera (leaf feeders). Sugarcane pests may be indigenous or introduced alien species. In South Africa, the principal sugarcane pest is *Eldana saccharina* Walker (Lepidoptera: Pyralidae) (Leslie, 1981; Carnegie and Kfir, 1992) which is a moth indigenous to Africa. This pest bores into the sugarcane stalks in the larval stage and causes significant losses to the industry. Scarab beetles commonly known as “White Grubs” are indigenous root feeding pests which often causes highly sporadic, localised damage to sugarcane stools (Carnegie 1988). *Chilo sacchariphagus* Bojer (Lepidoptera: Pyralidae) is an example of a recently introduced stalk borer from the Far east attacking Mozambique sugarcane which is currently classified as a biohazard threat to the South African sugarcane industry (Anonymous 2006). It is therefore clear that in a South African and worldwide context sugarcane is subject to attack by a wide range of insects with vastly different characteristics which often have a significant impact on industry performance.

Grasshoppers and locusts (Orthoptera: Acrididae), commonly termed acridids, attack sugarcane in various parts of the world including Indonesia (Lecoq 1999), West Africa (Maiga et al. 2008), India (Easwaramoorthy et al. 1989) and southern Africa (Whellan 1968, Price & Brown 1999, Bakker 1999). These insects defoliate the leaves and destroy the growing points of the plant thereby reducing its photosynthetic capabilities (Williams et al. 1969, Easwaramoorthy et al. 1989). Acridid pests such as these pose continuing threats to crops, pastures and rural communities in developing countries around the world (Belayneh 2005). Williams et al. (1969) lists 62 species of Acridodea which are known to attack sugarcane worldwide. Locusts and grasshoppers are regarded as major economic pests of crops and grasslands throughout the world's dry zones (Lomer 1999), however, their sporadic and unpredictable nature has possibly been the reason why still relatively little is known about the identity, ecology and biology of many economically important species.

1.1.2 Locusts as crop and pasture pests

All locusts are grasshoppers but not all grasshoppers are locusts (Simpson & Sword 2006), the fundamental difference lies in the ability to exhibit density dependant phase polyphenism and form marching hopper bands or flying swarms which can develop into plagues (Sword et al. 2010, Lecoq et al. 2011a). Four species of plague locust occur in southern Africa, namely the Brown Locust, *Locustana pardalina* (Walker) (Orthoptera: Acrididae: Oedipodinae) the red locust, *Nomadacris septemfasciata* (Serville) (Orthoptera: Acrididae: Cyrtacanthacridinae) the African Migratory Locust, *Locusta migratoria migratorioides* (Linné) (Orthoptera: Acrididae: Oedipodinae) and the Southern African Desert Locust, *Schistocerca gregaria flaviventris* (Burmeister) Orthoptera: Acrididae: Cyrtacanthacridinae) (Faure 1935, Price & Brown 1999). However, the two chief destructive species in this region are *L.pardalina* and *N. septemfasciata* (Uvarov 1928). In South Africa, sugarcane growing areas lie within the invasion area of two locust species: *N. septemfasciata* and the African migratory locust (Whellan 1968). Although there are no recognised *N. septemfasciata* outbreak areas in South Africa (Bahana 1999) it is mentioned as an occasional problematic species along the eastern seaboard of Kwazulu-Natal (Faure 1935, Picker et al. 2004). *Nomadacris septemfasciata* is highly mobile and swarms are known to travel over 100km in a day (COPR 1972, McGavin 2001). In southern Africa, three major plagues of *N. septemfasciata* have occurred in recent history (Bahana 1999) (Table 1.1), the last one started in 1929 and lasted until 1944 and affected most of Africa, South of the equator. During this plague, Northern Natal was heavily invaded from 1933-1937 and cost the sugarcane industry approximately £1 million in 1934 alone (De V. Minnaar 1990).

Table 1.1: History of red locust *Nomadacris septemfasciata* (Serville) outbreaks in Africa.

Reference	Outbreak occurrence	Duration (Years)	Outbreak interval (Years)	Origin of outbreak	Extent
Gunn 1952	1847-1854	7	38	Unknown	KZN, South Africa
Smee 1940	1915-1919	4	10	Uganda	Uganda
Bahana 1999	1929-1944	15	50	Tanzania	Sub Saharan Africa
Bahana & Ngazero 1999	1994-1996	2	13	Buzi Plains, Mozambique	Southern Africa
FAO 2010	2009-2010	1		Tanzania, Malawi, Mozambique	Tanzania (contained)

It was discovered that the source of the locusts causing the last plagues was in several areas in Central and East Africa leading to the recognition of eight or nine relatively small, seasonally flooded, grassland outbreak areas (Gunn1960, Bahana1999)(Figure 1.1). An outcome of these particular plagues which lasted 15 years was the creation of the International Red locust Control Organisation for Central and Southern Africa (IRLCOCSA). Its mandate was to identify the source of plagues and control hopper bands and incipient swarms. There have been no major outbreaks since 1944, possibly because of our improved knowledge of locust outbreak dynamics, insecticide technology, application techniques and intervention strategies (Whellan

1968, Price & Brown 1999, Lecoq et al. 2011a, Bahana 2000). Consequently, it is regarded as a pest of the past. This opinion however, should be treated with caution as recent population upsurges in Africa have occurred, for example in eastern Africa (FAO 2009) and have demonstrated that outbreaks are difficult to detect in the remote and often inaccessible flooded grasslands where swarms persist (Brown & Price 1997). In 1996 swarms escaping from the Buzi plains in northern Mozambique flew over 2000km through Zimbabwe, Botswana and South Africa, where they were observed over Pretoria for the first time in 50 years (Price & Brown 1999). Locust outbreaks therefore remain a serious problem in southern Africa, especially outbreaks of Brown, African Migratory and *N. septemfasciata* which still threaten sustainable agricultural production in southern Africa (Price & Brown 1999, Lomer 1999). More recently, in 2008 large build ups of *N. septemfasciata* populations were reported in eastern Africa, which prompted a full locust control campaign lasting from May - July 2009 and costing an estimated US$ 2 million (FAO 2010). In 2012 and 2013 Madagascar experienced the worst plague of Malagasy migratory locust (*Locusta migratoria capito*) (Sauss) (Orthoptera: Acrididae: Oedipodinae) since the 1950's, threatening the livelihoods of 13 million people (FAO 2013). This situation reminds us that although locust outbreaks are often sporadic and of little concern, if the right conditions exist they can become plagues with huge humanitarian significance.

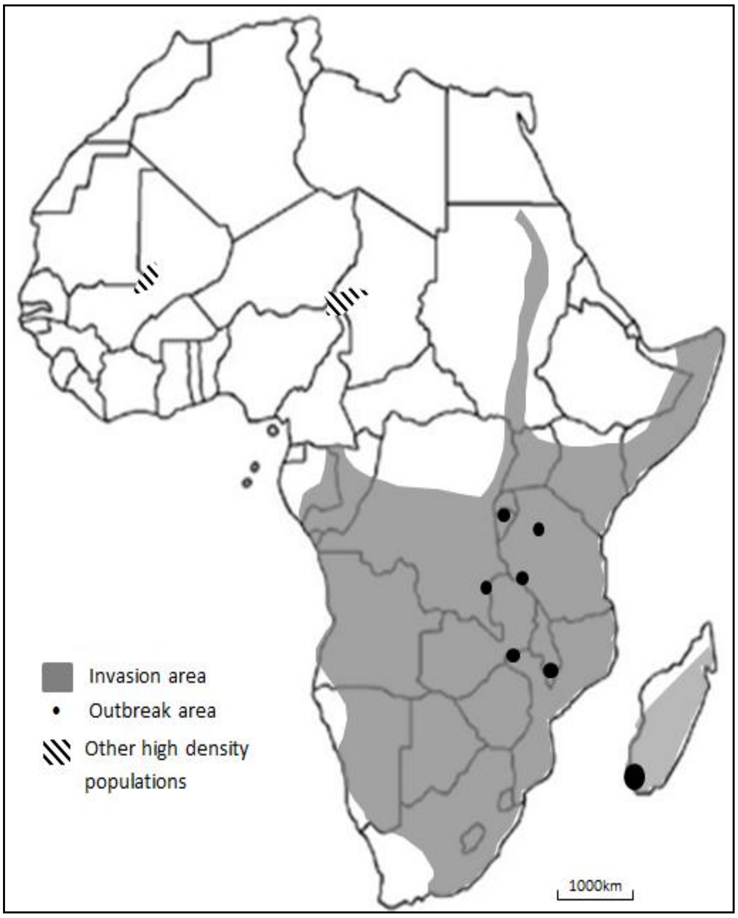

Figure 1.1: Recognised outbreak and invasion areas of the red locust *Nomadacris septemfasciata* (Serville). (Adapted from Bahana 1999).

1.1.3 Grasshoppers as crop and pasture pests

Grasshoppers have periodically exhibited both local and large-scale outbreaks on six continents and have been reported on throughout recorded history (Branson et al. 2006). In spite of their potential ecological and economic importance, little is known about the mechanisms and interactive effects that underlie grasshopper population dynamics and outbreaks (Joern and Gaines 1990, Lockwood 1997, Joern 2000).

Grasshoppers do not have gregarious habits and therefore do not swarm and migrate even in years of mass outbreaks, they remain pests of purely local importance with no immediate threat to neighbouring districts (Uvarov 1928). Grasshopper outbreaks have occurred sporadically in southern Africa and apart from the Elegant grasshopper *Zonocerus elegans* (Sjöstedt) (Orthoptera: Pyrgomorphidae)which attacks a wide range of wild and crop plants (Nyambo 1991) little information is available for other species. Outbreaks in sugarcane have been equally as sporadic as documented in the insect collection at the South African Sugarcane Research Institute (SASRI). Generally, long recession periods exist between short population upsurges so consequently there seems to be no concerted research undertaken on grasshoppers as sugarcane pests.

In terms of control, what applies to locusts does not necessarily apply to grasshoppers (Uvarov 1928). Grasshoppers, unlike locusts generally do not have definite or relatively restricted breeding grounds and they usually breed over very large areas, so to attempt the same control strategy used against locusts would be too expensive and not justifiable (Uvarov 1928). Chemical pesticides have for decades been the mainstay of control operations against locusts and grasshoppers for decades (Steedman 1990). However, the disadvantages of chemical control are recognised, and have been addressed thoroughly (e.g. Ritchie & Dobson (1995) and Berger (1991). Pest control tactics are economically justifiable if the value of the benefit (yield loss averted) exceeds the cost of the pesticide and its application (Economic threshold level) (Stern et al. 1959). Determining the economic threshold level, however, is difficult particularly for mobile and polyphagous insects such as grasshoppers and locusts. In combination with other highly variable interactions, the economic threshold is even more difficult to calculate (Hewitt & Onsager 1982).

1.2 Distribution, pest status and biology of important locust and grasshopper species

There are five species attacking Empangeni sugarcane which warrant attention. These species all belong to the family Acrididae. Within the acridid family they belong to three different subfamilies namely: Cyrtacanthacridinae (3 species), Tropidopolinae (1 species) and Eyeprepocnemidinae (1 species). The common factor between these species is the presence of a conspicuous spur or spine ventrally between the front legs, therefore, this group of grasshoppers are sometimes called the ‘spur-throated grasshoppers’. A study done by Bazelet (2011) in which grasshopper assemblages of indigenous vegetation corridors were assessed in the Zululand region of KwaZulu - Natal, a total of 33 individuals of the four most abundant species currently found in Empangeni sugarcane, were caught during that entire study. There is every reason to believe, therefore, that sugarcane crops are supplying an optimal habitat for certain species and is the reason why these relatively uncommon grasshopper species have become more abundant and are now regarded as serious pests in the region.

1.3 Morphometric studies

Traditional morphometrics and geometric morphometrics are used in this study. Morphometrics is the study of shape variation and its covariation with other variables (Dryden & Mardia, 1998). Traditional morphometrics uses multivariate statistics to describe patterns of shape variation between different groups usually using linear distance measurements, counts, ratios or angles (Adams et al. 2004). Geometric morphometrics, a fairly recently-developed method which became popular in the late 1980's, includes a number of approaches for the multivariate statistical analysis of coordinate data, usually limited to landmark point locations (Zelditch et al. 2004). These two methods will be used in conjunction with one another as tools to investigate phase polymorphism in *N. septemfasciata* in Empangeni sugarcane.

1.4 Grasshopper population dynamics – factors affecting population upsurges

The environmental factors regulating grasshopper populations – like all other insects is incredibly complex (Joern & Gaines 1990) and the biotic and abiotic processes involved are numerous as illustrated in figure 1.2.

The primary processes regulating insect population dynamics are often considered to be external factors such

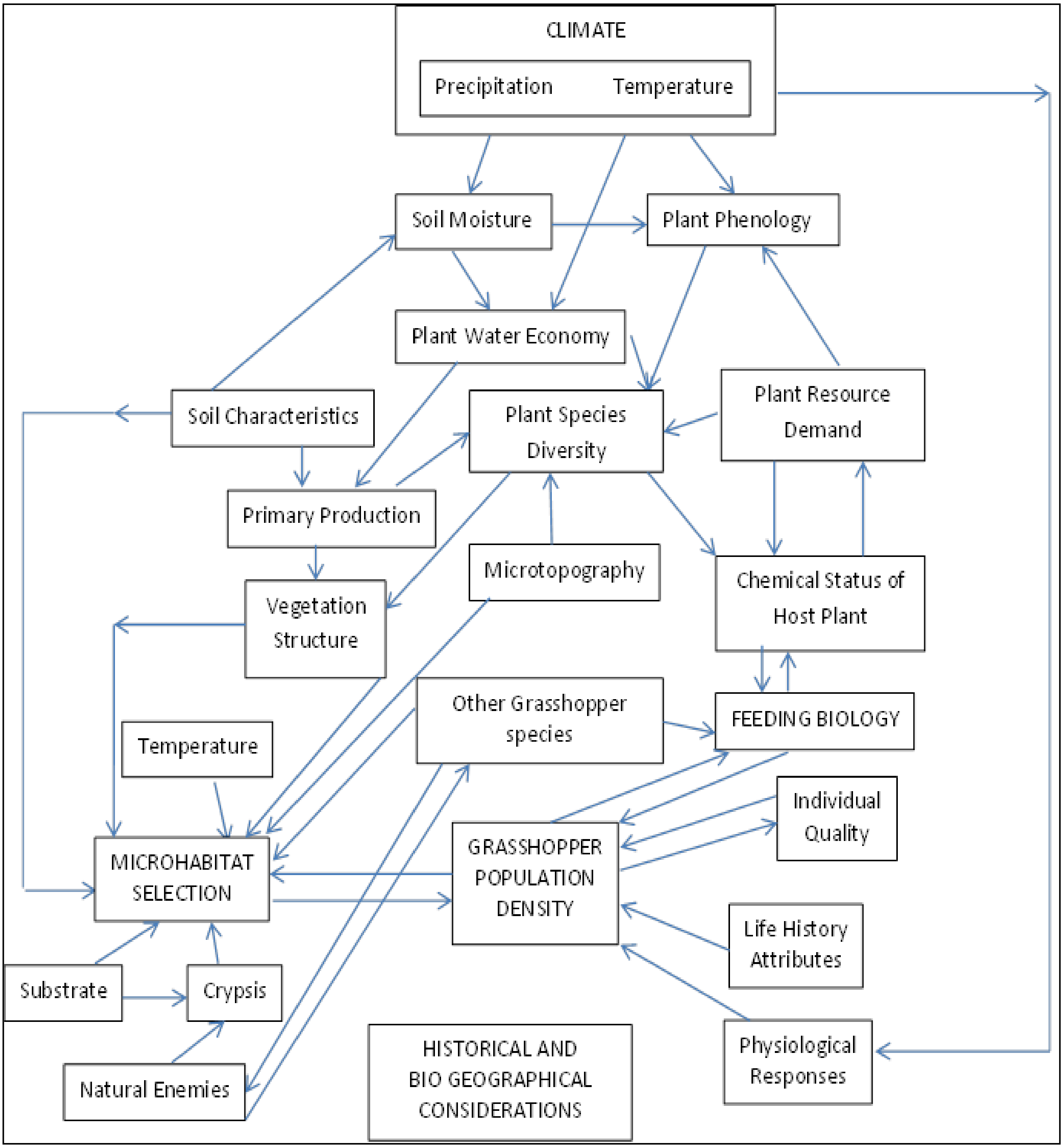

Figure 1.2: Biotic and abiotic factors involved in population dynamics of grasshoppers (adapted from Chapman & Joern 1990).

as climate, especially temperature and rainfall with density dependant biotic factors also being mentioned occasionally (Andrewartha & Birch 1954, Milne 1957, Dempster 1963). Grasshopper population dynamics can be considered to be consistent with the general patterns and processes of other insect herbivores (Joern & Gaines 1990) however, density dependant factors, which are often important regulating mechanisms in many insect orders (Speight et al. 2008) are often less important with regards to Orthoptera, particularly during plagues, possibly due to their mobile nature and migrating tendencies. Although climatic factors play an integral part in grasshopper population regulation, climate alone cannot be regarded as the primary influence resulting in outbreaks (Uvarov 1928). It is most likely a complex network of biotic and abiotic interactions influencing population dynamics (Joern & Gaines 1990).

1.4.1 Climate

Fluctuations in grasshopper abundance are commonly attributed to changes in weather (Andrewartha & Birch 1954, Dempster 1963, Uvarov 1966, Kemp 1987) and rainfall is recognised as a major factor influencing reproductive success which has been well documented, see: Symmons 1959, Joern & Gaines 1990. Many studies mention how “favourable climatic conditions led to large scale outbreaks” [see: Millist and Abdalla (2011) and Lomer et al. (1999)] although they do not mention what these favourable conditions were. Lecoq et al. (2011a) compared the reproduction rates of *N. septemfasciata* in Madagascar to the abundance and distribution of rainfall and concluded that rainfall has a strong impact on both egg and hopper survival and, in fact, low or patchily distributed rainfall, long dry periods as well as excessive rainfall can significantly reduce hopper survival. Lecoq (2011a) proposed that during egg development (September and October) about 15 cm of rain per month is required for successful egg development and rainfall above 30cm per month can become unfavourable, resulting in egg mortality. Various other authors found that a key mortality factor was a dry period during the vulnerable egg stage (Lea & Webb 1939, Albrecht 1957, Gunn 1956) which certainly has management implications in terms of predicting the reproductive success of the next generation of hatchlings. Outbreaks often develop due to consecutive seasons of favourable weather acting directly or indirectly (food supply) on insect populations (Speight et al. 2008). However these “favourable” conditions differ drastically between different species and between different areas. The relationship between weather and *N. septemfasciata* population upsurges has been addressed since the 1950’s (Rainey et al. 1957) but the conditions favouring upsurges have remained highly debated. Recently, Chiconela et al. (2003) developed a model to predict outbreaks in the Buzi plains of Mozambique. The model predicts that consecutive years of below average rainfall in the area are favourable for outbreaks such that the longer a dry spell continued, the more likely it was that an outbreak would occur. This also seems to be the case in outbreak areas of Tanzania where it was found that consecutive dry years result in a slow population build up over time and if followed by a season of favourable rains, egg survival increased and ample forage was available for emerging hoppers, resulting in population increases and eventually outbreaks (Peter Spurgin pers. Comm.). Although there is no doubt that weather patterns have a major effect on insect population dynamics, it cannot be the only factor responsible, as Uvarov (1928) explains, climatic factors alone are not sufficient for an explanation of periodicity of outbreaks. The very sensitive link between temperature, rainfall and locusts also opens up the debate on climate change in relation to locust outbreaks.

1.4.2 Impact of natural enemies

Grasshoppers are attacked by a wide range of natural enemies which includes pathogens, parasites, parasitoids and predators (Dempster 1963) of all life stages (Greathead 1966). The relative importance of mortality factors in relation to population density is illustrated in figure 1.3.

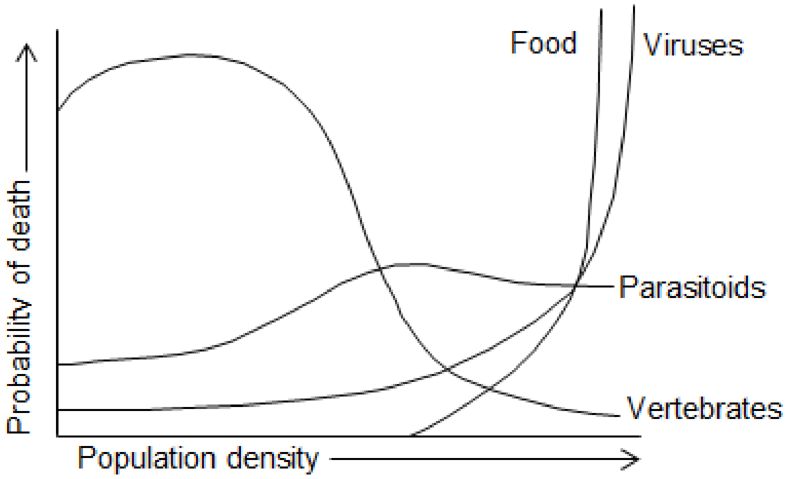

Figure 1.3: Relative importance of various mortality factors in relation to population density (Adapted from Berryman 1987).

Population size can certainly be altered by the presence of natural enemies. However, there is no clear consensus as to whether their impact is an important regulating factor or not (Joern & Gaines 1990) and it has been argued that natural enemies merely dampen the peaks of population fluctuations rather than have any significant impact in regulating them (Dempster 1963). In contrast, fungal pathogens in particular have been shown to significantly impact grasshopper populations (Chapman & Page 1979) which is possibly the reason why a number of entomopathogenic fungi such as *Metarhizium anisopliae* (Metchnikoff) (Hypocreales: Clavicipitaceae) or Green Muscle™ are commercially manufactured as bio control agents against grasshoppers. In most cases, the ability of natural enemies to regulate acridid populations has not been proven (Joern & Gaines 1990) so clearly, in order to try quantify this, more studies are needed. Many studies list the potential parasitoids and predators of acridids (See: Dempster 1963, Rees 1973). Although it has been shown that both can significantly regulate grasshopper population numbers during non-outbreak periods, their ability to supress population numbers during an outbreak (making them useful bio-control agents) is unclear. Some argue that predator and parasitiod numbers are simply too few to have a significant impact during outbreak periods (Dempster 1963).

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1.4.3 Insecticide Resistance

Synthetic acridicides are the most widely used control method against grasshoppers and locusts in Africa (Bahana 1999, Price & Brown 1999) and it is the current scenario in Empangeni sugarcane where control is exclusively based around these crop protection tactics. As such, populations of insects against which these are used can become resistant to them, especially if they are not used as recommended. Field-evolved resistance is defined as a "genetically based decrease in susceptibility of a population to a toxin caused by exposure of the population to the toxin in the field" (National Research Council 1986, Tabashnik 1994). Growers which have experienced high grasshopper populations and damage have used various insecticide products and methods in an attempt to control them, using their own intuition with some guidance from extension officers and others. Due to the recent nature of the grasshopper problem there is a lack of information and data with regards to how to control them and consequently crop protection measures have been carried out on a hap hazard and ill-informed basis. These issues have resulted in many ineffective spray applications and possibly led to the build-up of resistance among acridid populations in the Empangeni region. However, development of pesticide resistance in African acridids has never been documented (Maiga et al. 2008).

1.4.4 Soil characteristics

For the red locust, most kinds of soil appear to be suitable for egg laying as long as the soil is fairly moist (Faure 1935, Whellan 1968). Faure (1935) states that in South Africa, during the 1930's outbreak, egg packets were laid in a great variety of soils including black turf, red sand, gravel, stony koppies and even in hard packed roads. The main factor influencing oviposition area therefore does not seem to be soil type but rather the presence of bare soil. Symmons and Carnegie (1959) in the Rukwa valley, Tanzania, found that in terms of oviposition choice, *N. septemfasciata* shows a very significant preference for ground which was burnt the previous season and therefore was bare at the time of oviposition. They also found that where females did lay in grassy, vegetated soil, oviposition and egg-pod survival were least successful. Soil moisture plays an important role in the success of acridid breeding (Chapman & Joern 1990). Gunn (1956) found that in the Rukwa valley of Tanzania, low *N. septemfasciata* populations were often associated with high rainfall within the catchment presumably due to high egg mortality. Similarly, in studies of lake levels in association with incidences of swarm formation, Gunn (1955) concluded that persistent flooded plains are not favourable to *N. septemfasciata* population upsurges as excessive moisture decreases egg survival.

1.4.5 Host plant influences

Host plants play an important role in grasshopper dynamics through their influence on key population parameters (Joern & Gaines 1990). Many insect herbivores, including grasshoppers and locusts, have higher rates of fecundity when protein or nitrogen levels in their host plants are high (Awmack & Leather 2002) and naturally occurring plants may be generally poorer in quality compared to some crops (Joern & Gaines 1990). Therefore, the possibility of native grasshoppers switching from natural grassland plant hosts to an ever expanding sugarcane crop is certainly conceivable (as is the case with *Eldana sacchariana)*. According to Lecoq (1999), sugarcane plantations have a high potential for supplying good locust breeding habitats in terms of food, shelter and oviposition sites.

1.5 Laboratory rearing of selected species

Samples of all species occurring in the study area were collected alive in order to establish laboratory populations. They were bred by conferring to the techniques and equipment used by Albrecht (1953) and Harvey (1990). From these populations, aspects of their ecology such as-life cycle parameters and lengths can be determined under controlled conditions. Structured investigations to test the effects of certain biotic and abiotic factors on populations and individuals will be carried out in this study. These studies will further increase our understanding of the species and provide knowledge of their life histories which will be crucial in the development of IPM strategies which is a knowledge intensive management method (Matson, 1997).

To my knowledge, the only species present in Empangeni sugarcane which has been bred under laboratory conditions before is *N. septemfasciata* (Albrecht 1953).

1.6 Aim, objectives and thesis structure

1.6.1 Aim

The main aim of this study was to gain an understanding of the species composition, biology and ecology of the acridid species assemblages causing damage to sugarcane in the Empangeni region of Kwazulu-Natal, South Africa.

1.6.2 Objectives

This was achieved through a number of objectives including:

- A year-long population survey to determine the species composition and relative abundance of these species associated with these outbreaks.
- Determination of their population dynamics and life history traits.
- Determine the pest status of the different species in terms of their economic significance to affected growers
- Make inferences on the possible causal factors for outbreaks
- Determine the potential for swarming and to measure and understand the morphological differences between geographically separated populations using morphometrics.

1.6.3 Thesis structure

Chapter 1: is a general introduction and literature review of grasshoppers and locusts as crop and pasture pests and links are provided as to why this group could have become problematic in Empangeni sugarcane.

Chapter 2: reveals the species composition, and their ecology and biology on the effected farms and adjacent semi-natural grassland in the area. (Appendix 1 and 2 provide identification keys which were designed to facilitate correct identification of important species for growers, extension officers and researchers).

Chapter 3: Investigates the use of morphometrics as an indicator of swarming in the red locust.

Chapter 4: Describes feeding trials used to determine the pest status of the two most prominent acridid species found in Empangeni sugarcane. These trials were used to correlate with population densities and observed field damage in order estimate threshold levels. The feeding potential of the relevant species described in this chapter provides evidence for the identification of priority species in terms of potential damage caused to sugarcane

Chapter 5: Provides general conclusions about the relevance of the findings of this study and recommendations are made for further research. The objectives of this study have been designed to fit into the future development of an integrated pest management plan (IPM) with the goal that it will be implemented in the affected areas in order to manage acridid infestations so that they do not exceed the economic injury level of the sugarcane crop. Each of the chapters has been written up as separate chapters for publication purposes therefore some repetition may occur.

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Chapter 2: Population Surveys of grasshopper assemblages in Empangeni sugarcane

2.1 Introduction

Insect grazers such as grasshoppers and locusts are common native components of grasslands world- wide (Branson et al. 2006). The vegetation type of the Empangeni region in the KwaZulu-Natal province of South Africa is described as KwaZulu-Natal Coastal Belt Grassland (Mucina & Rutherford 2006) and grasshopper species seem to be a common component of grasslands in this region, (Bazelet 2011). These acridid species play a vital role in grassland ecology and functioning in terms of nutrient cycling, primary production and as a food source for many predators (Lockwood 1993).

Figure 2.1: A large swarm of *Nomadacris septemfasciata* adults along a cane break in a commercial sugarcane farm in Empangeni, there are approximately 350 individuals in this screen shot. B. Recently emerged *N. septemfasciata* hopper band in sugarcane in the same region.

In addition, they exhibit periodic local and large scale outbreaks which may affect a range of crops or compete with ungulate grazers for forage (Branson et al. 2006). In Empangeni, population upsurges have been reported for a number of years at increasing infestation levels which has prompted further investigation (Figure 2.1). For effective integrated pest management (IPM) tactics to be developed, a knowledge base must be built up which means obtaining a thorough knowledge of the target pest's life cycle, its ecological and behavioural interactions with its environment and natural controlling factors (Conlong and Rutherford 2009). This information is then used to develop a more integrated and sustainable approach for control as opposed to only relying on chemical crop protection strategies such as insecticide spraying (Belayneh 2005, Branson et al. 2006).

Almost every serious attempt to study natural or applied insect control of Orthoptera in the field has involved population sampling (Morris 1960), after the species causing the outbreaks have been identified. Surveys have generally been used to estimate population intensity in the field for conservation purposes (Gardiner et al. 2002) but also for studies relating to pest species (O' Neill et al. 2003). Variation between study sites in terms of environmental conditions and species assemblages make it hard to compare the results of one survey to another (Gardiner et al. 2005). Almost all population studies on grasshopper pests seem to have been undertaken in natural vegetation such as rangelands (see: Woodrow 1965, Branson 2005) and it seems very few studies have quantified population densities within an affected crop. Where an attempt of this has been made it has often been reliant on qualitative data, (see: Vestal 1913) which has obvious drawbacks. A similar scenario has occurred with the grasshopper problem in Empangeni where population densities and related damage have been reported based on qualitative visual estimates and opinions of various concerned parties. When attempting to study animal populations, quantitative population surveys based on rigorous capture methods are needed to gain a more accurate understanding of the situation (Clarke 1948, Southwood & Henderson 2000). Gathering quantitative data also means that it can be used for comparison over time and to correlate with other factors such as weather which is not possible at the moment. The most commonly used methods to determine acridid density are sweep netting and quadrat counts (Gardiner et al. 2005). Grasshopper populations in tall vegetation are very difficult to sample (Gardiner et al. 2005), which is the situation in sugarcane in Empangeni for the majority of the year. The only method which supposedly might work is night trapping (Gardiner et al. 2005) which would not have been practical in sugarcane fields due to lack of a nearby power source and the problem of theft. This is the reason why drive netting was developed. This was regarded as the only fairly accurate way to sample adult fliers within a sugarcane matrix when the sugarcane is tall. The mixed sugarcane age during sampling meant that three different methods were applied during the sampling period.

This chapter was, therefore, to identify the species of acridids making up the complex attacking sugarcane in the Empangeni area, and which species were the most 'common' species in these complexes. Secondly, determine the most efficient and practical method to accurately estimate population densities of adults and nymph populations. Thirdly, it provides an understanding of the biology of these species which can be used to improve management of these acridid pests in the affected sugarcane fields.

2.2 Materials and Methods

2.2.1 Site descriptions

Population surveys took place in the Empangeni region of KwaZulu- Natal, South Africa (28°44’56.74’’S 31°53’59.24’’E) from 30th May 2012 until 30th May 2013. 56 surveys took place during this period. Four farms which reported significant damage and high population densities were chosen as study sites (Figure 2.2). Magazulu farm (Tedder) (28°44'9.54"S 31°52'16.60"E) is situated within 2km of Empangeni town and is the most southern site which was surveyed. GSA farms (28°40'54.94"S 31°54'51.98"E) and Crystal Holdings, owned by Tongaat Hullett (28°40'0.50"S 31°54'47.37"E) are situated close to each other, roughly eight km from Empangeni town and Jengro (28°37'30.84"S 32° 0'52.68"E) is the most northerly site, situated roughly 18km from Empangeni town.

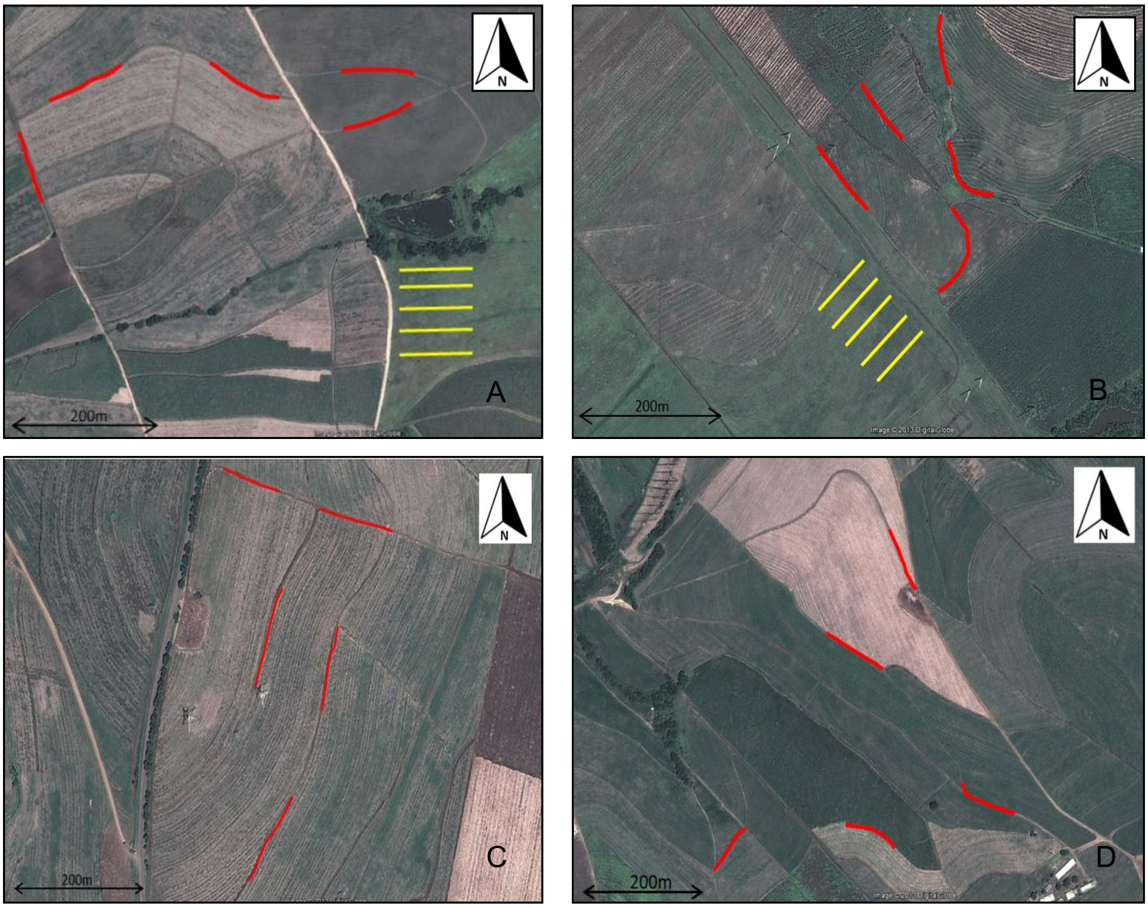

Figure 2.2: Aerial view of four farms where surveys took place and indicating the five 100m transects per farm (red lines) Yellow lines indicate the two veld survey areas. A) Tedder. B) Crystal Holdings. C) GSA farm. D) Jengro.

methods were used at certain times of the year because adults and hoppers responded differently to various trapping techniques and the annual sugarcane harvesting regime present in Empangeni also contributed to the choice of sampling method. Quadrat counts were not used because they are only suitable for low density populations within short vegetation (Gardiner et al. 2005) and sweep netting was found to be an effective method to use for hoppers and adults (adapted to drive netting for the latter). When the sugarcane was young, conventional sweep netting was used as it allowed the standard 180^o sweep to be done (see Gardiner et al. 2005), however as the cane got taller (above hip height) standard sweep netting became impractical and drive netting was then used.

From May 2012, drive netting was used to catch adult fliers, sugarcane age varied between the different farms, but survey sites were chosen specifically so that age was as similar as possible at the four sites. Mean cane age at the start of the survey was five months. A specific route was driven along cane breaks (Figure 2.2) while simultaneously brushing the insect net (Bugdorm cages & traps- 60cm diameter Product #: DC0005, Taiwan) against the sugarcane canopy. The route consisted of five 100 metre transects completed at a constant speed of 20km per hour and repeated on each farm. This method was used because tall cane forms very dense stands and has a closed canopy which makes conventional sweep netting within the sugarcane field impossible as it restricts the standard sweep netting movement (Bomar 2001). Sugarcane harvesting started in July 2012 at some study sites. Drive netting was thus impossible because the harvesting operations caused acridid populations to disperse more widely over the survey area. A visual line transect method was used in the harvested sugarcane in order maintain sampling accuracy. This method was used from the beginning of August until the end of November and involved walking five 100 metre transects per farm which were measured using a Garmin global positioning system (GPS). Line transects were completed as close as reasonably possible to the original drive netting transects in figure 2.2. A single transect involved walking 100m between a sugarcane row while counting each acridid which was disturbed in the row in which the counter was walking and the row either side of the counter (i.e. three rows - a width of approximately three metres). A hand held tally counter (Upgreen counters®, UK) was used to record the number of grasshoppers disturbed per transect which was added to the total amount for all five transects per farm. At the beginning of November, the new generation of hoppers started to emerge and sweep netting using the same net used for drive netting was used. Sweep netting was done along the same walked transects and conditions as mentioned above (during visual transects). Sweep netting was done by the operator sweeping across the tips of the cane in a full arc from his left to his right as described in (Gardiner et al. 2005). The methods described above are summarized in Table 2.1.

Table 2.1: Summary of the survey methods used to measure acridid abundances during population surveys on the four chosen survey sites.

Survey Methodology				
Method	From	To	Approx. Sugarcane Age	Growth Stage
Drive netting	30-May-12	12-Sep-12	8 months	Tall (6 ft. +)
Visual transects	20-Sep-12	03-Dec-12	12 months	Tall (6 ft. +)
Sweep netting	21-Nov-12	10-Jan-13	3 months	Moderately tall (2-5 ft.)
Drive netting	17-Jan-13	15-May-13	5 months	Moderately tall (2-5 ft.)

2.2.3 Data collection

During each field trip, rainfall and temperature was recorded for that day. An attempt was made to conduct field trips only during sunny, dry days in order to minimize sample bias due to climatic factors. One area on each farm, where population densities were high was selected as the designated survey site for that farm. Therefore, regardless of time of year or survey method being used, surveys were completed in the same general area. During direct sampling methods (netting), transparent, plastic, marked, perforated tubs (Height: 27cm, Width: 14cm) were used to store insects separately per survey area to be brought back to the laboratory alive for identification and counting. Once in the laboratory, insects were either killed by freezing to aid in counting or ‘cooled’, then counted and kept alive in order to be added to laboratory colonies when necessary. During indirect methods (visual transects) disturbed individuals were identified and recorded without being caught. Identifying individuals during flight was not difficult after spending a period of time in the field, it was also made easier by the fact that while visual transects were undertaken only two species were present (reproductive diapause type – Table 2.5). Individuals collected were sorted into morphologically similar groups, and identified by a specialist (Corinna S Bazelet, PhD, Postdoctoral Fellow, Stellenbosch University, Department of Conservation Ecology and Entomology, Private Bag X1, Matieland 7602, South Africa). Basic identification keys were constructed to assist farmers and extension officers to identify the acridid species found in the Empangeni area, to assist in decision making regarding pest management (See appendix 1 & 2).

2.2.4 DNA Extraction

Grasshopper species identified by Dr. Bazelet using morphological characters were then identified molecularly in the department of biotechnology at SASRI, the aim being to check if any species have been sequenced before and if not, load these sequences onto online databases for future reference.

DNA was extracted from the muscle of the hind femur, using the KAPA Express Extract DNA Extraction kit (Kapa Biosystems, South Africa) according to the manufacturer’s instructions.

PCR using Cytochrome Oxidase gene primers

PCR amplification was conducted using the KAPA 2G Robust PCR Kit (Kapa Biosystems) with 1µl DNA template. The final reaction conditions were as follows: 1X Kapa2G Buffer A, 0.2 mM dNTP mix, 0.5 µM each COI Forward and COI Reverse primer and 0.5 units Kapa2G Robust DNA Polymerase.

The DNA primer sequences were:
COIF
5’ AATTGGGGGGTTTGGAAATTG 3’
COIR
5’ GCTCGTGTATCAACGTCTATTCC 3’

PCR reactions were conducted in an Applied Biosystems Veriti Thermal Cycler using the following thermal cycling profile: 94°C for 2 min, followed by 35 cycles of 94°C for 30 sec, 55°C for 50 sec and 72°C for 90 sec. Final extension was at 72°C for 10 min. PCR products were purified using the DNA Clean and Concentrator kit (Zymo Research, USA) according to the manufacturer’s instructions.
DNA sequencing was conducted using the BigDye Terminator v3.1 Cycle Sequencing kit (Applied Biosystems, USA) according to the manufacturer’s instructions. Sequencing reactions were conducted in an Applied Biosystems Veriti Thermal Cycler using the BigDye Terminator v3.1 kit recommended thermal cycling profile. Sequencing products were purified using the BigDye XTerminator Purification Kit (Applied Biosystems) according to manufacturer’s instructions.

Uploading of DNA sequences to online database

After running a search on two DNA barcoding websites namely BOLD systems (www.boldsystems.org) and the National Centre for Biotechnology Information (www.ncbi.nlm.nih.gov) it was found that none of the species’ DNA had been submitted to these websites. Sequences were then submitted to BOLD systems and Genbank.

2.2.5 Damage rating index

The level of damage as a result of grasshopper feeding was observed on a weekly basis to generate a damage rating index for the period: May 2012-May2013. During weekly population surveys, five random sugarcane stools within the sugarcane survey sites were chosen and a damage rating from 1-5 was allocated to the stool depending on the amount of damage present, estimated as the percentage of leaf area lost on the youngest top five green leaves of a randomly chosen stalk in the stool (Table 2.2).

Table 2.2: Criteria used as a guideline to assess damage in order to obtain a damage rating index to correlate against population abundance data.

Rating	% Damage estimate
0/5:	0%
1/5:	1-20%
2/5:	21-40%
3/5:	41-60%
4/5:	61-80%
5/5:	81-100%

The five values per transect were then averaged to get a mean weekly damage rating per farm. The four mean weekly damage ratings were combined and averaged to get a monthly damage rating index and then plotted against the other farms over the entire year.

2.2.6 Grassland surveys

Grassland surveys were completed as a means of comparing grasshopper population densities and species composition in grassland sites to sugarcane survey sites. Four sites of natural grassland adjacent or nearby to the sugarcane survey sites on each farm were sampled for five months from October 2012 to February 2013. The objective was to compare natural grassland to sugarcane in terms of species abundance and diversity. Due to unforeseen circumstances, two of the grassland survey sites had to be abandoned therefore, only two grassland sites were used for comparison, from the period: 21 November to 17 January (7 weeks). During this period, all acridid species sampled were in the hopper stage. Grassland surveys were completed using the same sweep net method used in the sugarcane study sites. Five 100m transects were walked per site while sweeping the net over the top half of the grass sward in an 180^0 arc (Maps: A & B in figure 2.2). Captured specimens were then placed in separate marked tubs as described above and brought back to the laboratory for identification and counting.

2.2.7 Data analysis

Rank abundance calculation

Rank abundance was calculated by obtaining the total count of each of the five acridid species captured during the entire population survey period. These values were then logged to obtain a log abundance value which designated each species a ranking from 1-5 according to their total abundance in Empangeni sugarcane.

Correlation of relative abundance with the damage rating index

Monthly relative abundance was calculated by dividing the total count for all five species by the individual count of each species. The relative contribution of each species to the total count as a percentage was then plotted on a graph. In an attempt to correlate relative abundance of acridid population densities with observed damage, a Gamma rank correlation analysis was performed. For the purposes of this data set, the Gamma analysis is preferable over the Spearman's R analysis as the data contains many tied observations, which the Gamma analysis explicitly accounts for. Where a correlation between species abundance and the damage rating index was found, a pairwise comparison was done (two species showed a correlation with damage). All analyses were completed in Statistica 11.0. (StatSoft Inc., Tulsa, USA)

Correspondence analysis

To compare whether farms were associated with any particular species of grasshopper, a simple correspondence analysis was used with grasshopper species as column variables and farms as row variables. Likewise, a simple correspondence analysis was used to compare habitat type (sugarcane vs. grassland) with grasshopper species, over a seven week sampling period with habitat type as column coordinates and species as row coordinates. During this period an additional farm known as Kelly farm (see Figure 4.2) was sampled for the purposes of the correspondence analysis (Figure 2.11) using the same methods which were described in section 2.2.2. No supplementary row variables were used in either analysis. The analyses were conducted in Statistica 11.0. (StatSoft Inc., Tulsa, USA).

2.3 Results

A number of grasshopper species were recorded in Zululand sugarcane, including a few *Orthocta* sp. (Orthoptera: Acrididae) and *Zonocerus elegans* Thunberg (Orthoptera: Acrididae) however, five species were of particular concern due to their high population densities and consequently they are discussed here.

2.3.1 *Nomadacris septemfasciata* (Serville 1838) (Orthoptera: Acrididae: Cyrtacanthacridinae)

Nomadacris septemfasciata or *N. septemfasciata* is the only true migratory locust among this assemblage of grasshoppers and is a well-known pest of central and southern Africa (COPR 1982) and an occasional problematic species along the eastern seaboard of Kwazulu-Natal (Faure 1935, Picker et al. 2004). The scientific name '*septemfasciata'* refers to the seven dark, transverse bands on the forewings of adults (COPR 1972). The overall body colour of adults is a mixture of beige and brown, they are a relatively large (Males: 70mm Females: 80mm) and slender looking species and are very strong, mobile fliers. Two distinguishing characters in adults are the saddle shaped pronotum and the basal red/purple colouration of the hind wings in mature adults (Faure 1935) (Figure 2.3).

Nomadacris septemfasciata prefers tropical and subtropical regions (Faure 1935). In outbreak areas which can be considered as ideal *N. septemfasciata* habitat, Vesey- Fitsgerald (1955) described the following common characters between all of these areas:

- An extensive mosaic of tall and short grasses dominated by the species of *Echinochloa pyramidalis*, *Cynodon dactylon*, *Hyparrhenia* spp.
- Flood plains with impeded or closed drainage systems
- A distinct dry and wet season
- Fairly wide diurnal temperature fluctuations

Nomadacris septemfasciata is univoltine. A significant feature in the life cycle of *N. septemfasciata* is the very long life of the adult which can live for up to 10 months. Faure (1935) and Burnett (1951) provide detailed descriptions of the life cycle of the red locust. Hoppers emerge from November through to January and the first adults appear after about two months (February and March). For approximately eight months adults exist in a reproductively immature state (reproductive diapause). Mating and egg laying coincided with the commencement of summer rains in October/November. The incubation period is roughly one month after which the next generation of hoppers will emerge. Each female lays three egg packets on average with each containing between 20 and 100 eggs which explains why populations are able to increase so markedly. The general rate of increase of a population in ideal conditions is estimated at 100% per annum (Robertson 1958). The life cycle can be summarized as follows:

Egg- Laying: November-January
Hopper stages: December-May
Adult fliers: April-January

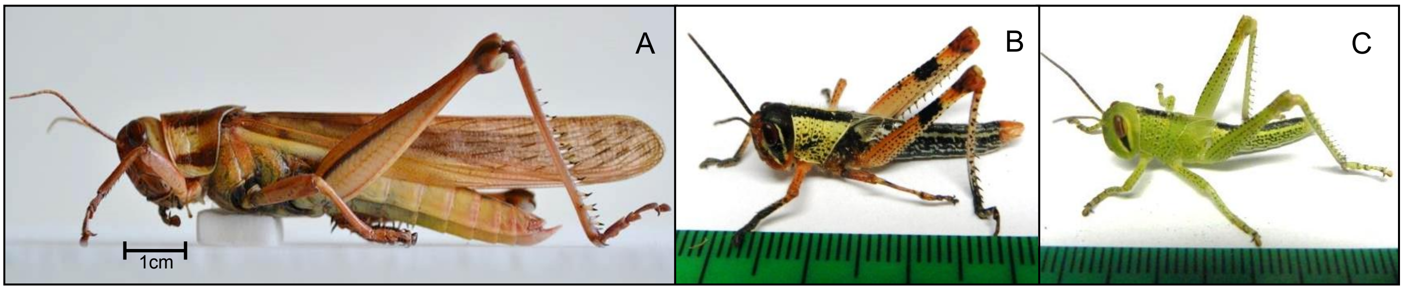

Figure 2.3: *Nomadacris septemfasciata* adult male (A), gregarious type hopper (B) and solitarious type hopper (C).

2.3.2 *Petamella prosternalis* (Karny, 1907) (Orthoptera: Acrididae: Tropidopolinae)

Petamella prosternalis is a relatively large and slender grasshopper with a fine rugose integument, the body is a uniform tan/brown colour (Dirsh 1965) (Figure 2.4). Apart from species accounts and general descriptions from Dirsh (1965), very little information is available on this species. According to the Orthoptera species file (http://orthoptera.speciesfile.org) its distribution includes much of North Africa, from Senegal in the west to Sudan in the east as well as Mozambique. It also occurs in the Zululand region (Bazelet 2011) and midlands region of KwaZulu- Natal (own observations) therefore it is presumed that this species is fairly widely distributed over much of Africa. Incidental collections in South African sugarcane have occurred since 1994 as documented in the SASRI insect collection and identified by the Agricultural Research Council – Plant Protection Research Institute (ARC-PPRI), Pretoria (SASEX no. 1869). This indicates that this grasshopper has been a sporadic pest of sugarcane in the past. *P. prosternalis* mate and lays their eggs before the onset of the dry winter months (April), the eggs then enter a diapause of up to six months. At the onset of the wet summer months, hoppers emerge and undergo seven moults before becoming adults after about two months.

Figure 2.4: ***Petamella prosternalis*** **adult male (A), brown coloured hopper (B) and green coloured hopper (C). The majority of brown coloured hoppers are males while the majority of green coloured hoppers are female.**

2.3.3 *Ornithacris cyanea* (Stoll 1813) (Orthoptera: Acrididae: Cyrtacanthacridinae)

The bird locust *Ornithacris cyanea* is one of five closely related and morphologically similar species within the genus *Ornithacris* (Dirsh 1965). It is the largest and strongest flying grasshopper found along South Africa’s eastern Grassland region (Rehn 1945) with conspicuous violet-fuchsia coloured hind wings (COPR 1982)(Figure 2.5). Its habitat preferences include woodland and wooded grassland (COPR 1982). *Ornithacris cyanea* has a univoltine life cycle, mating and laying its eggs just before the onset of the wet season. Eggs then hatch after about one month around October/November. Nymphs will moult five or six times before becoming adults after roughly two months. The adults disperse and survive the winter dry period as immature adults (reproductive diapause) until the onset of the summer wet season when they become reproductively active again. The bird locust is known to cause damage on graminaceous and other crops including coffee, cotton, corn and millett (COPR 1982)

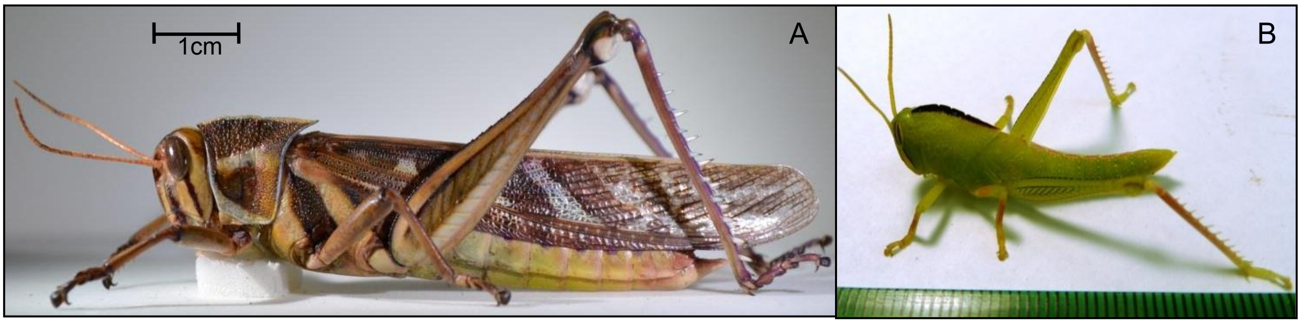

Figure 2.5: ***Ornithacris cyanea*** **adult male (A) and hopper (B).**

2.3.4 *Cataloipus zuluensis* (Sjostedt, 1929) (Orthoptera: Acrididae: Eyeprepocnemidinae)

The Genus *Cataloipus* includes 15 described species which are morphologically very similar. They are widely distributed in central and southern Africa. *Cataloipus zuluensis* is relatively large with long slender femora, dorsally the colouration is generally brown (COPR 1982). Body colour is yellow ventrally (Figure 2.6). A green stripe runs along either side of the pronotum, extending onto the forewings (COPR 1982). The hind tibiae are uniformly blue with black tipped white spines. Owing to the relatively short wings (extending to the end of the abdomen), *Cataloipus zuluensis* is a fairly weak flier(COPR 1982), often crawling deeper into sugarcane stools when disturbed as opposed to flying away like the other species. This species is generally univoltine with overwintering eggs, the hoppers go through 6 instars during the rainy season (Whellan 1975). Damage to crops has been reported widely over Africa on crops such as cotton, maize, sorghum, rice and soya (COPR 1982)

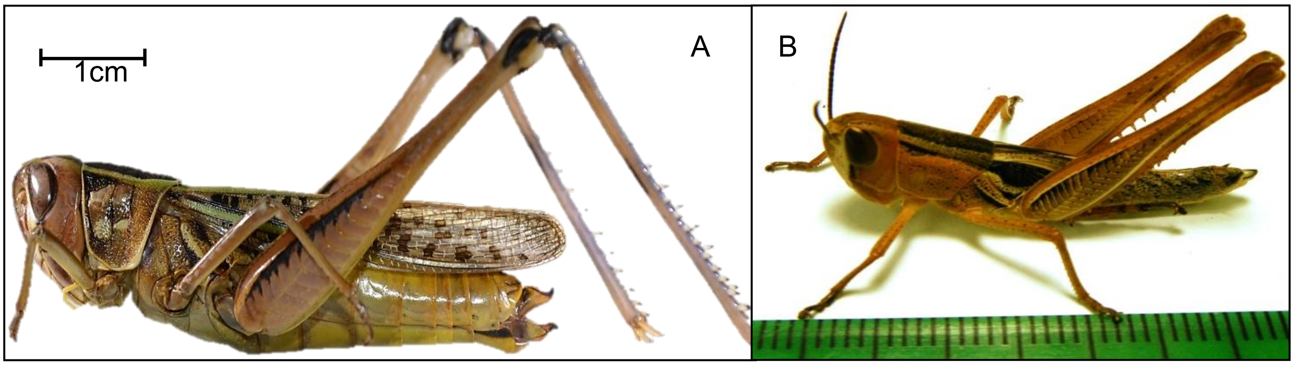

Figure 2.6: *Cataloipus zuluensis* adult female (A) and hopper (B).

2.3.5 *Cyrtacanthacris aeruginosa* (Stoll 1813) (Orthoptera: Acrididae: Cyrtacanthacridinae)

Cyrtacanthacris aeruginosa occurs throughout southern and central Africa (COPR 1982, Picker et al. 2004). It is a large species (body length 55mm) with a stocky appearance. Body colour is predominantly green with a thin white line along the forewings, the hind wings are pale yellow (Figure 2.7). The hind tibiae are bright red with dark tipped white/yellow spines (COPR 1982). They are arboreal and can be found in shrubby vegetation, forest edges as well as grasslands (Picker et al. 2004). It is regarded as a univoltine species. Picker et al. (2004) reports that immature adults survive through winter in a sexually immature state (reproductive diapause) while a previous study suggests that this species has an egg diapause over winter (Jago 1968). Robertson (1958) found this species to lay eggs between March and May. *C. aeruginosa* is regarded as a minor pest of vegetables and tobacco (Picker et al. 2004). Sporadic damage has also been reported in numerous central and southern African countries attacking a wide range of crops including sugarcane (Le Pelley 1959).

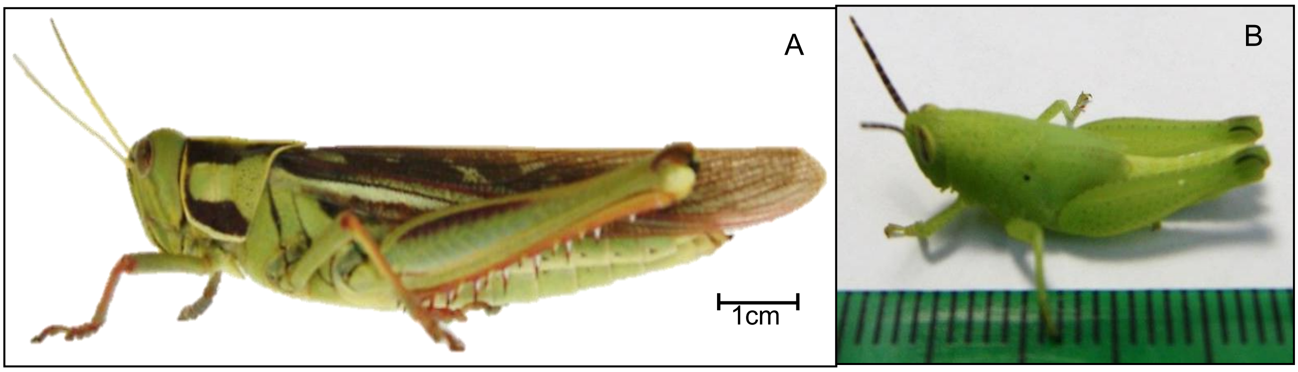

Figure 2.7: *Cyrtacanthacris aeruginosa* adult (A) and hopper (B).

2.3.6 Molecular identification

The results of the molecular identifications indicate that none of the species DNA match the sequences previously loaded onto GenBank or the BOLD websites as indicated by the different species names generated by the databases and the poor % matches of DNA sequences as indicated in table 2.3.

All five specimen sequences were loaded onto the BOLD website and submitted to Genbank. The Genbank accession numbers are as follows:

Nomadacris septemfasciata: BankIt1690897 SASRI1001-13.COI-5P KJ130657

Cyrtacanthacris aeruginosa: Bank1t1690897 SASRI1002-13.COI-5P KJ130656

Petamella prosternalis: Bank1t1690897 SASRI1003-13.COI-5P KJ130659

Cataloipus zuluensis: Bank1t1690897 SASRI1004-13.COI-5P KJ130655

Ornithcris cyanea: Bank1t1690897 SASRI1005-13.COI-5P KJ130658

Table 2.3: Molecular identification of the five major species found in Empangeni sugarcane illustrating poor matches on two different databases.

Sample name	Most significant match in GenBank	% overlap	Most significant match in BOLD	% overlap
Cyrtacanthacris aeruginosa	*Chondracris rosea*	82	*Schistocerca literosa*	89
Nomadacris septemfasciata	*Nomadacris apicerca*	91	*Nomadacris apicerca*	94
Ornithacris cyanea	*Chondracris rosea*	89	*Chondracris rosea*	89
Cataloipus zuluensis	*Diexis varentzovi*	86	*Tharra* spp.	57
Petamella prosternalis	*Melanoplus scudderi*	89	*Cacatua galerita*	57

2.3.7 Identification Keys

Identification (ID) keys were developed for these five species for both life stages using specimens obtained from the field. The purpose of the ID keys was to provide a quick and easy identification guide for growers, extension officers and researchers which would allow them to identify specimens in the field. The adult ID key was based on large and easily distinguishable morphological characters such as forewing length, hind tibia colour, hind wing colour and general body colour of the specimen (Appendix 1). The only morphological characters for hoppers which were fairly easy to distinguish were antennae and overall body colour. These two characters were thus used for the hopper ID key (Appendix 2)

2.3.8 Most abundant species in the complex

Figure 2.8 illustrates the rank of each species in terms of total abundance in all four Empangeni sugarcane sites combined. The log abundance value indicates that *P. prosternalis* had the highest overall abundance over a one year period, followed by *N. septemfasciata*. *Cataloipus zuluensis* is the third most abundant species followed by *C. aeruginosa* and *O. cyanea* at much lower densities.

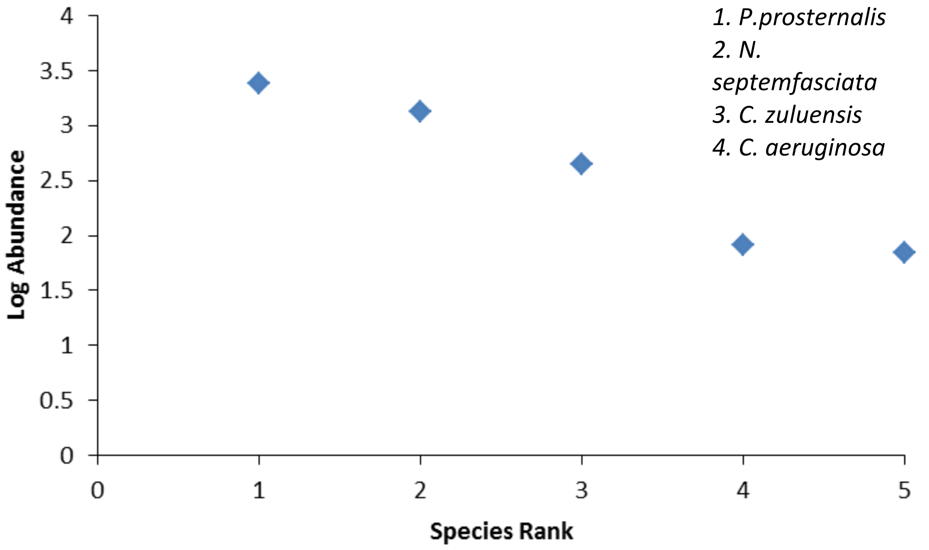

Figure 2.8: Rank abundance plot of the five most prominent Orthopteran species found in Empangeni sugarcane, based on population surveys carried out from May 2012 to May 2013 in four study sites.

2.3.9 Population Surveys and Damage Rating

At the commencement of population surveys in May 2012 *P. prosternalis* was by far the most dominant species (Figure 2.9), the other four species were found at much lower abundance levels. As the year progressed into winter *P. prosternalis* numbers started to decrease and the abundance of *N. septemfasciata* and *O. cyanea* increased in relation to *P. prosternalis.* At the beginning of August only *N. septemfasciata* and *O. cyanea* were still present until October 2012 when the next generation of hoppers emerged in a fairly synchronised manner. Hoppers were present for roughly 3 months until about January 2012. During this period *N. septemfasciata* and *P. prosternalis* were the dominant species while *C. zuluensis* made up roughly 20% of the hoppers surveyed while hoppers of *C. aeruginosa* and *O. cyanea* were found in very low numbers. *P. prosternalis* again was the dominant species from December 2012 to May 2013 when the same trend started to occur as in the previous year. During the period: May 2012-May 2013 the damage rating index fluctuated substantially indicating that damage varies in relation to population density and possibly the season and growth stage of the sugarcane plant. Damage is initially at 1.4 but starts to increase as the season progresses into winter. Damage then starts to taper off and decrease to a level of 1 at roughly the same period when *P. prosternalis* numbers start to decrease. Damage then remains fairly low and constant until mid-November which is possibly due to the hopper populations which would be in their 3rd or 4th instar and increasing body size rapidly as well eating voraciously which explains the increase in damage over this time.

Damage reaches a peak at the end of January 2013 which is the time period where grasshopper population density is the greatest, most individuals have undergone their final moult to become adults and the effects of natural mortality over time are small.

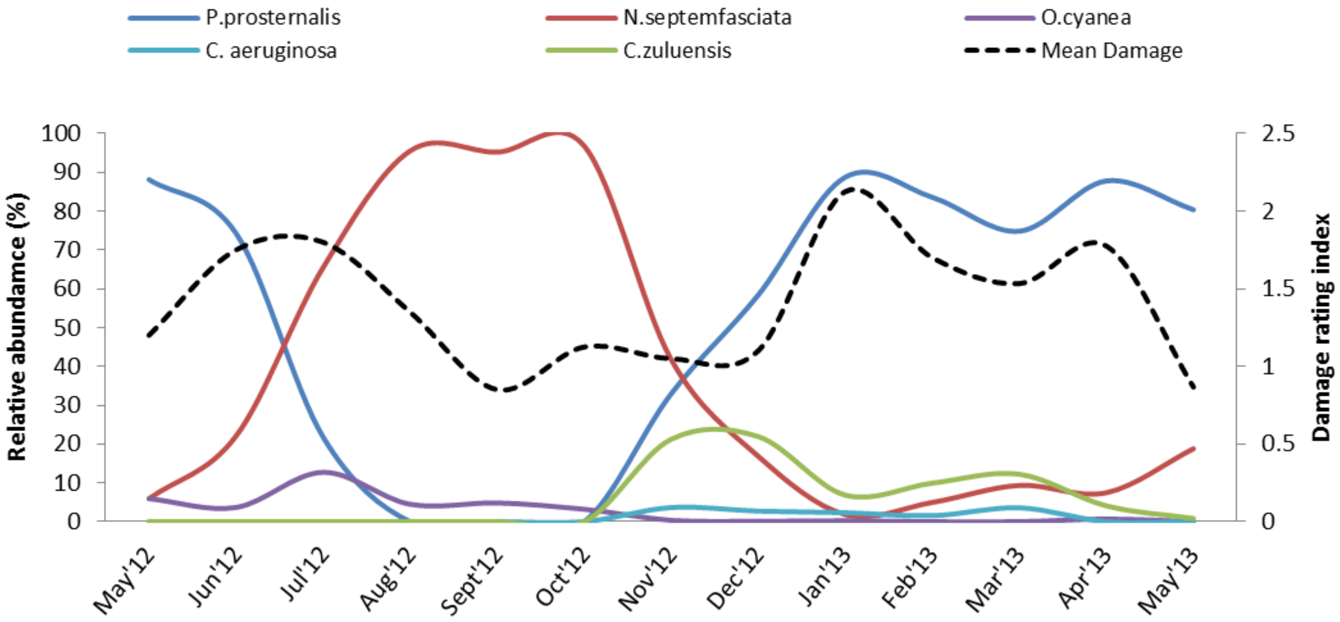

Figure 2.9: Population survey showing the relative abundance of the five most prominent acridid species in Empangeni sugarcane in relation to the damage rating index on the secondary y axis.

The damage rating index was significantly correlated with the fluctuations of *P. prosternalis*, and *N. septemfasciata* indicating the close relationship between *Petamella prosternalis* population density and damage to sugarcane (Figure 2.9 & Table 2.4).

Table 2.4: Relationship between acridid species abundance and damage rating in the study areas. Gamma statistic marked with a * are significant at P<0.05 showing that *P. prosternalis* and *N. septemfasciata* population densities are significantly correlated with observed damage.

Species	Damage rating
P. prosternalis	0.429143*
N. septemfasciata	0.250408*
O. cyanea	0.111739
C. aeruginosa	-0.190004
C. zuluensis	0.152348

2.3.10 Life cycles

Population surveys and laboratory observations were generally consistent with previous literature which indicated that the five main species in Empangeni sugarcane are all univoltine, meaning that they complete one life cycle per year. All species have an overwintering or diapause period although the life stage which enters into diapause differs between the species (Table 2.5). *Nomadacris septemfasciata* and *O. cyanea* overwinter in the adult stage. The immature adults enter a reproductive diapause at the onset of the dry season only becoming reproductively active five or six months later at the onset of the rainy season again (October). *Petamella prosternalis*, *C. aeruginosa* and *C. zuluensis* exhibit a different overwintering strategy whereby the adults mate and lay their eggs before the onset of the dry season in April or May. The eggs lie dormant in the soil for up to 7 months over winter until the rains arrive towards October or November (Table 2.5)

Table 2.5: Simplified summary of the two life cycle types present in the complex of grasshoppers attacking Empangeni sugarcane (Findings from population surveys and laboratory studies).

	Life cycle type	
Month	Egg diapause type: *P. prosternalis, C. aeruginosa, C. zuluensis*	Reproductive diapause type: *N. septemfasciata, O. cyanea*
January	Hoppers	Hoppers
February	Hoppers	Hoppers
March	Mating/oviposition	Hoppers
April	Mating/oviposition	Reproductive diapause
May	Mating/oviposition	Reproductive diapause
June	Egg diapause	Reproductive diapause
July	Egg diapause	Reproductive diapause
August	Egg diapause	Reproductive diapause
September	Egg diapause	Reproductive diapause
October	Egg diapause	Mating/oviposition
November	Hoppers	Mating/oviposition/Hoppers
December	Hoppers	Oviposition/Hoppers

2.3.11 Species composition

Figure 2.10 illustrates the mean abundance of six species found at the four sugarcane study sites and the two grassland study sites. The graph shows that species abundance, diversity and composition differ substantially between the six study sites. *Nomadacris septemfasciata* was most abundant on GSA and Crystal Holdings. Very few *N. septemfasciata* individuals were recorded in grassland sites. *P. prosternalis* was most abundant on GSA and Jengro during this period while in grassland sites, very few individuals were recorded. *Orthocta* species had much higher abundance levels in the grassland sites compared to sugarcane. *Cataloipus zuluensis* was most abundant on GSA and Jengro but an appreciable number were also recorded in the two grassland sites. *Zoncerus elegans* was almost exclusively found in grassland sites indicating that it certainly prefers natural veld over sugarcane as a host plant. *Cyrtacanthacris aeruginosa* was recorded in low numbers everywhere except one of the grassland sites were high numbers were seen.

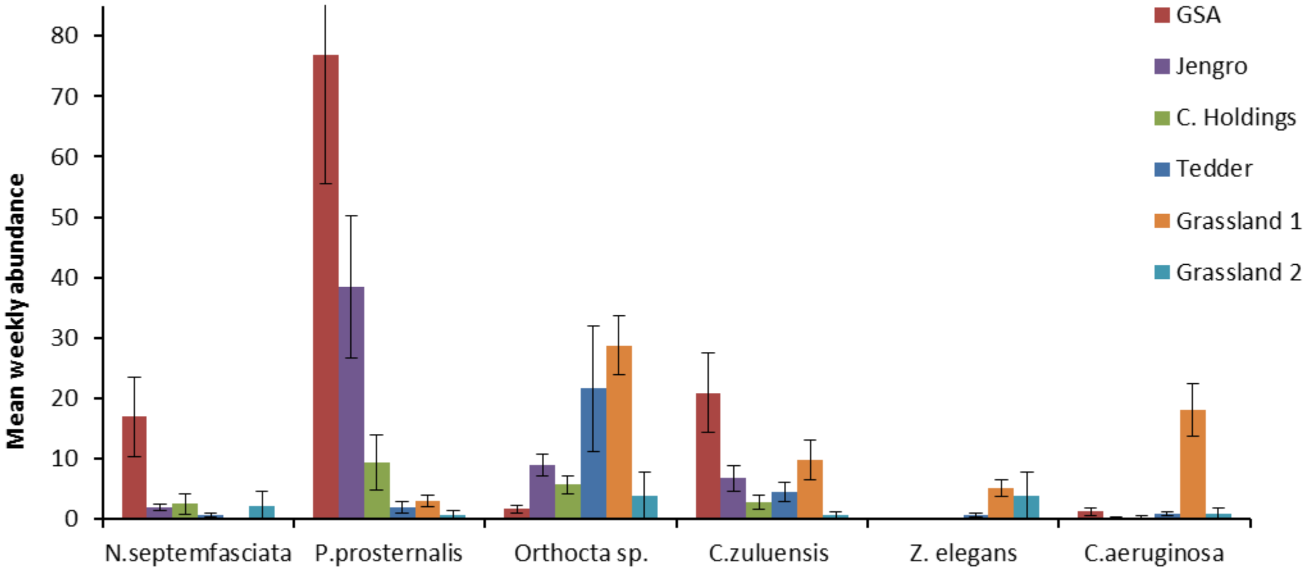

Figure 2.10: Mean weekly abundance (± SE) of six species of grasshoppers surveyed at the four sugarcane sites and the two grassland sites for the period: 21 November 2012 – 19 February 2013.

2.3.12 Correspondence analysis

Figure 2.11 illustrates the association between the four sugarcane sites and species abundance. Dimension 1 accounted for 83.91% of the variation in the graph, indicating a strong association between Jengro and *P. prosternalis* as they are closely situated to each other on the positive side of the axis, at a high order of magnitude.

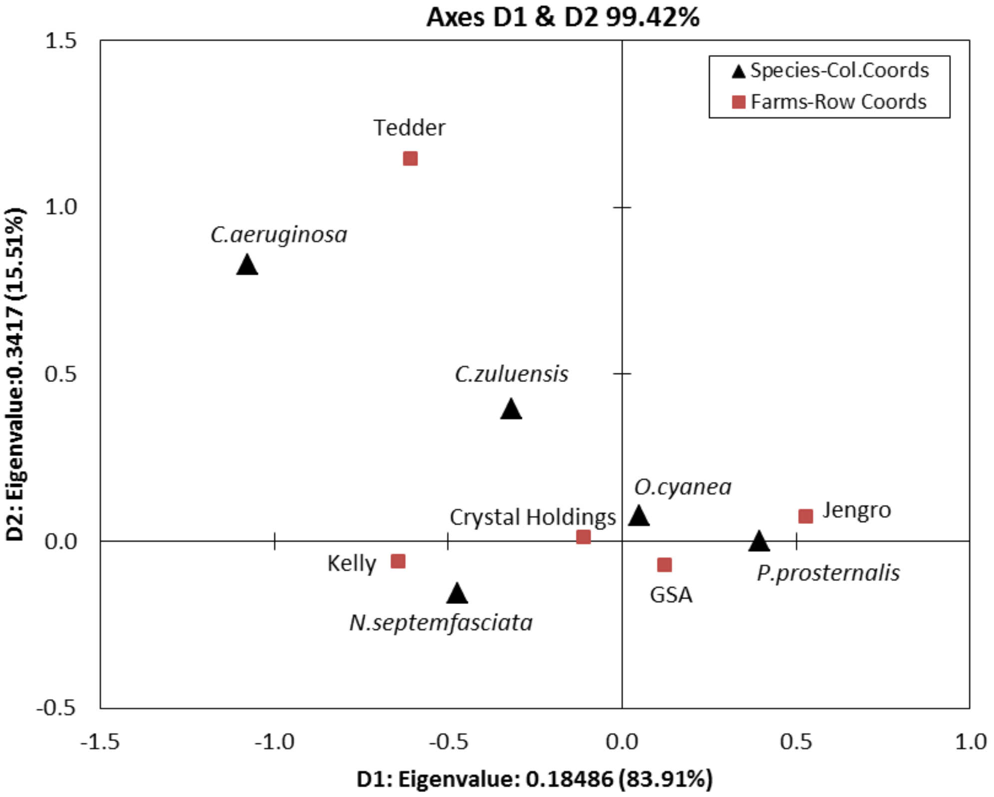

Figure 2.11: Correspondence analysis illustrating the association between sugarcane study sites and acridid species abundance for the period: 21 November 2012 – 19 February 2013.

Crystal Holdings and GSA are both found close to the point of origin meaning that they do not account for much variation in the data and therefore were not closely associated with one particular species. Kelly farm was most closely associated with *N. septemfasciata*, at a fairly high level of magnitude which agrees with what was observed in the field: swarms of *N. septemfasciata* were present on Kelly farm for about 7-8 months of the year. *Petamella prosternalis* was most closely associated with GSA and Jengro farms. *Ornithacris cyanea* was about equally associated with GSA and Crystal holdings. Tedder farm was somewhat of an outlier with a high, positive order of magnitude in top left quadrat, this farm being associated with *C. aeruginosa* and *C. zuluensis*.

Zonocerus elegans, Orthocta sp. and *C. aeruginosa* were closely associated with grassland sites which are reliable as dimension 1 accounts for 46.62% of the inertia (Figure 2.12). Dimension 2 (whilst only capturing 23.82% of inertia indicates that *N. septemfasciata* is closely associated with sugarcane sites at a high order of magnitude and similarly *P. prosternalis* shows a strong association with sugarcane sites but at a low order of magnitude. *C. zuluensis* seemed to be more closely associated with sugarcane sites. However this species also shows an appreciable association with the grassland sites but *C. zuluensis is* fairly close to the point of origin and therefore these associations are not as strong as the other species- site associations.

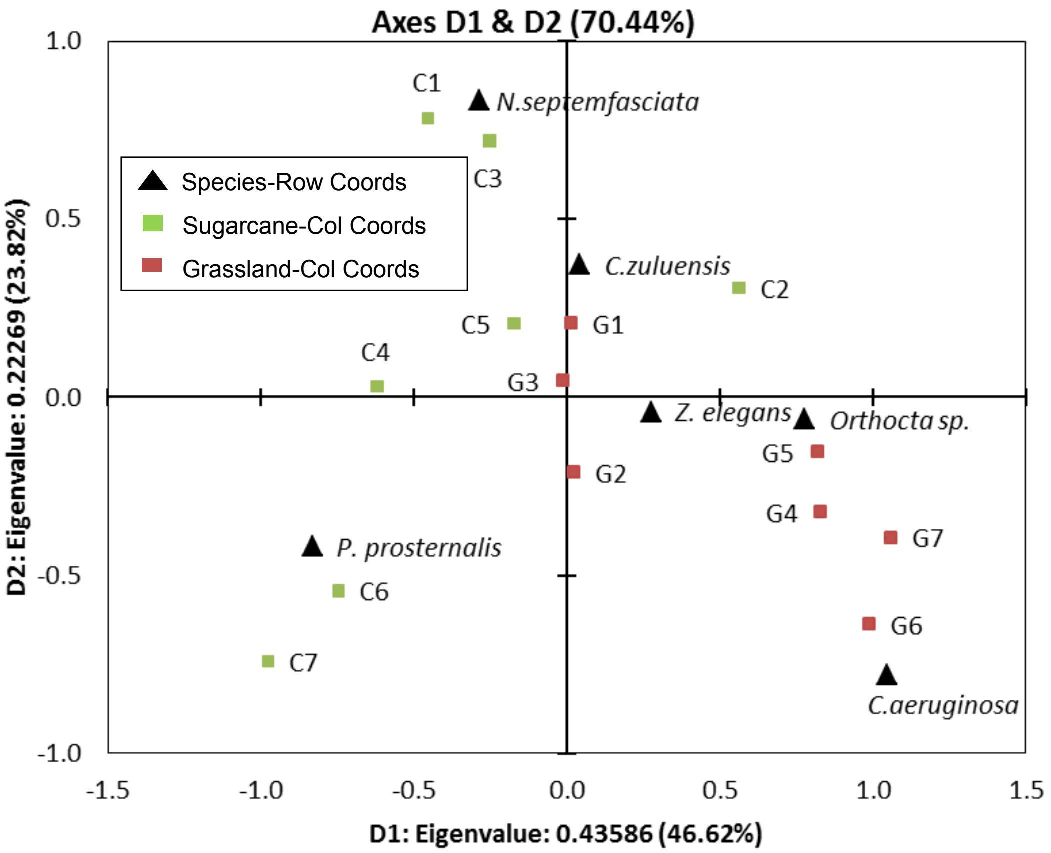

Figure 2.12: Correspondence analysis showing the association between grasshopper species in relation to sugarcane and grassland survey sites sampled over a seven week period from 21 November 2012 – 19 February 2013.

2.4 Discussion

2.4.1 Rank abundance

Overall, *P. prosternalis* was the most abundant species in Empangeni sugarcane, although *N. septemfasciata* was recorded at very high densities. Their extremely mobile nature of the latter species (Faure 1935) and particularly clumpy distribution (Rainey et al. 1957) meant that sampling may have underestimated their abundance in relation to the former species. The other species were more evenly distributed over the sampling area and generally easier to catch during the drive netting period of sampling. *Petamella prosternalis* and *N. septemfasciata* therefore, are the most important species in terms of the threat posed to sugarcane due to the sheer numbers observed in sugarcane. The other three noteworthy species are found at much lower abundance levels and consequently are of less importance in terms of the threat they pose as sugarcane pests. Acridid species always occurred as a species assemblage, it was never observed that only one species occurred in a particular area although species density variation was readily evident.

2.4.2 Population dynamics and related damage

Understanding the population dynamics of pest species is an important part of developing knowledge based, integrated pest management strategy (Luckman & Metcalf 1994). The population dynamics of *N. septemfasciata* observed in Empangeni are quite comparable with the findings of Lecoq et al. (2006, 2011) in Madagascar. Both of these studies define three different geographical areas where *N. septemfasciata* populations migrate to and from as a result of seasonal changes. The biological cycles observed in each of these designated areas which best matched the biological cycle observed in Empangeni is termed the ‘Main rainy season breeding zone’ which therefore indicates that Empangeni is certainly a *N. septemfasciata* breeding area but the difference being that in Madagascar long range migrations have been proven to occur while in Empangeni, this has not been proven. The likelihood of significant migrations occurring in the Zululand region however, are slim given the fact that reports of locust infestations have been concentrated around Empangeni and population surveys have shown that adults remain in the vicinity throughout the dry winter period. These findings do however, disprove the statement by Faure (1935) that “the Union (South Africa) does not serve as a permanent breeding ground of *N. septemfasciata* in its solitary phase”.

Information pertaining to *O. cyanea* has been largely limited to taxonomic studies, however Bazelet (2011), who worked in natural veld sites in the Zululand region, states that this univoltine species mates, lays eggs and dies before the onset of cold, dry weather in winter. The findings of this study, however, showed individuals of this species being caught throughout Overall, *P. prosternalis* was the most abundant species in Empangeni sugarcane, although *N. septemfasciata* was recorded at very high densities. Their extremely mobile nature of the latter species (Faure 1935) and particularly clumpy distribution (Rainey et al. 1957) meant that sampling may have underestimated their abundance in relation to the former species. The other species were more evenly distributed over the sampling area and generally easier to catch during the drive netting period of sampling. *Petamella prosternalis* and *N. septemfasciata* therefore, are the most important species in terms of the threat posed to sugarcane due to the sheer numbers observed in sugarcane. The other three noteworthy species are found at much lower abundance levels and consequently are of less importance in terms of the threat they pose as sugarcane pests. Acridid species always occurred as a species assemblage, it was never observed that only one species occurred in a particular area although species density variation was readily evident.

before winter which refutes the statement by Picker et al. (2004) who reports that (reproductively) immature adults survive through winter in a sexually immature state (reproductive diapause), but corroborates the findings of Jago (1968), which suggests that this species has an egg diapause over winter. It can be assumed that the biological cycle of all five species will be fairly constant regardless of external factors. Lecoq et al. (2011) found that during an eight year study, the biological cycle of *N. septemfasciata* repeated with great regularity. Although the diapause cycles of all species generally coincide with the change in seasons and onset of summer rains – thus causing early researchers to presume rainfall was the main factor involved, (see: Robertson 1958, Franc & Luong-Skovmand 2009) it has recently been found that photoperiod is the factor which is responsible for initiation and cessation of diapause(Lecoq et al. 2011). This finding could possibly explain why initiation and cessation of diapause is earlier in Empangeni *N. septemfasciata* populations compared to Madagascar populations due to the differences in latitude which affects photoperiod.

Damage to sugarcane was most closely and significantly correlated with *P. prosternalis* population fluctuations which suggest that this species is responsible for the majority of the damage observed on Empangeni sugarcane due to acridids. During feeding trails (Chapter 3), *P. prosternalis* also showed a higher feeding potential when compared to *N. septemfasciata*. These results therefore suggest that the combination of this species' relatively high population densities and high feeding potential means that it currently poses the biggest threat to Empangeni sugarcane (not taking into account the potential of *N. septemfasciata* to swarm). A shortcoming of the damage rating index is that is does not take into account the growth rate of the plant being analysed over time. Three of the four survey sites were dry- land sugarcane farms therefore a decrease in rainfall over winter may slow down plant recovery after feeding and therefore cause damage to be overestimated during winter months.

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Information pertaining to *O. cyanea* has been largely limited to taxonomic studies, however Bazelet (2011), winter proving that it has a similar biological cycle to *N. septemfasciata*, which is a closely related species in the sub family Cyrtacanthacridinae. Another closely related species within this sub family is *C. aeruginosa.* According to this study *C. aeruginosa* had a different biological cycle compared to its two closely related species as it was found to mate and lay its eggs and die before winter which refutes the statement by Picker et al. (2004) who reports that (reproductively) immature adults survive through winter in a sexually immature state (reproductive diapause), but corroborates the findings of Jago (1968), which suggests that this species has an egg diapause over winter It can be assumed that the biological cycle of all five species will be fairly constant regardless of external factors. Lecoq et al. (2011) found that during an eight year study, the biological cycle of *N. septemfasciata* repeated with great regularity. Although the diapause cycles of all species generally coincide with the change in seasons and onset of summer rains – thus causing early researchers to presume rainfall was the main factor involved, (see: Robertson 1958, Franc & Luong-Skovmand 2009) it has recently been found that photoperiod is the factor which is responsible for initiation and cessation of diapause(Lecoq et al. 2011). This finding could possibly explain why initiation and cessation of diapause is earlier in Empangeni *N. septemfasciata* populations compared to Madagascar populations due to the differences in latitude which affects photoperiod.

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2.4.3 Species composition in relation to sugarcane and grassland study sites

Acridid species exhibit a wide preference range in host plant choice (Behmer & Joern 2008) and microclimate choice (Gardiner & Hassall 2009) like many other insect herbivores. These statements also hold true for acridid species in Empangeni where it was shown that some species prefer sugarcane crops while others show a preference for natural grasslands. *Nomadacris septemfasciata* and *P. prosternalis* showed a clear preference for sugarcane sites. In a study by Bazelet (2011) in natural grasslands in the Zululand region, over two years, no *N. septemfasciata* and only 22 individuals of *P. prosternalis* were caught in those study sites. This indicates that these two species possibly prefer sugarcane to grasslands in this area as a habitat. Michelmore (1947) in the Rukwa Valley, Tanzania found that hoppers of *N. septemfasciata* showed a marked preference for tall dense grass. Lea and Webb (1939) also found that *N. septemfasciata* hoppers, when disturbed in short grass would immediately make for the shelter of tall clumps of grass. In the Rukwa valley, adult *N. septemfasciata* were practically completely restricted to tall grass (Burnett 1951). Generally, sugarcane gets much taller than surrounding natural grasslands, especially over winter therefore these two findings could explain why *N. septemfasciata* hoppers prefer tall sugarcane over shorter grassland areas. *Orthocta sp., Z.elegans* and to a degree, *C. aeruginosa* preferred grassland sites to sugarcane sites, however, Bazelet (2011) recorded no *Orthocta sp.,* one *C. aeruginosa* and two *Z.elegans* individuals' during her study. *Cataloipus zuluensis* acted like a generalist feeder, as it was recorded in grassland and sugarcane in roughly equal abundances.

Nomadacris septemfasciata is capable, over time to cover distances of over 1000 miles or more (Rainey et al. 1957) but currently all species including *N. septemfasciata* are confined to relatively small areas in Empangeni. Sugarcane may be the factor causing *N. septemfasciata* populations to remain quite sedentary contrary to their mobile nature (Lea 1935). Grasshoppers will always remain pests of local importance and currently it seems *N. septemfasciata* may remain a pest of local importance but the potential for population upsurges resulting in significant outbreaks should not be underestimated.

2.5 Conclusion

Species identification and population surveys are the first steps in trying to develop an integrated pest management plan. Population surveys have shown that *P. prosternalis* is the most abundant species in Empangeni sugarcane followed by *N. septemfasciata*. These two species are our main suspects in terms of the potential damage they can cause and should be considered as the primary targets for any type of control which may be implemented. The other three species; *C. zuluensis*, *C. aeruginosa* and *O. cyanea* were found at lower numbers, but should not be ignored as their potential for population increase is not well known but certainly possible. All species studied are univoltine which meant that in order to correlate population fluctuations with weather variables as done in previous literature, (see: Chiconela et al. 2003) surveys will have to be completed over a longer period of time in order to determine the effects of climate on acridid populations. The findings also show that the distribution of species among farms and veld areas is not uniform, some species being found at higher densities on certain farms and similarly some species were found to have higher densities in grassland sites compared to sugarcane plots. *Nomadacris septemfasciata*, *P. prosternalis* and *O. cyanea* were found in higher densities in sugarcane sites while *C. aeruginosa* was more common in grassland sites.

Cataloipus zuluensis was found in sugarcane and grassland at roughly equal density levels. These findings provide important information for managers and growers verifying which species are of particular concern for them and therefore they are able to develop a more species-specific management plan accordingly.

These findings indicate which species prefer sugarcane over natural veld which is very important in terms of management as it gives one a better understanding of the distribution and abundance of different species and indicates which the priority species in terms of control are. Species such as *Z.elegans, C. aeruginosa* and *Orthocta* sp. are not a major threat and therefore controlling them is not necessary. If appreciable numbers are found in sugarcane (which has been the case previously), they either move out of cane into veld areas or die off to such an extent that they do not require control measures.

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Chapter 3: The feeding potential of the red locust *Nomadacris septemfasciata* (Serville) and *Petamella prosternalis* (Karny) on sugarcane

3.1 Introduction

Exotic crops are highly vulnerable to attack by local insect pests (Williams et al. 1969, Pimentel 1986), and sugarcane, grown extensively in KwaZulu-Natal is no exception (Bakker 1999, Ramashala 2012). There are many pests which attack sugarcane, but grasshoppers are generally not regarded as a pest of serious economic importance as evidenced by the little attention they receive in comparison to other pests of sugarcane, (Williams et al. 1969, Bakker 1999, Ramashala 2012). Regionally, they are a pest of minor importance but locally, they are able to cause extensive damage, (Bakker 1999, Fortmann 2012 pers. comm.). Grasshoppers feed on the leaves of sugarcane, thereby affecting the photosynthetic capability of the plant (Easwaramoorthy et al. 1989). When infestations are severe, defoliation may be so severe that only the mid-rib of the plant is left (Figure 3.1).

Outbreaks of *Petamella prosternalis* (Karny) (Orthoptera: Acrididae: Tropidopolinae) have occurred at least since 1994 in the Zululand region as documented by the insect collection in the entomology department of SASRI. However, due to the fact that *P. prosternalis* is not regarded as one of the plague locusts and the sporadic nature of its outbreaks, little attention has been given to this species. Recent population upsurges in Empangeni seem to be much larger in terms of density, extent and persistence (Chapter 2), and thus have prompted more attention being given to it as a potential crop spoiler.

The red locust *Nomadacris septemfasciata* Serville (Orthoptera: Acrididae: Cyrtacanthacridinae) is regarded as a major crop and pasture pest in central and southern Africa (FAO 1967, COPR 1982) and has eight recognised outbreak areas in Tanzania, Malawi, Zambia and Mozambique (Bahana 1999). Two major outbreaks have affected South Africa, the last one starting in 1929 and lasting until 1944 (Morant 1947). In 1934 it was estimated that there was a 300 000 ton or 25 % reduction in output of sugarcane in the Zululand region as a result these massive plagues originating from the north (De V Minnaar 1990). Dymond (1934) estimated a decrease in sugarcane yield of 33% as a result of *N. septemfasciata* swarms attacking sugarcane estate in Mozambique. It is thus evident that this species rarely affects South Africa on a regional scale but when it does, it is a pest of tremendous importance (Faure 1935).

The effect these two acridid species have on sugarcane yield is not well known. The extent of damage will depend on the age of the sugarcane when defoliation occurred (Gordon 1935), variety attacked (Dymond 1934, Tothill 1940) and the number of defoliation incidents (Anonymous 1955, Bakker 1999). The measurement of yield loss due to acridid damage is difficult because valid comparisons between damaged and undamaged cane are usually impossible (Williams et al. 1969). As the sugarcane needs time to re-establish its

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canopy, growth over a given period of time will be limited. In addition to these losses in yield, the purity of the cane juice may well be lowered than otherwise would have been the case (Bakker 1999). Artificial defoliation experiments in South Africa have also shown that cane quality (recoverable sugar per ton) could also be negatively affected by locusts (Williams et al. 1969). There is thus a need to investigate the effects of grasshopper feeding on sugarcane and their economic injury level should be determined as done with other pests in South African sugarcane (see: Leslie 2009). Economic-injury level (EIL) is defined as the 'lowest population density of a pest that will cause economic damage' and economic threshold (ET) is the 'the amount of injury that will justify artificial control measures' (Stern et al. 1959). The first step in determining these levels is to perform feeding trials. Feeding trials should form the basis of crop loss assessments and in estimating economic thresholds (Coop & Croft 1993).

Figure 3.1: Damage to sugarcane as a result of acridid feeding in Empangeni, KwaZulu-Natal, showing that only the mid-rib is left on some leaves. Damage is exacerbated by the fact that leaves are often cut down during feeding without actually being consumed.

Determining economic threshold levels i.e. when to take action against a pest is dependent upon a number of factors such as acridid density, crop productivity, climatic factors, area affected and the efficacy of treatment options (Davis et al. 1992). A fair amount of research has been conducted on the economic threshold of grasshoppers in rangelands and natural grasslands (Davis et al. 1992, Hewitt & Onsager 1982) but information pertaining specifically to food crops seems less prevalent. **T**he U.S. Department of Agriculture's Animal and Plant Health Inspection Service (APHIS) uses a threshold value of eight grasshoppers per square yard as guideline for control operations (Davis et al. 1992). Legg and Lockwood (1995) suggest that control measures are rarely justified at grasshopper densities less than 27 per m^2. Both of these studies dealt with grasshopper pests in rangelands of the USA, therefore their applicability in the current context are questionable. In a more applicable study, Gordon (1935) investigated the damage done by Oriental migratory locust *Locusta migratoria manilensis* (Meyen) Orthoptera: Acrididae: Oedipodinae) to sugarcane in the Philippines and found mean leaf damage of 13.3% cause by this species.

In an attempt to obtain similar information, feeding trials were conducted with *P. prosternalis* and *N. septemfasciata* collected from the Empangeni area, to calculate their feeding potential over a specified time, and to relate this to their respective population densities. The ultimate aim was to estimate the economic injury level for these pests from this data. This will help to make more accurate and effective decisions when the need to control them arises. The FAO (2009) estimates that a *N. septemfasciata* adult can consume roughly its own weight, about two grams, in fresh food in 24 hours and further predicts that a very small part of an average swarm (or about one ton of locusts) eats the same amount of food in one day as around 2 500 people. Clearly, there is a need to understand the effects of grasshopper and locust feeding on Empangeni sugarcane to avoid relying on ambiguous statements such as the one above. This study attempts to provide baseline feeding data on the two most common sugarcane acridid species which may aid determining their impact on Empangeni sugarcane and in making informed management decisions in the future.

3.2 Materials and Methods

Two acridid species; *N. septemfasciata* and *P. prosternalis* were selected for trials to ascertain their feeding potential in relation to sugarcane. These species were chosen because they are the most abundant in the species complex of the region (Chapter 2). It was thus presumed that they would be responsible for the most damage to sugarcane. Two methods of assessing feeding damage were completed. The first was completed in the laboratory and the second in the field.

3.2.1 Laboratory studies

Feeding trials took place in the Insect Rearing Unit, South African Sugarcane Research Institute (SASRI) in controlled environment rooms at 25.5^0C (±1.5^0C) at a relative humidity of 65-70%. Fourth and fifth instar hoppers of both species were obtained from field populations in Empangeni (28^o44’56.74’’S 31^o53’59.24’’E), each species was placed in separate 30cm x 30cm x 30cm Bugdorm® (UK) insect rearing cages and kept according to the lab conditions described in Chapter 1. One trial per week was conducted from 22 January to 21 May 2013 adding up to a total of 20 trials. Additional specimens were collected when necessary to account for mortality during the trial period (mortality was minimal). The first four trials were completed with all specimens still in the hopper stage, after which the final ecdysis into adults started to take place.

Trial procedure

After being starved from the previous evening (±15 hours) for logistic reasons, 16 individuals (eight male and eight female) of both species were taken from their communal cages and placed in separate clear plastic containers with perforated lids (height: 18cm, diameter: 8.5cm) and allowed to feed on sugarcane leaf pieces for seven hours. Once the trial was completed they were placed back into their separate communal cages. Sugarcane leaves used in the trials were obtained from field 13B at the SASRI experimental farm, Mt Edgecombe (29°42'22.77"S 31° 2'47.31"E). The variety used was NCo376 and it was seven month old plant sugarcane at the start of the trial. For each trial, five green leaves from three separate stools, were removed at the stem and cut into 15cm lengths. Care was taken to use the basal section of the first fully expanded leaves below the spindle leaves to ensure that cut lengths were equivalent as possible. These lengths were then measured using a LI-COR® LI-3100C Area Meter to obtain their total leaf area before feeding. They were then placed into the separate plastic containers with each individual used in the trial. Each container contained 50cm^2 of moist building sand in its base to prevent desiccation of the leaf during the trial. After seven hours the leaves were removed and re-measured with the area meter to obtain the percentage leaf area injured per specimen. This procedure made up one trial, a total of 20 trials were completed at the rate of one per week.

Calculation of conversion formulas

To develop a conversion formula for converting leaf area (LA) injured/eaten to leaf weight injured, the method by Coop & Croft (1993) was used to convert leaf area to leaf weight. 15 leaves from five different stools were selected at random from the same block of cane used in the feeding trials. 50 pieces of 10cm^2 (5cmx2cm) cane leaf were randomly cut out of these leaves and immediately weighed using a METTLER TOLEDO ML54 analytical balance to obtain a specific leaf weight (SLW) for a 10cm^2 piece of fresh cane leaf, converted to an SLW for 1cm^2 of cane leaf in grams/centimetre^2 for the purposes of our calculations. In order to convert feeding potential per seven hour trial, to feeding potential per day, the results were multiplied by 3.43 (24hours÷7hours). It was presumed that, at constant temperature, the feeding rate of individuals in the trial be constant throughout a 24 hour day. This assumption was made by referring to previous literature which states that *N. septemfasciata* eats just as voraciously at night compared to the day (Lea 1935, Burnett 1951, de V Minnaar 1990). This was necessary in order obtain a standardised feeding damage value in grams per day.

Weighing of grasshopper specimens

Weighing of specimens was completed in order to establish feeding potential in relation to body weight. 60 *P. prosternalis* and 60 *N. septemfasciata* adult specimens (30 male and 30 female of each species) collected from the field in May 2013 were starved for five hours to purge them of their gut contents. Specimens were then killed by freezing and placed in a drying oven at 60^{o}C for 48 hours. A METTLER TOLEDO ML54 analytical balance was used for weighing in order to obtain a mean (±SE) specimen weight in terms of species, life stage and gender (Table 3.2).

Data analysis

For figure 3.4 and 3.5, feeding trial data were averaged over the entire trial period for each respective factor used in the analysis. Data analyses were performed using Genstat 15.0 (VSN International LTD, UK). Data were tested for normality of frequency distribution using the Shapiro-Wilks test. All data used were found to be homoscedastic, therefore the interaction of the two species in terms of gender and life stage was evaluated using univariate General Linear Model (GLM) analysis of variance (ANOVA). Where the interactions were found to be significant, a Turkey's post-hoc test was performed to separate the means.

3.2.2 Field Studies

Damage rating index

The level of feeding damage on sugarcane as a result of the acridid complex was observed in the study areas once a week during population surveys (see: materials and methods in Chapter 2). This was done by randomly selecting five sugarcane stools during survey transects for observation. From these observations, a damage rating index was generated by allocating a rating of 1-5 to each stool, depending on the severity of the observed damage. The damage rating was calculated by estimating the percentage leaf area lost on the top five green leaves, below the still furled spindle leaves of a randomly selected stalk in the stool. The five estimates per transect were then averaged to obtain a mean weekly damage rating which was averaged again to obtain a mean monthly damage rating per farm (Figure 3.2). Relative abundance data for *N. septemfasciata* and *P. prosternalis* was then plotted on the same graph as a secondary axis in order to illustrate the correlation between feeding damage and population density. The criteria used to estimate damage, resulting in a damage rating index is displayed in Table 3.1. For the purposes of this study, damage rating data from Jengro farm(28°37'30.84"S 32° 0'52.68"E), GSA farm(28°40'54.94"S 31°54'51.98"E) and Kelly farm(28°41'29.75"S, 31°54'26.92"E) was only included as damage on the other two surveyed farms was minimal.

Table 3.1: Criteria used as a guideline to assess damage in order to obtain a damage rating index to correlate against population abundance data.

Rating	% Damage estimate
0/5:	0%
1/5:	1-20%
2/5:	21-40%
3/5:	41-60%
4/5:	61-80%
5/5:	81-100%

3.3 Results

3.3.1 Field studies

The damage rating index illustrated the extent of spatial and temporal damage to sugarcane on the three affected farms in relation to relative abundance of *N. septemfasciata* and *P. prosternalis* (Figure 3.2). Overall mean damage rating from feeding over the entire period was 2.063 which according to Table 3.1, equates to a mean observed damage of between 21-40%.

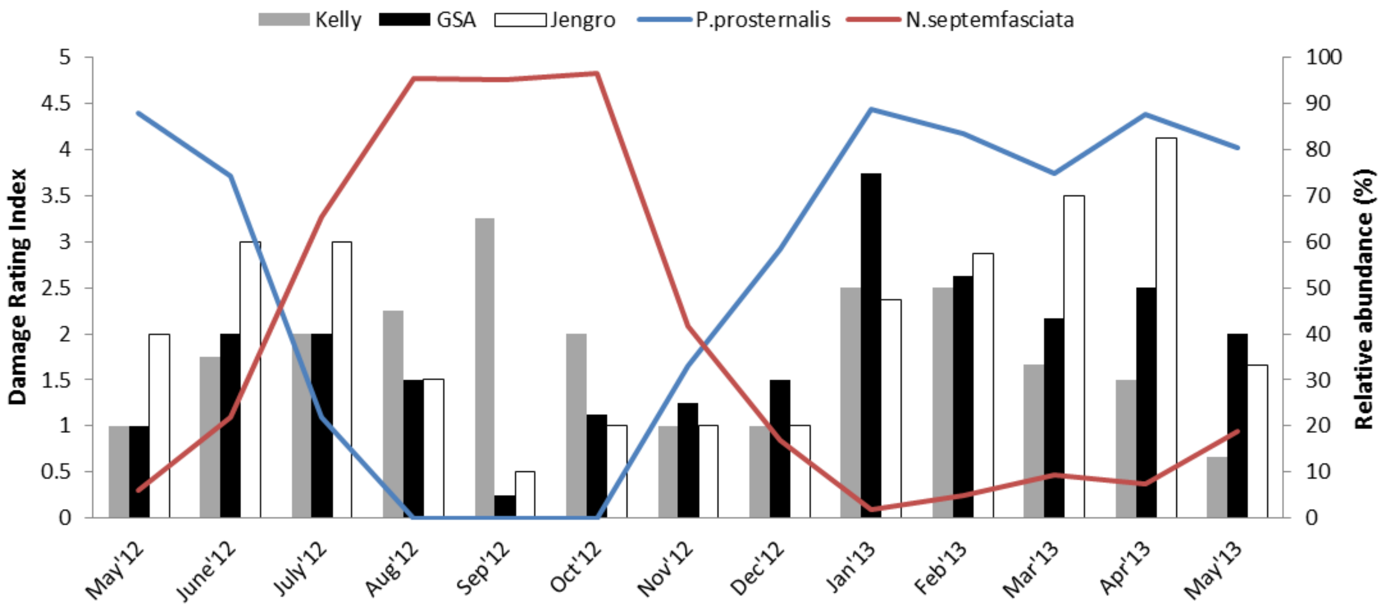

Figure 3.2: Mean monthly damage expressed as a damage rating index at three study sites and relative abundance of *Nomadacris septemfasciata* and *Petamella prosternalis* on the secondary axis, from May 2012 to May 2013

3.3.2 Laboratory studies

Feeding damage fluctuated quite substantially during the trial period (Figure 3.3). An overall mean of 16.29% leaf damage was calculated for both species combined. *Petamella prosternalis* had a higher overall mean feeding damage value (19.7%) compared to *N. septemfasciata* (13.37%).

The first five trials (22 January – 12 February) were completed while specimens were in the hopper stage, after which all specimens had moulted to become adults. Initially, *N. septemfasciata* hoppers had a higher feeding potential than *P. prosternalis* hoppers, but this quickly changed after the second trial (22 January – 28 January) after which *P. prosternalis* usually exhibited higher feeding damage per trial. At the beginning of February, hoppers start to undergo their last moult to become adults and *P. prosternalis* adults continue have a higher feeding potential than *N. septemfasciata* adults. In mid-March the difference in feeding rate between the two species increases further which can be ascribed to the fact that *P. prosternalis* starts to become reproductively active at this point in time (see Table 2.5 in chapter 2), which may require them to consume more forage (Chapman & Joern 1990). At the same time *N. septemfasciata* starts to enter a winter reproductive diapause (see Table 2.5 in chapter 2) which may result in decreased feeding.

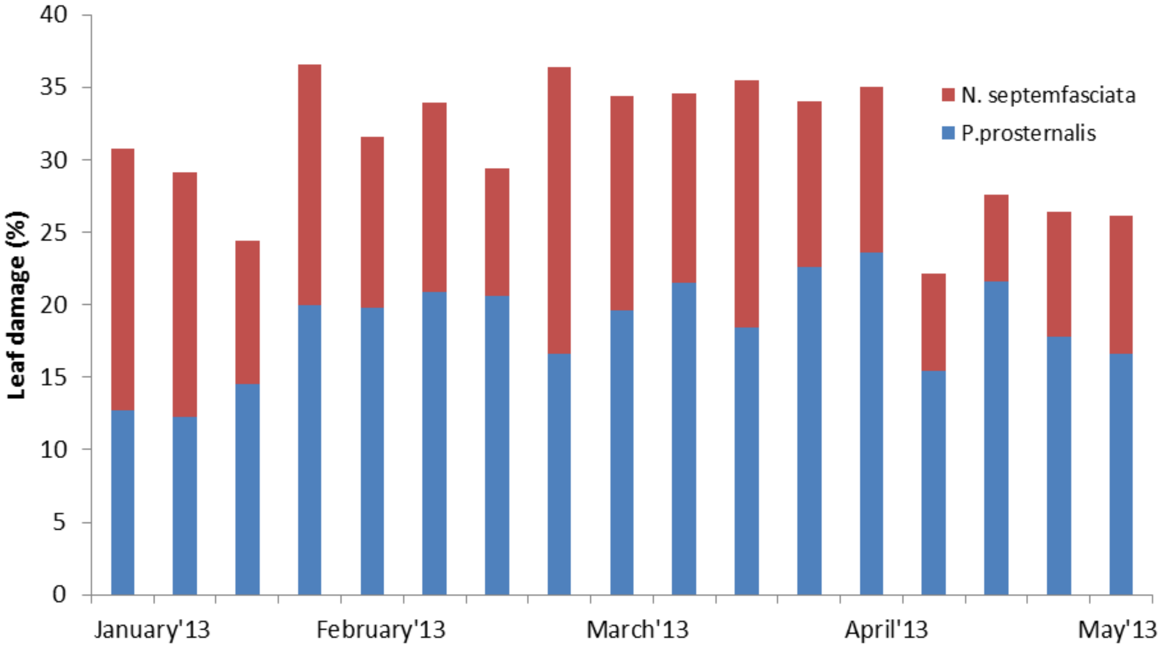

Figure 3.3: Comparison of % leaf damage by *Nomadacris septemfasciata* and *Petamella prosternalis* from January 2013 to May 2013.

Feeding damage of the two species (regardless of life stage) varied significantly ($F_{1,\,638} = 66.75$; $P<0.001$) with *P. prosternalis* eating the most (figure 3.4), consuming a mean of 32% more leaf area than *N. septemfasciata* per trial. Insect life stage (hopper or adult, regardless of species) did not significantly affect % leaf damage ($F_{1,\,638} = 0.15$; $P=0.698$), however, the interaction between the two species and life stage was statistically significant ($F_{1,\,638} = 4.54$; $P=0.034$). This significant interaction indicates that in one species (*N. septemfasciata*) the difference in feeding rate between life stages was large while in the other species (*P. prosternalis*) the difference was small.

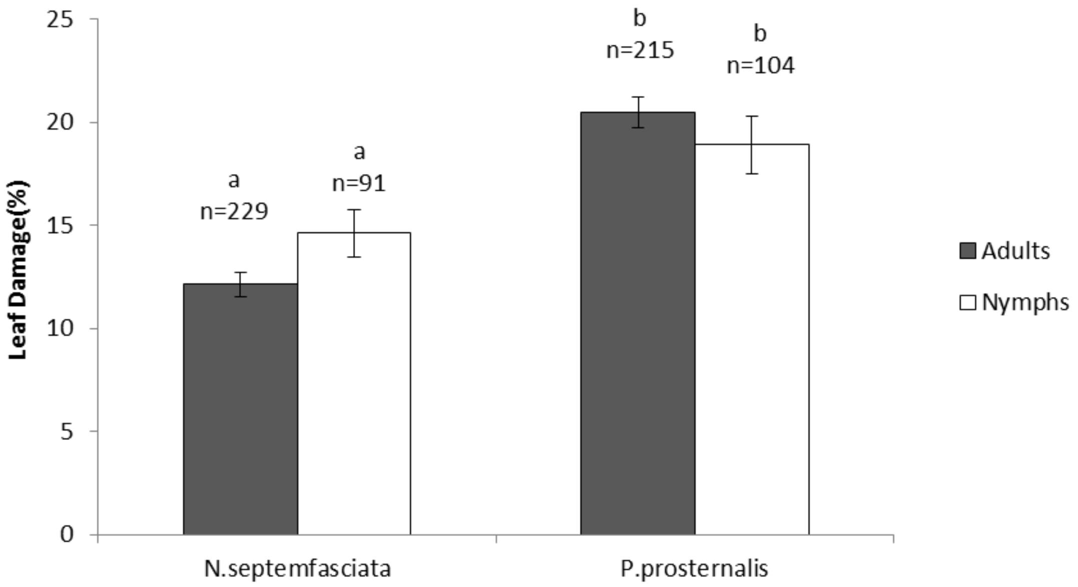

Figure 3.4: Percent leaf damage (±SE) caused by feeding of different life stages of *Nomadacris septemfasciata* and *Petamella prosternalis* on sugarcane leaves in laboratory trials. The means (after 2 way ANOVA) with the same letters are not significantly different (P>0.05) after Tukey's honestly significant difference test (HSD).

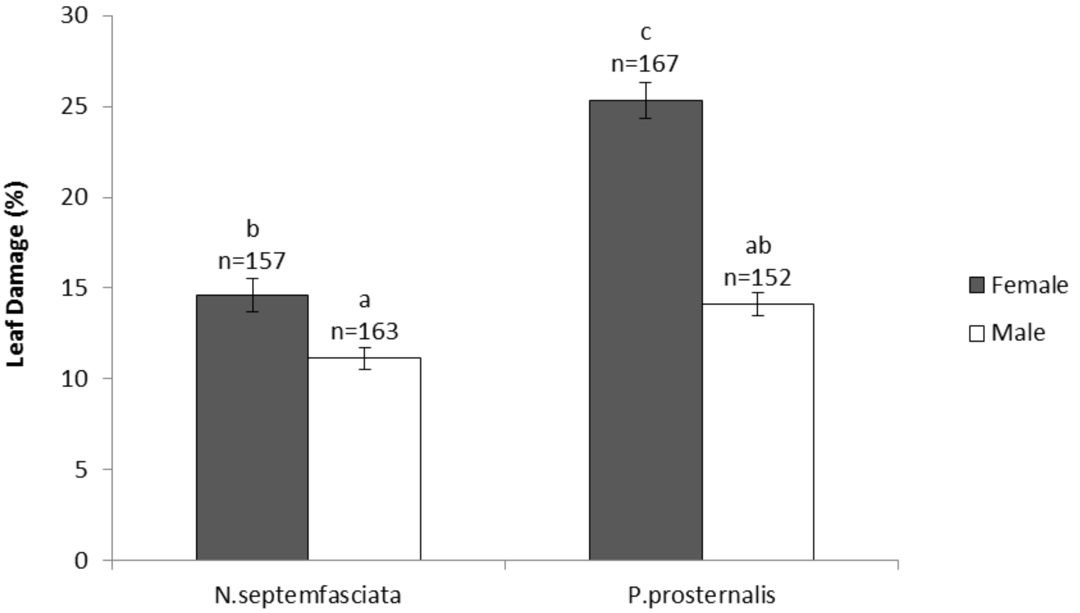

Figure 3.5: Percentage leaf damage and ±SE by male and female specimens of *Nomadacris septemfasciata* and *Petamella prosternalis* on sugarcane leaves in laboratory trials. The means (after 2 way ANOVA) with the same letters are not significant (P>0.05) after Tukey's honestly significant difference test (HSD).

Feeding damage (% leaf damage) between gender (regardless of species) was highly significant ($F_{1,\ 638}$=81.88; P=<0.001) with females of both species eating on average 37% more leaf area per trial (Figure 3.5). The interaction between the two species and gender was statistically significant ($F_{1,\ 638}$ = 22.82; P=<0.001).

This significant interaction also indicates that in *P. prosternalis* the difference in feeding damage in terms of gender is much larger than in the red locust. By subtracting % leaf damage of male *P. prosternalis* by that done by female *P. prosternalis* and multiplying by 100, it is evident that female *P. prosternalis* consumed approximately 44% more leaf area than males while *N. septemfasciata* females only consumed 24% more leaf area compared to their male counterparts.

Cumulative damage potential was computed on the four groups (male and female of each species) by adding mean leaf damage of the previous trials to the mean leaf damage of the subsequent trial (Figure 3.6). The cumulative difference in feeding potential is readily evident among males and females and among smaller and larger individuals. *Petamella prosternalis* females caused the most damage to sugarcane leaves with a total cumulative damage of 296.7 cm^2 of sugarcane leaf eaten over a period of 20 trials (feeding for seven hours per day). *Nomadacris septemfasciata* females were second with a total cumulative damage potential of 168cm^2 followed by *P. prosternalis* males with 161.9 cm^2 and *N. septemfasciata* males with 126.5 cm^2 of leaf area eaten. It is thus evident that females have a higher potential to cause damage compared to males. Coop and Croft (1993) found a significant relationship between body size and feeding potential of grasshoppers feeding on pearl millet in Mali therefore it can be said that females have a higher feeding potential due to their heavier weight and larger body size (Table 3.2). *Petamella prosternalis* (in both sexes) is a smaller grasshopper (dry weight) than *N. septemfasciata* but the results show that it has a higher feeding potential relative to its body size and thus has the potential to do more damage per gram of body weight. Therefore, body size is not well correlated with damage potential. Figure 3.6 demonstrates a theoretical maximum damage potential of each species over 20 days of feeding, it must be remembered that feeding only took place for seven hours each day therefore total cumulative damage is very likely to be considerably underestimated.

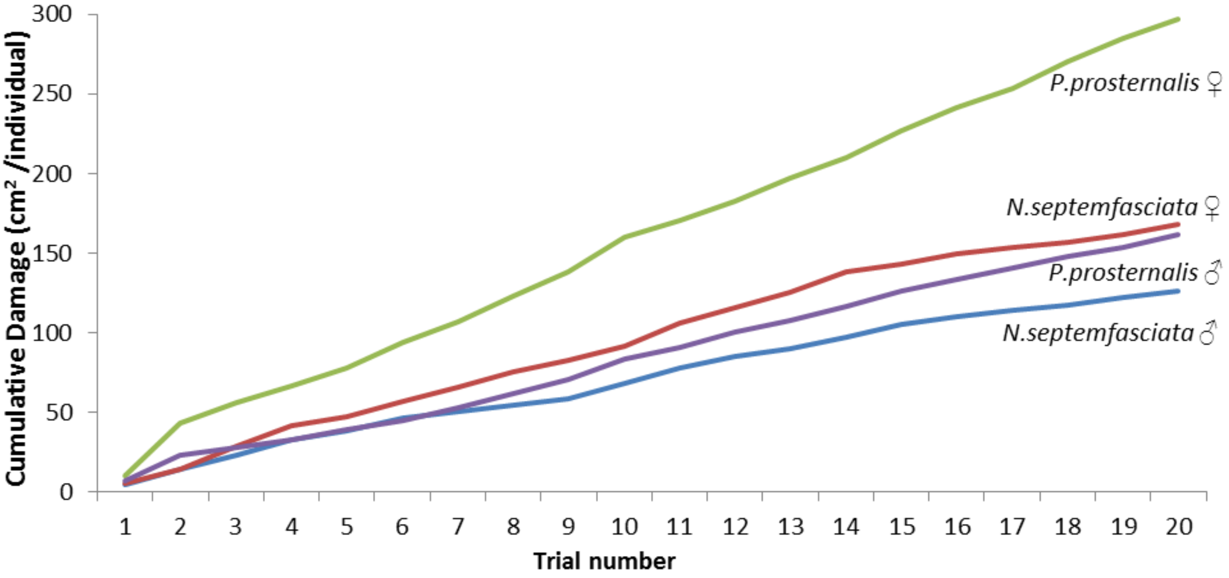

Figure 3.6: Mean cumulative sugarcane leaf damage by *Petamella prosternalis* and *Nomadacris septemfasciata* species in laboratory trials (feeding 7 hours per day) during 20 trial days between 22 January 2013 and 21 May 2013.

Mean mass (±SE) of adult *N. septemfasciata* and *P. prosternalis* as well as maximum and minimum specimen mass is shown (Table 3.2). It is clear that *N. septemfasciata* is generally larger than *P. prosternalis*. Females are also larger than males in both species.

Table 3.2: Dry mass of female and male adult specimens of *Petamella prosternalis* and *Nomadacris septemfasciata* collected from Empangeni sugarcane study sites in May 2013.

	N. septemfasciata		*P. prosternalis*	
	♀	♂	♀	♂
Mean (g)	1.24953	0.908113	0.85188	0.317391
Standard Error	0.027438	0.013991	0.036826	0.008598
Minimum (g)	0.9533	0.7926	0.4212	0.2167
Maximum (g)	1.5691	1.0398	1.2609	0.3889
(n)	30	30	30	22

The specific leaf weight (SLW) calculated for the variety NCo 376 was: 0.04625 grams per cm^2. (See: materials and methods). In order to convert leaf area injury (LAI) in cm^2/ seven hours to leaf weight injury in grams/day the following formula was used: (*LAI x 3.43 x SLW = Leaf weight injury in grams/day*) (Table 3.3).

Table 3.3: Estimated leaf weight injury by *Nomadacris septemfasciata* and *Petamella prosternalis* calculated by converting leaf area injured to leaf weight injured using the calculated specific leaf weight (SLW) value of sugarcane variety NCo 376.

	Leaf area injury (cm^2/7hrs)	Leaf weight injury (g/day)
P. prosternalis ♂	7.95	1.26
P. prosternalis ♀	15.15	2.40
N. septemfasciata ♂	6.36	1.00
N. septemfasciata ♀	8.31	1.32

Nomadacris septemfasciata adult (male and female averaged) can consume roughly 1.16 grams of fresh cane leaf in 24 hours, (about 108% of its own body weight in a day while *P. prosternalis* (male and female averaged) can consume approximately 1.83 grams of leaf in a day- Table 3.2) while *P. prosternalis* (male and female averaged) can consume approximately 1.83 grams of leaf in a day, (about 300% of its own body weight in a day -Table 3.2).

3.4 Discussion

3.4.1 Laboratory studies

In order to determine the ET for acridids in this study a number of factors need to be considered. These include: feeding potential of the pest, acridid population density, crop productivity, climatic factors, cost and return of control relationships, and the efficacy of treatment options (Torell & Huddleston 1987, Davis et al. 1992). It is clear that a lot of this information is lacking for the current study. Laboratory feeding data is an important first step. Coop & Croft (1993) found that the use laboratory feeding data to estimate potential damage levels and economic thresholds in a field situation is limited. In terms of this study, the factors which limit this data include: 1) the containers which each individual was placed in greatly restricted movement of which the effects on feeding behaviour are unknown. 2) There was no alternative food source, only sugarcane was made available to them during the trials. 3) The two species used in the trials have different life cycles (see Table 2.5 in chapter 2) and during the trial period, *P. prosternalis* was entering into its reproductive period, while the *N. septemfasciata* was entering into the period of reproductive diapause which may affect feeding behaviour (Coop & Croft 1993) and thus skew the results.

Insect feeding rates are also temperature dependent (Hammond et al. 1979, Ferro et al. 1985, Lactin & Johnson 1995). Lactin & Johnson (1995) found that in laboratory trials *Melanoplus sanguinipes* Bruneri (Acrididae: Melalopplinae) showed maximum feeding rates at 40°C in third, fourth and fifth instars. If we consider that the Empangeni region has experienced a mean annual temperature of 21.6^0 C and a mean wet season (1 September – 28 February) temperature of 23.2^0 C over the last 10 years (SASRI WeatherWeb) it becomes apparent that acridid populations in the field will be exposed to temperatures during the day similar to the temperature at which the current trials were conducted for a significant period of their life. Feeding behaviour is also an important factor to consider. Coop & Croft (1993) found that at higher densities, O.*senegalensis* tends to band in groups (gregarize), thus causing a potentially non-uniform pattern of injury compared with other grasshopper species. *Nomadacris septemfasciata* is likely to behave in the same manner as it has the ability to gregarize (Lecoq et al. 2011) which potentially could lead to very high injury levels in localised areas whereas a solitary species such as *P. prosternalis* may cause less conspicuous damage over a larger area. This phenomenon has already been observed in Empangeni sugarcane.

Feeding trial results were fairly erratic and tended to fluctuate substantially from trial to trial. A possible explanation is that individual specimens were taken from communal cages and placed in separate containers during the trials which may have had an effect on their feeding behaviour. Increasing growth in hoppers is sustained by increased consumption of food (Chapman & Joern 1990). According to the present study, this phenomenon is true with respect to *P. prosternalis* but is not the case for *N. septemfasciata*. During the first two weeks of laboratory trials (22 January – 8 February) feeding damage by *N. septemfasciata* were higher than in the next five feeding trials (8 February – 30th February). Hill et al. (1968) and McCaffery (1975) found that female acridids increase consumption in order to produce eggs, suggesting that during a female's reproductive period, feeding rate will be elevated. Leather (1993) states that insects which undergo an adult winter diapause rely mostly on nutritional reserves for sustenance, however periodic feeding does still occur, presumably at a lower rate. These statements may explain why *N. septemfasciata* feeding was lower than *P. prosternalis* feeding; during March/April *N. septemfasciata* enters into a period of winter reproductive diapause while *P. prosternalis* becomes reproductively active (see Table 2.5 in chapter 2)

Coop & Croft (1993) and Hewitt & Onsager (1982) found that grasshopper dry weight is positively correlated with feeding potential. However, in this study the opposite was observed. The largest specimen (*N. septemfasciata* female) did not have the highest feeding potential. Similarly, the smaller *P. prosternalis* males had a higher feeding potential than *N. septemfasciata* males (Table 3.2 & 3.3). This result means that there must be another factor other than body size which is causing this discrepancy. It is possible that the more sedentary and 'tame' nature of *P. prosternalis* was conducive to higher feeding rates being observed during laboratory trials. It has been shown that gregarious phase *Schistocerca lineata* Scudder (Orthoptera: Acrididae) hoppers exhibit a preference for toxic host plants, which when ingested, result in them being toxic to predators (Simpson & Sword 2006). In the gregarious phase *N. septemfasciata* may also exhibit a preference for toxic host plants to maintain its toxicity to predators therefore resulting in a reduction in feeding when only presented with sugarcane leaves during laboratory trials. Easwaramoorthy et al. (1989) found that during laboratory trials of two acridid species feeding on sugarcane in India, three times more leaf damage was recorded by one species even though the two species tested had almost identical weights. This finding shows that certain species are more voracious feeders than others, and thus have a higher feeding potential relative to their weight. These findings add validity to the present study which showed similar results, indicating that *P. prosternalis* is a more voracious feeder compared to *N.septemfasciata*.

The statement by Williams et al. (1969) and FAO (2009) which estimates that 'the red locust (*N. septemfasciata)* adult can consume roughly its own weight, about two grams, in fresh food in 24 hours' has some measure of reality, as the present study confirmed that *N. septemfasciata* is able to consume roughly its own weight in a day, but this weight of forage is about half (1.16 grams) as much as their estimate. However, *P. prosternalis* can consume three times its own weight in a day, thus posing a substantial risk to the sugarcane in the Empangeni region.

3.4.2 Field studies

The damage rating index shows that the variation in observed damage corresponds quite closely to the fluctuations in relative abundance of *N. septemfasciata* and *P. prosternalis*. This proves that damage is intrinsically linked to the life history of the acridid populations on the sugarcane farms in the study area. Feeding damage on Jengro farm, where *P. prosternalis* was the dominant species closely relates to the population fluctuations of *P. prosternalis* (Chapter 2). The same observation can be made for Kelly farm in relation to *N. septemfasciata* (Chapter 2), while GSA farm is not closely linked to the trends of either species but instead shows relatively high damage throughout the year, possibly because neither species was dominant on this farm - high densities of both species were observed there. Where populations of *P. prosternalis* were dominant (Jengro), the damage rating index values were higher than where *N. septemfasciata* was dominant (Kelly). This suggests that laboratory

does compare well to observed field data, in that *P. prosternalis* has a higher potential to cause sugarcane damage compared to *N. septemfasciata*.

Sugarcane growth rates and acridid population densities are the two most critical elements needed to calculate ET and EIL. Calculating population density in a high density population is notoriously difficult (Burnett 1951). Gardiner (2005) stated that “there are few methods that can be effectively utilised in tall vegetation particularly at high population densities”. This is the predicament which is faced in the sugarcane fields in the study sites in Empangeni. Nevertheless, it should still be possible to obtain reasonably accurate density estimates if suitable equipment and manpower is provided. Therefore the data shown here and in Chapter 2 should be regarded as the first step towards obtaining accurate EIL’s in order to determine the ET’s of the acridid species in Empangeni sugarcane.

Although the determination of ET’s is based on the accumulation of much quantitative data, at present program managers and other stakeholders do exercise a considerable amount of qualitative judgment and discretion before control treatments for high acridid numbers are employed (Davis et al. 1992). At present, the ultimate decision of control must be made by the grower based on the amount of forage he feels he can afford to lose (Hewitt & Onsager 1982).

3.5 Conclusion & Recommendations

It is clear that in terms of feeding potential, regardless of other factors involved, *P. prosternalis* has a higher feeding potential than *N. septemfasciata* but both species can cause significant damage which at times will require action against them in order to limit damage. The laboratory results obtained in this study mirror the results of the studies field observations. Unfortunately, with the current available data from feeding trials, it is not possible to make specific recommendations with regards to ET and EI levels of the acridid complex. However, the combination of laboratory and field data from this study provides crucial baseline data to assist in understanding the impact of acridid feeding on Empangeni sugarcane. Before applying any control measures growers should take into consideration the species they are dealing with, its life stage, and level of infestation, age of the sugarcane and the time of year when the high populations occur, which all play a part in the resultant damage they cause. For future crop loss assessments, the growth rate of sugarcane leaves (in grams/day) needs to be determined which can be used to compare to the rate of defoliation (grams/day) by the relevant acridid species, which has been established in this study.

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Chapter 4: An analysis of the phase status of the red locust, *Nomadacris septemfasciata* (Serville) in Empangeni, Kwazulu-Natal, South Africa

4.1 Introduction

The *Nomadacris septemfasciata* (Serville) (Orthoptera: Acrididae) is a true locust species (Lecoq et al. 2011a) and consequently exhibits density-dependent phase polymorphism. This phenomenon was first described by Boris Uvarov in 1921 and was later re-described as phase polyphenism, defined as the ability to form marching hopper bands and/or flying swarms which can potentially lead to outbreaks and plagues (Lecoq et al. 2011a). This form of phenotypic plasticity is in response to changes in local population density and affects the expression of numerous behavioural, physiological and morphological traits (Loher 1990, Simpson & Sword 2006, Sword et al. 2010). Phase polyphenism is limited by two extreme phenotypes, namely, phase: *Gregaria* (swarming, gregarious form) and phase: *Solitaria* (sedentary, isolated form). However, polyphenism occurs on a continuous scale and individuals can occur in an intermediate or *transiens* phase and are then described by the terms *congregans* or *dissocians*, depending on the direction of the current phase change (Albrecht 1956, Simpson et al. 2006, Sword et al. 2010, Lecoq et al. 2011a). Behavioural change is the most labile of responses to crowded conditions and is mediated by direct physical contact between individuals (Simpson & Sword 2006). These promiscuous encounters provide positive feedback for continued local crowding and subsequent change in behaviour, from avoidance to active aggregation that can lead to other phenotypic changes such as changes in colouration and morphology, which develop more slowly (Sword et al. 2010). The irregular fluctuations between remission and upsurge periods requires knowledge about the gregarization process, and understanding this process can allow for early detection and prediction of the severity of the locust outbreak (Lecoq et al. 2011a). In Empangeni sugarcane, incipient swarms have been observed at increasing densities. Hopper density and colouration also indicate that these populations are showing gregarious tendencies, possibly indicating a shift from solitarious individuals to gregarious individuals.

Three different techniques exist to determine the phase status of *N. septemfasciata*. The first technique uses hopper body colour as a phase determinant. The gregarization threshold for hoppers is estimated to be 100 000 hoppers/ha, after which individuals start to behave in a gregarious manner and change from a general green body colour to a combination of orange, yellow and black (Faure 1935, Burnett 1951). For adults the phase transformation threshold is estimated at 5000 individuals/ha (Lecoq et al. 2011a). The second technique uses morphological measurements of the legs and wings. Numerous studies have used traditional morphometric ratios to determine phase status, such as in Burnett (1951), Rainey et al. (1957) and Franc et al. (2005). In these studies the ratio of elytron length (E) to hind femur length (F) was compared, resulting in an E/F ratio which can be compared to other studies. An E/F ratio of 2.0 marks the limit between the two forms, i.e. an E/F value above 2.0 generally means a population is in a gregarious state (Franc et al. 2005). The third technique uses eye stripes as an indication of phase change. According to Burnett (1951a) adults of the phase *solitaria* should exhibit eight eye stripes while adults of the phase gregaria should exhibit seven eye stripes which therefore allow them to be separated by these criteria. Geometric morphometrics has not been used previously to assess phase polyphenism in locusts in general. In this study, the results of the above mentioned established techniques will be compared to geometric morphometric results in an attempt to draw comparisons between them and to determine whether geometric morphometrics is a suitable method to use for assessing phase changes in locusts

Body colour as an indicator of phase change

The colour of locust hoppers varies according to the population density at which they occur and has been used as an indication of phase status in a number of locust species including in *N. septemfasciata* (Faure 1935). According to Lecoq et al. (2011a) a conspicuous black spot on the hind femur is the first sign of gregarious tendencies but the following criteria were used to classify hoppers into the separate phases.

- Solitarious (low density populations): general green background colouring on all body parts with a general lack of black pigmentation as in other phases. Eye stripes are still clearly visible in older instars. No prominent black spot on the hind femur
- Transiens: Great variations in colour between individuals but generally hoppers have a larger proportion of yellow/orange on them as well as increase in melanised areas. The overall green colour disappears and eye stripes are still visible but less conspicuous. Prominent black spot on hind femur is often visible.
- Gregarious: High degree of melanisation, body marked with highly developed black spots, eye stripes are invisible and general background colour of the hopper is orange. Black spot on hind femur is conspicuous. More than 5000 hoppers per Hectare.

Traditional Morphometrics

Morphometrics is the study of shape variation and its covariation with other variables (Dryden & Mardia, 1998). Traditional morphometrics uses multivariate statistics to describe patterns of shape variation between different groups usually using linear distance measurements, counts, ratios or angles (Adams et al. 2004). Criteria values such as the ratio between the Elytra (E) and Femur (F) length can be used for characterizing the phases of a locust (Burnett 1951, Rainey et al. 1957, Franc et al. 2005) and this parameter has proven to accurately separate populations of field collected specimens of *N. septemfasciata*, *Schistocerca gregaria* (Forskal) (Orthoptera: Acrididae) and the migratory locust *Locusta migratoria capito* (Sauss) (Orthoptera: Acrididae). With regards to *N. septemfasciata*, elytra length is always longer than the length of the femur and the difference between these two values results in a ratio which generally falls in the region from 1.8-2.2. The higher the ratio, the more gregarious an individual is, therefore a low E/F ratio (e.g. 1.81) means an individual is in the solitary phase, a medium E/F ratio (e.g. 1.97) means an individual is in the transiens phase and a high E/F ratio (e.g. 2.16) means an individual is in the gregarious phase. In morphological terms, the longer the wings and shorter the hind legs (proportionately) the greater the chance that the individual is gregarious and therefore able to migrate farther. Table 4.1 displays the E/F ratio from previous studies of gregarious populations of *N. septemfasciata* at various locations.

Table 4.1: Comparison of the E/F ratios of *Nomadacris septemfasciata* from various studies in Africa and Madagascar.

Location	Sex	E\F Ratio	Reference
D.R. Congo	M	2.02	Bredo (1938)
	F	2.14	
Zimbabwe	M	2.11	Michelmore & Allan (1934)
	F	2.11	
Chad	M	1.85	Tetefort & Wintrebert (1967)
	F	1.82	
Madagascar	M	1.88	Tetefort & Wintrebert (1967)
	F	1.84	
Tanzania	M	1.84	Burnett (1951)
	F	1.82	
Madagascar	M	2.06	Franc et al. (2005)
	F	2.03	
Empangeni	M	2.02	Present study
	F	1.99	

Counting eye stripes

It has been shown that swarming *N. septemfasciata* of the gregarious type undergo six larval instars before becoming winged adults while solitarious individuals go through one extra moult (Burnett 1951a, Albrecht 1957). This moulting polymorphism thus allows one to determine the phase status of the adult locust by counting the number of dark vertical eye stripes present on the eye (Figure 4.1). According to Burnett (1951a) adults of the phase solitaria should exhibit eight eye stripes while adults of the phase gregaria should exhibit seven eye stripes which therefore allow them to be separated into phases by the number of eye stripes present.

Figure 4.1: Example of the dark vertical eye stripes present in adult *Nomadacris septemfasciata* eyes which are used to determine the number of instars an individual passed through and therefore the phase status of the specimen.

Geometric Morphometrics

There are a number of shortcomings of traditional morphometrics such as size correction issues and the fact that you can obtain the same set of distance measurements from two different shapes because their relative position to one another is not taken into account (Adams et al. 2004). Due to these shortcomings, geometric morphometrics was developed as an alternative way of quantifying and analysing morphological shape variation. Landmark-based geometric morphometrics starts with the imaging of the chosen study subject after which digitizing takes place to collect two or three dimensional coordinate data. Coordinates are obtained from definable, homologous landmarks on a variety of organisms or appendages such as insect wings, fish fins or mammal mandibles (see: Mahalanobis et al. 1934, Baylac et al. 2003, Petit et al. 2006). In order to analyse these data the effects of variation in position, orientation, and scale of the specimens (non-shape effects) must be removed. Statistical analysis and graphical representation of the data is then possible using a standard procedure which can be summarized as a generalized procrustes analysis (GPA), followed by projection of the aligned coordinates onto a linear tangent space for multivariate analyses, and the graphical visualization of results in terms of the configurations of landmarks (Adams et al. 2004). The use of geometric morphometrics in studying locust species is limited to the investigation of male genetalia (Song & Wenzel 2008) and it has not been used to study phase polyphenism. There is substantial potential to use geometric morphometrics as a method of analysing wing shape variation in relation to phase polyphenism.

The aim of this study is to determine the density-dependant phase polyphenism of populations of *N. septemfasciata* occurring in Empangeni sugarcane study sites, based on traditional and geometric morphometric measurements, eye stripes and field observations of hopper colour. This will provide information to determine the potential for these populations to form swarms and outbreaks.

4.2 Materials and Methods

4.2.1 Traditional morphometric measurements

N. septemfasciata specimens were collected at three locations, Jengro (28°37'30.84"S 32° 0'52.68"E), Kelly Farm (28°41'29.75"S, 31°54'26.92"E) and GSA farms (28°40'54.94"S 31°54'51.98"E) during the sampling period from May 2012 to May 2013 (Figure 4.2). Three body characters were measured on 594 individuals (Table 4.2) using a digital calliper (Marshal Tools) to the nearest 0.05mm. Body length, from frons to the end of the forewings, length of left forewing (elytra) and length of right hind femur were measured. These characters are ideal for morphometric measurements as they do not change after the insect dies or is dried (Bazelet 2011). Measurements were made according to the method as stipulated at the 4th International Locust Conference in Cairo (Egypt, 1937).

Figure 4.2: Map showing the three sampling locations where adult specimens of *Nomadacris septemfasciata* were obtained for morphometric data analysis.

Traditional morphometric data analysis

Analyses were performed using Genstat 15.0 (VSN International LTD, UK). Data were tested for normality of frequency distribution using the Shapiro-Wilks test. The data were found to be homoscedastic. Univariate General Linear Model (GLM) analysis of variance (ANOVA) was used to test the interactions between populations in terms of location and time. Where the interactions were found to be significant, a Tukey's post-hoc test was performed to separate the means.

A discriminant function analysis (DFA) was conducted to determine the major differences between groups (i.e. phases: solitarious, transiens and gregarious) relative to the variation within these groups (Fisher 1936). The E/F values were divided into three groups based on the criteria from Rainey et al. (1957) as follows: solitarious: 1.8-1.87 transiens: 1.88-2.05 gregarious: 2.06 -2.15. For the analysis, the grouping variable selected was phase, while the independent variables were the morphometric measurements (LTOT, E, F and E/F) of each specimen, with a stepwise analysis selected. The analysis was performed in Statistica version 11.0 (StatSoft Inc., Tulsa, USA).

4.2.2 Eye stripes

Individuals collected for morphometric measurements were also observed under a dissecting microscope (Zeiss- 2.5X magnification) to count the dark vertical stripes present on the eye. Eye stripes of the left eye were counted wherever possible and recorded as seven or eight eye stripes.

4.2.3 Geometric morphometric measurements

Following results from traditional morphometrics, it was decided to establish whether geometric morphometrics would be a useful tool to use in establishing phase polyphenism. Since the specimens used in traditional morphometric measurements had already been destroyed, 100 new specimens (25 male and 25 female) from two locations were collected within 1 week of each other in May 2013. The two locations were Jengro farm (28°37'30.84"S, 32° 0'52.68"E) and Kelly Farm (28°41'29.75"S, 31°54'26.92"E). These two farms were selected because they were a considerable distance apart (Figure 4.2), reducing the chance of immigration and emigration between the two locations, therefore possibly providing greater differences between the two populations. The distance between these two locations is approximately 12.8km (in a straight line), and traditional morphometric measurements had found significant differences between individuals from these two locations. Left and right forewings of the 100 specimens were removed at the base using scissors. Individual wings were mounted between two standard microscope slides (25.4x76.2mm, 1.0-1.2mm thick), and were taped together and labelled according to replicate number, locality, gender and side. Images were taken of each wing between the slides using a tripod mounted Canon 50D, with 100mm macro lens (f2.8L), positioned inside a photographic light box (FalconEyes® Model #: FLB 416AB). The tps utility programmes (Rohlf 2008) were used to prepare files for data analysis (Stonybrook morphometrics). To generate files in the correct format, tpsUtil 1.52 was used. From the individual images, 9 common landmarks (Figure 4.3) were selected and positioned on specific vein intersections or terminations of the wing using tpsDIG 2.16. Vein intersections were selected based on prominence and to obtain the best coverage over the entire wing. Lastly, tpsRelw 1.11 was used to generate a consensus set of x-y coordinates for each wing.

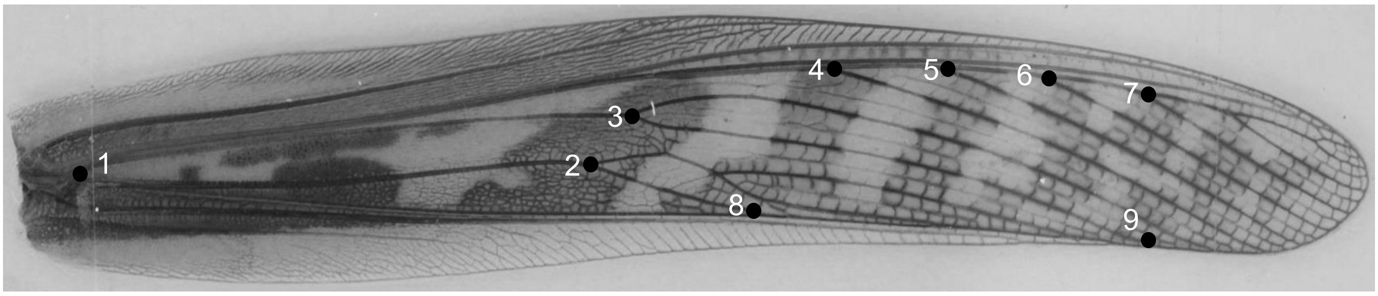

Figure 4.3: Indication of the nine landmarks used in geometric morphometric studies on the forewing of adult *Nomadacris septemfasciata,* viewed dorsally.

Geometric morphometric data analysis

MorphoJ version 1.05c (Klingenberg 2011) program was used for further analysis. From the x-y coordinates, shape information was extracted with a full procrustes fit, aligned by principal axes, which removes information for size, position and orientation, leaving only centroid size and shape information for each specimen (Rohlf & Slice 1990). To ensure that the error as a result of the imaging and digitizing processes was within the acceptable limits, a Procrustes ANOVA {$2k - 4$, where k = number of landmarks in two dimensions (x-y coordinates) minus four degrees of freedom (two are lost during translation for position, one for scaling to a standard size and one for rotating to a standard position) (Klingenberg & McIntyre 1998)}, was performed on a subset (40 specimens) of the complete data in a separate analysis.

On the total dataset, a new procrustes fit was performed, again aligned by principle axes. This was followed by a principal components analysis (PCA), based on a covariance matrix of landmark coordinates, to explore the variation associated with the shape changes by illustrating the percentages of the total shape variation for which the eigenvalues account. This was followed by a discriminant function analysis (DFA) ,to determine whether group differences existed between the two populations at Jengro and Kelly (solitarious and gregarious, respectively, based on E/F ratios) (Iversen et al. 1976). In MorphoJ, Fisher's classification rule is used for the DFA, which sets the cut-off point at zero. Left and right wings were averaged to exclude asymmetry. The classifiers which define each individual (such as gender, location etc.) were then selected and results displayed graphically using a canonical variate analysis (CVA) to visualise the relationship between different groups. Specimens were sorted according to gender using the description of the reproductive organs by Faure (1935).

4.3 Results

4.3.1 Hopper body colour as an indicator of phase status

Using Lecoq et al. (2011a) and Faure (1935) as guidelines, the colouration of hoppers in Empangeni on a qualitative basis indicated that individuals were either in the transiens or gregarious phase and very few hoppers matching the description of a true solitarious individual were found (Figure 4.4). During population surveys (Chapter 2) the vast majority of hoppers caught matched the description of gregarious hoppers in a number of ways such as behaviour, colouration and density, which in one severely infested area in November 2012 reached approximately 72 000 hoppers per hectare.

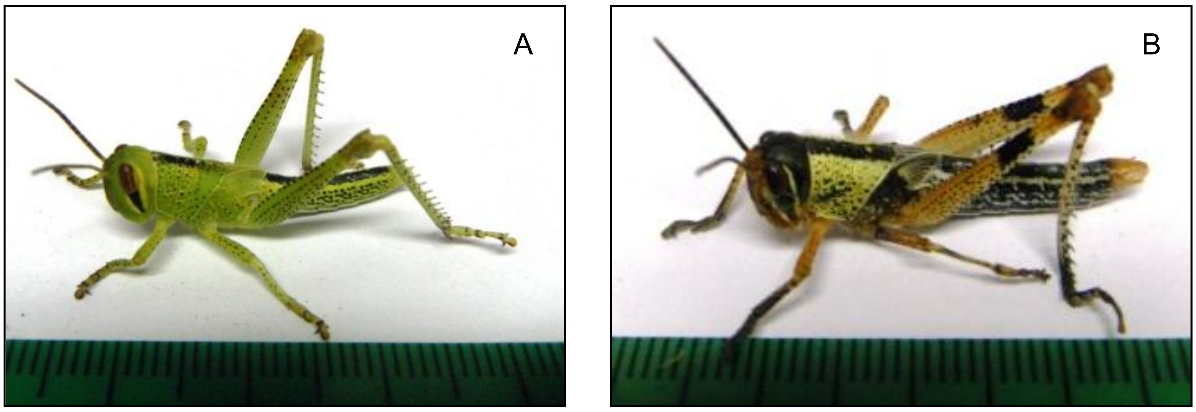

Figure 4.4: An example of a solitarious (A) and gregarious (B) hopper of *Nomadacris septemfasciata* found in Empangeni sugarcane.

4.3.2 Traditional Morphometric measurements

Table 4.2 displays mean total length, elytron length, femur length and the E/F ratio and associated sample variance (the average of the squared differences from the mean) of the different populations separated firstly by location and then by year. Females were larger than males. Kelly and GSA harboured more gregarious populations than Jengro and males generally had a higher E/F ratio compared to females.

Table 4.2: Morphometric measurements of *Nomadacris septemfasciata* samples collected from the study sites in the Empangeni region showing the mean total length, elytron length, femur length and the elytron/femur (E/F) ratio as an indication of the phase status of each population.

	Total Length (mm)				Elytron length (mm)				Femur length (mm)				E/F Ratio			
	♂		♀		♂		♀		♂		♀		♂		♀	
	Mean	Var.	Mean	Var.	Mean	Var.	Mean	Var.	Mean	Var.	Mean	Var.	Mean	Var.	Mean	Var.
Kelly(n=136)	70	7.56	79.7	4.29	57.6	2.5	66.1	3.43	28.3	0.55	33.1	1.22	2.0318	0.0028	1.9969	0.0028
GSA(n=82)	69.8	3.14	79.4	5.29	57.6	2.5	66	3.16	28.3	0.77	32.8	1.08	2.0341	0.0026	2.0104	0.0029
Jengro(n=108)	68.7	2.68	79.5	6.22	56.5	2	65.8	4.31	28.3	0.78	33.5	1.6	1.999	0.0022	1.9656	0.0016
2012(n=216)	70.1	3.66	79.9	4.81	57.3	2.8	66.1	3.42	28.5	0.63	33.3	1.37	2.007	0.003	1.9875	0.0032
2013(n=378)	69.4	2.92	79.5	4.69	57.3	2.4	66	3.4	28.3	0.63	33	1.3	2.0265	0.0026	1.9998	0.0027

The mean E/F value (males and females combined) for GSA was 2.024 and for Kelly farm it was 2.0165. The mean E/F value of the Jengro population was 1.9851. The E/F ratio varied significantly according to site ($F_{2, 513} = 17.77$; $P < 0.001$) and time ($F_{1, 513} = 23.84$; $P < 0.001$) (Figure 4.5). The E/F ratio for GSA and Kelly were significantly higher compared to Jengro indicating that *N. septemfasciata* populations at GSA and Kelly were more gregarious than at Jengro. E/F ratio was higher in 2013 compared to 2012 indicating that overall, the F1 generation were more gregarious than the parent populations.

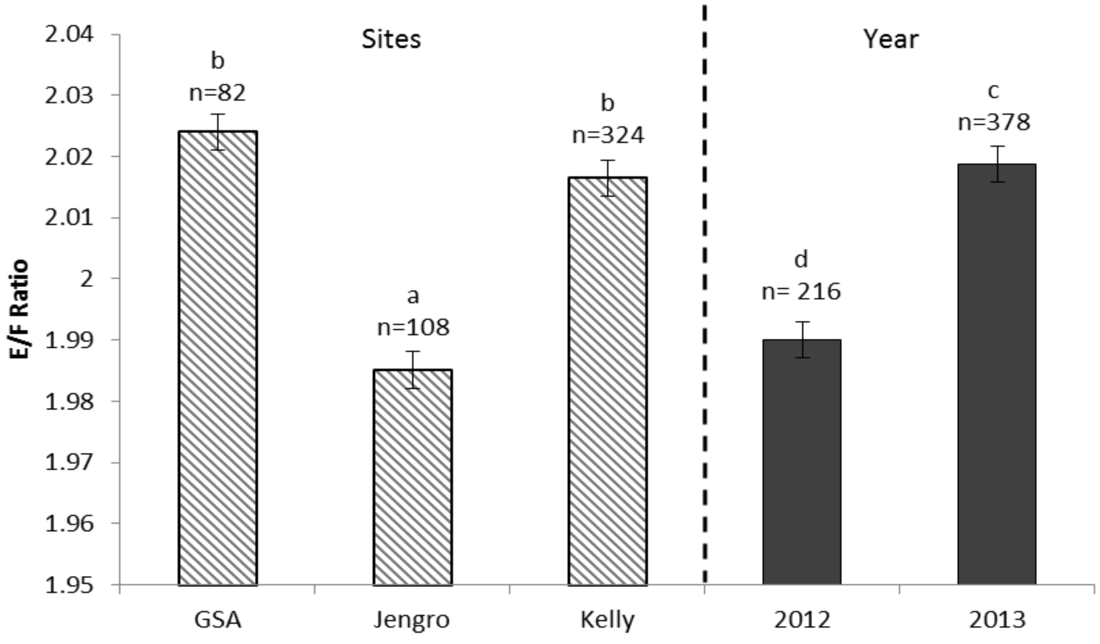

Figure 4.5: Comparison of E/F values of the *Nomadacris septemfasciata* for three sites and over two generations (±SE; n= 594). Lower case letters above the bars indicate significance between categories if the letter is different.

Discriminant function analysis

The following E/F values, as in Rainey et al. (1957) were used to delimit specimens into their respective categories: Solitaria: 1.8-1.87, Transiens: 1.88-2.05 Gregarious: 2.06 -2.15 for the DFA. The stepwise-forward analysis was completed in one step and removed LTOT, E and F as variables (Table 4.3). Overall, the discrimination of E/F was highly significant (*Wilks' lambda* = 1.0000; $F_{(2,478)} = 302.85$; $p<0.0000$). The classification functions are given in Table 4.4. With these functions, the transiens and solitary phases were classified correctly in 100% of cases, while the gregarious phase was classified correctly in 92% of cases. Of the total specimens measured, 359 were of the transiens phase, which was over-predicted by 2.7%, 119 were of the gregarious phase, which was under-predicted by 8.4% and 3 were of the solitarious phase, which was correctly predicted, based on E/F ratios alone. Means of E/F ratios of observed values for each phase were 1.85 (solitary), 1.99 (transiens) and 2.08 (gregarious). The DFA confirms that the E/F ratio is the best trait to use in separating specimens according to phase in Empangeni sugarcane.

Table 4.3: Discriminant Function Analysis of variables (morphometric measurements of *Nomadacris septemfasciata*) not in the model.

Variable	Wilks' lambda	Partial Wilks' lambda	F to enter(2, 477)	p-value	Tolerance
Total length (LTOT)	0.441082	0.999998	0.000569	0.999432	0.993073
Elytron length (E)	0.440935	0.999664	0.080275	0.922875	0.993958
Femur length (F)	0.440808	0.999377	0.148661	0.861901	0.905586

Table 4.4: Classification functions for the character E/F ratio required to *classify Nomadacris septemfasciata* into solitary, transiens or gregarious phases.

Measurement	Solitary	Transiens	Gregarious
E/F ratio	1382.9	1488.81	1556.37
Constant	-1284.13	-1482.76	-1621.45

4.3.3 Eye stripes

90.2% of specimens had seven eye stripes meaning they underwent six larval instars while 8.5 % had eight eye stripes. Therefore, according to this method that the vast majority of specimens observed were in the gregarious phase (Table 4.5).

Table 4.5: Results of adult eye stripe counts per locality and over two years for *Nomadacris septemfasciata*. These results illustrate the spatial and temporal variation in number of instars, indicated by the number of eye stripes undergone between different populations.

Site/Year	Sex	8 eye stripes (%)	7 eye stripes (%)
Kelly (n=196)	♂(n=113)	3.5	98.2
	♀(n=83)	2.4	95.2
GSA (n=18)	♂(n=8)	12.5	87.5
	♀(n=10)	10.0	90.0
Jengro (n=109)	♂(n=58)	19.0	81.0
	♀(n=51)	17.6	70.6
2012 (n=67)	♂(n=34)	0.0	100.0
	♀(n=33)	0.0	100.0
2013 (n=257)	♂(n=155)	8.4	91.6
	♀(n=102)	11.8	88.2
Mean (%)		8.5	90.2

The results suggest that Kelly farm held the most gregarious population and Jengro had the least gregarious population, which compliment morphometric data from individuals collected from the two farms. In 2012 no individuals were found with eight eye stripes, while in 2013 about 10% had eight eye stripes. This result is in conflict with the morphometric E/F data which indicated that the 2013 generation was more gregarious than the 2012 generation. However, it should be remembered that roughly four times more specimens were measured in 2013 compared to 2012. Table 4.6 displays the number of instars passed through by *N. septemfasciata* under laboratory conditions (±27^{0}C, 60% RH). Eggs collected from the field hatched in the laboratory on 20 November 2012. Hoppers and eventually adults were kept in cages at low densities (three individuals per cage). The progeny of these eggs would inherit the phase which their parents experienced because the effect of parental crowding is passed on through a chemical agent introduced into the foam surrounding the eggs (Simpson & Sword 2006). This laboratory data confirmed that *N. septemfasciata* in Empangeni is able to exhibit the "six larval instar" phase type typical of swarming *N. septemfasciata* (Burnett et al. 1951a, Albrecht 1957).

Table 4.6: Results of laboratory rearing of the *Nomadacris septemfasciata* from field collected eggs indicating the number of instars undergone by one individual from November 2012 until reaching the adult stage.

	Instar							
	Hatched	1	2	3	4	5	6	7(Adult)
Date	20-Nov	27-Nov	05-Dec	13-Dec	24-Dec	15-Jan	26-Jan	08-Feb
Duration (Days)		7	3	8	11	22	16	13

4.3.4 Geometric morphometrics

The Procrustes ANOVA indicated that the mean square (MS) and *F* values for imaging error and digitizing error were much smaller than the individual*side interaction (the first biological interaction) ,which indicates that the variation present in the data as a result of imaging and digitizing is negligible (Table 4.7). No significant difference existed between left and right wings (side) therefore this data were combined for all further analyses.

Table 4.7: Procrustes ANOVA for shape effects quantifying measurement error at two levels for *Nomadacris septemfasciata* forewings.

Effect	SS	MS	df	F	P
Side	0.00029446	0.0000210330	14	0.0002	0.9473
Ind*Side	0.03409083	0.0002705621	126	28.87	<0.0001
Imaging error	0.00262392	0.0000093711	280	2.43	<0.0001
Digitizing error	0.00216062	0.0000038582	560		

The principal component analysis showed that the great majority of the variation was accounted for by the first principal component (PC) (40.653%), with the first and the second making up 59.7% of the variation (Figure 4.7). The shape changes associated with the PCs represent specific shifts of landmarks in various regions of the wing (Figures 4.8 and 4.9). The transformation grid of the first PC indicates that almost all the variation observed was in a lateral direction, indicating that wing shape differed in terms of their length more than their width (Figure 4.8). The landmark situated at the proximal end of the wing (landmark 1) contributed substantially The Procrustes ANOVA indicated that the mean square (MS) and *F* values for imaging error and digitizing error were much smaller than the individual*side interaction (the first biological interaction) ,which indicates that the variation present in the data as a result of imaging and digitizing is negligible (Table 4.7). No significant difference existed between left and right wings (side) therefore this data were combined for all further analyses.

The principal

to overall shape variability. Similarly, the distal, anterior part of the wings (represented by landmarks 5, 6, 7 and 9) also contributed greatly to overall variation.

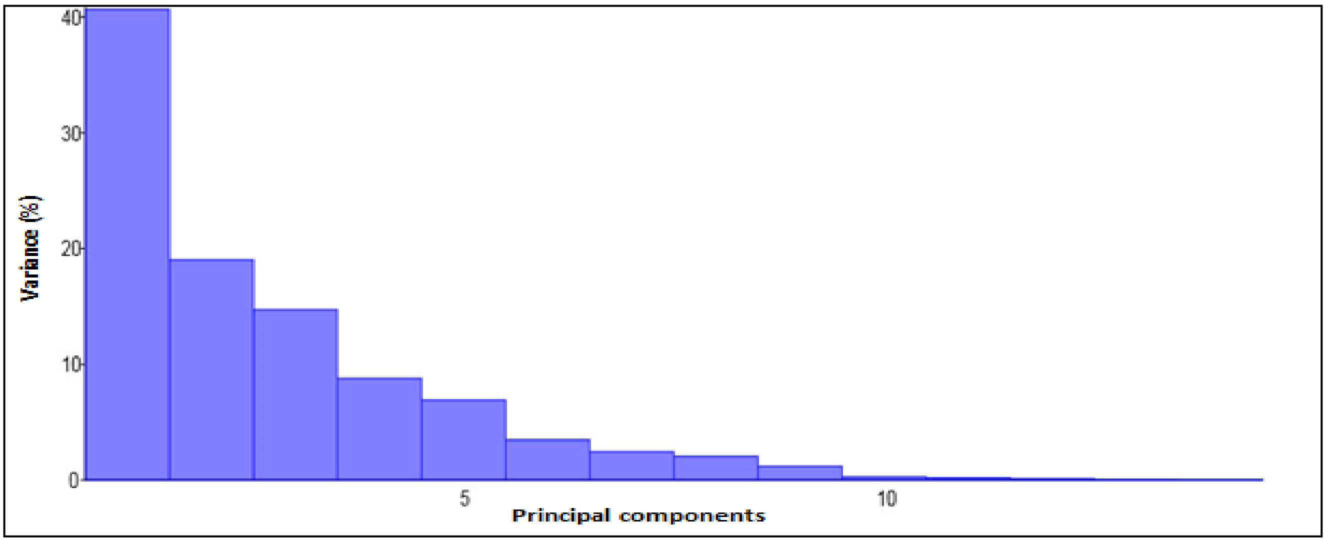

Figure 4.7: Principle components analysis (PCA) illustrating that the first two PC's contribute to the majority of the variation in wing shape in Nomadacris septemfasciata.

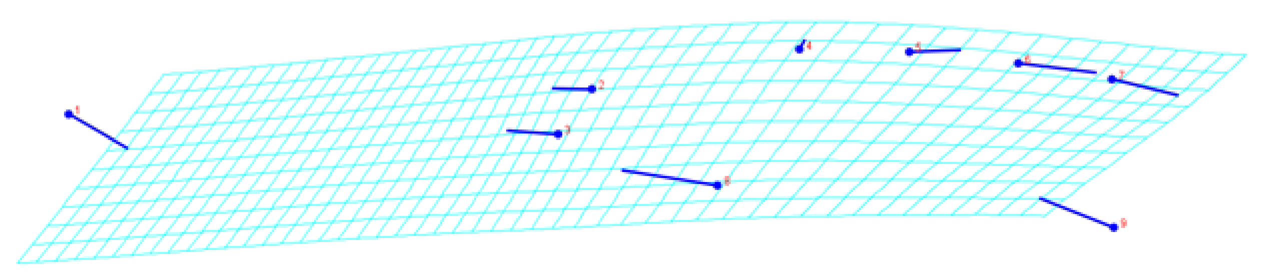

Figure 4.8: PC 1 transformation grid showing the shape change, using the thin plate spline, of nine landmarks (at a scale factor of 2) on the total forewing dataset for Nomadacris septemfasciata collected from Jengro and Kelly. The dot represents the consensus shape, while the line represents the shift of the landmark associated with that particular shape change.

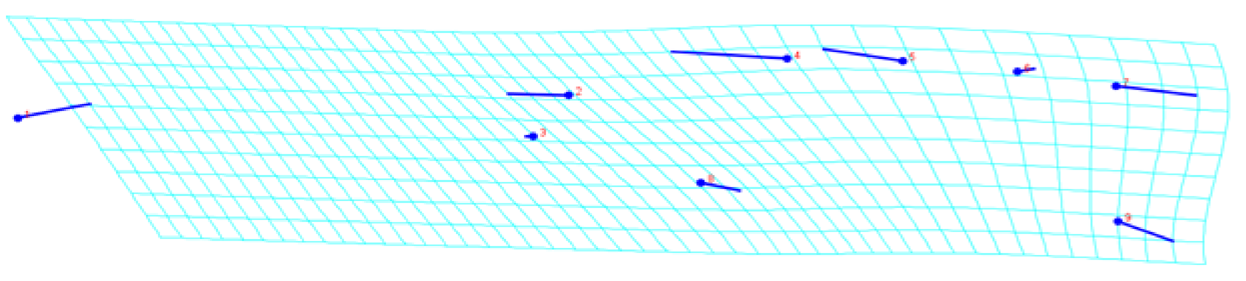

Figure 4.9: PC 2 transformation grid showing the shape change, using the thin plate spline, of nine landmarks (at a scale factor of 2) on the total dataset forewing for Nomadacris septemfasciata collected from Jengro and Kelly. The dot represents the consensus shape, while the line represents the shift of the landmark associated with that particular shape change.

Differences between populations according to location were significant according to the DFA, however separation of groups was not very precise, although significant at the 5% level (Table 4.8). The separation of samples of the original scores seem to be somewhat inflated, however, compared to the cross-validation scores (Table 4.8).

Table 4.8: Classification matrix from Discriminant Function Analysis comparing two *Nomadacris septemfasciata* populations by location with gender excluded as a variable.

Discriminant Function Analysis			
Comparison:	Jengro-Kelly		
T-square	34.3183		
P-value	0.0179		
From discriminant function:			
	Jengro (transiens)	Kelly (gregarious)	Total
Transiens	35	15	50
Gregarious	16	34	50
From cross-validation:			
	Jengro	Kelly	Total
Transiens	27	23	50
Gregarious	23	27	50

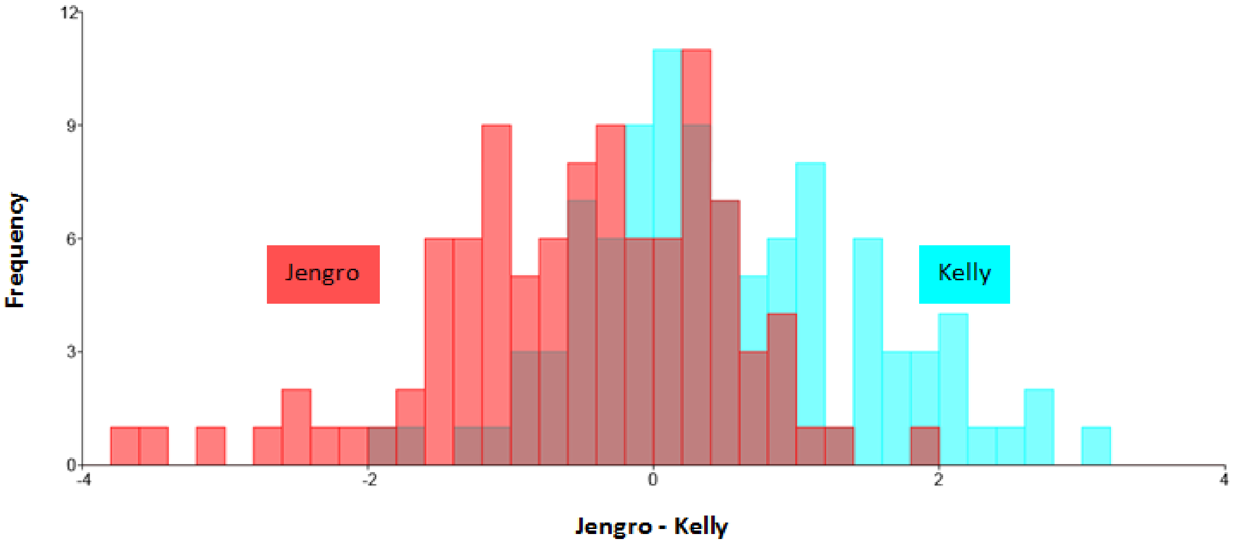

Figure 4.10: Discriminant analysis graph illustrating the difference in wing shape of *Nomadacris septemfasciata* between Jengro and Kelly.

The discriminant analysis (Figure 4.10) illustrates a significant degree of separation between the two populations, however this separation is not as distinct compared to the level of separation seen in terms of gender (Table 4.9). The eigenvalue was 0.261, which represented 100% of the variation, meaning that all of the separation was achieved using the first canonical variate. According to the results of the DFA, a significant difference existed between wing shape of males and females, as also indicated by group allocation (Table 4.9).

The discriminant analysis (Figure 4.11) illustrates very clearly the large separation of groups in terms of gender and cross-validation scores are more similar to the original analysis than the case with separation according to location. The eigenvalue was 1.634, accounting for 100% of the variance.

Table 4.9: Classification matrix from Discriminant Function Analysis comparing two *Nomadacris septemfasciata* populations by gender with location excluded as a variable.

Discriminant Function Analysis			
Comparison:	Male-Female		
T-square	180.4715		
P-value	<0.0001		
From discriminant function:			
	Male	Female	Total
Male	46	4	50
Female	5	45	50
From cross-validation:			
	Male	Female	Total
Male	42	8	50
Female	9	41	50

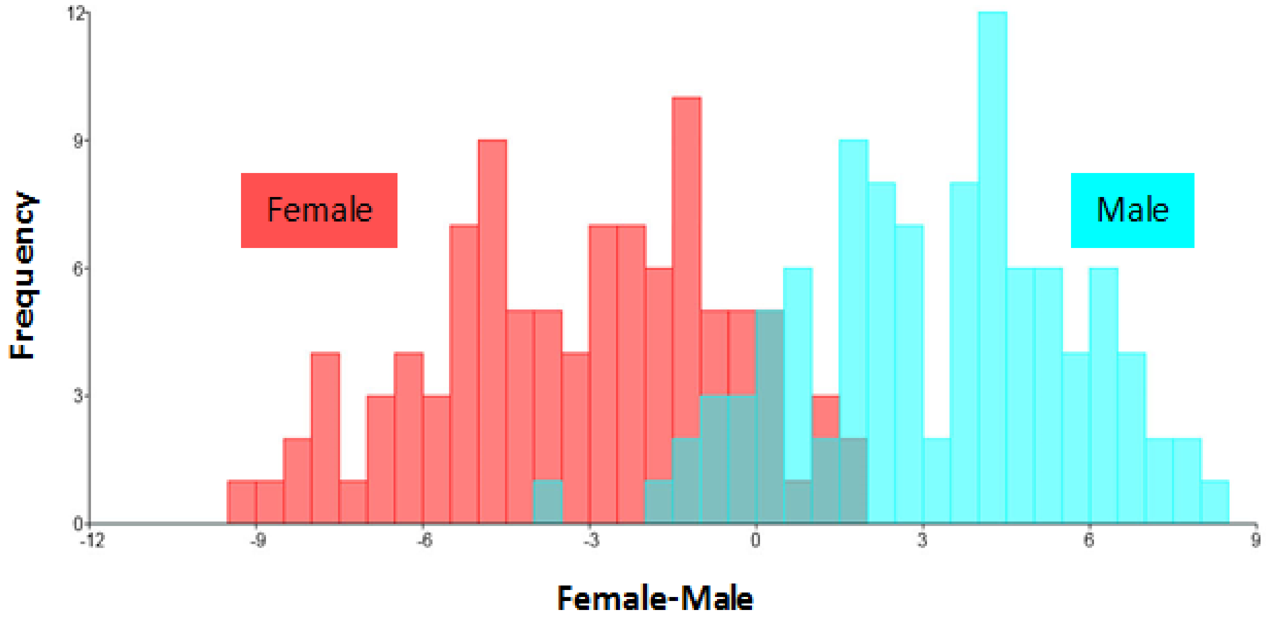

Figure 4.11: Discriminant analysis graph illustrating the difference in wing shape between male and female *Nomadacris septemfasciata* collected in Empangeni.

4.4 Discussion

4.4.1 Hopper colouration

The vast majority of hoppers observed exhibited colours matching the description of gregarious or transiens individuals. A semi quantitative method as used by Lecoq et al. (2011a) was used as a guideline in this when relating hopper colour to phase status. Observations indicated that highly melanised orange/yellow hoppers with the black spot on the hind femur (Figure 4.4) were seen very often, indicating that the transiens/ gregarious phase hoppers were by far, in the majority.

4.4.2 Traditional Morphometrics

The close similarity in the mean E/F value between GSA and Kelly farms is most likely because they are situated within 2km of each other. The environmental conditions may thus be similar and the possibility of individuals migrating from one farm to another is large. It is well known that *N. septemfasciata* adults are very mobile (Faure 1935). Rainey et al. (1957) and Franc et al. (2005) both found that E/F ratios are higher in males than in females, as is the case in this study. Rainey et al. (1957) also found that low variances in E/F ratio are typical of either of the extreme phases (gregarious or solitarious). With regard to this study, variances were equal or lower compared to the findings of Rainey et al. (1957), which strengthens the argument for gregariousness in the Empangeni *N. septemfasciata* populations. It must be mentioned though that present measurement techniques may be more accurate than in 1957, which may have contributed to the low observed variances. Burnett (1951b), in the Rukwa valley, Tanzania compared populations of *N. septemfasciata* over two generations and concluded that they were becoming more solitary due to the decreasing E/F ratio. By comparing the E/F ratio over two seasons (2012 vs. 2013) in Empangeni, it is evident that the current generation (2013) of *N. septemfasciata* in Empangeni is becoming more gregarious than the previous generation, meaning they may exhibit a higher propensity for swarming and further outbreaks. According to the classification functions used by Rainey et al. (1957) the majority of specimens measured in this study were in the transiens phase. The classification functions used can be considered fairly conservative considering that Franc et al. (2005) used an E/F ratio of 2.0 to separate gregarious forms from solitarious forms, while in the present study a value of 1.87 was used as the limit between solitarious and transiens and 2.05 was the limit between transiens and gregarious.

4.4.3 Eye stripes

Eye stripe data indicates that the vast majority of specimens observed were of the ‘seven stripe gregarious type’ as in Burnett (1951a). In 2013, more specimens were found with the extra eye stripe, typical of the solitarious individual compared to 2012, which therefore seem to contradict the E/F ratio results. This difference however, was fairly small. Current literature searches reveal that no studies have used eye stripes as an indicator of phase since the 1950’s (Burnett 1951, Albrecht 1957) which may suggest that it is not a very reliable indicator of phase status. According to this literature, eye stripes are only indicative of the gregarious and solitary phase, the present E/F ratios indicate that the majority of specimens measured are in the transiens phase which results in this method being fairly limited in its ability to accurately determine the phase status of *N. septemfasciata*.

4.4.4 Geometric morphometrics

There was a highly significant separation in wing shape between males and females according to the DFA (Figure 4.11). This indicates that sexual dimorphism (both in size and shape) is prominent in this species, supported also by the similar cross-validation scores to the original analysis. *Nomadacris septemfasciata* males tend to be more gregarious than females, often exhibiting higher E/F ratios than their female counterparts (Michelmore & Allan 1934, Bredo 1938, Burnett 1951 Rainey et al. 1957, Franc et al. 2005). This could possibly be the reason why males and females showed such a large disparity in wing shape. Very little research has been conducted on the use of geometric morphometrics in Orthoptera and even less on gender differences (see Klingenberg et al. 2010, Beasley et al. 2012, working on cricket and grasshopper wings, respectively). However, in stingless bees (Apidae: Meliponini), it is known that males and workers have different behaviours and flight activities during their lifespan which could lead to differentiation in the patterns of wing venation (Francoy et al. 2009). Similarly, Glanville fritillary butterfly's *Melitaea cinxia* Linneaus (Lepidoptera: Nymphalidae) dispersal ability and forewing shape is associated with the different life-history traits of the sexes (males may need to fly further than females), resulting in different selection pressures being enforced on wing shape in each of the sexes (Breuker et al. 2007).

Differences in wing shape when comparing individuals from Jengro and Kelly locations were investigated, with the predetermined assumption that any shape differences observed would be due to the different phase status of the populations at the Jengro and Kelly farms. A significant difference did exist (although small) between the two locations. Kelly harbours more gregarious populations while Jengro harbours a more solitarious population (See Figure 4.5/Table 4.2). The cross-validation results were very different from the original analysis, which indicates that the separation is not that clear, either due to the actual separation between gregarious and transiens phases not being that clear-cut between the farms or due to sample size not being large enough (100 specimens). The results of this chapter, however, clearly indicate that between Jengro and Kelly populations, a statistically significant difference in the E/F ratio (from traditional morphometrics) and wing shape (from geometric morphometrics) does exist. The potential for long distance dispersal in this species is high, given their highly mobile nature and therefore it could be presumed that an appreciable amount of dispersion occurs between these populations. The results, however, seem to indicate that these populations are quite separate in terms immigration and emigration due to their significantly different E/F ratios even though they are only separated by a few kilometres It has already been stated that *N. septemfasciata* populations at all survey sites are quite sedentary, with minimal overall movement throughout the season, a phenomenon also noted by Rainey et al. (1957) who 'observed the often static nature of populations' in the Rukwa valley, Tanzania. Lea (1935) also noted that in 1934, 'a large *N. septemfasciata* swarm originating from the north settled on a 3000 acre sugar estate in Entumeni, Zululand and spent at least six weeks confined to this area which was in stark contrast to the mobility of swarms in uncultivated bush country further north.' Therefore it is proposed that the current behaviour of swarms and the geographical separation of these two populations are far enough to ensure that they develop in relative isolation to one another and therefore are likely to show differences in terms of their E/F ratios and wing shape. Population densities at Kelly farm are also a lot higher than at Jengro farm (Chapter 2).

4.4.5 Resource quality and distribution in relation to swarming and phase change

It is well known that favourable weather plays a key role in promoting crowding and subsequent population increases, swarming and plagues (Camuffo & Enzi 1991) in locusts. Habitat structure is also a critical factor which initially promotes behavioural gregarization and ultimately leads to local crowding and population increases (Sword et al. 2010). A heterogeneous habitat with patchily distributed host plants tends to concentrate populations and this promotes crowding (Simpson & Sword 2006). Michelmore (1947) found that *N. septemfasciata* adults showed a marked preference for tall dense grass while hoppers preferred shorter grass, but upon being disturbed they immediately made for tall clumps of vegetation for shelter, which may act as concentrating factors. A sugarcane field is certainly not a heterogonous habitat and cannot be described as having a clumpy distribution of host plants, therefore this finding cannot explain why *N. septemfasciata* seems to be showing gregarious tendencies in Empangeni.

Features of a habitat influence the distribution of populations in terms of finding food and microclimate for basking and shelter (Sword et al. 2010). Duranton (1997) noted that agriculturally induced environmental modifications lead to better ecological conditions for *N. septemfasciata*, and could result in more frequent gregarization in the future, while clumping of resources such as food plants, roosting/basking sites or areas of favourable microclimate encourages solitarious locusts to come together and behaviourally gregarize (Bouaïchi et al. 1996). It has been observed in Empangeni on numerous occasions that sugarcane breaks (the roads intersecting blocks of sugarcane) seem to be acting as an attractant to individuals for basking purposes. *Nomadacris septemfasciata* swarms will typically descend from the sugarcane plants at dawn and sit on the road flanking the sun until the temperature has increased enough to allow them to fly. Similar behaviour was observed in the Rukwa valley by Rainey et al. (1957) in natural grasslands. The reason for this occurrence is presumably because sugarcane is very dense and does not provide an optimal basking site and therefore they move onto the road which often gives 100% exposure to the sun from fairly early in the morning. Observations from the present study indicated that basking locusts were commonly found on three sugarcane breaks at Jengro, Kelly and GSA farms in particular, the common factor between the three being their orientation to the sun. All of the roads roughly face in a north-eastern direction, which presumably will receive the sun the earliest as the sun rises in the east. Simpson et al. (2006) states that gregarization potential depends critically on the fine scale distribution and quality of resources, therefore it is possible that the cane breaks are acting as the 'heterogeneous', aggregating factor which *N. septemfasciata* require within this environment to start to gregarize. Lea (1935) states that the sugarcane fields of Zululand 'may be regarded as grassland habitats comparable in general facies with the long grass stands of the Rukwa valley.'

An empirical rule which has been repeatedly proven to be correct (see: Golding 1934, Michelmore 1947) is that *N. septemfasciata* swarms are more active in short grasslands and bush compared to long grassland habitat. This possibly explains why *N. septemfasciata* populations in Empangeni seem to be quite sedentary and no significant migrations have been observed. We can thus assume, as a general rule, that populations of this species in sugarcane fields are likely to remain confined to the areas where they are initially observed. The International Red Locust Control Service (1937) (presently the IRLCOCSA) considers that 'the movement of a swarm out of long grass and into surrounding short grass, very often precedes movement on a much larger scale, out of the general area'. Opinions such as these should be noted, and incorporated into monitoring protocols in the Empangeni region. The relationship between hopper colour and hopper density is also very important as it indicates when a population may reach the gregarization threshold and start to swarm, therefore this factor should also be incorporated into future survey protocols for this species.

4.4.6 Harvesting regime and oviposition sites

Sugarcane burning prior to harvesting is common in the Empangeni growing region, and this normally occurs from April through to late November/early December. Symmons and Carnegie (1959) found that *N. septemfasciata* significantly preferred areas of burnt grass over unburnt grass in terms of oviposition preference. Lea (1935) found that ground covered by an accumulation of trash was not favoured for oviposition. Faure (1935), Whellan (1968) and Lea (1935) all found that the *N. septemfasciata* females will lay eggs in practically any type of soil. Therefore, it is probable that burning is the critical factor in this region, providing exposed areas of soil, thus providing ideal oviposition sites for *N. septemfasciata* females in sugarcane fields. The oviposition period of *N. septemfasciata* from October to December (Chapter 2) coincides with the height of the harvesting (and thus burning) season in Empangeni, which exacerbates the problem. Although it seems that fire will not kill *N. septemfasciata* (Lea & Webb 1939), it may influence their distribution by breaking up swarms (Burnett 1951b) or possibly causing widely distributed populations to come together on unburnt sugarcane areas (personal observations). The possibility of both of these scenarios occurring is likely, given the vastly different situations on different farms in terms of harvesting scheduling and geography of the farm.

4.5 Conclusion and Recommendations

Nomadacris septemfasciata populations in Empangeni sugarcane exhibit characters closer to the gregarious phase in terms of morphometric data, eye stripes and hopper colour, although population densities and adult behaviour tend to indicate a lower degree of gregariousness. The data indicating a high degree of gregariousness is based on quantitative measurements, and therefore should be regarded as more reliable. The phase state of *N. septemfasciata* populations is sometimes given on the basis of an expert, but qualitative, opinion (Lecoq et al. 2011a). This can be problematic as populations can be miscategorised purely on the basis that they exhibit gregarious tendencies. It is thus important that quantitative data is captured, which ensures the validity of the observations made. A technique offering the relevant quantitative data collection and analysis for phase analysis is geometric morphometrics. It is recommended, however, that in order to propose geometric morphometrics as a suitable method to assess phase status, a study be conducted with more samples (>100) and that wing shape (e.g. centroid size) and E/F ratios be measured on the same individuals. This analysis can therefore combine both measurements and therefore provide more accurate recommendations.

The chance of Empangeni *N. septemfasciata* populations to start swarming, resulting in outbreaks should be regarded as possible. The likely outbreak areas will be in the vicinity of Kelly and GSA farms, where N. *septemfasciata* populations are currently most dense. Whether it happens or not depends on a number of factors becoming favourable for them. These include climate, resource distribution, management practices and the crop protection measures which are used.

Environmental factors which promote gregariousness and swarming are already present in Empangeni as has been discussed. Therefore, it is paramount that methodical population monitoring takes place, so that population upsurges can be recognized as soon as possible to allow for effective mitigating actions to be implemented in a timeous manner. The role of fire as a possible aggregating or dispersing factor should be further investigated, as it has the potential to significantly affect the distribution of *N. septemfasciata* swarms on particular farms.

4.6 References

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Chapter 5: Conclusions and Recommendations

The results of this study provide the first detailed identification and account of acridid outbreaks in Empangeni sugarcane. The impact of this acridid complex in terms of damage to sugarcane is discussed and the potential for swarming and large scale outbreaks to occur in *N. septemfasciata* populations. In the general introduction, the principles and characteristics of acridid outbreaks were discussed. The difference between grasshoppers and locusts was defined and the known causes for population upsurges and outbreaks to occur were outlined. Possible reasons for the outbreaks occurring in Empangeni sugarcane were highlighted. Chemical insecticides have been the mainstay of acridid management for decades, however, the concept of integrated pest management (IPM) in relation to acridid pests is slowly gaining ground as the limitations of chemical insecticides (non-target effects, environmental pollution, high economic costs) are realised (Lomer 1999, Belayneh 2005, Bitzadze 2013). Therefore, the data obtained from this study will form the basis on which to develop an integrated pest management (IPM) plan against these pests. The aim of this study was to investigate the biology and ecology of acridid outbreaks in Empangeni sugarcane. Each major objective will now be discussed.

5.2 Objective 1: A year-long population survey to determine the species composition and relative abundance of the species associated with these outbreaks and Objective 2: Determination of their population dynamics and life history traits.

Literature shows that population sampling is imperative in order to study any insect either in conservation studies or in a crop pest context (Morris 1960, Gardiner et al. 2005). Chapter 2 therefore, deals with the morphological and molecular identification of the acridid species complex present in Empangeni sugarcane. It was shown that species composition and abundance differs substantially both spatially and temporally within sugarcane fields. Species abundance and composition in adjacent grassland sites also differed substantially. Correspondence analyses showed that *N. septemfasciata* and *P. prosternalis* were closely associated with sugarcane sites while species such as *Z.elegans* and *C. aeruginosa* were more closely associated with grassland sites, indicating that they pose a lesser threat to sugarcane growers. This is consistent with other studies which showed that acridid species exhibit choice preferences within an agri-ecosystem (Coop & Croft 1993). Identifying and quantifying the acridid species complex in Empangeni and surrounding grassland was an important first step in gaining knowledge of the population dynamics and life history traits of target species allowing for more accurate and effective management decisions to be made. From this study it was found that all species are univoltine but two different types of life cycles were observed: *Nomadacris septemfasciata* and *O. cyanea* exhibited an adult winter reproductive diapause type of life cycle while *P. prosternalis, C. aeruginosa* and *C. zuluensis* exhibited a winter egg diapause type of life cycle.

5.3 Objective 2: Determine the pest status of the different species in terms of their economic significance to affected growers

Literature detailing the effect of acridid feeding on crops is limited (Coop & Croft 1993) and in terms of sugarcane, even more limited (Williams et al 1969, Easwaramoorthy 1989). According to field observations, damage as a result of acridid feeding varied between farms and in relation to the time of year. The fluctuations in damage over time were closely associated with the population density of acridid species on each farm. Thus, by knowing the life cycle of the acridid species it is possible to determine which species are responsible for the damage depending on what time of year defoliation occurred. Feeding trials are the basis for crop loss estimates and to determine the economic threshold (ET) and economic injury level (EIL) (Coop & Croft 1993). Feeding trials were completed in the laboratory for the two principal species in Empangeni, *N. septemfasciata* and *P. prosternalis*. Results indicated that during the time the trials were conducted, *P. prosternalis* had significantly higher feeding rate in males and females compared to those of *N. septemfasciata*. Hoppers of *N. septemfasciata* had a higher feeding rate than the adults, while *P. prosternalis* hoppers had a lower feeding rate compared to the adults. Females of both species had higher feeding rates compared to males. Feeding rate has been shown to be significantly, positively correlated with acridid dry weight (Hewitt & Onsager 1982, Coop & Croft 1993). This finding held true with regards to *P. prosternalis* hoppers versus adults and for males versus females of both species. It did not hold true when comparing *N. septemfasciata* hoppers versus adults and when comparing *P. prosternalis* adults to *N. septemfasciata* adults (of both sexes). The feeding potential of these two species of acridids was thus quantified, which provides useful insights into the potential damage each species is able to cause on sugarcane. However, to make accurate ET and EIL calculations, data on growth rate of sugarcane and acridid density within sugarcane fields is required. Field trials should also be conducted as the applicability of data from laboratory trials to a field situation is problematic. Field trials could be conducted as in Coop & Croft (1993) who used plants within a crop, covered with netting. This would ensure that the conditions under which trials took place would be comparable to the real situation.

5.4 Objective 3: Make inferences on the possible causal factors for outbreaks

Chapter 1 discussed the main factors responsible for insect outbreaks in general, and acridid outbreaks specifically. The factors involved are often very complex and have a number of direct and indirect interactions (Joern & Gaines 1990). It seems that abiotic factors such as precipitation and temperature more often than not play a critical role in population regulation (Andrewartha & Birch 1954). This study was unable to specify the complex causes for orthopteran outbreaks in sugarcane fields over a two year period, as this was the first time that their abundance was quantified on a regular basis. However, long term population surveys will likely result in a link being found between weather patterns and population fluctuations, as has been proven in other studies (Chiconela et al. 2003, Maiga et al. 2008).. From seven week sampling period comparing sugarcane to semi natural grasslands, it was found that certain species within the acridid complex significantly prefer sugarcane. Sugarcane may be supplying optimal habitat for these species and may be acting as either an ecological source or sink for these species causing their abundance to be higher in sugarcane than in other nearby habitats. Furthermore, these optimal conditions may cause the acridids reproduction to be elevated in sugarcane., These optimal conditions could be related to improved host plant quality, better microclimate, decreased predators or a host of other factors It may not always be possible, however, to manipulate certain factors. Understanding the basic life history of the species is paramount to making an informed decision on the most effective way to manage them, as a first step in IPM.

5.5 Objective 4: Determine the potential for swarming and to measure and understand the morphological differences between geographically separated populations using morphometrics.

Chapter four deals with the phase status of *N. septemfasciata* populations in Empangeni and uses both traditional and geometric morphometrics as well as two other known techniques to try to determine this. As in previous studies, the measured E/F ratio (Rainey et al. 1957, Franc et al. 2005, Lecoq et al. 2011) was found to be the most important character to use in determining phase status., The number of eye stripes (Burnett 1951, Albrecht 1957) and hopper body colour (Faure 1935, Lecoq et al. 2011) have been used as indicators of phase status of the red locust, but were not found to be accurate characters to use in this study. The data collected over two seasons show that *N. septemfasciata* are consistent with a transiens or gregarious population, shifting in the direction of becoming more gregarious. Geometric morphometrics, a relatively new technique of measuring wing shape changes, was tested on *N. septemfasciata* populations to draw comparisons between the results from traditional morphometrics in an attempt to evaluate the efficacy of using geometric morphometrics as a method of phase determination. Although this method was highly sensitive to assessing sexual dimorphism in wing shape of this species, the sample size was most likely not large enough to assess phase changes, although some differences could be established.

5.6 Development of an integrated pest management plan.

The knowledge acquired about the acridid complex in Empangeni should be used in the development of an IPM plan. IPM strategies against locusts and grasshoppers have been successfully implemented in Australia (Hunter 2010), in areas of North America (Branson 2006) and currently are the subject of various studies into its efficacy and sustainability in all areas affected by acridid outbreaks (Lomer 1999, Peveling 2001, Belayneh 2005). An integrated approach to acridid control in Empangeni should involve the strategic use of synthetic insecticides and, guided by a scouting and monitoring programme of acridid populations. Growers should also incorporate management practices which are deemed to decrease acridid populations, for example providing a cover on recently harvested sugarcane to reduce acridid oviposition sites, and judicious burning practices, especially during the oviposition period of the problem acridid species. An integrated approach such as this has the best chance of sustainably managing acridid populations to keep them below their EIL. How the results of this study can be implemented within a management strategy will now be briefly discussed in the sections below.

5.6.1 Monitoring protocol

A monitoring programme should be based around the goal of detecting hopper emergence, clear identification of the species, and if the species is deemed problematic to sugarcane, spraying them with an approved insecticide, as indicated on the insecticide label, as soon as possible. Early stage hoppers are the most susceptible to insecticides and will be fairly clumped after emergence. This enables more targeted spray applications. Designated monitors (growers, extension officers or pest and disease officers) should observe acridid populations throughout the year. A critical period of monitoring is the wet summer months from October – February. *Nomadacris septemfasciata* oviposits during September-November, often in concentrated areas, therefore every effort should be made to scout for oviposition sites. The oviposition site is very likely to be close to the site where the swarm is currently present. Locating the oviposition site will ensure the early detection of hopper emergence sites which is crucial in an effective spray application, or other more benign control operations, such as ploughing of the oviposition site (Symmons & Carnegie 1959) resulting in high hopper mortality, reduced non-target effects and decreased costs. Monitoring of the other non-swarming species is also very important but oviposition will not occur in clumped areas like *N. septemfasciata*, therefore emergence of hoppers is likely to be distributed over a wider area. Recording the location of oviposition and emergence sites over time should be done. This will allow growers to detect high risk areas over time, resulting in more accurate and effective spray applications and or other management options such as ploughing, in the future.

5.6.2 Management practices

It is known that areas of bare ground are favourable for *N. septemfasciata* oviposition (Symmons & Carnegie 1959). Therefore growers should make an effort to reduce the amount of bare ground on their farms during harvesting. This can be done by trashing sugarcane instead of burning, a proposition already put forward by Lea (1935). By reducing the amount of bare ground on a farm will decrease the chance of *N. septemfasciata* ovipositing there and even if they do, egg survival may be decreased by the presence of a vegetation layer on the soil surface (Lea 1935, Symmons & Carnegie 1959). Weed management in sugarcane is very important, from an agronomic perspective. Coop & Croft (1993) found that good weed management was associated with decreased numbers of a host of acridid pest species that they studied, since many acridid species are polyphagous (Whellan 1968). Therefore, weed management should be prioritized due to the dual benefit it may provide.

5.6.3 Control Strategies

The effectiveness of synthetic pesticides as an immediate crop protection method is well known and accepted (Lomer et al. 1999), therefore they will remain the first choice of control operations for the foreseeable future, although they need to be used more efficiently and in conjunction with other control options. Currently in the

Empangeni area, they are unfortunately sprayed quite indiscriminately, regardless of acridid species, life stage, time of year, time of day or application method used. The acquired knowledge from this study and from past control measures should be used to streamline the insecticidal spraying of acridids. South Africa has had a proactive history of introducing safer and more environmentally benign acridicides (Price 1999) and in Empangeni there is a clear need to investigate other control options as the exclusive and widespread use of synthetic pesticides is becoming less effective, more costly and possibly causing unnecessary and avoidable environmental consequences. A case in point is the observed aggregation of high numbers of *N. septemfasciata* adults along sugarcane breaks. The swarms aggregate here to bask in the early morning sun. This phenomenon/behaviour can be exploited by exploring the possibility of using a contact insecticide/biocide along these sugarcane breaks, which may well prove to be a very efficient method of control. Other control options such as bran baits, which have been widely used for many years (Uvarov 1928, Foster et al. 1996), should be considered. Uvarov (1928) stated that: “the poison bait method is exceedingly simple and very effective and is the best method to use against solitary grasshoppers”. Fungicides Bio-insecticides such as the commercially available Green Muscle® which consists of spores of the fungus *Metarhizium flavoviride* Gams & Rozsypal (Deuteromycotina: Hyphomycetes) (Langewald 1999) should also be seriously considered for extensive trials in Empangeni. Green Muscle® is known as an effective biological pesticide against grasshoppers and locusts which has been extensively tested in the field. It has been shown to be effective and persist (Lomer et al. 1999). *Beauveria bassiana* (Bassi) (Hypocreales: Clavicipiticeae), identified by the Agricultural Research Council – Plant Protection Research Institute ARC-PPRI (Accession number: M183/788a) was recorded in October 2012 by the author of this thesis as the cause of death for a number of field collected adult *N. septemfasciata* specimens. This entomopathogenic fungus has already been proposed as a possible bio-control agent against *N. septemfasciata* (Schaefer 1936, Foster et al. 1996), therefore its use should be further investigated. Crop protection tactics do not stop the locust breeding cycle (Showler 1991), therefore the main objective of the IPM plan should be based around strategic management, aiming to manage acridid populations by targeting the sexually immature hopper stages to break the breeding cycle.

This study therefore managed to establish some basic biological information to initiate a management plan against these acridid pests on sugarcane in Empangeni. It was found that the problem is persisting and is likely to become worse if current control strategies continue.

The following recommendations are made for future research:

- Continuation of population surveys over the long term in order to relate to climate data.
- Increase survey area to gain a better understanding of population abundances, regionally.
- Investigate the use and efficacy of Bio – pesticides such as Green Muscle®.

- Undertake host plant preference trials to ascertain acridid feeding preferences.
- Conduct feeding trials in a field situation so that results are more comparable.
- Investigate alternate control techniques such as destroying acridid egg beds, insect growth regulators and poison baits

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APPENDICES

Appendix 1

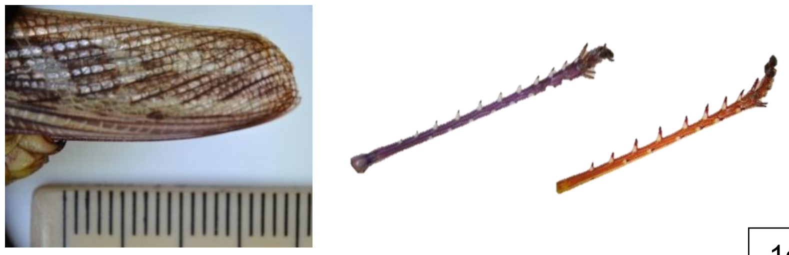
Adult Identification Key for Five Acridid Species in Empangeni Sugarcane

1a) Wings extend far (>/= 10mm) past the tip of the abdomen and hind tibia red or purple… 2a
1b) Wings do not extend far (</= 5mm) past the tip of the abdomen. Hind tibia either brown or blue… 4a

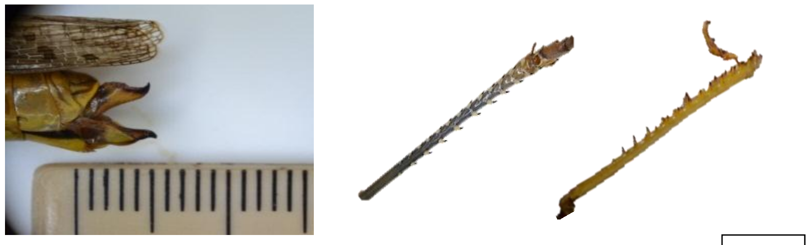
2a) General body colour green, hind wings are transparent, yellow basally… *Cyrtacanthacris aeruginosa*
2b) General body colour not green… 3a

3a) Pronotum is distinctly convex. Starting at the base of wing, majority is vermillion red… *Ornithacris cyanea*
3b) Pronotum is not convex, rather flat or concave. Hind wing pink/purple, basally… *Nomadacris septemfasciata*

4a) Hind tibia blue/grey, abdomen yellow ventrally, dorsal surface with two green longitudinal stripes… *Cataloipus zuluensis*
4b) Hind tibia not blue, general body colour a uniform beige/brown… *Petamella prosternalis*

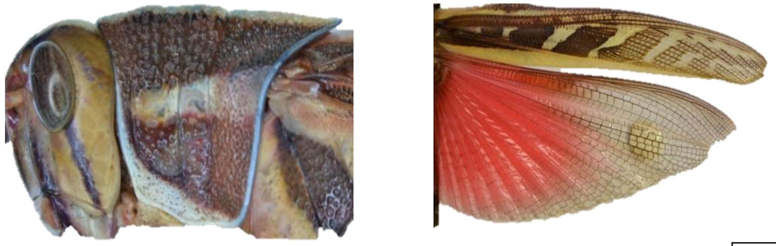


1a

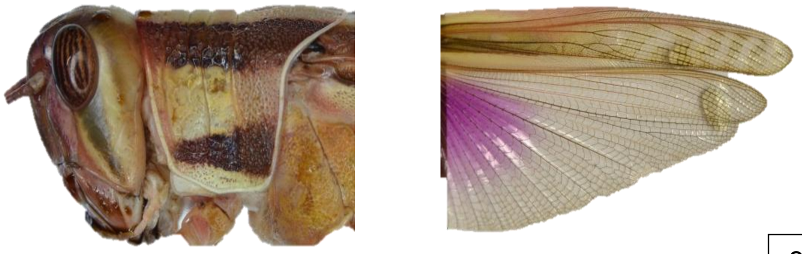


1b

2a



3a



3b

4a

Appendix 2

Hopper Identification Key for Five Acridid Species in Empangeni Sugarcane

1a) Antennae are black… 2a

1b) Antennae are not black… 5a

2a) Prominent subocular stripe present under the eye… 3a

2b) No subocular stripe present under the eye… 4a

3a) General background colour green, eye stripes present, may have varying amounts of black spots over entire body …*Nomadacris septemfasciata* (Solitarious phase)

3b) General background colour orange, with prominent hind femur spot. Varying mixture of black orange and yellow...*Nomadacris septemfasciata* (Gregarious phase)

4a) Relatively stout, round body with general green background colouring and eye stripes visible … *Cyrtacanthacris aeruginosa*

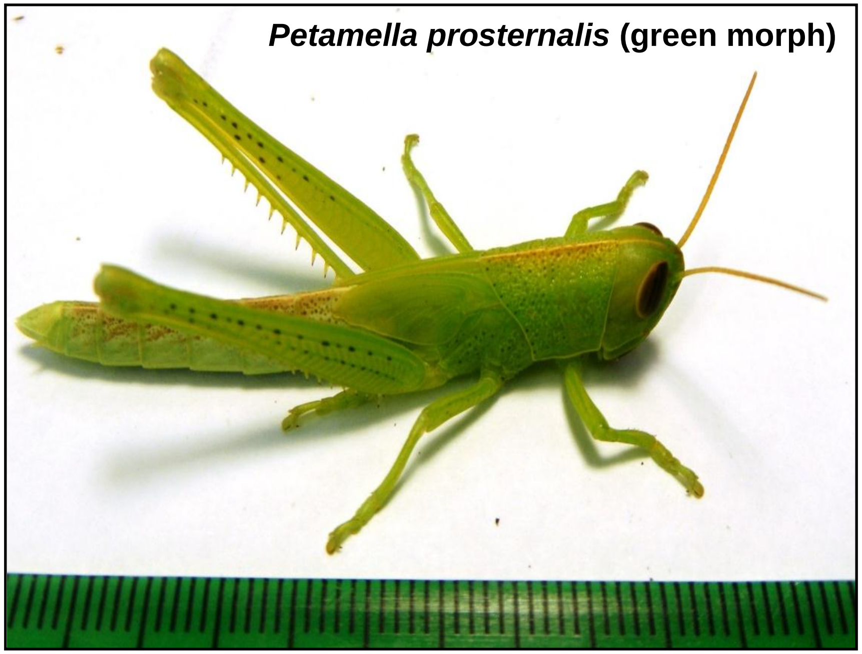
4b) General body colour brown with relatively long hind femur and no eye stripes visible… *Cataloipus zuluensis*

5a) Prominent subocluar stripe and prominent ridge on pronotum... *Ornithacris cyanea*

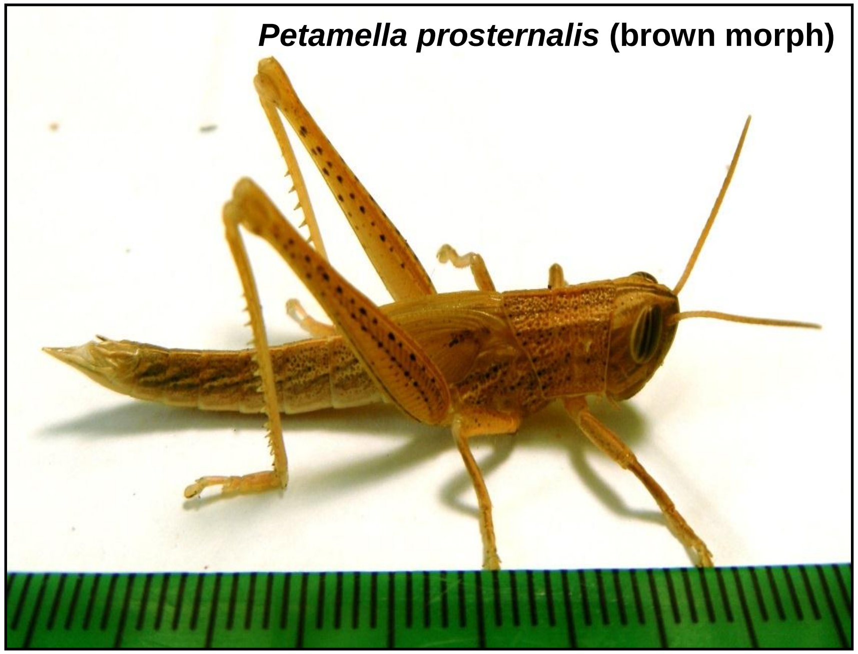
5b) No subocular stripe, pronotum not ridged, rather flat… 6a

6a) General background colour green… *Petamella prosternalis* (Green morph)

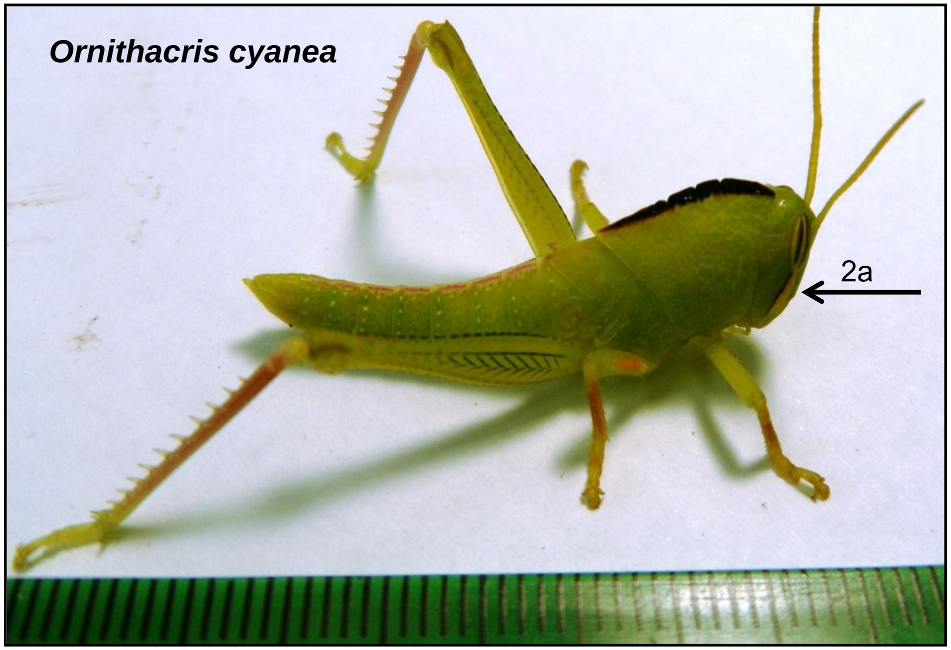
6b) General background colour beige/light brown... *Petamella prosternalis* (Brown morph)



***Petamella prosternalis* (green morph)**

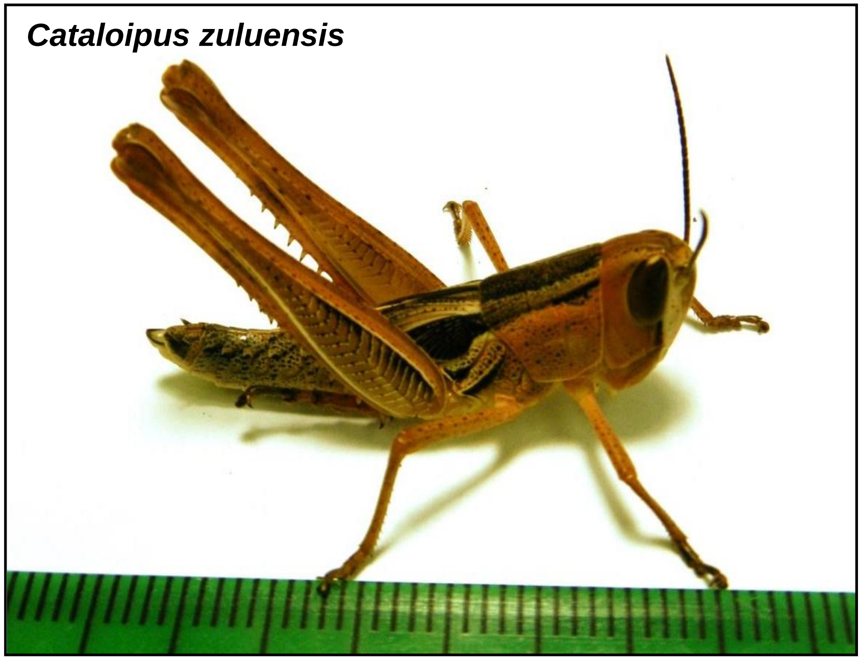


***Petamella prosternalis* (brown morph)**

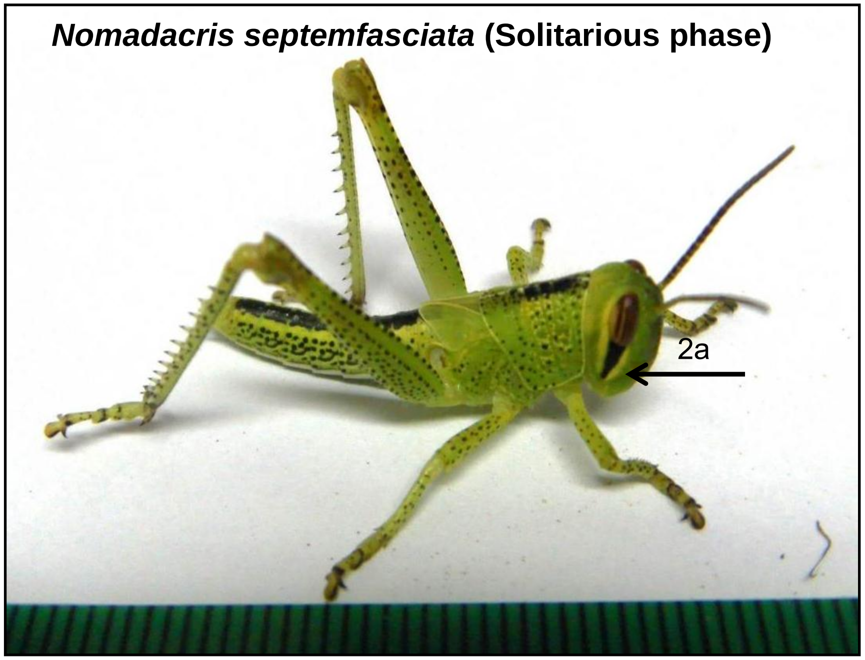


Ornithacris cyanea

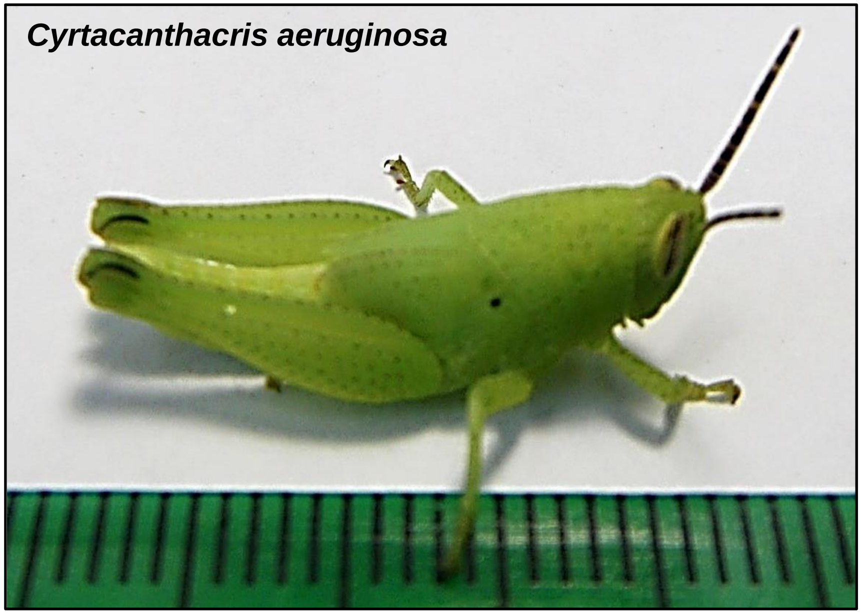
Cataloipus zuluensis



***Nomadacris septemfasciata* (Solitarious phase)**



Cyrtacanthacris aeruginosa



***Nomadacris septemfasciata* (Gregarious phase)**