

Distribution of pests of blueberries in South Africa and re-description and behaviour of the blueberry bud mite *Acalitus vaccinii* (Eriophyidae)

By

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Declaration

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Summary

Blueberries (*Vaccinium* spp. (Ericaceae)) are native to North America. Their production in South Africa was established about 32 years ago. South Africa exports about 80% of produced blueberries to the northern hemisphere. The blueberry industry is small compared to other fruits and challenges facing this industry includes limited use of pesticide on soft fruits and the lack of research on this crop. The aim of the present study was to document the main factors that affect arthropod species richness, abundance and communities in blueberry orchards of South Africa. Factors investigated included orchard location, season, production type (open fields versus production in tunnels) and pesticide usage (organic versus inorganic farming). Arthropods were collected from six farms in the Mpumalanga and Western Cape Provinces using vacuum and clipping sampling methods. Arthropods were grouped into dominant feeding guilds to assess the impact of these factors on pests and beneficial taxa. There was a significant variation in arthropod numbers throughout the year with numbers of all taxa peaking during warmer months. Different localities often had different arthropod species, numbers and composition. Fields with fewer predators and parasitoids tended to have higher number of plant feeding species. Production of blueberries in tunnels did not necessarily reduce pest numbers, but rather, different pest species reacted differently towards these production methods. Organic fields housed similar numbers of phytophagous species as inorganic fields indicating that pesticide usage does not necessarily aid in pest control on blueberries. Organic production is therefore advocated to maximise numbers of beneficial species while also resulting in greater profitability. As the South African blueberry industry grows, more pests will likely be reported. In 2014, the blueberry bud mite, *Acalitus vaccinii* Keifer (Acari: Trombidiformes: Eriophyidae) was reported for the first time at one farm in Mpumalanga Province. Internationally it is known as one of the most significant pests of blueberries. In South Africa it caused nearly 80% reduction in fruit yield within two years of detection. Identification of eriophyoid mites requires a high level of expertise and this challenge is intensified by a lack of identification keys and good quality species descriptions. The species description of *A. vaccinii* was published in 1939 and no longer meets modern standards for species description in this group. I therefore revised the original descriptions, and described all stages of the mite including the female, male and immatures. The male was described here for the first time. The original description was improved by addition of information and characters using new microscopy techniques such as phase contrast light microscopy and scanning electron microscopy. I also compiled a key to all Eriophyoidea species known on blueberries around the world to aid future identification. The biology of *A. vaccinii* in South Africa is discussed.

This study investigates the role of beneficial taxa and pest species in blueberry orchards in South Africa. It thus provides baseline information to aid in development of pest management strategies

in blueberry orchards in South Africa, with particular emphasis on the newly introduced blueberry bud mite.

Opsomming

Bloubessies (*Vaccinium* spp. (Ericaceae)) is inheems tot Noord Amerika. Produksie in Suid Afrika het omtrent 32 jaar gelede begin. Suid Afrika voer ongeveer 80% van sy bloubessie produksie uit na die noordelike halfrond. Die bloubessie industrie is klein vergeleke met ander vrugte en uitdaagings met hierdie industrie sluit die gelimiteerde gebruik van gifstowwe en die gebrek van navorsing op hierdie gewas in. Die doel van die huidige studie was om die hoof faktore wat n effek het op artropood spesie rykheid, hoeveelheid en populasies in bloubessie boorde in van Suid Afrika te dokumenteer. Ondersoekte faktore het boord lokaliteit, seisoen, produksie tipe (oop veld teenoor produksie tonnens) en gifstof gebruik (organies teenoor anorganiese boerdery) ingesluit. Arthropoda was versamel vanaf ses verskillende plase in die Mpumalanga en in die Wes Kaap provinsies met behulp van 'n blaas suigtoestel en knip-versamel metodes en gegroep in dominante voedingsgroepe. Daar was merkbare variasie in artropood getalle gedurende die jaar met meete taxa wat gepiek het gedurende die warmer maande. Verskillende lokaliteite het meestal verskillende spesies, getalle en komposisies gehad. Velde met minder predatore en parasitoïdes was geneig om hoër getalle plantvoedende spesies te huisves. Produksie van bloubessies in tonnens het nie noodwendig laer getalle gehad nie, maar eerder, verskillende pes spesies het verskillend gereageer op verskillende produksie metodes. Organiese velde huisves soortgelyke hoeveelhede plantvoedende spesies as anorganiese velde. Dus, die gebruik van kiemdoder help nie noodwendig met pes beheer op bloubessies nie. Organiese produksie word dus geadviseer om getalle voordelige spesies te optimaliseer en terselfdertyd groter winsgewendheid te bevoordeel. Soos die Suid Afrikaanse bloubessie bedryf groei, sal daar waarskynlik meer peste gerapporteer word. In 2014, was die bloubessie bot myt, *Acalitus vaccinii* Keifer (Acari: Trombidiformes: Eriophyidae) gerapporteer op een 'n plaas in die Mpumalanga Provinsie. Internasionaal is dit bekend as een van die mees beduidende peste op bloubessies. In Suid Afrika het dit amper 80% reduksie in vrug opbrengs in slegs twee jaar van monitoring veroorsaak. Identifikasie van eriophoied myte benodig hōe vlak van kundigheid en dit word verder bemoeilik deur 'n tekort aan goeie identifikasie sleutels. Die spesie beskrywing van *A. vaccinii* was gepubliseer in 1939 en voldoen nie aan modrne standarde vir spesie beskrywing nie. Dus het ek die oorspronklike beskrywings hersien en alle lewensfasas, insluitend vroulike, manlike en onvolgroeïdes, herbeskryf. Die manlike vorm word hier vir die eerste keer beskryf. Die oorspronklike beskrywing was verbeter deur die byvoeging van informasie en karaktertrekke met behulp van nuwe mikroskoop tegnieke soos fase kontras lig mikroskopie en elektron mikroskopie. Ek het ook n sleutel vir alle Eriophyoidea spesies bekend vanaf bloubessies ter wereld saamgestel. Die biologie van *A. vaccinii* in Suid Afrika word bespreek.

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Preface

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Chapter 1

General introduction

Blueberry is a group of native North American perennial flowering plants in the family Ericaceae and genus *Vaccinium* L. (Robinson and Fernald 1908; Galletta 1975). Plants grow as shrubs with alternating oval leaves. Stems are yellow or green in summer and red in winter. When in bloom, clusters of 8-10 pink or white flowers are borne at the tips of stems and can turn red in winter (Figure 1). Currently, there are three groups or types of blueberry planted around the world: highbush, lowbush and rabbiteye. Culturing of the species group 'rabbiteye' started in 1893 (Hancock and Draper 1989) and that of 'highbush' started in the early 1900's in New Jersey, United State of America (USA) (Eck 1966). Highbush varieties are derived from species indigenous to the Central USA States of New Jersey, Michigan and Washington and have been developed primarily from *Vaccinium australe* Small and *V. corymbosum* L. for sunny, acidic and swampy areas on the eastern coast of North America. Lowbush varieties are derived from species such as *V. augustifolium* Aiton and *V. myrtilloides* Michaux which are indigenous to Canada and colder regions of the USA. Rabbiteye varieties originate from species such as *V. ashei* Reade that are indigenous to Southern Georgia, South Carolina and Florida. Rabbiteye varieties are drought resistant, with short chilling requirements and can tolerate a wide soil pH range and high temperatures and are therefore normally the first choice for plant breeders (Eck et al. 1990). Fruit size varies between varieties, with highbush producing the largest at 3-4g per berry. Therefore it is also the most common commercial variety in North America. It comprises 75% of total blueberry plantations, with the rest shared between rabbiteye and lowbush. In 2010, North America had over 40 000 ha of blueberries (Brazelton 2007; Brazelton 2011). Approximately one-third is marketed as fresh fruit while the rest are sold as frozen produce or juiced. This supply needs to be maintained throughout the year.

The blueberry industry in South Africa

The blueberry industry in South Africa was established about 32 years ago by importing propagation material from North America. The primary aim of blueberry production in South Africa is to export to the Northern Hemisphere during their winter season (Meyer and Prinsloo 2003). There are three varieties in South Africa; northern highbush, southern highbush and rabbiteye, of which 61% is planted under net, 25% in open fields and 14% under shade (or in plastic tunnels). The field choice is largely driven by retail and export markets, which require minimal use of pesticides and high quality produce free from damage and scarring. Important characters for choosing a variety include berry size, colour and firmness, resistance to cracking,

the tendency of fruit to drop when ripe, keeping of high quality aroma, flavour and the specific ripening season (Eck1966).



Figure 1: Flowering blueberry plants in South Africa a) Jewel variety b) Emerald variety

Although it is a small industry in South Africa compared to other fruit, by 2012 production had grown by approximately tenfold over the previous 5 years (Erasmus 2013). The production continues to increase as global demand increases. Currently, South Africa has 1 300 ha of planted blueberry fields, which is expected to grow to 2 000 ha by 2020. In 2015/16 South Africa produced 3 117 tons of blueberries, which was expected to increase by approximately 90% in 2016/17 due to new agricultural practices in place, newly imported genetically modified varieties and 90% of new plants starting to bear fruit during that year. The Western Cape Province is the biggest blueberry producer in South Africa, accounting for approximately 60% of production, followed by the Limpopo Province at 15%, North West Province at 10%, Gauteng Province at 8%, Eastern Cape Province at 4%, Free State Province at 2% and Mpumalanga Province at 1% (Sikuka 2017; Figure 3). South Africa's main competitors in the export markets are Argentina which produces about 25 000 tons, Peru that produces 40 000 tons and Chile that produces ca. 140 000 tons of blueberries annually. To ensure a profitable blueberry industry, continuous production of high quality fruit is essential. Consumers and processors of blueberries have a zero-tolerance policy for pest-contaminated fruit (Garcia-Salazar 2002). For this reason, growers must be able to effectively monitor pest populations and be ready to counteract infestations with appropriate measures when needed.

Numerous challenges remain ahead of the blueberry industry. Climate change is one of the biggest challenges as all production depends on specific climatic conditions. For example, heat stress and lack of chilling hours severely negatively affects yield. In addition, climate change may lead to changes in the distribution of pests and diseases, making the exclusion of these from isolated production areas difficult. Pests and diseases, like with other fruit industries, remain the biggest challenge to an expanding blueberry production market (see section below). This problem is amplified in South Africa by a very limited number of registered pesticides, as currently it is not seen as economically viable for chemical companies to invest in this small industry (Erasmus 2013). The use of pesticides and other control mechanisms is hampered even further by the lack of studies on current pests of blueberries in South Africa, largely due to its fairly recent establishment in the country. Greater investment in research and innovation is required to help growers solve current pest problems under local production conditions, and help curb future economic losses.

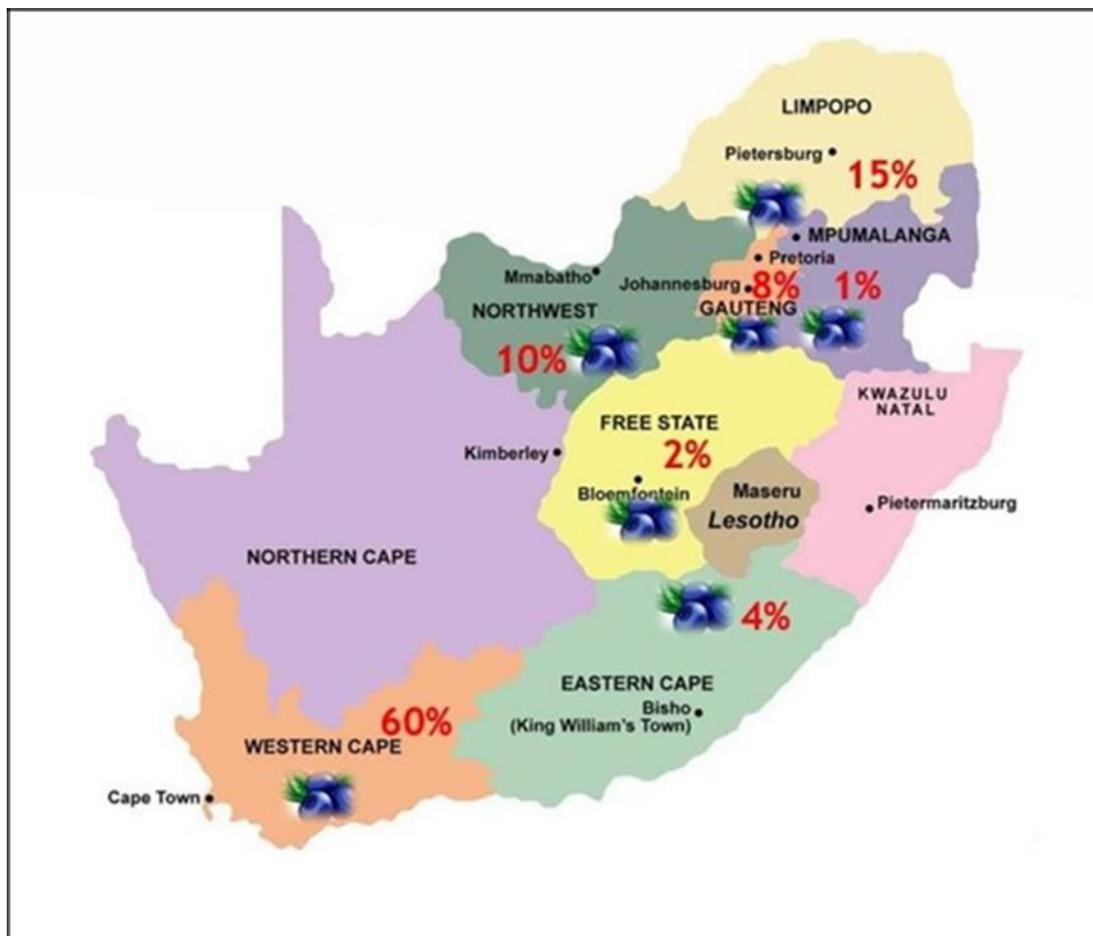


Figure 3: Blueberry production areas in South Africa. (Source: Hortgro, 2018, South Africa)

Since blueberry is not native to South Africa, all commercial propagation material was originally imported from the USA. Importation followed strict biosecurity measures to minimize the risk of introducing known pests or diseases (Saccaggi et al. 2016). Imports are guided by South Africa's entry requirements to which the exporter and importer must agree and comply with, including inspection (where necessary) for associated pests or diseases. The biggest importers of blueberry propagation material are situated in the Western Cape Province as it is also the biggest blueberry production area (DAFF, unpublished data). Plant material is supplied as either seedlings or tissue culture shoot tips which is propagated in quarantine laboratories and later sold to producers all over the country as plug plants. Methods of cultivation include hardwood and softwood cuttings and micropropagation techniques such as tissue culture, depending on the blueberry variety. Other methods such as budding, mound layering and seed propagation may be used for special purposes (Eck 1988). Although South Africa's import regulations are usually strictly applied, human error may occur. Due to budget constraints and challenging working conditions, it is difficult to employ and retain trained, experienced, dedicated and motivated personnel. This is a major concern in biosecurity systems for agricultural defense, as it makes it more difficult to monitor the import of all regulated material. In addition, some importers may decide not to adhere to regulations, increasing the risk of introduction of pests and diseases

In South Africa, the blueberry industry faces a unique threat in terms of the likelihood of pests that will seek habitat on *Vaccinium* in the future. Most productive farmlands are situated in the Greater Cape Floristic Region. The Cape Floristic Region (CFR) is a biodiversity hotspot of 90 000 km² area (Goldblatt and Manning 2000; Myers et al. 2000). It is comprised of approximately 9000 plant species, and is home to numerous vertebrates, invertebrates and microorganisms, of which about 70% are endemic to the region (Usher 1972; Schlettwein and Giliomee 1987; Giliomee 2003; Rouget et al. 2003; Botes et al. 2006; Stander 2016). Fynbos is one of the defining vegetation types in this biome (Figure 4). It is largely comprised of Proteaceae, Ericaceae and Restionaceae (Naveh and Whittaker 1980; Cowling 1994; Davis et al. 1996). The Ericaceae is a family of flowering plants found in acid and infertile soils. It has 124 genera including economical important *Vaccinium* (i.e blueberry, huckleberry, cranberry). The genus *Erica* is the most species-rich relative of *Vaccinium* with about 690 species in the CFR (Pirie et al. 2011). Research has shown that plant diversity favors arthropod diversity, notably herbivorous arthropods, and that these often exhibit host specificity at the plant family level (Siemann et al. 1998; Lewinsohn and Roslin 2008; Castagneyrol and Jactel 2012; Dinnage et al. 2012). This implies that with time, these herbivorous arthropods may move from the wild flora (notably *Erica* species) to closely related cultivated species (such as *Vaccinium* species). In addition, the sclerophyllous nature of leaves and chemical defenses against herbivores in many

plants of the CFR vegetation may result in herbivorous arthropods searching for a more palatable host (Johnson 1992; Giliomee 2003). It is unfortunate that the majority of biodiversity research in the region has been done on plant diversity whereas the ecology and diversity of arthropods have been neglected (Johnson 1992; Braschler et al. 2012; Matenaar et al. 2014). We have very limited knowledge of the current status of the arthropod diversity in the Cape floristic region compared to our agricultural fields and many herbivorous species, even including undescribed taxa, could become pest species in the future.



Figure 4: Cape Floristic Region vegetation containing a multitude of plant species including *Erica* (Source: Bjørkan, 2012)

Changing natural areas for agricultural purposes simplifies complex ecosystems by replacing diverse plant assemblages with dense stands of crop monocultures (Altieri 1999; Krebs et al. 1999). Non-native crops such as blueberries have become dominant components of many landscapes, including fruit orchards. Like any other non-native crops, the introduction of blueberries changes environmental conditions in terms of food resources for the communities of arthropods present in natural ecosystems. When a crop is introduced into a new region, it is without natural enemies, after which the arthropod communities will be structured from the regional composition of species, the spatiotemporal heterogeneity of the landscape and the spatial arrangement of habitat elements (Jeanneret et al. 2003). The survival and abundance of these arthropods on a crop depends on the suitability of the habitats and the characteristics of the surrounding landscape (Jeanneret et al. 2003). The arthropods that normally adapt more quickly are the native or exotic species that have established in disturbed nearby areas, and most of them will be polyphagous (Strong et al. 1984) and ectophagous (Kennedy and Southwood 1984). The absence of natural enemies in such invasions could lead to re-investment of costly defence mechanisms such as a chemical control to meet production requirements (Crous et al. 2017). Spatio-temporal heterogeneity of the production landscape

and the optimal spatial arrangement of habitat elements are essential for maintaining species diversity (Burel 1992). Maintenance of biodiversity has been successfully used in agricultural landscapes to promote natural enemies and suppress pests and associated crop damage (Landis et al. 2000; Gurr et al. 2003). Several studies have documented the positive effect of increasing habitat diversity on the abundance and diversity of natural enemies in agricultural systems (Bianchi et al. 2006; Chaplin-Kramer et al. 2011). Loss of agrobiodiversity therefore often has immediate risks in terms of costs for producers and long-term effects on agricultural productivity as well as jeopardizing food security.

In addition to suppression of pests, enhanced agricultural biodiversity provides ecosystem services such as fertility and nutrient enhancement, water retention and pollination (Thrupp 2000). Farmers have employed different practices to enhance and conserve biological diversity in order to produce crops in traditional farming systems (Thrupp 2000). Strategies include the use of biological control agents that consume insect pests, thereby eliminating or reducing pesticide use through integrated pest management (IPM) (Thrupp 2000). IPM aims to incorporate knowledge of the crops, their related pests and beneficial species into management programmes which are designed to reduce synthetic chemical usage as much as possible while supporting the use of ecosystem services, thereby enhancing crop production (Stern et al. 1959; Sandhu et al. 2007).

The first step in pest control is to understand the specific factors that influence the richness and abundance of pests and the damage they cause. Arthropod populations may vary from one year to the next and between different sites depending on various factors such as prevailing weather conditions, differences in agricultural practices, presence and abundance of other species such as predators or parasitoids and numbers of competitors (Eck et al. 1990; Marucci 1966). Agricultural practices such as open or closed fields alter the microclimate and thus have an influence on the arthropods present. The major feeding guilds usually found associated with cultivated crops are phytophages, predators, parasitoids, detritivores and pollinators. Phytophagous insects (plant feeders) serve as a food source for beneficial arthropods such as parasitoids and predators. Predators and parasitoids are well known for keeping pest populations at a manageable economic level by balancing natural systems. Detritivores help regulate nutrients in the soil for plants. Pollinators are essential for fruit set in fruit bearing crops such as blueberries and maximize the yielding potential of the crop (Isaacs et al. 2009). Understanding these interactions would, therefore, help to refine crop management for optimal production and promotion of the beneficial taxa.

Pests of blueberry in North America

A high incidence of pests and diseases of blueberry have been reported in the main production areas of North America, especially in Michigan, New Jersey, North Carolina and Maine. It has been noted that the older the plantation, the more diseases and pests are associated with it (Hancock and Draper 1989). As this is a fairly new crop in South Africa, it is expected that pests and disease-causing organisms will start to accumulate on these from native and non-native areas (Roubos et al. 2014). Three key pests in North America are briefly discussed below as examples of future South African threats, whilst other important pests are presented in Table 1 (Hancock and Draper 1989).

The spotted-wing drosophila fly, *Drosophila suzukii* Matsumura (Diptera: Drosophilidae) was first detected in California in 2008 and has subsequently spread to the rest of the USA and into Canada and Mexico (Bolda et al. 2010; Wise et al. 2015). It infests fruit from the time of ripening up to the time of harvest. Adult insects lay eggs within developing fruit using a serrated ovipositor. The larvae develop within the fruit causing extensive damage. This species has a high reproduction rate, completing a generation in only two to three weeks. If left uncontrolled it could cause up to 80% crop losses on a diverse range of berry crops including raspberry, blackberry, grape and cherry (Lee 2011). Currently, integrated pest management programmes for this species on blueberries rely on baited traps and foliar-applied insecticides to monitor and control adults (Wise et al. 2015). *D. suzukii* has not been reported in Africa yet (dos Santos et al. 2017). However, considering the cryptic nature of the larvae, favorable conditions in South Africa and the rate of imports and exports, the chance of it being introduced and establishing here are high. *Drosophilla suzukii* is adapted to mild temperatures and year round rainfall. Dos Santos et al. (2017) showed that the eastern and western parts of South Africa are suitable for establishment of *D. suzukii*. Infestation has been reported to cause significant loss of revenue. For example, in the main production areas of small fruit (blueberry, raspberry, blackberry, cherries and strawberries) in the Pacific Coast States (California, Oregon and Washington) of the USA the total revenue loss for blueberry was estimated at an average of \$18 million in each of the states totalling \$421 million for all five crops (Bolda et al. 2010; Walsh et al. 2011; Farnsworth et al. 2017).

The Japanese beetle, *Popillia japonica* Newman (Coleoptera: Scarabaeidae), was first discovered in the United States of America in 1916. It feeds on approximately 300 plant species including *Vaccinium* (Fleming 1972; Potter and Held 2002; Van Timmeren and Isaacs 2009). Adults usually occur in very high numbers during harvest time, causing extensive foliage and fruit damage. High population numbers also increase the risk of contaminating the fruits during harvest. Larvae feed on roots of the grass surrounding the target crop, but seldom feed on the

crop itself, and there is no evidence of the larvae feeding on the blueberry roots. The larval population may be reduced by tillage, consequently reducing the adult's population (Szendrei et al. 2005). The larval population can also be controlled by entomopathogenic nematodes. However, even with all these options, insecticides are still the primary control method for the beetle. The beetle can travel long distances by flight, hitchhiking and as fruit contaminants. However, its establishment is limited by suitable soil temperature. Unfortunately for South Africa, according to Match index predictions that were used to analyze and predict invasibility into other regions, South Africa is at a risk of invasion by this pest (Fleming 1972; Allsopp 1996; Potter and Held 2002; Cheraghian 2014). For south-western Michigan counties (Muskegon, Ottawa, Berrien, Allegan, and Van Buren) the median economic loss due to the Japanese beetle has been estimated at \$180 per hectare ranging from \$25 to \$2500 overall lost per hectare per year (Szendrei and Isaacs 2006). Therefore, crop losses due to invasion by this insect may be substantial.

The blueberry bud mite, *Acalitus vaccinii* Keifer (Acari: Trombidiformes: Eriophyidae) was first reported as an economic pest by Fulton in 1940. These mites are microscopically small and may stay undetected until physical damage to the blueberry plant is observed. Even with suspected symptoms, its presence can only be detected or confirmed by inspecting the buds using a microscope. Mites overwinter in the buds of the plants, and feeding damage becomes evident in spring when bud tissues turn red. The mites have multiple generations throughout spring and summer and feed on developing tissues, causing reduced vegetative growth that negatively impacts the following year's crop. Bud infestations of up to 80% have been reported in some cultivars (Neunzing and Galletta 1977). No blueberry species are fully resistant to the blueberry bud mite, though some varieties have been observed as free of mites under field collections (Neunzing and Galletta, 1977). Effective control is extremely difficult since the mites live deep inside developing buds and are therefore protected by layers of bud scales. It is essential that application of acaricides on plants in the field is done post-harvest and be applied at high pressure and at high quantities to obtain effective coverage and penetration (Cromroy and Kuitert 2001; Isaacs et al. 2004)). In addition to acaricides, *Hirsutella thompsonii* Fisher, a mesothermic mycopathogen of various invertebrates, has been proven to be active against *A. vaccinii* (Weibelzahl and Liburd 2009).

| Table 1: Symptoms caused by some of the major economically important pests on blueberries in North America | | |
|---|---|---|
| Plant Part | Pest (Family name) | Symptoms |
| Buds and blossoms | <i>Anthonomous musculus</i> Say (Curculionidae) | Dwarfed leaves, unopened flowers |
| | <i>Frankliniella tricità</i> Fitch (Thripidae) | Scared fruits, fruit abortion |
| | <i>Frankliniella bispinosa</i> Morgan (Thripidae) | Scared fruits, fruit abortion |
| | <i>Frankliniella occidentalis</i> Pergande (Thripidae) | Scared fruits, fruit abortion |
| | <i>Acalitus vaccinii</i> Keifer (Eriophyidae) | Red blisters on buds, malformed flowers, small fruits |
| Fruits | <i>Rhagoletis mendax</i> Curran (Tephritidae) | Soft and mushy berries |
| | <i>Conotrachelus nenuphar</i> Herbst (Curculionidae) | Dropping of immature green berries |
| | <i>Acrobasis vaccinii</i> Riley (Pyralidae) | Soft and mushy berries, fruit absence |
| | <i>Grapholita packardi</i> Zeller (Tortricidae) | Soft and mushy berries |
| | <i>Drosophila suzukii</i> Matsumura (Drosophilidae) | Soft and pulpy fruit, rotten fruit |
| Foliage | <i>Scaphytopius magdalensis</i> Provancher (Cicadellidae) | Stunt disease |
| | <i>Popillia Japonica</i> Newman (Scarabaeidae) | Skeletonized foliage, leaf drop |
| Stems | <i>Aspidiotus ancyclus</i> Putman (Coccidae) | Weakened plant, reduced yield, shortened life |
| | <i>Lecanium nigofasciatum</i> Pergande (Coccidae) | Weakened plant, reduced yield, shortened life |
| | <i>Hendecaneura shawiana</i> Kearfott (Tortricidae) | Shoot dieback |
| Crowns and roots | <i>Cryptorhynchus obliquus</i> Say (Curculionidae) | Death of twigs, branches, and shoots |
| | <i>Otiorhynchus ovatus</i> Linnaeus (Curculionidae) | Death of twigs, branches and shoots |
| | <i>Otiorhynchus rugosotriatus</i> Goeze (Curculionidae) | Death of twigs, branches and shoots |
| | <i>Otiorhynchus sulcatus</i> Fabricius (Curculionidae) | Death of twigs, branches and shoots |

Blueberries Pests in South Africa

Despite the rapid growth of the blueberry industry in South Africa, little is known regarding blueberry pests present in South Africa or their impact on production. Meyer and Prinsloo (2003) assessed the potential of blueberry production in South Africa and at that time no pests were reported. They expected that more pests and diseases would be reported as the blueberry industry grew in South Africa. A few years later, Barnes et al. (2015) reported 16 insect pests on blueberries in South Africa but admitted that this was not a comprehensive list. To date, no comprehensive survey of arthropods present on blueberries in South Africa has been undertaken. The pests that are known primarily affect leaves, stems and fruits and are mostly only damaging at very high populations.

Leaves

The Western flower thrip, *Frankliniella occidentalis* Pergande (Hemiptera: Thripidae), is a sporadic pest between December and March that has been reported in Worcester, Villiersdorp, and Porterville of the Western Cape. Thrips have piercing mouthparts and feed by sucking up

the sap from epidermal cells. Feeding causes post-harvest scarring leading to stunted growth affecting the following season's crop. It can be controlled by natural enemies such as lacewings, ladybird beetles, hoverflies and anthocorid bugs. In addition, contact pesticide and cultural control by not disturbing flowering plants close to blueberry plantations will help attract and retain thrips away from the blueberry crop.

The weevils, *Eremnus atratus* Sparrman, *Eremnus horticola* Marshall, *Eremnus setulosus* Bohemian, *Phlyctinus callosus* Schönherr, and *Sciobius tottus* Schönherr (Coleoptera: Curculionidae) are all international phytosanitary pests that have been reported in Porterville in the Western Cape. They cause shot-holing of leaves and leaf notching. High populations may result in complete loss of young leaves leading to crop loss. Feeding activity by *P. callosus* and *E. atratus* can extend to nearby apple, nectarine and grape orchards. *Sciobius tottus* can also affect apples, plums and pears. Weevils are difficult to control because of their hard bodies and the ineffectiveness of environmentally friendly chemicals that are often used. Integrated pest management using chemical, physical, biological and cultural control is usually recommended. Physical control includes the use of trunk barriers preventing adults from accessing the plant. The larval and pupal population has been controlled effectively by entomopathogenic nematodes and fungi.

The moth, *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae), is a polyphagous pest attacking all field crops including vegetables and ornamental flowers. It is a sporadic pest and larva feed on young leaves. Serious damage to blueberries plants has been reported in Vyeboom and Porterville regions in the Western Cape Province. It can be controlled with biological control agents such as wasps, rove beetles, ladybird beetles, spiders, ants, earwigs, anthocorids, mirid bugs, and lacewings. Despite such a wide range of natural enemies, pest populations often cannot be kept under the economic threshold, requiring additional control by pesticides. The pear leaf roller, *Epichoristodes acerbella* Walker (Lepidoptera: Tortricidae), another moth species, has been known as an important pest on carnation and chrysanthemums. This species and *Lozotaenia capensana* Walker (Lepidoptera: Tortricidae) were reported for the first time on blueberries by Barnes et al. (2015).

The greedy scale, *Hemiberlesia rapax* Comstock (Hemiptera: Diaspididae) and the cotton cushion scale *Icerya purchasi* Maskell (Hemiptera: Monophlebidae) are sap-sucking pests. Infestation by these pests may lead to leaf fall, wilting, dieback, discoloration and stunted growth. They occur primarily on bark and twigs but can also be found on leaves during heavy infestations. During feeding, honeydew is secreted on leaf surfaces. The honeydew promotes the growth of fungi that blocks the light and air, affecting photosynthesis and eventually leading to drying up of shoots, defoliation and stunted growth. *Icerya purchasi* affects more than 60 host

plants and it is also a serious pest of citrus. It can be effectively controlled by biological control. It has been easily controlled in citrus of California and pomegranate fields in China.

Flowers

The larvae of the Western flower thrip, *F. occidentalis* feeds on ovaries and developing berries just after flowering, resulting in russet like scarring. The Pear leaf roller, *E. acerbella* has for a long time been known to feed on carnation and chrysanthemums, and was recently reported on blueberry flowers, causing desiccation and tangling of petals by silk. All previously mentioned weevils may also be found feeding on blueberry flowers.

Fruits

As mentioned before, feeding by the Western flower thrip may result in russetting of fruits. Weevils also cause scarring of fruits rendering them unmarketable. Larvae of the pear leaf roller, *E. acerbella* feed on fruits, leaving open wounds that are prone to excessive rotting. High infestations with giant scale, *I. purchasi* may also result in colonization of fruits, causing fruit discoloration and premature fruit abscission.

Stems

Thrips also feed on the stem of the crop, causing scarring and stunted growth. Weevils may cause stem notching and scale insects can cause stem dieback.

Roots

The larvae of the banded fruit weevil *P. callosus* may feed on roots of blueberry crops.

The blueberry bud mite in South Africa: an example of an emerging threat fraught with taxonomic difficulties

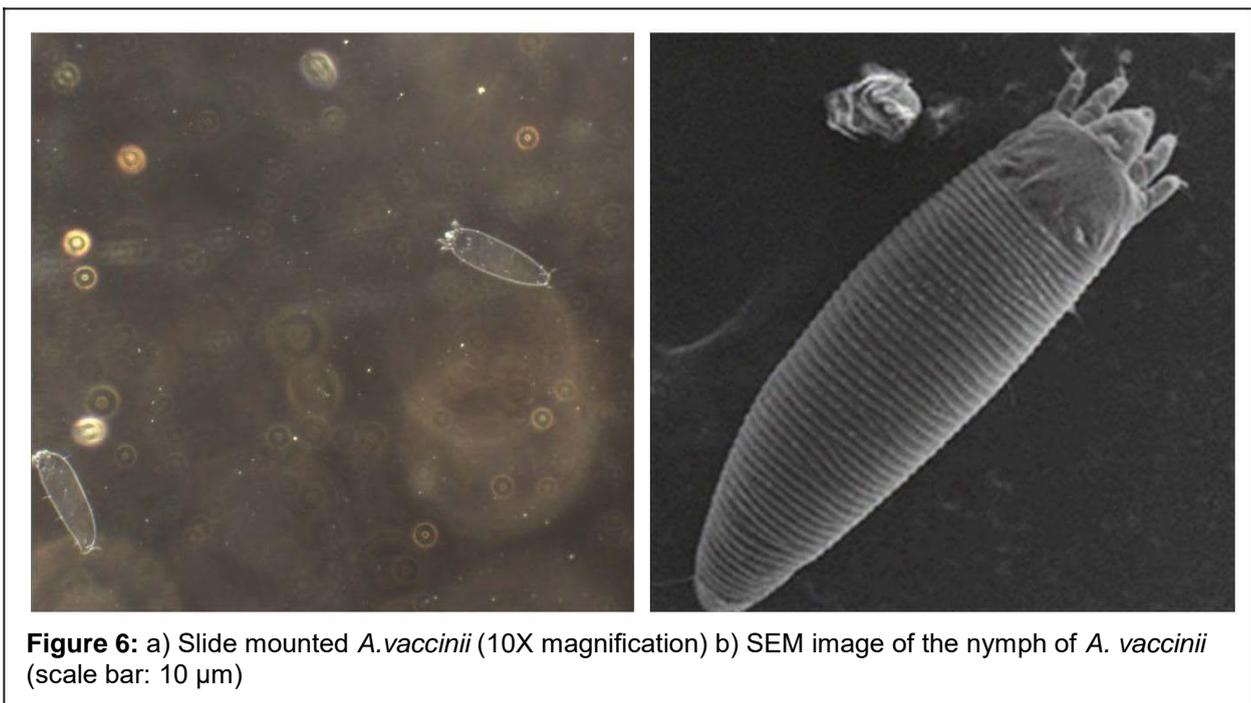
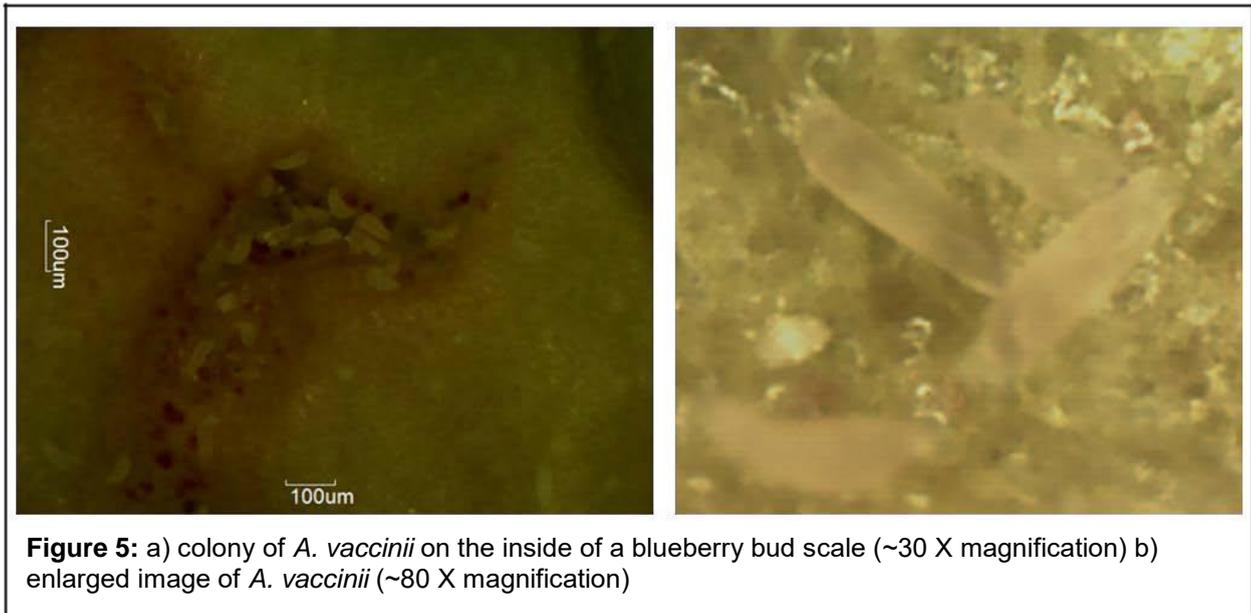
In 2014, the Department of Agriculture Forestry and Fisheries (DAFF) was alerted to the presence of the blueberry bud mite, *A. vaccinii* (Acari: Trombidiformes: Eriophyidae), on a farm in Mpumalanga. *Acalitus vaccinii* is one of the world's most important blueberries pests, with devastating effects on production. This was the first time that this pest had been reported outside North America. The symptoms were first observed by a farmer in 2012, who initially thought it was due to winter damage. When symptoms persisted and became more severe, he took a sample to the Agricultural Research Council (ARC) for testing, where the presence of *A. vaccinii* was demonstrated. The identification was confirmed by Dr. C. Craemer, a leading eriophyoid taxonomist at the ARC. Identification of *A. vaccinii* was a tedious process as no comprehensive key to *Acalitus* species nor to eriophyoid species on *Vaccinium* is available (only

a key to African *Acalitus* species was available). We had to rely on the original description by Keifer (1939) and an additional short description by Baker and Neunzig (1970), both of which are lacking detail. By 2014 (less than 2 years since the first symptoms were observed), blueberry bud mite spread to all blueberry plants on the farm. Buds and flower damage levels were up to 90%, causing a drastic reduction in yield from 50 tonnes to 10 tonnes (80%) in two years (C. Craemer; pers comm). Early in 2015, the DAFF initiated a survey in blueberry growing areas in South Africa to determine the extent of occurrence of blueberry bud mite in the country. To date, blueberry bud mite has only been detected in the Mpumalanga Province, and the main growing region, the Western Cape Province, remains free of this pest for now.

Eriophyoid mites are a very distinctive group of mites. They are microscopically small, 0.1 to 0.3 mm long and have elongated worm-like annulated bodies. Both the adults and juveniles have only two pairs of legs. The Eriophyoidea is the second most economically important superfamily of crop pests among the Acari and comprises three families: the Phytoptidae, Eriophyidae, and Diptilomiopidae. All eriophyoid mites are plant feeding and they are associated with blisters, rusts, galls, erineum, leaf curling, witches broom and bud malformation symptoms. These symptoms are very specific to the association between the mite and the plant as the mites are host specific or have a very narrow host range (Lindquist and Oldfield 1996). The Eriophyidae is the largest family in the superfamily of Eriophyoidea with approximately 227 genera and over 3,000 species reported by Amrine et al. (2003). Many of the species in this family are of economical importance due to the symptoms they induce and their ability to transmit plant pathogens.

Due to their tiny size and cryptic nature, eriophyoid mites have fewer body characters to use for morphological assessments compared to other groups of Acari, making their identification particularly challenging. Eriophyoid mites can only be detected under a microscope at least 30 X magnification (Figure 5). Identification requires slide-mounting and examination under at least 1000 X magnification with high contrast lighting. Popular methods for studying slide mounted eriophyoids are Phase Contrast Light Microscopy (PCLM) and/or Differential Interference Contrast Microscopy (DIC). PCLM provides a clear and bright view of a semi-transparent specimen contrasted against a darker background (Figure 6a). This allows for greater clarity of the edges of the specimen, except when structures are fairly complex or the specimen is not very transparent. Under such conditions, DIC microscopy would be preferred. DIC provides a false 3D image without a bright diffraction halo. However, the use of slide mounted specimens and associated microscopy techniques come with various problems and artifacts which stand the danger of being carried over into descriptions. On slide-mounted specimens, for example, fine characters are sometimes distorted, obscured and otherwise not clearly visible. To overcome these limitations and ensure correct identification, new methods of studying

eriophyoid mites are constantly explored. For example, Low Temperature Scanning Electron Microscopy (LTSEM) is now regularly used to make minute characters clearly visible and undistorted (Craemer 2010; Figure 6b). Similarly, the use of Confocal Laser Scanning Microscopy (CLSM) has enabled the study of internal anatomy, which helped pave the way to the discovery of new characters useful for discerning between species (Chetverikov et al. 2012; 2013).



Eriophyoid systematics is dependent on high quality specimens for morphological descriptions. As the mites are very small and delicate it is very difficult to preserve specimens on slides as permanent mounts. These slides do not last very long due to several factors including drying out of the water-based mounting medium, precipitation of medium on the slide and continual clearing of the specimen by chemicals on the slide. The average time that a slide mounted specimen remain usable is about 25 years. Twenty-five years does not give enough time allowance to amend any errors that might have occurred during the initial identification and description if need be, nor to compare with closely related new species that may be discovered in the future. As a result of morphological misinterpretations, the error can then be carried over to ecological studies and eventually to pest management programmes (Bortolus 2008). It is therefore essential that type specimens are captured in other forms such as detailed published descriptions, drawings and digital images. Such forms of descriptions and images then serve as the type specimen if the original specimen has deteriorated.

For a long time, the amount of detail captured in new eriophyoid species descriptions varied between authors. As a consequence, many of the older descriptions are not of a high taxonomic standard, which makes species identification and comparison difficult. In 1996, Amrine and Manson published a monograph to set a standard for the description of Eriophyoidea. They argued that all features need to be drawn, all characters need to be measured, and biological data needs to be included. This was echoed by de Lillo et al. (2010) that set a detailed presentation standard for new eriophyoid species descriptions. If a new species description follows the standards as set out in these texts, misinterpretations and incorrect classification can be minimized.

The description of the blueberry bud mite, *A. vaccinii* by Keifer in 1939 is a good example of a description that needs improvement. The description is 79 years old. The original type specimen has completely deteriorated and it is therefore not possible to compare with newly collected specimens. As there is no type specimen, the line drawing is the only reference to the type. Although the original description has line drawings of the type specimen, which is one of the requirements for the current standard, the drawings are small and of low quality, and not all the characters were included. Microscopic equipment and techniques have greatly improved since 1939, making it possible to see all the tiny structural details not possible in that initial description and to ensure long lasting evidence for the characters of the specimens in the form of micrographs. Another shortfall of the original description is the lack of male, larva, nymph and deutogyne details. In short, the original descriptions lack measurements of many morphological characters that are required to meet the current standards outlined by Amrine and Manson (1996) and de Lillo et al. (2010).

Line drawings

Line drawings in the style of H.H Keifer should form the core of species descriptions, as they are permanent and universally understandable (Amrine and Manson 1996; de Lillo et al. 2010). Semi-schematic line drawings illustrate the most important morphological characters of the mites. As specimens are often distorted during the mounting process, several specimens must be studied. This also helps capture possible variation in more plastic characters. At least five drawings should be made, illustrating the following characters: 1. dorso-ventral view of the whole mite; 2. lateral view depicting gnathosomal details, setae, annuli and microtubercles; 3. prodorsal shield showing ornamentation; 4. coxal area, showing coxae, genital coverflap and genitalia of both sexes and 5. lateral view of legs (Amrine and Manson 1996). Included with these line drawings are all measurements and counts as outlined in de Lillo et al. (2010).

Microphotographs

Microphotographs such as that provided by Scanning Electron Microscopy (SEM) and Phase Contrast Light Microscopy (PCLM) can be added to descriptions to enhance the quality of the publication. A PCLM image is taken from a slide mounted specimen, making it a clear visual representation of what a specimen actually looks like (Figure 6a). However, PCLM can only show a two-dimensional image and distorts some three-dimensional features. SEM is used to study structural details by focussing a beam of high-energy electrons which generates a three-dimensional image of the surface of the specimen (Oatley et. al 1966), giving clear detail on the shape of the features (Figure 6b). This method is very useful for Eriophyoidea mites as it shows details and shape of complex structures such as the empodium and the shape of microtubercles. However, its uses are limited by costs of the equipment and it cannot be used for diagnostic purposes if only a limited number of specimens are available. Two types of SEM are normally used for mites: Low Temperature Scanning Electron Microscopy (LTSEM) (Echlin 1970; 1978) and Ambient Temperature Scanning Microscopy (ATSEM) (Achor et al 2001). LTSEM is best for studying the morphology of mites because of its ability to preserve different structures.

OBJECTIVES OF THE STUDY

This thesis sets out to document the diversity of arthropods associated with the cultivation of blueberries across South Africa and on clarifying taxonomic issues with the blueberry bud mite. Data is presented in two separate chapters, each intended for publication in scientific journals.

In chapter 2, I assess the diversity of arthropods on blueberry plants in the Mpumalanga and Western Cape Provinces. The influence of specific production practices (open vs closed, sprayed vs organic), differences in season and differences in cultivar are also investigated. All

dominant feeding guilds were included to gain a complete picture of how these variables may influence both pests and beneficial taxa.

In chapter 3, I review all available literature on the ecology and taxonomy of the newly-introduced blueberry bud mite, *Acalitus vaccinii* Keifer in South Africa, and provide an up to date detailed morphological re-description. Counts and measurements of all important morphological characters of females, males and immatures are provided in illustrations and micrographs. I also note differences between published data and those observed in this study. This re-description will improve future accurate identification of this mite species and will contribute to an improvement of quality and utility of eriophyoid descriptions for taxonomic purposes in general.

I conclude this dissertation with a short concluding chapter in which I highlight my main findings and provide guidance for future studies.

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Chapter 2

The effect of cultivation practice on arthropods associated with blueberries in South Africa.

ABSTRACT

Blueberries, *Vaccinium* species (Ericaceae) are an international fruit crop exotic to South Africa, established about 32 years ago for international trade. Challenges facing the blueberry industry include limited use of pesticide on soft fruits and the lack of research on this crop. A high incidence of pests has been reported on blueberry in North America. However, little is known about the status of pests in South Africa. The aim of the study was to understand the main factors that affect arthropod species richness, abundance and communities in blueberry orchards in South Africa. The factors in question were orchard location, season, field type (open vs. tunnel) and pesticide usage (inorganic vs organic). The various factors were assessed at six farms in the Mpumalanga and Western Cape Province using the vacuum and clipping methods over a year. Arthropods were placed into general feeding guilds in order to understand their role and to describe communities. Production of blueberries in tunnels may have a positive influence depending on the taxa. Organic production maybe favoured as it does not lead to a large increase in pest species, while possibly promoting the diversity of predators. Most noticeable was variation in number of arthropods at different farms largely due to different agricultural practices. Farms with less predators and parasitoids tended to have higher number of plant feeding species. The choice of production system depends on different factors to be considered by each producer. This study provides baseline information to aid in development of pest management strategies on blueberry orchards.

INTRODUCTION

Blueberries, *Vaccinium* spp. (Ericaceae), are an internationally cultivated crop native to North America (Robinson and Fernald 1908; Galletta 1975). South Africa had ~1300 hectares of planted blueberries which produced ~2500 tons in the 2015/16 production season. Around 78% of the yield is exported annually (Sikuka 2017). The largest markets for this seasonal crop are in the northern hemisphere, with Europe and the USA being particularly profitable (Meyer and Prinsloo 2003). However, production is restricted to the warmer months of the year, occurring from June to August in the northern hemisphere and December to March in the southern hemisphere (Hancock & Draper 1989, Lobos & Hancock 2015). There is consequently the advantage of counter-cyclical seasonality in the industry that boosts international prices.

Compared to other fruits, the blueberry industry in South Africa is small, but is fast-growing with production increasing tenfold between 2009 and 2012. It is believed that production will continue to increase as global demand increases. Production areas in South Africa include all nine provinces. The current largest production area is in the Western Cape, accounting for at least 60% of national production. Mpumalanga accounts for about 1% of national production. This disparity may be explained by the difference in duration of the winter season between the two provinces. Blueberry plants require low winter chilling conditions for optimal production and Cape Town has longer chill hours (number of cold winter hours) than other provinces (Meyer and Prinsloo 2003).

Monitoring and control of blueberry pests is important to ensure good quality fruit, high yields and to maintain a growing and profitable industry. Numerous arthropod pests have been reported on plants grown in the northern hemisphere (Elsner and Whalon 1985; Hancock and Draper 1989; Roubos et al. 2014; Carroll et al. 2015). The most notable of these include the spotted-wing drosophila fly, *D. suzukii* Matsumura (Diptera: Drosophilidae), the Japanese beetle, *Popillia japonica* Newman (Coleoptera: Scarabaeidae), and the blueberry bud mite *Acalitus vaccinii* Keifer (Acari: Trombidiformes: Eriophyidae). Damage caused by the spotted-wing drosophila fly leads to significant loss of revenue from berry crops in the USA totalling \$421 million (Bolda et al. 2010; Walsh et al. 2011; Farnsworth et al. 2017). The Japanese beetle causes an estimated loss of \$2.5 per hectare (Fleming 1972; Potter and Held 2002; Van Timmeren and Isaacs 2009; and Szendrei and Isaacs 2006). Infestation of the blueberry bud mite results in reduced growth and yield. They feed on buds, damaging developing tissues which may cause extensive crop losses (Fulton 1940; Neunzing and Galletta 1977; Cromroy and Kuitert 2001).

In the southern hemisphere where blueberries are non-native, there have been relatively few reports of associated pests. The South African blueberry industry is fairly young (started in the early 1990s) and the only pest insects currently reported are in the Hemiptera, Thysanoptera, Lepidoptera and Coleoptera (Barnes et al. 2015). For example, the Western flower thrip, *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae), has been reported to be a sporadic pest in the Western Cape Province (Barnes et al. 2015). *Phlyctinus callosus* Schönherr (Coleoptera: Curculionidae) is an international phytosanitary pest that also causes damage in some areas of this province. However, South Africa is also home to a massive diversity of native Ericaceae (e.g. 609 species in the genus *Erica* L.) which could house numerous potential pests pre-adapted to feed on *Vaccinium* spp. It is expected that, as the industry matures, pest species on blueberries will increase over time as these shift hosts from both native and other non-native plants. Growers should therefore be aware of future potential pests occurring in their area.

Insect populations vary between years, seasons and sites. These changes depend on various factors such as prevailing weather conditions, differences in agricultural practices, and presence and abundance of other species such as predators, parasitoids and competitors (Eck 1966). Similarly, specific cultural practices may also influence arthropod numbers as these manipulate microclimatic conditions (e.g. open production versus production in tunnels results in differences in moisture regimes, air flow and temperatures) (Meyer and Prinsloo 2003; Demchak 2009; Ogden and Van Iersel 2009; [Sikuka 2017](#)). Records of fluctuating numbers of all arthropod groups provide essential information in production landscapes. A first step in pest control is understanding the factors influencing population size. This helps to streamline monitoring programmes and informs timing of pesticide applications, avoiding unnecessary spraying and increasing profitability. Appropriate application of pesticides may also avoid unnecessary disruption of natural biological control organisms and those associated with productive soil (e.g. detritivores) that play a vital role in healthy crops (Eck 1966; Elsner and Whalon 1985; Roubos et al. 2014). Importantly, all of the abovementioned variables may influence the numbers of pollinators that are essential for fruit production (Isaacs et al. 2009). Knowledge of fluctuation in numbers of arthropods from all feeding guilds would therefore help refine crop management practices for optimal production and minimal impact on beneficial taxa.

The present study sets out to document the diversity of arthropods associated with the production of blueberries in the Mpumalanga and Western Cape Provinces of South Africa. The seasonal variation in arthropod diversity according to production practices in the Western Cape was also examined. The influence of production practices are investigated by comparing arthropod numbers and community composition between different farms and between fields in the open versus those covered with plastic or netting, and between organic and inorganic

systems. All dominant feeding guilds are included to gain a complete picture of how these variables influence both detrimental and beneficial taxa.

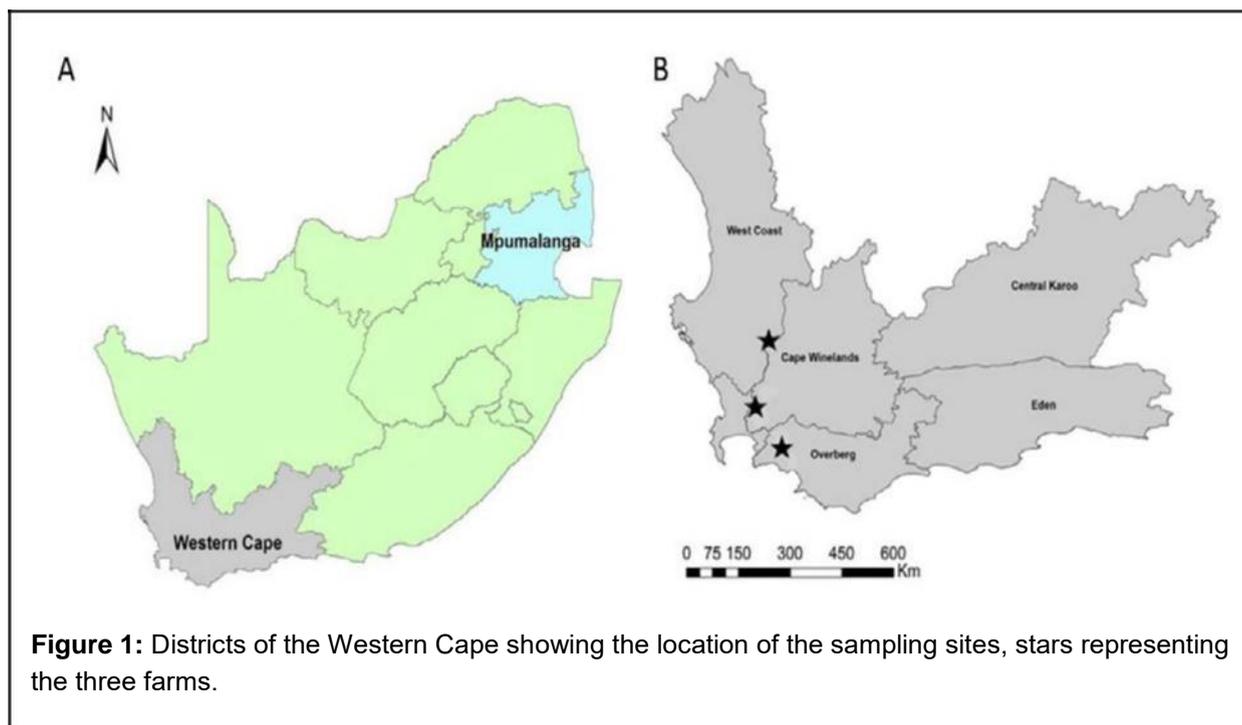
MATERIALS AND METHODS

Study sites

The study was conducted on three farms in the Western Cape Province and three farms in the Mpumalanga Province of South Africa.

In the Western Cape, Lushof in Saron; Gelukstroom in Botrivier; and Backsberg in Sonop were studied (Figure 1). Three highbush blueberry cultivars, namely Jewel, Emerald and Star, are the dominantly planted cultivars in the region and are usually inter-planted to promote cross pollination. The Jewel cultivar was planted on all farms due to its high yielding capacity. The size of the farm, age of the plants when surveyed, and their cultivars are presented in Table 1. At each farm, two blocks were surveyed, one in the open and another that was planted under plastic or shading. These covers are used to manipulate the ripening time and to protect against fungal diseases and/or pests (Figures 2, 3 & 4). Growers follow a set fertilising and spraying programme throughout the year, but these differ between farms. Lushof farm had organic sites on both open and closed fields. This alters the taste of the fruit and takes advantage of higher profits fetched by organic fruits. Organic sites on open and closed fields were also surveyed to observe the effect of pesticide usage and field type on the production of blueberries.

In Mpumalanga, samples were collected on three farms near the towns of Amsterdam, Lydenburg and Dullstroom (identity of the farms can not be revealed, therefore towns are hereafter referred to as farms). All three farms planted different cultivars to those in the Western Cape sites including Centurion, Berkley, Spartan, Elliot, Climax and De lite (Table 1). Samples at these farms were collected randomly between cultivars. All plants in Mpumalanga were planted under hail nets (Figure 3) and no pesticides were applied at Lydenburg and Dullstroom.



| Farm | Area | GPS co-ordinates | Blueberry area | Cultivars | Ages |
|-------------|-------------|---------------------------|-----------------------|---|--------------------------|
| Lushof | Saron | 33.158878S, 19.009280E | 68.0 hectares | Jewel Emerald | 2-4 years 2 years |
| Gelukstroom | Botrivier | 34.089474S, 19.167592E | 44.0 hectares | Jewel Star | 7 years 6 years |
| Sonop | Klapmuts | 33.803759S, 18.894952E | 12.5 hectares | Jewel Emerald | 4 years 4 years |
| Amsterdam | Amsterdam | | 10.0 hectares | Bluecrop, Berkley, Elliot Spartan | 14 years |
| Dullstroom | Dullstroom | | 4.0 hectares | Elliot | 4 & 8 years |
| Lydenburg | Lydenburg | | 3.0 hectares | Climax, de lite | 25 years |



Figure 2: Open field of star cultivar blueberries at Gelukstroom farm, Botrivier, Western Cape.



Figure 3: Hail net over A) star and jewel blueberry cultivars at Backsberg farm, Sonop, Western Cape; and B) Elliot blueberry cultivar in Dullstroom, Mpumalanga.



Figure 4: Plastic cover over jewel cultivar crops at backsberg farm, Sonop, Western Cape.

Arthropod sampling

Vacuum sampling

Arthropods associated with above-ground plant parts were collected using a Stihl SH 86 vacuum sampler (Stihl, Germany) with a 30cm collection nozzle fitted with a collection net as described by Dietrick et al. (1959). At each site, the nozzle of the vacuum sampler was inserted into the foliage 100 times with 3-4 'pokes' on each neighbouring plant. Samples were collected randomly about 2.5 meters away from the edges, and replicated at 10 points ($n = 10$) in each production block to maximise catches (Richmond and Graham 1969). The collection of litter and soil was avoided by keeping the nozzle horizontal to the soil surface. To investigate the effect of season, sampling was repeated bi-monthly between October 2015 and October 2016 on all three farms in the Western Cape Province. On each farm, sampling was conducted in one block that was not covered (open), and in one block that was covered by either netting or plastic (closed). To investigate the combined effect of production type (open vs. closed) and pesticide use (organic vs. inorganic), additional sampling was conducted on the farm Lushof, ten sites in each of four blocks were sampled as described before in the following combinations: Open and organic, open and inorganic, closed and organic, closed and inorganic. Sampling was conducted only once in

November 2015 in the Mpumalanga Province. Samples (from each sampling point) were placed separately in re-sealable plastic bags and transported to the laboratory where arthropods were collected and preserved in 70% ethanol until sorting. All collected arthropods were sorted to morphospecies (Oliver and Beattie 1996) and assigned to a feeding guild based on mouth parts and prominent feeding behaviour of members of the family. Identifications were done to order and family with the aid of Scholtz and Holm 1985 and Picker et al. 2002, or to species where possible using relevant taxonomic literature for the group. Identifications of selected groups were done by entomologists W. Pieterse (Hemiptera: Sternorrhyncha) and H. Ramukhesa (Coleoptera) at the Department of Agriculture, Forestry and Fisheries in Stellenbosch, and taxonomists M. Stiller (Thysanoptera), R. Stals (Coleoptera) and I. Miller (Hemiptera: Sternorrhyncha) at the Biosystematics Institute of the Agricultural Research Council in Pretoria.

Clippings method

For sampling of smaller arthropods such as mites, thrips and scale insects that would not be collected using the vacuum sampler, branches were removed from plants and microscopically inspected for arthropods. Samples of branches and vacuum samples were collected simultaneously from the same plots, but not from the same plants. A branch of 30cm was collected (using secateurs) from each of 30 randomly chosen plants per field and placed individually in a sealable plastic bag. Samples were kept cool in a cooler box and transported to the laboratory for inspection. Samples were kept at 4°C until inspection. Each branch was carefully examined for the presence of small arthropods using a Nikon stereo microscope under at least 30X magnification. Buds were inspected for the presence of the blueberry bud mite, *Acalitus vaccinii* (Eriophyidae), a known pest of *Vaccinium* spp. (for further details, see chapter 3).

All smaller arthropods were identified to family or species by professionals of a particular group as described above. Based on the identity and the behaviour of the family, arthropods were then assigned to a feeding guild. Mites were mounted on glass slides in PVA medium (Upton 1991; Evans 1992; Upton 1993), identified to family using Krantz and Walter (2009) and assigned to feeding guild based on published data and on the behaviour of the family. Where possible, mites were identified to species using relevant taxonomic literature for the specific group. Mite identifications were done by acarologist D. Saccaggi at the Department of Agriculture, Forestry and Fisheries in Stellenbosch.

Data analyses

For analysis of feeding guilds at different sites, morphospecies were assigned to six major feeding guilds: phytophages; predators; parasitoids; pollinators (including nectar feeding); detritivores and scavengers. Detritivores included fungivores for the vacuum method and scavengers included fungivores for clipping method. The Formicidae were treated as a separate guild and excluded in further analysis due to the low number collected.

Completeness of sampling was investigated using non-parametric and least biased species estimators as is recommended for small sample sizes that contain many rare species (Hortel et al. 2006). These were Chao2 and Jackknife2 using 9999 permutations in PRIMER 6 (PRIMER-E 2008). Estimations were done for all the farms, separately and combined per province, for open and closed fields, organic and inorganic systems, various guilds at each cultivation practice, and at all farms for both the vacuum and clipping sampling methods.

Similarity in Arthropod diversity between farms was assessed using the Jaccard index of similarity (C_j). The number of shared species, the numbers and percentages of unique species, and the Jaccard index of similarity were presented for comparisons between farms and the effect of cultivation practices (i.e. open and closed fields, organic and inorganic systems) using Venn diagrams. The Jaccard index of similarity (Magurran 2004) was defined as follows:

$$C_j = j / (a+b-j)$$

where j is the shared species between the sites, a = total number of species at site A, and b = total number of species at site B

To test the influence of farm, collection month and field type (open vs. closed and organic vs. inorganic) on species richness and abundance, I calculated Generalised Linear Mixed Models (GLMMs) with Poisson distribution (as this was count data), fit by a Laplace approximation (Bolker et al. 2009), and with cultivar included as a random factor using the R programme for statistical computing (R-3.4.2 for Windows) and packages *Lme4* and *multcomp*. Models for these analyses therefore followed the general formula (and only for applicable variables):

$$\text{alpha diversity} \sim \text{Farm} + \text{Month} + \text{Field} + \text{pesticide use} + (1|\text{Cultivar})$$

Best fitting models were selected based on AIC criteria and residuals were tested for overdispersion (Akaike 1973; Mazerolle 2006). The model that resulted in the lowest AIC value was considered the best. A χ^2 statistic and P-value (Bolker et al. 2009) were then calculated.

Separate models were run for species richness and abundance between the different feeding guilds. Posthoc analyses were performed on significant factors using Tukey posthoc tests in R (Hothorn et al. 2008). To assess the effect of locality between farms in the Mpumalanga Province on species richness and abundance of different feeding guilds, Kruskal–Wallis ANOVA's were performed in R followed by Tukey posthoc tests. When performing analyses on the combined effect of pesticide usage (organic vs. inorganic) and field type (open vs. closed) on species richness and abundance for various feeding guilds, calculations of GLMMs and post-hoc analysis generally followed methods described above, except that collection month was used as a random factor.

I also compared arthropod community composition of various guilds between farms, open and closed fields and organic and inorganic systems for arthropods collected using both the vacuum sampling and clipping methods using Permutational multivariate analysis of variance (PERMANOVA) in PRIMER 6 (PRIMER-E 2008). These analyses were performed to calculate F- and p-values as well as to do pairwise post-hoc testing, using 9999 permutations with collection month set as a random variable. Analyses were performed using the Bray-Curtis similarity measures with square-root transformed data to reduce the weight of common species (Anderson 2001). Data was further explored and visualised using canonical analysis of principal coordinates (CAP) (Anderson and Willis 2003; Anderson 2006). CAP analysis is effective in delineating particular gradients of interest within a multivariate dataset, despite the presence of other potentially important factors which were not measured.

RESULTS

Arthropod Species Richness

Overall species numbers

In the Western Cape Province, a total of 9842 individuals from 420 morphospecies were collected from the three farms using the vacuum sampler and 1941 from 55 morphospecies using the clippings method throughout the year (Table 2). Farm identity had a significant influence on species richness. However, this changed according to the collection method used. Using the clipping method Lushof farm had significantly lower observed and estimated species richness than Sonop and Gelukstroom farms (Tables 2, 3, 4, Appendix 1, Figure 5).

In the Mpumalanga Province, a total of 501 individuals from 98 morphospecies were collected using the vacuum sampler (Table 2). Farm identity also had a significant influence on arthropod numbers (Table 3). Amsterdam farm (where pesticides were used) had the highest observed overall and estimated species richness as compared to the other two farms (Table 2, Figure 5).

Guild species richness

In terms of feeding guilds and using the vacuum sampling method, the numbers of species per guild varied significantly between the different farms in the Western Cape Province (Tables 2, 3, Appendix 1, Figure 6). Gelukstroom generally had significantly less species in all guilds as compared to the other two farms, which were generally fairly similar. Using the clipping method, no significant differences were observed for the different feeding guilds most likely due to much smaller sample sizes (Tables 2, 3, 4, Appendix 1). In the Mpumalanga province, using the vacuum sampler, numbers of species per guild was similar for most guilds at all the farms (Table 2). Species richness of phytophagous arthropods, predators, parasitoids and pollinators did not significantly differ between the farms and the only significant differences found in species richness in this province was between the detritivores that were significantly higher at Amsterdam than at the other two farms, which were statistically similar (Table 3 & Figure 7).

| Table 2: Abundance, observed species richness and estimated species richness of arthropods collected using a vacuum sampler and clippings from blueberry crops from six farms in two provinces in South Africa. | | | | | |
|---|----------------|---------|-----------|---------------------|-------------|
| Locality and Method* | Feeding Guilds | Species | Abundance | Chao2 \pm SD | Jackknife 2 |
| Vacuum (MP) | All | 98 | 501 | 139.5 \pm 14.65 | 163.17 |
| Amsterdam farm (MP) | All | 71 | 262 | 10,0 \pm 0.0 | 10 |
| Lydenburg farm (MP) | All | 31 | 121 | 10 \pm 0.0 | 10 |
| Dullstroom farm (MP) | All | 31 | 118 | 10 \pm 0.0 | 9,1 |
| MP (overall) | Phytophagous | 21 | 135 | 35.4 \pm 11.18 | 39.215 |
| MP (overall) | Predators | 17 | 68 | 18.6 \pm 2.16 | 20.126 |
| MP (overall) | Parasitoids | 15 | 41 | 19.5 \pm 4.80 | 22.58 |
| MP (overall) | Detritivores | 18 | 150 | 31 \pm 8.34 | 35.41 |
| MP (overall) | Formicidae | 9 | 61 | 17 \pm 11.66 | 15.30 |
| MP (overall) | Pollinators | 18 | 46 | 22.57 \pm 4.24 | 26.83 |
| Vacuum (WC) | All | 420 | 9842 | 711,09 \pm 59,67 | 722,01 |
| Gelukstroom (WC) | All | 141 | 3372 | 253,23 \pm 38,65 | 253,82 |
| Sonop farm (WC) | All | 239 | 3553 | 375,81 \pm 36,55 | 406,37 |
| Lushof farm (WC) | All | 276 | 2917 | 401,28 \pm 32,86 | 440,47 |
| Open field (WC) | All | 350 | 5568 | 534,78 \pm 42,93 | 573,55 |
| Closed field (WC) | All | 280 | 4274 | 577,16 \pm 78,67 | 508,32 |
| WC (overall) | Phytophagous | 114 | 4405 | 209,56 \pm 35,69 | 210,48 |
| WC (overall) | Predators | 108 | 1617 | 165,54 \pm 24,85 | 172,75 |
| WC (overall) | Parasitoids | 57 | 404 | 115,6 \pm 35,70 | 100,58 |
| WC (overall) | Detritivores | 60 | 1926 | 90,79 \pm 16,15 | 95,86 |
| WC (overall) | Formicidae | 10 | 75 | 11,5 \pm 7,19 | 11,64 |
| WC (overall) | Pollinators | 73 | 1416 | 127,00 \pm 25,72 | 127,7 |
| Organic (WC) | All | 225 | 2385 | 299,26 \pm 21,44 | 344,14 |
| Inorganic (WC) | All | 62 | 1555 | 327,12 \pm 34,162 | 351,19 |
| WC (inorganic vs. organic) | Phytophagous | 69 | 320 | 126.8 \pm 28.0 | 126.31 |
| WC (inorganic vs. organic) | Predators | 62 | 367 | 85.14 \pm 14.8 | 90.71 |
| WC (inorganic vs. organic) | Parasitoids | 45 | 166 | 53.17 \pm 5.7 | 60.90 |
| WC (inorganic vs. organic) | Detritivores | 48 | 297 | 60.07 \pm 8.8 | 66.76 |
| WC (inorganic vs. organic) | Formicidae | 3 | 5 | | |
| WC (inorganic vs. organic) | Pollinators | 50 | 267 | 66.07 \pm 11.04 | 72.73 |

Table 2 (cont.): Abundance, observed species richness and estimated species richness of arthropods collected using a vacuum sampler and clippings from blueberry crops from six farms in two provinces in South Africa.

| Locality and Method* | Feeding Guilds | Species | Abundance | Chao2 \pm SD | Jackknife 2 |
|----------------------------|-------------------------------|---------|-----------|-------------------|-------------|
| Clipping (WC) | All | 55 | 1941 | 93.56 \pm 20.28 | 98.89 |
| Gelukstroom farm (WC) | All | 31 | 642 | 41,08 \pm 8,00 | 46,918 |
| Sonop farm (WC) | All | 35 | 1050 | 63,9 \pm 19,81 | 63,815 |
| Lushof farm (WC) | All | 21 | 249 | 21 \pm 39,84 | 38,63 |
| Open field (WC) | All | 35 | 548 | 89 \pm 40,92 | 67,75 |
| Closed field (WC) | All | 40 | 1393 | 16 \pm 11,66 | 14,762 |
| WC (overall) | Phytophagous | 21 | 62 | 29.64 \pm 6.82 | 35.79 |
| WC (overall) | Predators | 16 | 195 | 47.0 \pm 39.6 | 29.88 |
| WC (overall) | Scavengers | 10 | 364 | 10.0 \pm 1.1 | 11.99 |
| WC (overall) | Fungivores | 7 | 46 | 7.23 \pm 4.78 | 9.45 |
| Organic (WC) | All | 22 | 65 | 34 \pm 9,16 | 39,9 |
| Inorganic (WC) | All | 17 | 165 | 33 \pm 16,49 | 30,9 |
| WC (inorganic vs. organic) | Phytophagous | 12 | 60 | 36.5 \pm 31 | 24.38 |
| WC (inorganic vs. organic) | Predators | 7 | 35 | 11,5 \pm 7.2 | 11,70 |
| WC (inorganic vs. organic) | Scavengers (incl. fungivores) | 9 | 134 | 10.0 \pm 1.9 | 11.00 |

Table 3: Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage on alpha diversity of arthropods associated with blueberries collected using a vacuum sampler.

| Richness | | df | chi-square | p- | Posthoc | Dispersion (rdev/rdf) |
|-------------------|------------------|----|------------|--------|----------|-----------------------|
| All guilds (WC) | | | | | | 2,46 |
| | farm | 2 | 333,61 | <0,001 | fig. 5a | |
| | month | 5 | 330,96 | <0,001 | fig. 8a | |
| | field | 1 | 36,12 | <0,001 | fig. 10a | |
| | farm*month*field | 10 | 112,15 | <0,001 | | |
| Detritivores (WC) | | | | | | 0.76 |
| | farm | 2 | 72,643 | <0,001 | fig. 6d | |
| | month | 5 | 144,81 | <0,001 | fig. 9d | |
| | field | 1 | 0,0032 | 0,9551 | fig. 11d | |
| | farm*month*field | 24 | 58,509 | <0,001 | | |
| Phytophagous (WC) | | | | | | 1,20 |
| | farm | 2 | 10,33 | <0,01 | fig. 6a | |
| | month | 5 | 30,03 | <0,001 | fig. 9a | |
| | field | 1 | 7,505 | <0,01 | fig. 11a | |
| | farm*month*field | 27 | 122.73 | <0,001 | | |
| Predators (WC) | | | | | | 0,84 |
| | farm | 2 | 67,134 | <0,001 | fig. 6b | |
| | month | 5 | 69,466 | <0,001 | fig. 9b | |
| | field | 1 | 8,9702 | <0,01 | fig. 11b | |
| | farm*month*field | 10 | 78,206 | <0,001 | | |

Table 3 (cont.): Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage on alpha diversity of arthropods associated with blueberries collected using a vacuum sampler.

| Richness | | df | chi-square | p- | Posthoc | Dispersion (rdev/rdf) |
|--------------------------------------|------------------|----|------------|----------|-----------|-----------------------|
| Parasitoids (WC) | farm | 2 | 16,50 | <0,001 | fig. 6e | 0,57 |
| | month | 5 | 14,04 | <0,05 | fig. 9e | |
| | field | 1 | 3,000 | <0,1 | fig. 11e | |
| | farm*month*field | 10 | 25,22 | 0,237 | | |
| Pollinators (WC) | farm | 2 | 33,975 | <0,001 | fig. 6c | 0,99 |
| | month | 5 | 60,048 | <0,001 | fig. 9c | |
| | farm*month*field | 1 | 1,7668 | 0,1838 | fig. 11c | |
| Organic vs Inorganic all guilds (WC) | pesticide | 1 | 23,036 | <0,001 | not shown | 3,40 |
| | field | 1 | 64,685 | <0,001 | not shown | |
| | pesticide*field | 1 | 7,97 | <0,05 | fig. 12 | |
| Detritivores (WC) | pesticide | 1 | 4,2478 | 0,0393 | not shown | 1,40 |
| | field | 1 | 0,091 | 0,763 | not shown | |
| | pesticide*field | 1 | 0,158 | 0,691 | fig. 13a | |
| Phytophagous (WC) | pesticide | 1 | 12,829 | <0,0001 | not shown | 1,17 |
| | field | 1 | 20,534 | 0,0003 | not shown | |
| | pesticide*field | 1 | 0,6647 | 0,4149 | not shown | |
| Pollinators (WC) | pesticide | 1 | 8,7437 | 0,003107 | not shown | 0,73 |
| | field | 1 | 0,1145 | 0,735 | not shown | |
| | pesticide*field | 1 | 0,6128 | 0,4337 | not shown | |
| Predators (WC) | pesticide | 1 | 23,75 | <0,0001 | not shown | 0,84 |
| | field | 1 | 9,0508 | 0,0026 | not shown | |
| | pesticide*field | 1 | 0,2356 | 0,6274 | fig. 13b | |
| Parasitoids (WC) | pesticide | 1 | <0,0001 | 0,9791 | not shown | 0,72 |
| | field | 1 | 0,5284 | 0,4673 | not shown | |
| | pesticide*field | 1 | 0,0496 | 0,8237 | not shown | |
| All guilds (MP) * | farm | 2 | 17,348 | 0,000171 | fig. 5c | |
| Phytophagous (MP) * | farm | 2 | 5,753 | 0,05631 | not shown | |
| Predators (MP) * | farm | 2 | 1,1033 | 0,576 | not shown | |
| Parasitoids (MP) * | farm | 2 | 5,6743 | 0,058 | not shown | |
| Pollinators (MP) * | farm | 1 | 4,489 | 0,106 | not shown | |
| Detritivore (MP) * | farm | 2 | 11,596 | 0,003034 | fig. 7 | |

* analysis to test the effect of farm was done using Kruskal-Wallis ANOVA

| Table 4: The best fitting model for assessment of the influence of farm, collection month (season) and field type (open vs. closed) and pesticide usage (inorganic vs. organic) on alpha diversity of arthropods associated with blueberries. | | | | | | | |
|---|--------------------------------------|--------|--------|---------|----------|----------|--|
| Richness | Best fitting model (variables excl.) | AIC | BIC | logLik | deviance | df.resid | |
| Vacuum All guilds (WC) | | | | | | | |
| | none | 2219,5 | 2258,3 | -1099,7 | 2199,5 | 350 | |
| | farm*month*field | 2127,3 | 2205,1 | -1043,7 | 2087,3 | 340 | |
| Detritivores (WC) | field | 879,81 | 911,65 | -430,91 | 861,81 | 245 | |
| | farm*month*field | 871,3 | 991,6 | -401,6 | 803 | 220 | |
| Phytophagous (WC) | none | 886,28 | 920,67 | -433,14 | 866,28 | 220 | |
| | farm*month*field | 817,6 | 944,8 | -371,8 | 743,6 | 193 | |
| Predators (WC) | none | 1090,3 | 1127,2 | -535,13 | 1070,3 | 288 | |
| | farm*month*field | 1066,1 | 1202,9 | -496 | 992,1 | 261 | |
| Parasitoids (WC) | none | 432,59 | 461,79 | -206,3 | 412,59 | 127 | |
| | farm*month*field | 449,4 | 539,9 | -193,7 | 387,4 | 106 | |
| Pollinators (WC) | none | 766,33 | 796,5 | -374,17 | 748,33 | 202 | |
| | farm*month*field | 734,3 | 858,3 | -330,1 | 660,3 | 174 | |
| Vacuum Organic vs Inorganic | | | | | | | |
| All guilds (WC) | none | 931,47 | 942,62 | -461,74 | 923,47 | 116 | |
| | pesticide*field | 925,5 | 939,44 | -457,75 | 915,5 | 115 | |
| Detritivores (WC) | field | 367,67 | 374,85 | 180,83 | 361,67 | 78 | |
| | pesticide*field | 371,4 | 383,4 | -180,7 | 361,4 | 76 | |
| Phytophagous (WC) | none | 416,4 | 427 | -204,2 | 408,4 | 100 | |
| | pesticide*field | 417,8 | 431 | -203,9 | 407,8 | 99 | |
| Pollinators (WC) | field | 330,54 | 340,5 | -161,27 | 322,54 | 86 | |
| | pesticide*field | 331,9 | 344,4 | -161 | 321,9 | 84 | |
| Predators (WC) | none | 432,7 | 443,6 | -212,3 | 424,7 | 108 | |
| | pesticide*field | 434,5 | 448,1 | -212,2 | 424,5 | 107 | |
| Parasitoids (WC) | pesticide | 228,1 | 234,53 | -111,05 | 221,1 | 60 | |
| | pesticide*field | 232,1 | 242,8 | -111 | 222,1 | 58 | |
| Clipping All guilds (WC) | | | | | | | |
| | none | 2025,1 | 2074,9 | -1002,5 | 2005,1 | 1070 | |
| | farm*month*field | 1954,5 | 2138,9 | -940,2 | 1880,5 | 1043 | |
| Scavengers (WC) | month | 806,3 | 826,1 | -398,2 | 796,3 | 376 | |
| | farm*month*field | 860,6 | 990,8 | -397,3 | 794,6 | 348 | |
| Phytophagous (WC) | month | 131,87 | 142,17 | -60,932 | 121,86 | 53 | |
| | farm*month*field | 210 | 247 | -87 | 174,7 | 40 | |
| Predators (WC) | month | 383,56 | 399,38 | -86,78 | 373,56 | 170 | |
| | farm*month*field | 433 | 531,1 | -185,5 | 371 | 144 | |

Table 4 (cont.): The best fitting model for assessment of the influence of farm, collection month (season) and field type (open vs. closed) and pesticide usage (inorganic vs. organic) on alpha diversity of arthropods associated with blueberries.

| Richness | Best fitting model (variables excl.) | AIC | BIC | logLik | deviance | df.resid |
|--------------------------------------|--------------------------------------|--------|--------|---------|----------|----------|
| Clipping Organic vs Inorganic | | | | | | |
| All guilds | pesticide | 507,22 | 518,87 | -250,61 | 501,22 | 357 |
| | pesticide*field | 500,34 | 519,77 | -245,17 | 490,34 | 355 |
| Scavengers (WC) | field | 118 | 124 | -56 | 112 | 51 |
| | pesticide*field | 121,9 | 131,9 | -56 | 111,9 | 49 |
| Phytophagous (WC) | pesticide | 69,605 | 73,707 | -31,802 | 63,605 | 26 |
| | pesticide*field | 73,6 | 80,4 | 31,8 | 63,6 | 24 |
| Predators (WC) | field | 47,289 | 50,277 | -20,645 | 41,289 | 0,0121 |
| | pesticide*field | 51,3 | 56,2 | -20,6 | 41,3 | 15 |

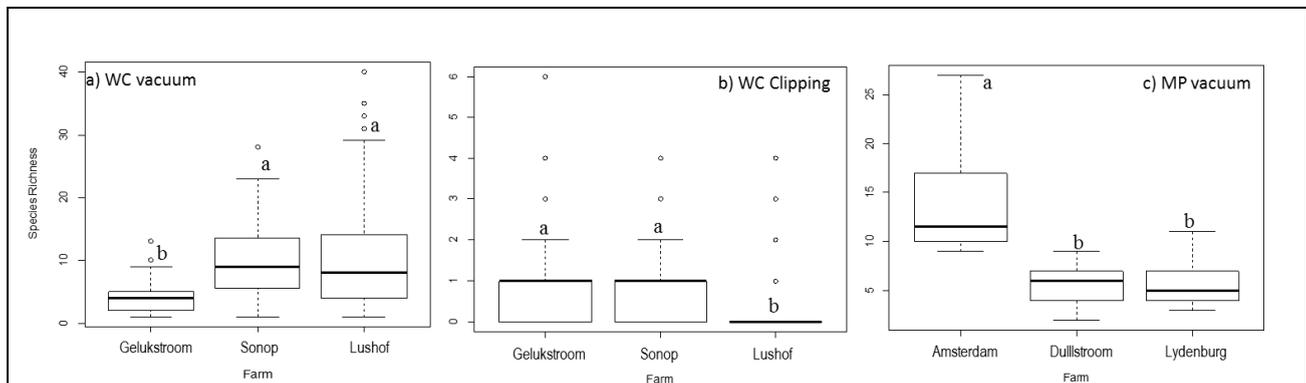


Figure 5: Median overall species richness of all arthropods collected per farm over one year using a) vacuum sampler, b) clipping in the Western Cape Province and c) Mpumalanga Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

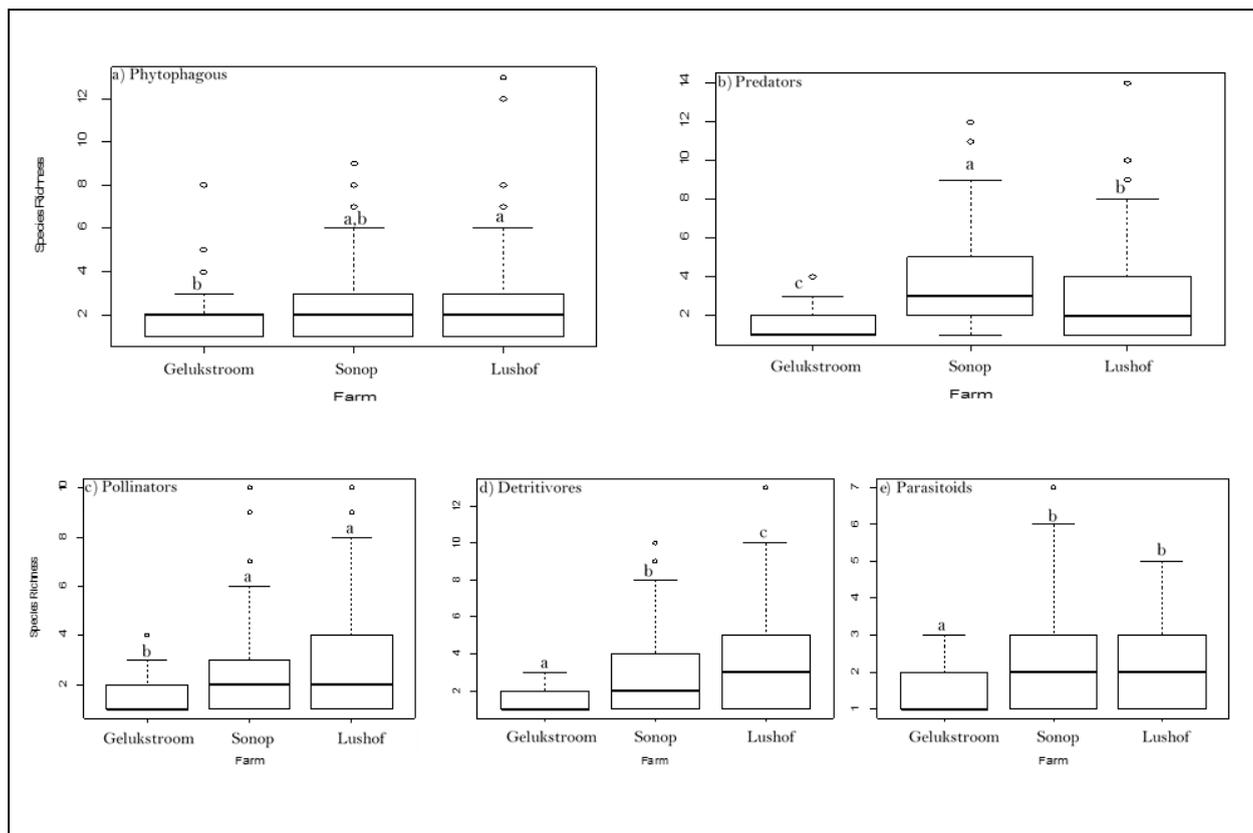


Figure 6: Median arthropod species richness of five feeding guilds collected over one year using a vacuum sampler at various farms in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

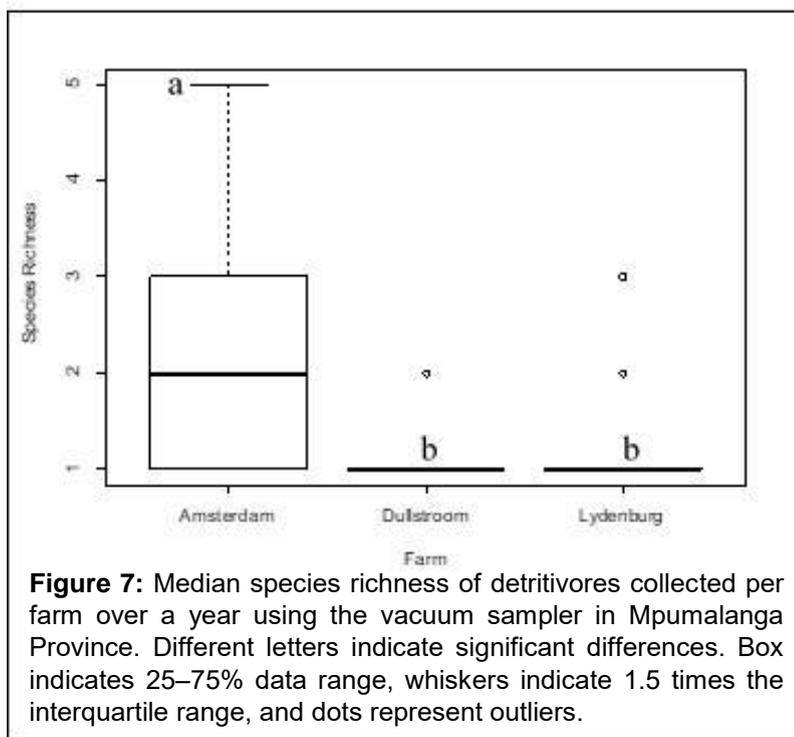


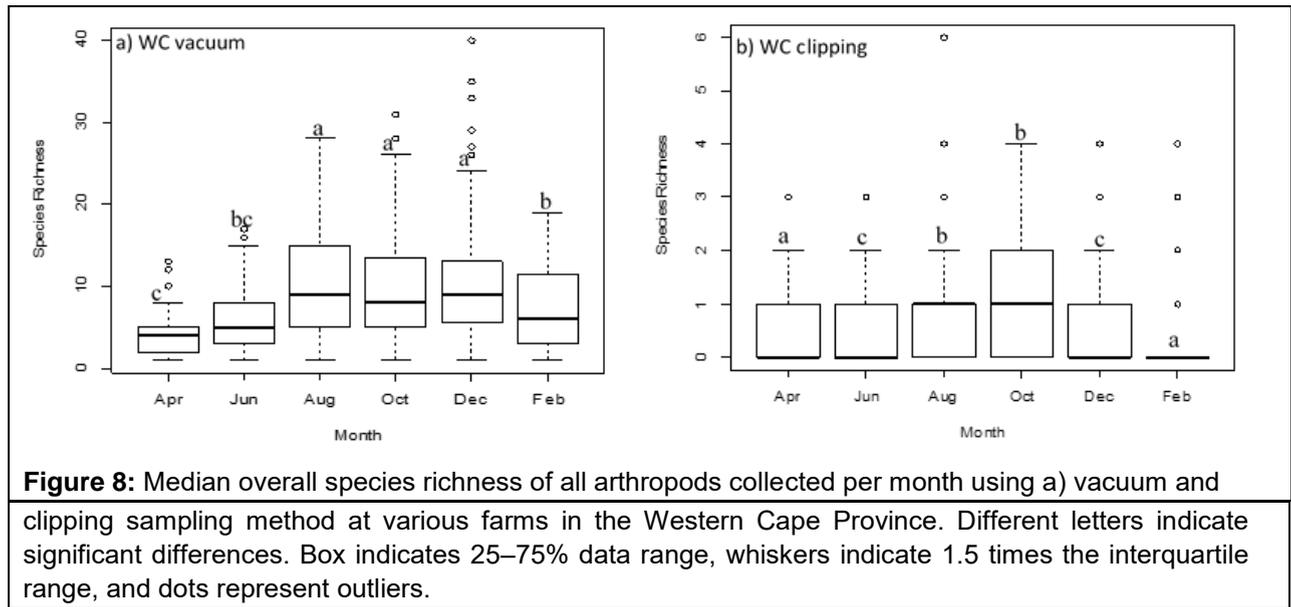
Figure 7: Median species richness of detritivores collected per farm over a year using the vacuum sampler in Mpumalanga Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

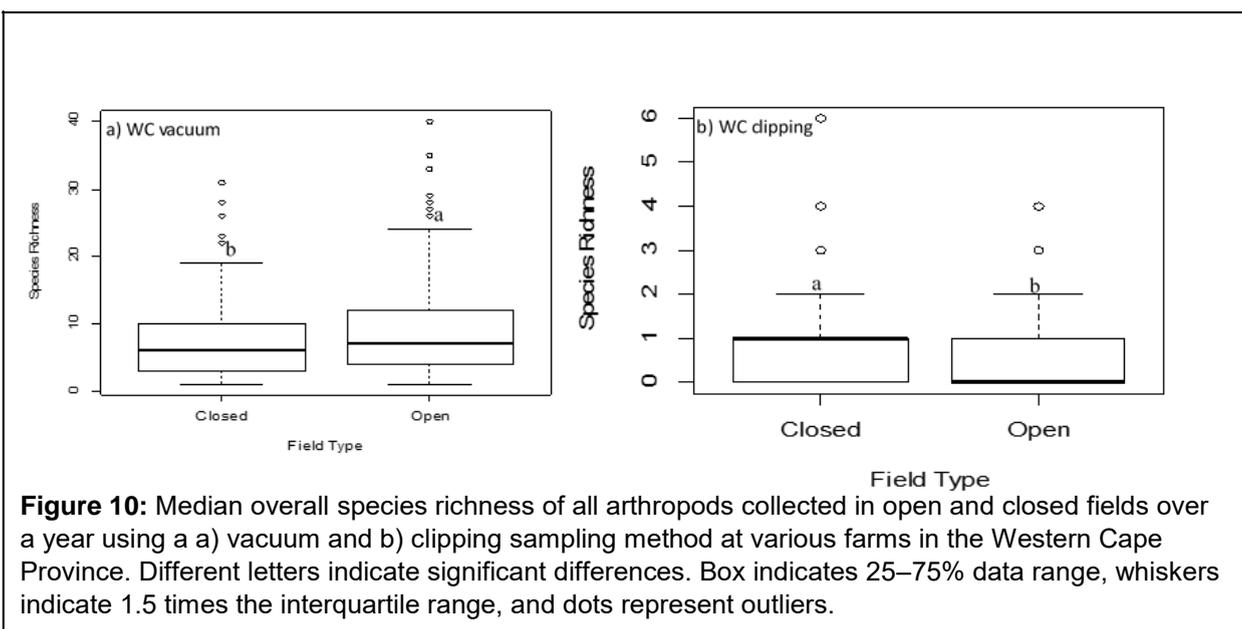
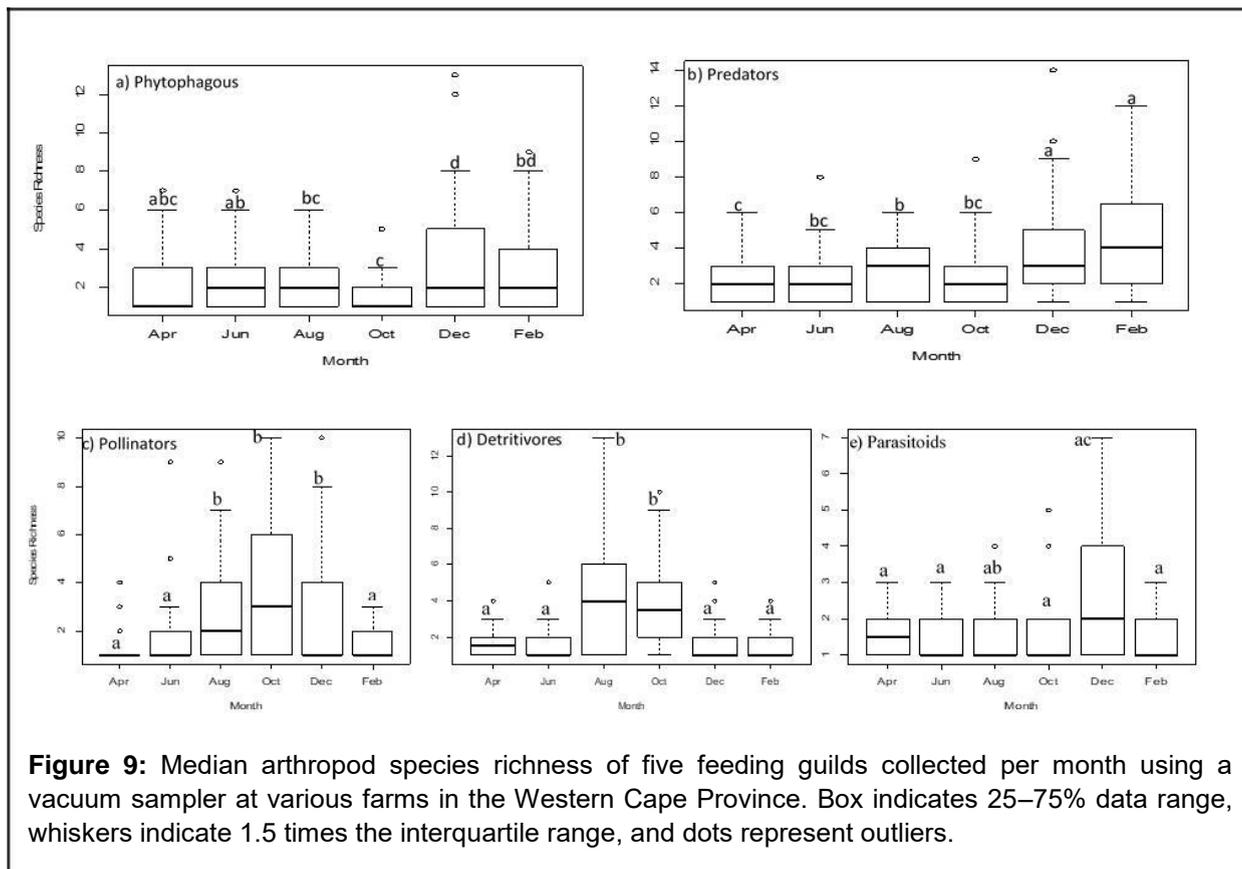
Seasonal species richness

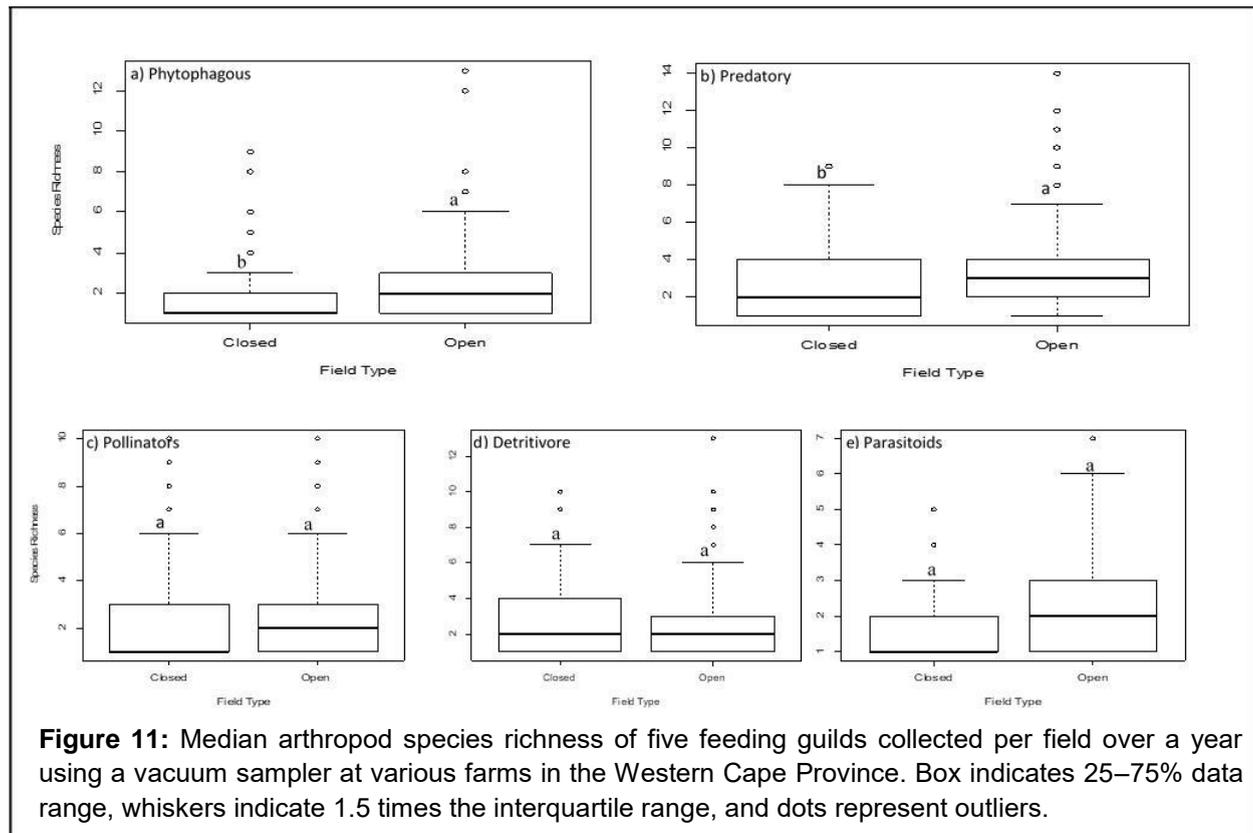
In terms of season, arthropod richness varied significantly throughout the year in the Western Cape Province (Table 3), with the highest number of species generally collected in August for vacuum and in October for clipping sampling methods (Tables 3, 4, appendix 1, Figure 8). Using the vacuum sampler, phytophagous arthropod richness was highest in December but not significantly more than in February (Table 3, Figure 9). Predator species richness was significantly high in February but was not significantly different than those collected in December (Table 3, Figure 9). Pollinator species richness was highest in October whereas detritivores were richest in August (Table 3, Figure 9). The number of parasitoid species remained similar throughout most the year but was slightly higher in December (Table 3, Figure 9). No significant differences were observed between the numbers of species in the different guilds throughout the year using the clipping method, likely to be due to very low sample sizes (Appendix 1). Notably, no phytophagous species were collected in April.

Species richness in open vs. closed fields

Significant differences in numbers of species of arthropods were observed when comparing the production of plants in open vs. closed systems in the Western Cape Province (Tables 2, 3 & 4, Figure 10). For overall species richness, open fields had a significantly higher number of arthropod species than closed fields (Figure 10). Closed fields had a higher number of predatory species whereas open fields had a higher number of phytophagous species. The rest of the guilds were statistically similar for both closed and open fields (Tables 3 & 4 and Figure 11). Using the clipping method, significant differences were observed between closed and open fields for overall collection, however, there were no significant differences for the different guilds (Tables 2, 4, Appendix 1).





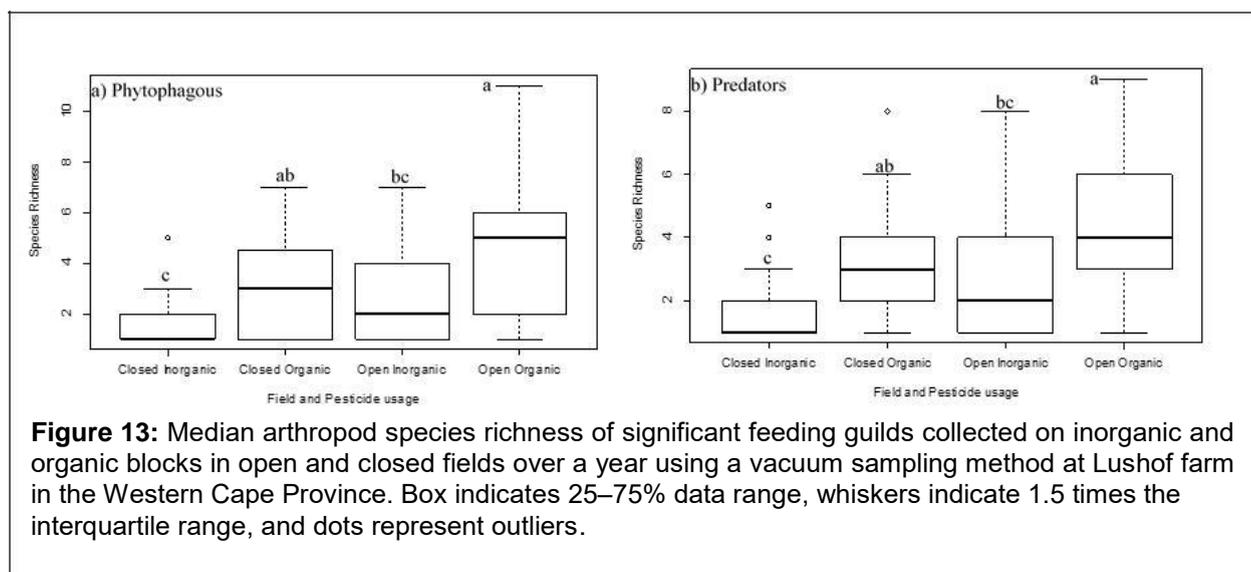
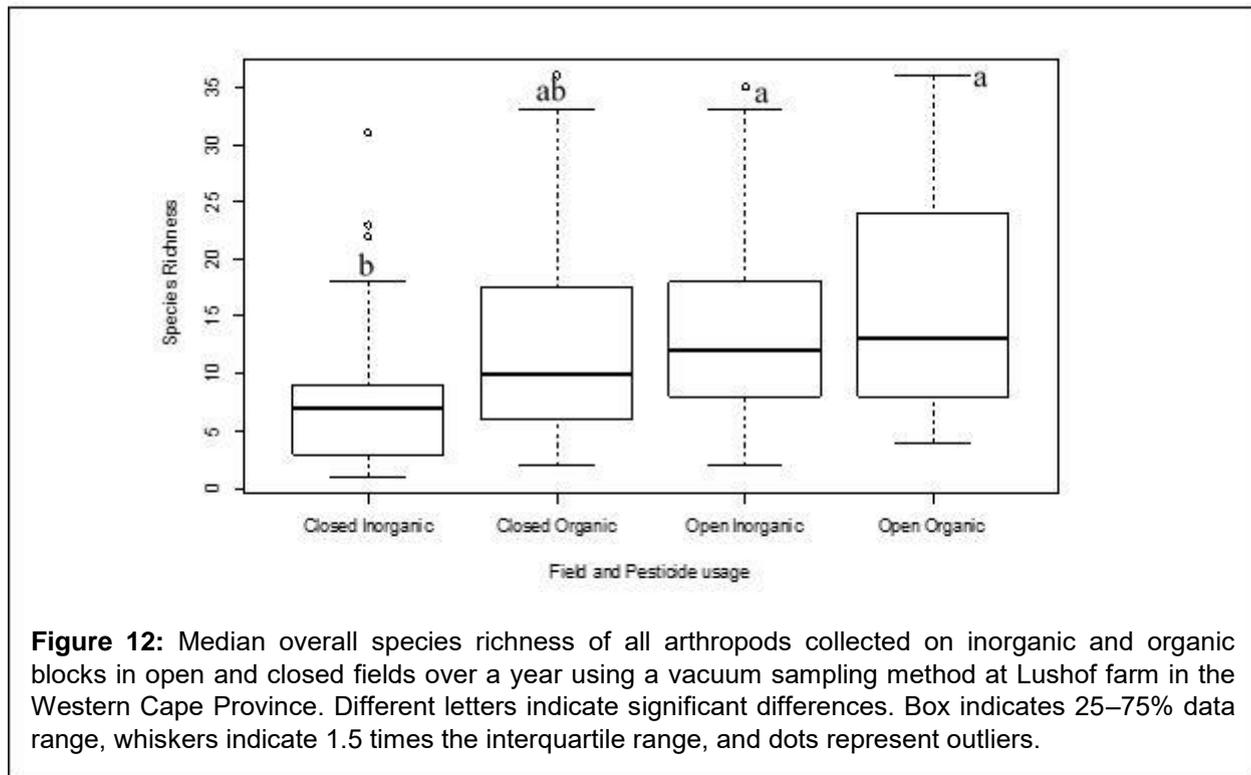


Species richness in organic vs. inorganic fields

A total of 225 and 65 morphospecies were collected from organic fields and inorganic fields, respectively, at Lushof farm using the vacuum sampler throughout the year (Table 2). Twenty-two and 17 morphospecies were collected using the clipping sampling method for these two fields, respectively (Table 2). Open vs. closed fields showed similar results for these production methods as was found when considering only the inorganic fields, in that open fields usually had significantly higher numbers of species (Tables 2, 3 & 4, Figure 12). For arthropods collected using the vacuum sampler, open organic systems had significantly higher overall and estimated species richness whereas closed inorganic systems had the least (Tables 2, 3 & 4, Figure 12). However, using the clipping method there were no statistical differences in the number of observed species between organic and inorganic systems (Tables 2 & 4, Appendix 1).

In terms of feeding guilds and using the vacuum sampling method, the numbers of species per guild varied between the organic and inorganic fields (Table 2). The number of species in the phytophagous and predator guilds were significantly higher in organic than in the inorganic fields. The number of species in the phytophagous and predator guilds were significantly higher in organic fields, particularly when these were open. Closed inorganic fields had significantly

lower number of species (Tables 2, 3 & 4, Figure 13). For the arthropods collected using the clipping sampling method, there were no statistical differences in the species richness of different guilds (phytophagous, predators and scavengers) between all field-type and pesticide-usage systems (closed-organic, closed-inorganic, open-organic and open-inorganic) (Tables 2 & 4, Appendix 1).



Arthropod Abundance

Overall arthropod abundance

A total of 9842 arthropod individuals were collected from the three farms in the Western Cape Province using the vacuum sampler and 1941 using the clipping sampling method throughout the year (Table 2). Farm identity had a significant influence on the number of arthropods collected. Using both the vacuum sampler and the clipping method, Sonop farm had a significantly higher number of arthropod individuals than both Gelukstroom and Lushof that were statistically similar (Table 2, 5 & 6, Appendix 2, Figure 14). In Mpumalanga Province, a total of 501 arthropod individuals were collected (Table 2). Farm identity also had a significant influence on arthropod numbers in this province (Table 6), with Amsterdam farm (using pesticides) having significantly higher numbers of arthropod individuals than Lydenburg and Dullstroom farms, which were statistically similar (Table 2, Figure 14).

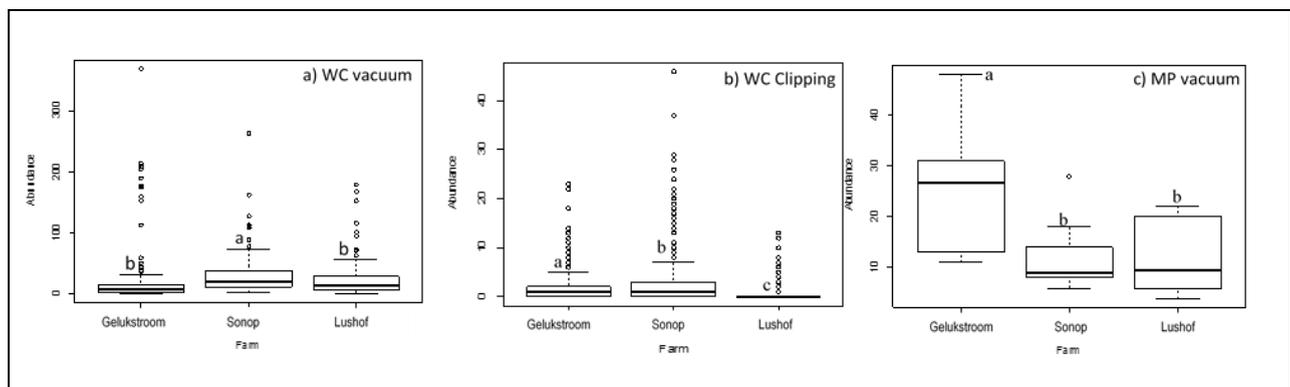


Figure 14: Median overall abundance of all arthropods collected per farm over one year using a a) vacuum sampler, b) clipping in the Western Cape Province and c) Mpumalanga Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

| Table 5: Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage on abundance of arthropods associated with blueberries collected using a vacuum sampler. | | | | | | |
|---|------------------|----|------------|---------|-----------|-----------------------|
| Abundance | | df | chi-square | p- | Posthoc | Dispersion (rdev/rdf) |
| All guilds (WC) | | | | | | 23,7 |
| | farm | 2 | 223,56 | <0,001 | fig. 14a | |
| | month | 5 | 5573 | <0,001 | fig. 17a | |
| | field | 1 | 485,39 | <0,001 | fig. 19a | |
| | farm*month*field | 27 | 2984,3 | <0,001 | | |
| Detritivore (WC) | | | | | | 5,466 |
| | farm | 2 | 314,59 | <0,001 | fig. 15d | |
| | month | 5 | 435,61 | <0,001 | fig. 18d | |
| | field | 1 | 0,4894 | 0,4842 | fig. 20d | |
| | farm*month*field | 24 | 315,75 | <0,001 | | |
| Phytophagous (WC) | | | | | | 20,61 |
| | farm | 2 | 1341,8 | <0,001 | fig. 15a | |
| | month | 5 | 4129 | <0,001 | fig. 18a | |
| | field | 1 | 60,503 | <0,001 | fig. 20a | |
| | farm*month*field | 27 | 1465 | <0,001 | | |
| Predators (WC) | | | | | | 3,72 |
| | farm | 2 | 206,11 | <0,001 | fig. 15b | |
| | month | 5 | 445,88 | <0,001 | fig. 18b | |
| | field | 1 | 43,174 | <0,001 | fig. 20b | |
| | farm*month*field | 27 | 264,06 | <0,001 | | |
| Parasitoid (WC) | | | | | | 1,55 |
| | farm | 2 | 30,33 | <0,001 | fig. 15e | |
| | month | 5 | 68,602 | <0,001 | fig. 18e | |
| | field | 1 | 7,2214 | <0,01 | fig. 20e | |
| | farm*month*field | 21 | 69,739 | <0,001 | | |
| Pollinators (WC) | | | | | | 5,25 |
| | farm | 2 | 69,788 | <0,001 | fig. 15c | |
| | month | 5 | 569,13 | <0,001 | fig. 18c | |
| | field | 1 | 0,0522 | 0,8193 | fig. 20c | |
| | farm*month*field | 27 | 295,09 | <0,001 | | |
| Organic vs Inorganic (WC) all guilds | | | | | | 19,4 |
| | pesticide | 1 | 186,14 | <0,001 | not shown | |
| | field | 1 | 220,96 | <0,001 | not shown | |
| | pesticide*field | 1 | 54,7 | <0,001 | fig. 21 | |
| Organic vs Inorganic (WC) Phytophagous | | | | | | 12,7 |
| | pesticide | 1 | 290,7 | <0,0001 | not shown | |
| | field | 1 | 4,4881 | 0,03413 | not shown | |
| | pesticide*field | 1 | 22,343 | <0,0001 | | |
| Organic vs Inorganic (WC) Predators | | | | | | 2,6 |
| | pesticide | 1 | 125,68 | <0,0001 | not shown | |
| | field | 1 | 60,845 | <0,0001 | not shown | |
| | pesticide*field | 1 | 0,873 | 0,3501 | fig. 22 | |

| Table 5 (cont.): Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage on abundance of arthropods associated with blueberries collected using a vacuum sampler. | | | | | | |
|---|-----------------|----|------------|---------|-----------|-----------------------|
| Abundance | | df | chi-square | p- | Posthoc | Dispersion (rdev/rdf) |
| Organic vs Pollinators | Inorganic (WC) | | | | | 2.6 |
| | pesticide | 1 | 125.68 | <0.0001 | not shown | |
| | field | 1 | 60.846 | <0.0001 | not shown | |
| Organic vs Parasitoids | pesticide*field | 1 | 0.873 | 0.3501 | not shown | 1.5 |
| | Inorganic (WC) | | | | | |
| | pesticide | 1 | 0.2 | 0.6548 | not shown | |
| Organic vs Detritivores | field | 1 | 3.325 | 0.06823 | not shown | |
| | pesticide*field | 1 | 1.204 | 0.2725 | | 5.25 |
| | Inorganic (WC) | | | | | |
| All guilds (MP) | pesticide | 1 | 0.5752 | 0.4482 | not shown | |
| | field | 1 | 0.8851 | 0.3468 | not shown | |
| | pesticide*field | 1 | 1.3315 | 0.2485 | | |
| Phytophagous (MP) | farm | 2 | 10.09 | 0.00644 | fig. 14c | |
| Predators (MP) | farm | 2 | 2,189 | 0.3348 | fig. 16a | |
| Parasitoids (MP) | farm | 2 | 0,677 | 0.7128 | fig. 16b | |
| Pollinators (MP) | farm | 2 | 8,07 | 0.01767 | fig. 16e | |
| Detritivore (MP) | farm | 2 | 5,745 | 0.05651 | fig. 16c | |
| | farm | 2 | 9,61 | 0.00816 | fig. 16d | |

* analysis to test the effect of farm was done using Kruskal-Wallis ANOVA

| Table 6: The best fitting model for assessment of the influence of farm, collection month (season) and field type (open vs. closed) and pesticide usage (inorganic vs. organic) on alpha diversity of arthropods associated with blueberries. | | | | | | | |
|---|------------------------------|--------|--------|---------|----------|----------|--|
| | Best model (variables excl.) | AIC | BIC | logLik | deviance | df.resid | |
| Vacuum Abundance | | | | | | | |
| All guilds (WC) | none | 9943,8 | 9982,6 | -4961,9 | 9923,8 | 350 | |
| | farm*month*field | 7013,5 | 7157,3 | -3469,8 | 6939,5 | 323 | |
| Detritivores (WC) | field | 2237,5 | 2269,3 | -1109,7 | 2219,5 | 245 | |
| | farm*month*field | 1971,2 | 2091,5 | -951,6 | 1903,2 | 220 | |
| Phytophagous (WC) | none | 5376,4 | 5410,8 | -2678,2 | 5356,4 | 220 | |
| | farm*month*field | 3956,4 | 4092,6 | -1945,7 | 3891,4 | 193 | |
| Predators (WC) | none | 2019,5 | 2056,5 | -999,8 | 1999,5 | 288 | |
| | farm*month*field | 1809,5 | 1946,3 | -867,7 | 1735,5 | 261 | |
| Parasitoids (WC) | none | 587,15 | 616,35 | -283,57 | 567,15 | 127 | |
| | farm*month*field | 559,4 | 649,9 | -248,7 | 497,4 | 106 | |
| Pollinators (WC) | field | 1745,2 | 1775,3 | -63,55 | 1727,2 | 202 | |
| | farm*month*field | 1506 | 1630 | -716 | 1432 | 174 | |

| Table 6 (cont.): The best fitting model for assessment of the influence of farm, collection month (season) and field type (open vs. closed) and pesticide usage (inorganic vs. organic) on alpha diversity of arthropods associated with blueberries. | | | | | | |
|---|------------------------------|--------|--------|---------|----------|----------|
| | Best model (variables excl.) | AIC | BIC | logLik | deviance | df.resid |
| Vacuum Organic vs Inorganic | | | | | | |
| All guilds | none | 2900,4 | 2911,6 | -1446,2 | 2892,4 | 116 |
| | pesticide*field | 2847,7 | 2861,6 | -1418,9 | 2837,7 | 115 |
| Detritivores (WC) | none | 726,56 | 733,74 | -360,28 | 720,25 | 77 |
| | pesticide*field | 728,3 | 740,3 | -359,2 | 718,3 | 76 |
| Phytophagous (WC) | none | 1663,6 | 1674,2 | -827,8 | 1655,6 | 100 |
| | pesticide*field | 1643,3 | 1656,5 | -816,6 | 1633,3 | 99 |
| Pollinators (WC) | none | 681,77 | 692,65 | -336,89 | 673,77 | 108 |
| | pesticide*field | 682,9 | 696,5 | -336,5 | 672,9 | 107 |
| Predators (WC) | none | 681,8 | 692,6 | -336,9 | 673,8 | 108 |
| | pesticide*field | 682,9 | 696,49 | -336,89 | 673,77 | 107 |
| Clipping Abundance | | | | | | |
| All guilds (WC) | none | 4936,4 | 4983,3 | -2458,2 | 4916,4 | 1070 |
| | farm*month*field | 4211,3 | 4395,7 | -2068,6 | 4137,3 | 1043 |
| Scavengers (WC) | none | 2179 | 2218,4 | -1079,5 | 2159 | 371 |
| | farm*month*field | 2085,9 | 2216,1 | -1010 | 2019,9 | 348 |
| Phytophagous (WC) | field | 195,35 | 211,83 | -89,674 | 179,35 | 50 |
| | farm*month*field | 210,67 | 247,75 | -87,333 | 174,67 | 40 |
| Predators (WC) | none | 821,1 | 852,8 | -400,5 | 801,1 | 167 |
| | farm*month*field | 764,6 | 863,1 | -351,3 | 702,6 | 146 |
| Clipping Organic vs Inorganic | | | | | | |
| All guilds (WC) | none | 805,47 | 821,01 | -398,74 | 797,47 | 356 |
| | pesticide*field | 780 | 799,4 | -385 | 770 | 355 |
| Scavengers (WC) | none | 107,5 | 113 | -49,8 | 99,5 | 25 |
| | pesticide*field | 109,4 | 116,3 | -49,7 | 99,4 | 24 |
| Phytophagous (WC) | none | 107,53 | 113 | -49,765 | 99,53 | 25 |
| | pesticide*field | 109,4 | 116,33 | -49,7 | 99,4 | 24 |
| Predators (WC) | field | 67,635 | 70,622 | -30,817 | 61,635 | 17 |
| | pesticide*field | 70,7 | 75,6 | -30,3 | 60,7 | 15 |

Guild Abundance

Using the vacuum sampling method, the numbers of arthropod individuals per guild varied significantly between the different farms in the Western Cape Province (Figure 15). The phytophagous guild overall contained the highest number of arthropod individuals, followed by the detritivore and predator guilds (Table 2). Herbivore and predator numbers were significantly different between all the farms. Phytophagous arthropods were most abundant at Gelukstroom, the farm that had the lowest numbers of predators. In contrast, at Sonop predator numbers were highest and herbivorous species numbers were lowest (Tables 5 & 6, Figure 15). Lushof farm had intermediate numbers of predators and herbivores. The number of pollinators and

detritivores were highest in Sonop but not significantly different from Lushof. Pollinator and detritivore guilds were the lowest in Gelukstroom farm. Parasitoids were significantly higher in Lushof than in Gelukstroom. Sonop had a lower number of parasitoids but not significantly different to Gelukstroom (Figure 15).

When using the clipping sampling method, scavengers were most abundant followed by the predators (Table 2). Number of individuals per guild varied significantly between different farms in the Western Cape Province, except for the phytophagous insect numbers that were similar for all the farms (Table 6). Predator and scavenger numbers were highest at Sonop farm (Appendixes 2, 3, & Table 6).

In Mpumalanga Province, the detritivore guild had the highest number of arthropod individuals when using the vacuum sampler, followed by the phytophagous guild (Table 2). The numbers of individuals for most guilds (phytophagous, predators and pollinators) was statistically similar between all the farms (Table 5, Figure 16). Numbers of detritivores and parasitoids were highest at Amsterdam farm, but only significantly so when compared to Lydenburg farm.

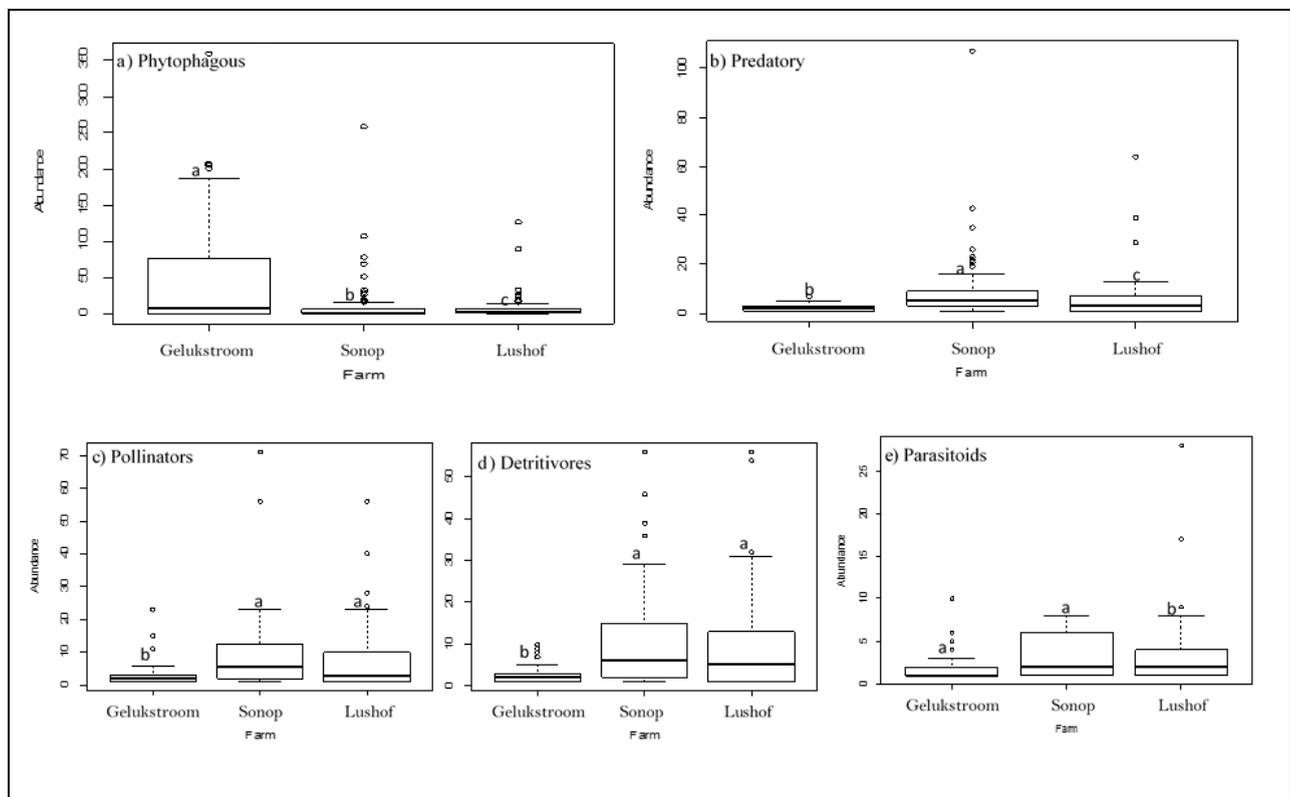


Figure 15: Median arthropod abundance of five feeding guilds collected over one year using a vacuum sampler at various farms in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

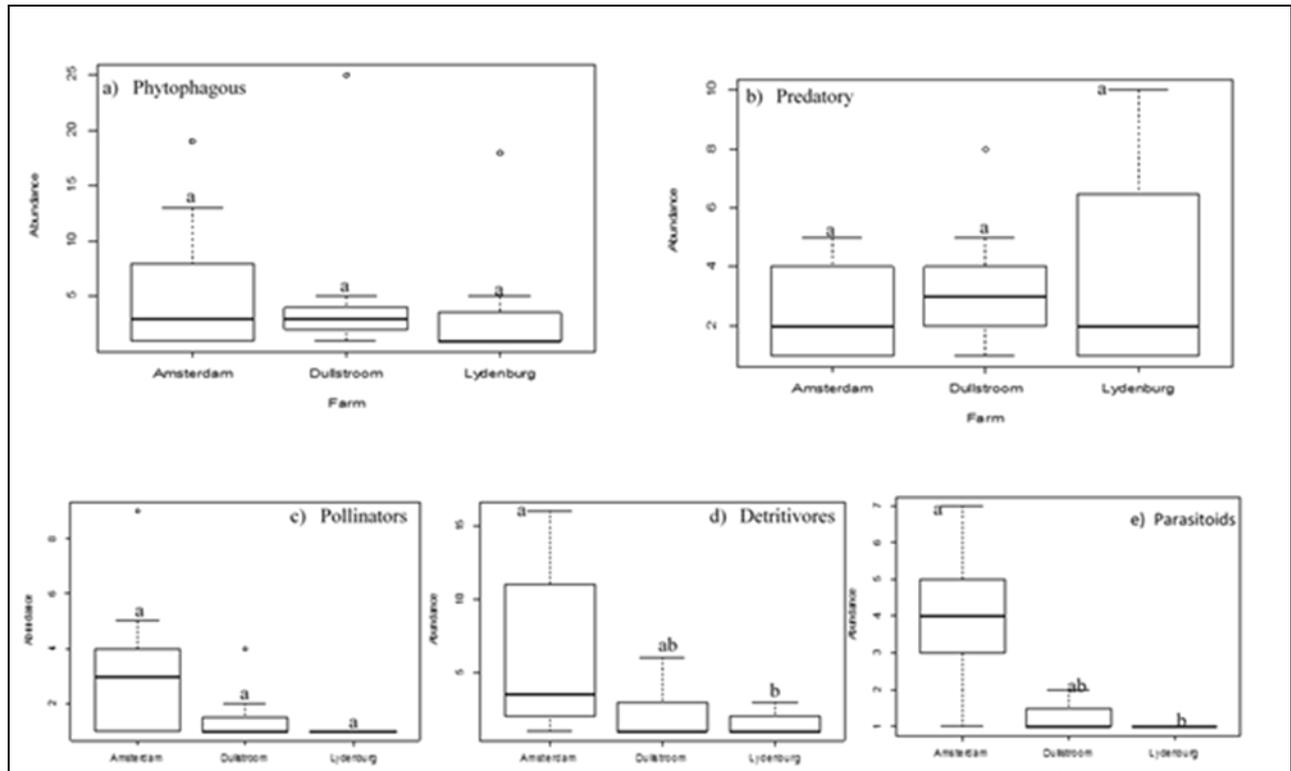


Figure 16: Median abundance of five feeding guilds collected per farm over a year using the vacuum sampler in Mpumalanga Province. Different letters indicate significant differences. Box indicates 25– 75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

Seasonal arthropod abundance

Using the vacuum sampling method, arthropod numbers varied significantly throughout the year in the Western Cape Province with significantly higher number of individuals in December due to high number of phytophagous individuals (Tables 5 & 6, Figure 17). Phytophagous arthropod numbers were lowest in August. Predator numbers peaked in February followed by December and were lowest in April, but not significantly so when compared to June and October (Tables 5 & 6, Figure 18). Pollinator numbers were significantly higher in October followed by August and lowest in April. Detritivores were most prevalent in August and their numbers were lowest in June and February (Tables 5 & 6, Figure 18). The number of parasitoids individuals was highest in December (Tables 5 & 6, Figure 18).

Using the clipping sampling method, the highest number of individuals were collected in October although it did not differ significantly to numbers of arthropods collected in August and December (Table 6, Appendix 2 & Figure 17). Phytophagous arthropod abundance was higher in February, but not significantly as compared to the other months (Appendix 2, 4) and no phytophagous arthropods were collected in April (Appendix 1 & 3). The number of predators was highest in December but it remained the same for all the other months (Appendix 2 & 4).

The number of scavenger individuals was highest in August, but it was not significantly different to June and October (Appendix 2 & 4).

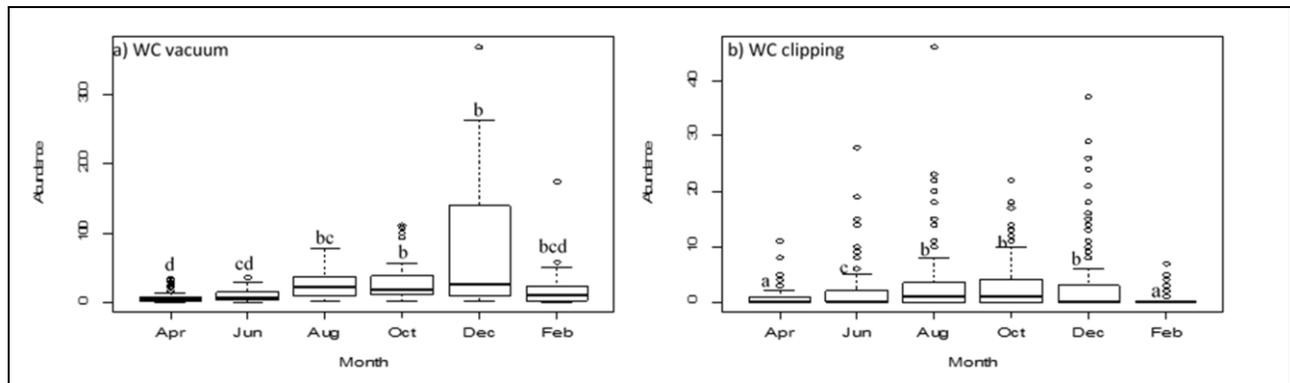


Figure 17: Median overall abundance of all arthropods collected per month using a a) vacuum and b) clipping sampling method at various farms in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

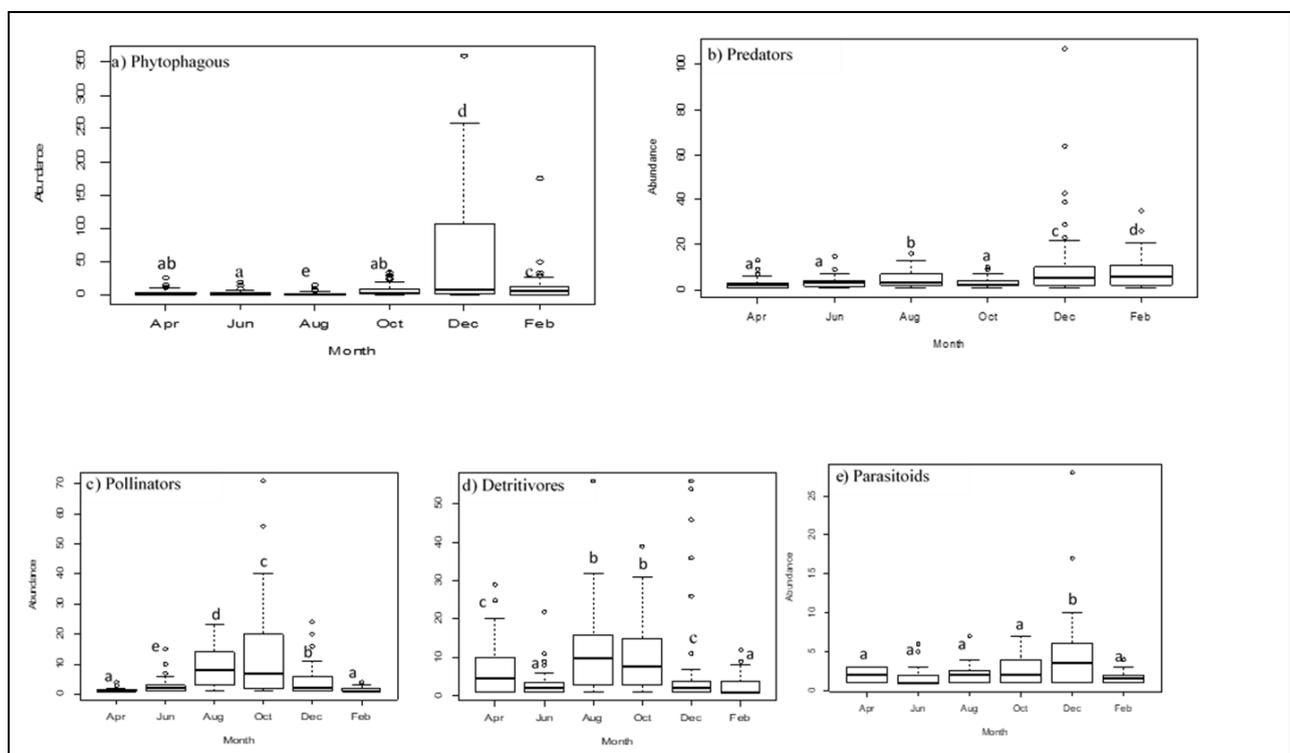
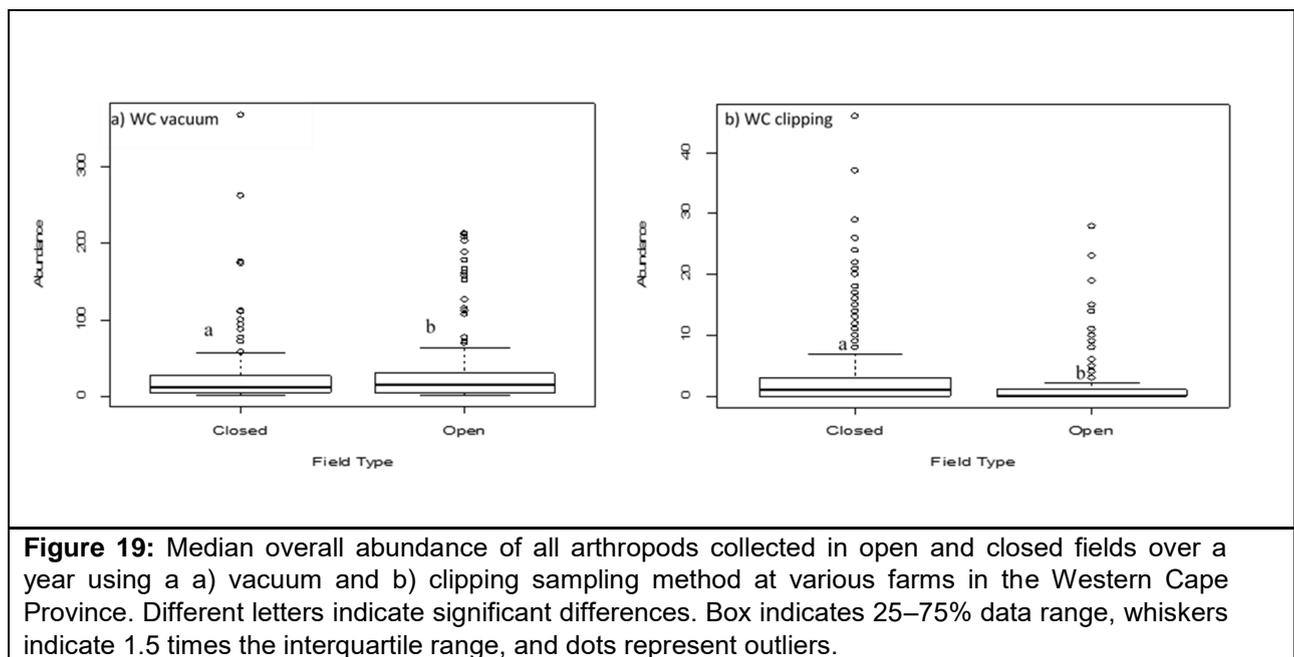


Figure 18: Median arthropod abundance of five feeding guilds collected per month using a vacuum sampler at various farms in the Western Cape Province. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

Abundance of arthropods in open versus closed fields

Some differences in numbers of arthropods individuals were observed when comparing the production of plants in open vs. closed systems in the Western Cape Province (Table 2, Figure 19). Open fields had a significantly higher number of arthropods than closed fields when considering all arthropods collected (Figure 19). Open fields also had a significantly higher number of individuals for the phytophagous, predator and parasitoid guilds (Tables 5 & 6, Figure 20). Pollinators and detritivore numbers were similar for both the field types (Table 5 & 6, Figure 20).

Using the clipping sampling method, closed fields had a significantly higher number of arthropods than open fields, but this was not true for vacuum sampling (Figure 19). The number of scavengers and predators were significantly different between the two systems, whereas phytophagous numbers were the same (Table 6, Appendix 2 & 5).



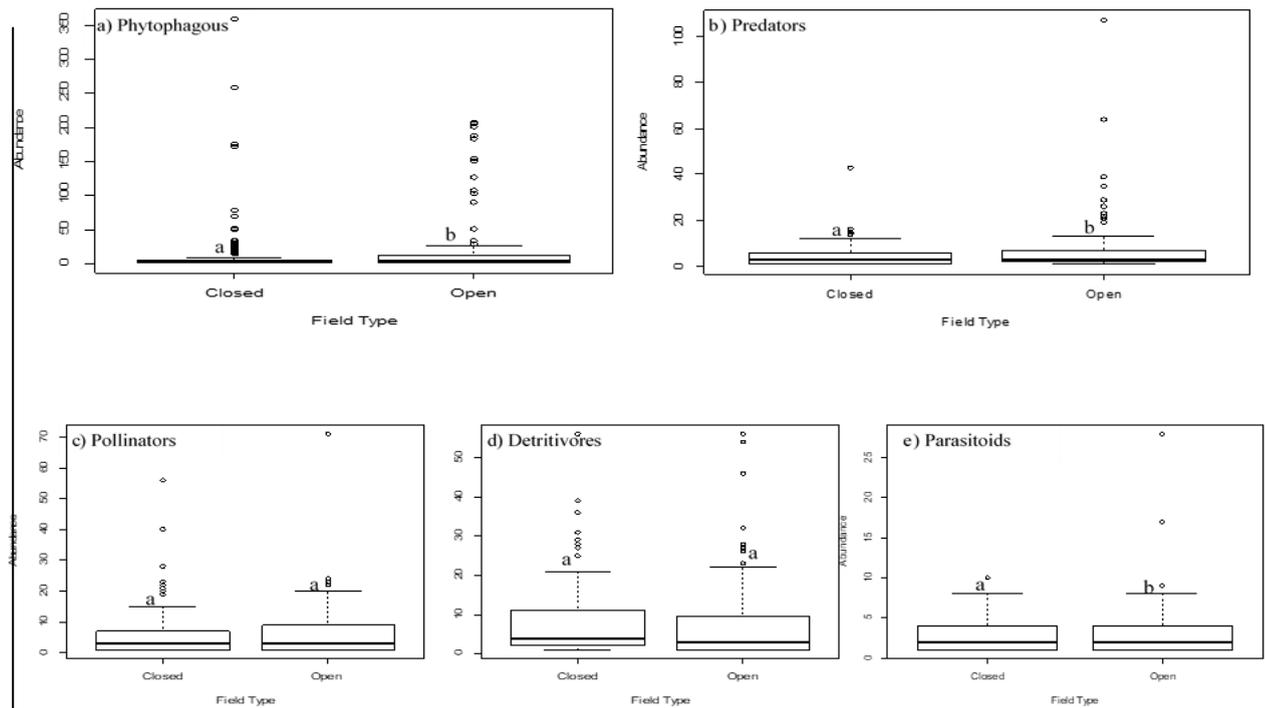


Figure 20: Median arthropod abundance of five feeding guilds collected per field over a year using a vacuum sampler at various farms in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

Abundance of arthropods in Inorganic vs organic fields

A total of 2385 and 1555 arthropod individuals were collected from organic fields and inorganic fields, respectively, at one farm (Lushof) in the Western Cape Province using the vacuum sampler throughout the year (Table 2). Sixty-five and 165 arthropod individuals were collected using the clipping sampling method for these two fields, respectively.

For arthropods collected using the vacuum sampler, organic fields had significantly high number of arthropod individuals than inorganic fields (Tables 2, 5 & 6 & Figure 21). Here, open fields had more arthropod individuals than closed fields (Figure not shown). When considering the interaction between pesticide usage and field type, there were no significance differences between all four systems (closed organic, closed inorganic, open organic and open inorganic) (Figure 21).

For the clipping method, there were no significant differences in the total number of arthropods between the organic and inorganic fields, but closed fields had a significantly higher number of arthropod individuals than the open fields (Tables 2 & 6, Appendix 2). The interaction between the two systems resulted in a higher number of arthropods in the closed inorganic fields as compared to open organic system (Appendix 6).

In terms of feeding guilds and using the vacuum sampling method, the numbers of individuals per guild varied between the organic and inorganic fields and between closed and open systems (Tables 5 & 6). There were no significant differences between all field and pesticide systems for all the guilds except for predator numbers that were significantly higher in open organic systems (Tables 5 & 6, Figure 22). For the arthropods collected using the clipping sampling method, there were no significant differences between the systems for predator and phytophagous guilds. The number of scavenger guild individuals was highest in closed inorganic systems and lowest in open organic systems (Tables 6 & 7, Appendix 2 & 7).

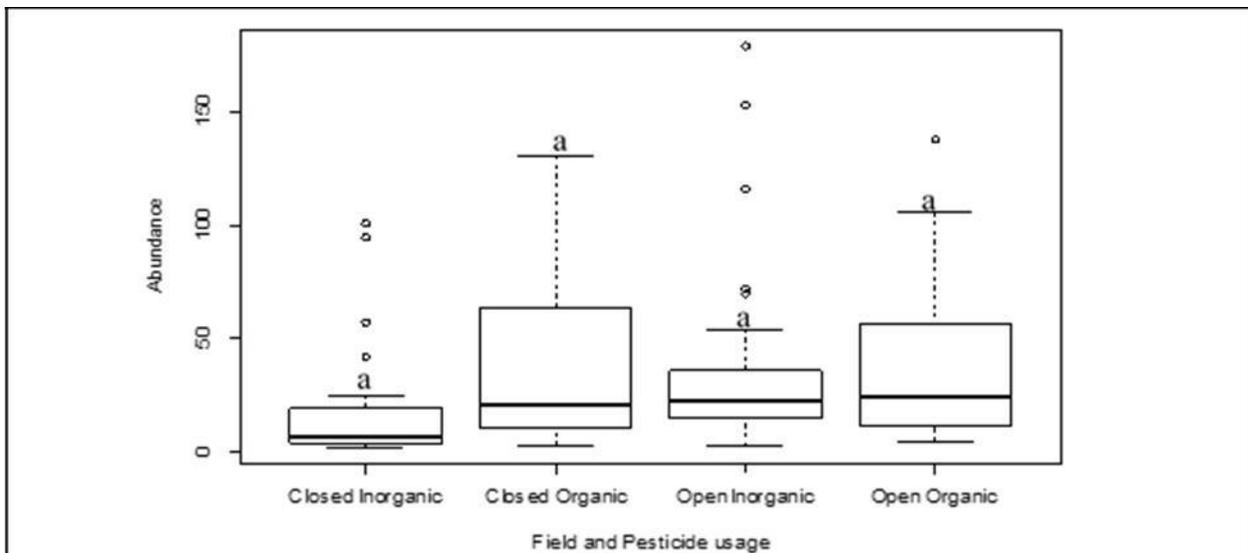


Figure 21: Median overall abundance of all arthropods collected on inorganic and organic blocks in open and closed fields over a year using a vacuum sampling method at Lushof farm in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

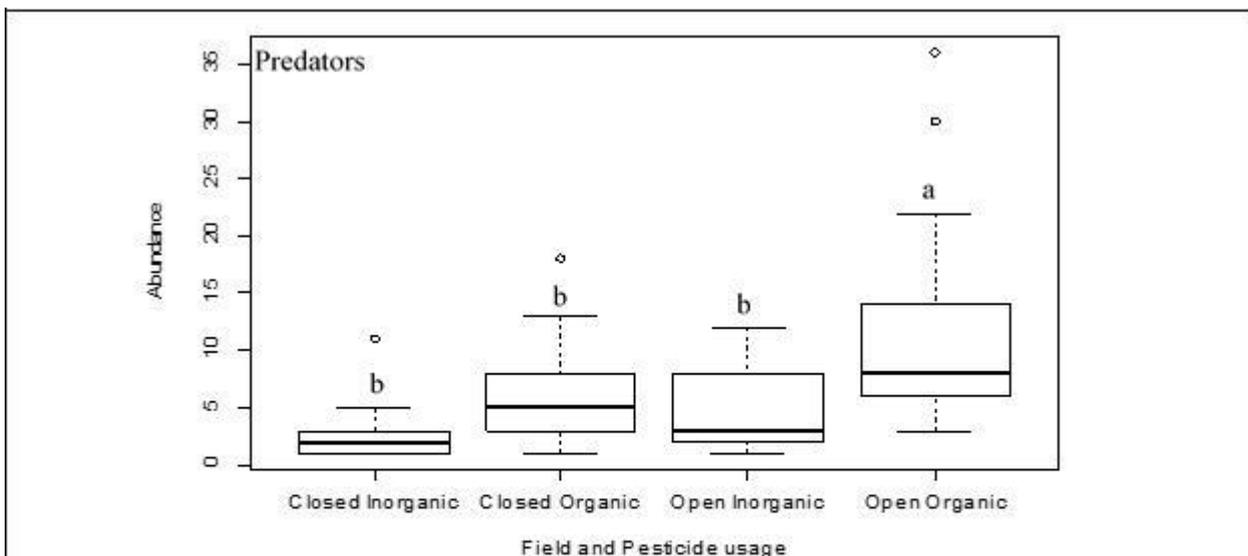


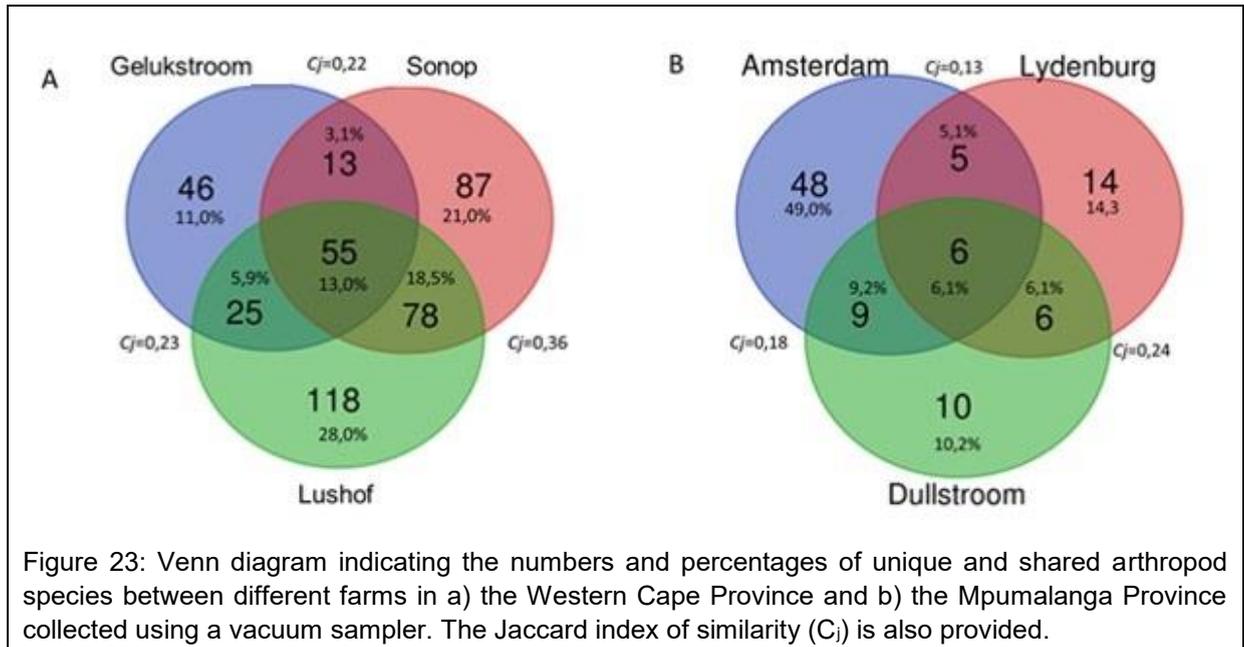
Figure 22: Median overall abundance of significant feeding guild, predators collected on inorganic and organic blocks in open and closed fields over a year using a vacuum sampling method at Lushof farm in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

Arthropod communities

Shared and unique taxa between farms

The bulk of the 422 species collected using the vacuum sampler in the Western Cape Province were unique to a specific farm. Only 13% of all species collected using this method were shared between the three farms. The most abundant of those shared were common pest taxa such as *Bemisia* species complex (Hemiptera: Aleyrodidae) (2593 individuals), a Sciaridae (Diptera) species (2139 individuals), a Miridae species (mirid 1: Hemiptera) (804 individuals) and a Chironomidae species (chiron 2, Diptera) (399 individuals) (Table 7). Communities between Sonop and Lushof were fairly similar according to the Jaccard's index (Figure 23).

Only 55 species were collected using the clipping method and a fair number of these were also unique per farm (Appendix 8). Ten of the 13 species shared between farms were mites, of which members of the Tydeidae were the most abundant. The scavenger *Tydeus grabouwi* (Acari, Tydeidae) was particularly abundant (Appendix 8, Table 7). As was found in the Western Cape Province, the bulk of species collected on different farms in the Mpumalanga Province were unique for each locality. Only 6 species were shared between the three farms, of which the most abundant species was a detritivore, COL D, (Scarabaeidae: Coleoptera) with 79 individuals. The inorganic farm, Amsterdam, had by far the highest number of unique species. The most abundant unique species at this site were the phytophagous COL A (Coleoptera: Chrysomelidae) with 32 individuals, followed by the pollinator, DIP 7 (Diptera) and parasitoid, HYM 2 (Hymenoptera), with 14 and 9 individuals, respectively. Lydenburg and Dullstroom were more similar to each other in terms of arthropod communities (as measured by the Jaccard coefficient) than either were to Amsterdam (Figure 23).



| Table 7: Numbers and percentages of unique and shared species of different guilds between farms, pesticide usage (organic vs. inorganic) and field type (open vs. closed) of all six farms, collected using vacuum and clipping methods. | | | | | | |
|---|-------------------|---------------------|------------------|---------------------|--------------------|--------------------|
| Variables | All guilds | Phytophagous | Predators | Detritivores | Pollinators | Parasitoids |
| Vacuum (WC) | | | | | | |
| GS,KM,LH | figure 23A | 11 (10%) | 13 (12%) | 9 (15%) | 10 (13%) | 12 (21%) |
| Lushof (LH) | | 36 (32%) | 27 (25%) | 11 (18%) | 27 (36%) | 17 (29%) |
| Sonop (KM) | | 38 (34%) | 20 (18%) | 9 (15%) | 12 (16%) | 9 (16%) |
| Gelukstroom (GS) | | 8 (7%) | 15 (14%) | 5 (8%) | 5 (7%) | 8 (14%) |
| Pesticide usage | | | | | | |
| inorganic, organic | 125(45%) | 26 (38%) | 32 (52%) | 23 (48%) | 23 (46%) | 21 (47%) |
| organic | 99(36%) | 31 (45%) | 23 (37%) | 13 (27%) | 16 (32%) | 13 (29%) |
| inorganic | 53(19%) | 12 (17%) | 7 (11%) | 12 (25%) | 11 (22%) | 11 (24%) |
| Field type | | | | | | |
| open, closed | 199(47%) | 35 (31%) | 55 (50%) | 37 (60%) | 41 (68%) | 30 (52%) |
| open | 146(35%) | 52 (46%) | 37 (34%) | 18 (29%) | 24 (32%) | 16 (27%) |
| closed | 77(18%) | 26 (23%) | 17 (16%) | 7 (11%) | 10 (13%) | 12 (21%) |
| Vacuum (MP) | | | | | | |
| AD, LB, DS | figure 23B | 0 | 2 (12%) | 2 (10%) | 1 (5%) | 1 (6%) |
| Amsterdam | | 11 (52%) | 4 (24%) | 10 (50%) | 10 (55%) | 11 (69%) |
| Dullstroom | | 2 (10%) | 3 (18%) | 0 | 4 (22%) | 1 (6%) |
| Lydenburg | | 8 (38%) | 0 | 4 (20%) | 0 | 1 (6%) |
| Clipping (WC) | | | | Scavengers | | |
| GS,KM,LH | appendix 8 | 2 (10%) | 3 (19%) | 7 (41%) | n/a | n/a |
| LH | | 2 (10%) | 2 (13%) | 1 (6%) | n/a | n/a |
| KM | | 8 (38%) | 6 (38%) | 3 (18%) | n/a | n/a |
| GS | | 7 (33%) | 4 (25%) | 2 (12%) | n/a | n/a |
| Pesticide usage | | | | | | |
| inorganic, organic | 10 (34%) | 1 (8%) | 3 (43%) | 5 (56%) | n/a | n/a |
| organic | 11(38%) | 7 (58%) | 2 (28%) | 2 (22%) | n/a | n/a |
| inorganic | 8(28%) | 4 (33%) | 2 (28%) | 2 (22%) | n/a | n/a |
| Field type | | | | | | |
| open, closed | 20(36%) | 4 (19%) | 5 (31%) | 10 (59%) | n/a | n/a |
| open | 15(27%) | 9 (43%) | 4 (25%) | 4 (23%) | n/a | n/a |
| closed | 20(36%) | 8 (38%) | 7 (44%) | 3 (18%) | n/a | n/a |

Shared and unique taxa between closed and open fields

Using the vacuum sampler, more arthropod species were collected in open fields than in closed fields at all farms. However, in general these different production methods also shared a fairly large percentage (47%) of morphospecies (Table 7). The most abundant were phytophagous species *Bemisia* spp. (Aleyrodidae: Hemiptera) with 2593 individuals followed by mirid 1 (Miridae: Hemiptera) with 804 individuals (Table 7). Unlike when using the vacuum sampling method, using the clipping sampling method revealed more species in closed fields than in open fields (Table 7). The scavenger, *Tydeus grabouwi* (Acari: Tydeidae) with 627 individuals was the most abundant, followed by the predator, Iolinidae species (Acari: Iolinidae), with 355 individuals.

Using the vacuum sampler, there were 146 species unique to open and only 77 species unique to closed fields (Table 7). Most abundant of the taxa unique to the open fields were the pollinator species Lep 09 (Lepidoptera) with 29 individuals and the phytophagous species Lygaeid 1 with 21 individuals (Table 7). Species unique to closed fields included the detritivore species sciarid 4 (Diptera: Sciaridae) with 10 individuals followed by the parasitoid species DIP B1 (Diptera) with 7 individuals and the phytophagous species miridjuv 5 with 7 individuals. Using the clipping method, there were no unique abundant taxa in the open field. Here, the scavenger species Triophtydeidae species (Acari: Triophtyidae) and a predatory (Acari: Anystidae) species had the greatest number of collected individuals. The most abundant taxa in closed fields were the phytophagous species *Scirtothrips auranti* (Thysanoptera: Thripidae) with 24 individuals and the scavenger species Tyd B (Acari: Tydeidae) with 13 individuals (Table 7).

Shared and unique taxa between organic and inorganic systems

For the arthropods collected using the vacuum sampler, species richness was higher in organic fields compared to inorganic fields. There were 99 species unique to organic and only 53 species unique to inorganic fields (Table 7). Most abundant of the taxa unique to the organic fields were the predator species Coccinelid 5 (Coleoptera: Coccinelidae) with 17 individuals and the detritivore species DIP T4 (Diptera: Psychodidae) with 13 individuals. The most abundant species unique to inorganic systems were the detritivore species DIP GG (Diptera), with 13 individuals, followed by the parasitoid species, HYM P (Hymenoptera) and the pollinator species DIP G (Diptera: Culicidae) (Table 7).

Using the clipping method, the highest number of individuals unique to organic fields were six for the phytophagous species Diaspididae (Hemiptera) and four for an *Aphis* sp. (Hemiptera: Aphididae). The highest number of individuals unique to inorganic systems were the phytophagous *Scirtothrips aurantii* (Thysanoptera: Thripidae) with 12 individuals and the

scavenger Tydeidjuv (Acari: Tydeidae) with four individuals (Table 7). Nevertheless, the bulk of species collected were shared between the two production types (Table 7). Using the vacuum sampling method, most abundant shared species included phytophagous species mirid 1 (Hemiptera: Miridae) with 484 individuals and the pollinator species chiron 2 (Diptera: Chironomidae) with 157 individuals. Using the clipping method, the most abundant taxa shared were the scavenger species Tydeidae species (Acari: Tydeidae) with 55 individuals and a *Tydeus* sp. (Acari: Tydeidae) with 31 individuals (Table 7).

Arthropod community composition

As expected, time of collection (season) had a strong effect on arthropod assemblage composition (Table 8). However, when controlling for the effect of season, PERMANOVA analysis indicated that farm identity influenced arthropod community assemblage composition significantly in both the Western Cape Province and in the Mpumalanga Province (Table 8). Farms also separated well after CAP analyses (Figures 24 & 25). This was true when considering samples collected using either the vacuum sampler (Table 8) or the clipping method in the Western Cape Province (Appendix 9 & 22).

When Western Cape farms were compared to each other in terms of overall assemblages, Sonop and Lushof were significantly different from Gelukstroom but similar to each other (Table 8). When comparing samples collected using the vacuum sampler, this was mostly driven by the phytophagous, predator and pollinator communities. Phytophagous species that were abundant were Aphid 2, *Bemisia afer* and mirid sp 1, 2 & 3. Abundant pollinator species were Chiron 2, DIP S, DROS and DIP OO, while abundant predator species were Chrysopid 1, COL 11, SP M, SP G and SP C. Using the clipping method in the Western Cape Province, overall assemblages differed significantly between Lushof and the other two farms, Sonop and Gelukstroom (Appendix 22). This was largely driven by their respective phytophagous and scavengers communities (Appendix 22). Phytophagous species that were abundant were thrip juveniles (Thyjuv) and the abundant scavenger species were Tydeidae species and *Tydeus grabouwi*. The abundant predator species were from families, Iolinidae and Phytoseiidae. Different guilds were often significantly different between farms in the Western Cape Province and in the Mpumalanga Province using either collection method. In the Mpumalanga province, all farms differed significantly in terms of their arthropod community assemblages which were largely driven by their respective phytophagous communities such COL A (Hemiptera: Chrysomelidae) (Table 8 & Figure 25). Results of CAP analyses for different guilds largely echoed results of PERMANOVA analyses. For results of CAP analyses of different feeding guilds please refer to appendix 1-16.

Interestingly, production of plants in terms of inorganic versus organic methods had little effect on most arthropod assemblages and guilds assessed (Table 8, Appendix 22).

However, after CAP analyses of overall arthropod communities from organic and inorganic fields separated more clearly for samples collected using the vacuum sampler (Figure 26) and those collected using the clipping method (Figure 27). Except for phytophagous communities (Appendixes 18a & 21), such separations of communities based on production type in terms of organic vs inorganic were not evident after CAP analyses of the various guilds separately (Appendix 17-21).

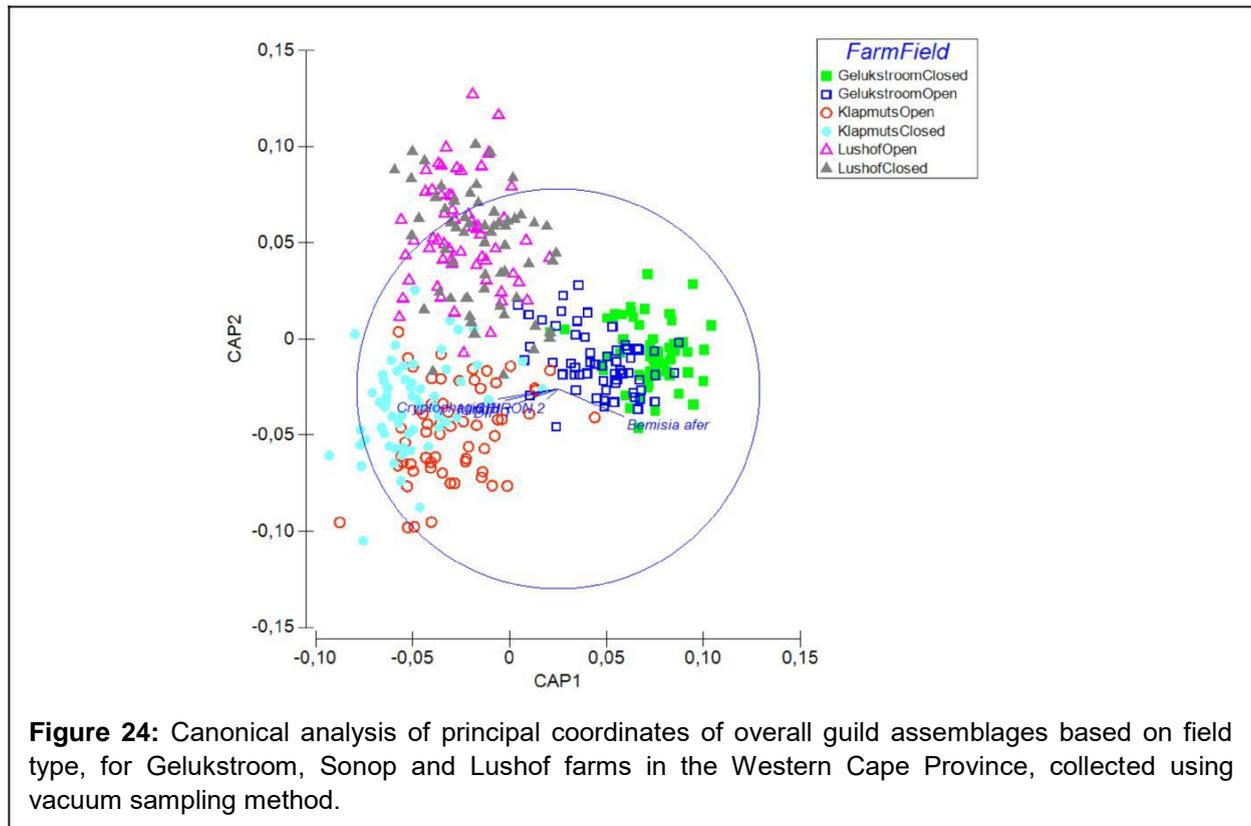


Figure 24: Canonical analysis of principal coordinates of overall guild assemblages based on field type, for Gelukstroom, Sonop and Lushof farms in the Western Cape Province, collected using vacuum sampling method.

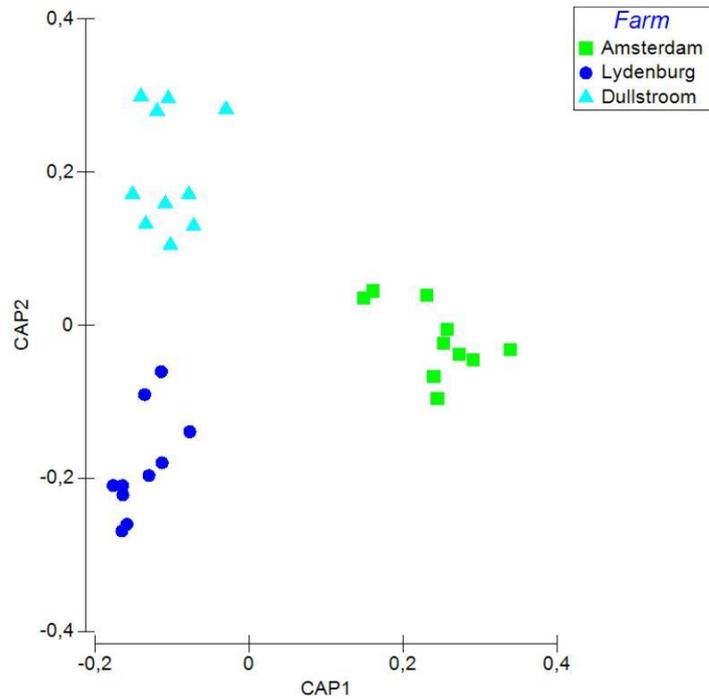


Figure 25: Canonical analysis of principal coordinates of the overall guild assemblages for Amsterdam, Dullstroom and Lydenburg farms in Mpumalanga province, collected using the vacuum sampling method.

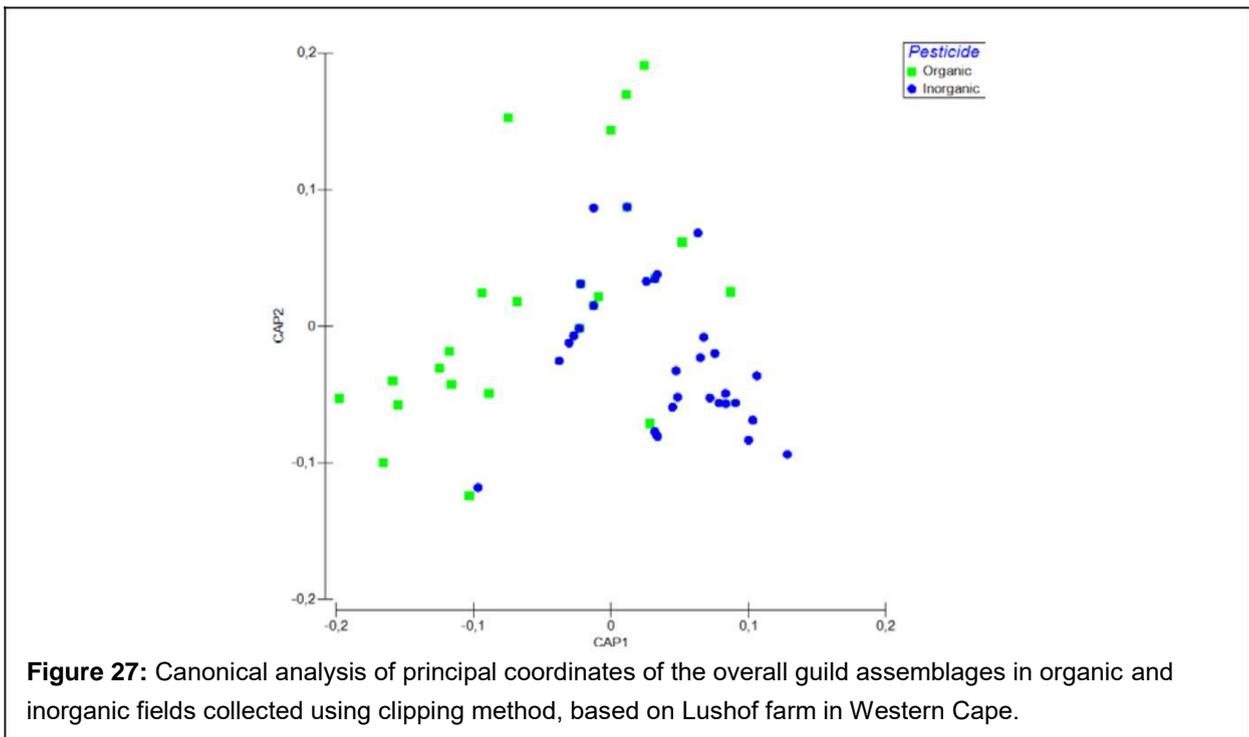
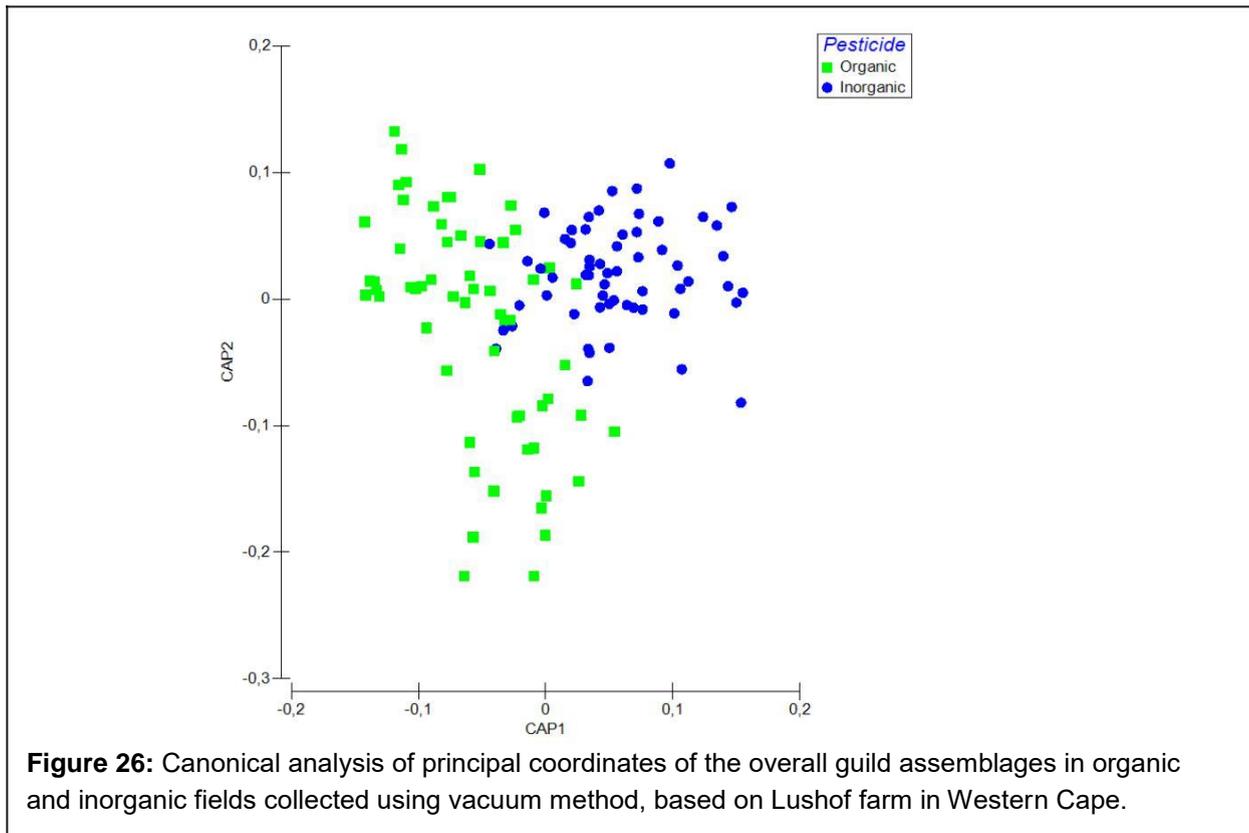


Table 8: Effect of farm, field type (open vs. closed) and pesticide usage (organic vs. inorganic) and their interaction on beta diversity of various feeding guilds on blueberry crops collected using vacuum method and a PERMANOVA pairwise test between farms

| | | Variables | df | Pseudo-F | P(perm) | Post hoc | |
|---------------------------|--------------|------------|----|----------|---------|-----------------------------|-----|
| WC vacuum | | | | | | | |
| All guilds | Farm | Field | 2 | 2,5749 | 0,0001 | GS≠KM; GS≠LH; KM=LH | |
| | | FarmxField | 1 | 1,3816 | 0,2404 | | |
| Phytophagous | Farm | Field | 2 | 1,1587 | 0,283 | | |
| | | FarmxField | 2 | 3,0548 | 0,0001 | GS≠KM;GS≠LH; KM=LH | |
| Predators | Farm | Field | 1 | 1,1283 | 0,3783 | | |
| | | FarmxField | 2 | 1,1559 | 0,2652 | | |
| Parasitoids | Farm | Field | 2 | 3,6085 | 0,0003 | GS≠KM;GS≠LH; KM≠LH | |
| | | FarmxField | 1 | 1,704 | 0,1218 | | |
| Pollinators | Farm | Field | 2 | 1,3074 | 0,1667 | | |
| | | FarmxField | 2 | 1,0572 | 0,2926 | | |
| Detritivores | Farm | Field | 1 | 1,0335 | 0,4326 | | |
| | | FarmxField | 2 | 0,93314 | 0,4106 | | |
| Formicidae | Farm | Field | 2 | 1,9737 | 0,0027 | GS≠LH;GS=KM; KM=LH | |
| | | FarmxField | 1 | 1,0454 | 0,4349 | | |
| Formicidae | Farm | Field | 2 | 0,98838 | 0,4081 | | |
| | | FarmxField | 2 | 1,6679 | 0,0374 | all comparisons significant | not |
| Formicidae | Farm | Field | 1 | 0,99741 | 0,4626 | | |
| | | FarmxField | 2 | 0,9951 | 0,4227 | | |
| Formicidae | Farm | Field | 1 | 2,7168 | 0,074 | GS≠KM? | |
| | | FarmxField | 1 | 1,8587 | 0,1476 | | |
| Formicidae | Farm | Field | 1 | 1,4315 | 0,2442 | | |
| | | FarmxField | 1 | 1,4315 | 0,2442 | | |
| <i>Field Type</i> | | | | | | | |
| Open field (WC) | All guilds | Farm | 2 | 1,7488 | 0,0082 | all similar | |
| | | Month | 5 | 6,594 | 0,0001 | | |
| Closed field (WC) | All guilds | Farm | 2 | 2,4383 | 0,0004 | GS≠KM;GS≠LH; KM=LH | |
| | | Month | 5 | 6,3659 | 0,0001 | | |
| <i>Pesticide usage</i> | | | | | | | |
| Organic vs Inorganic (WC) | All guilds | Field | 1 | 1,0059 | 0,4679 | | |
| | | Pesticide | 1 | 0,90926 | 0,5353 | | |
| Organic vs Inorganic (WC) | Phytophagous | Field | 1 | 0,77842 | 0,7123 | | |
| | | Field | 1 | 1,6946 | 0,1731 | | |
| Organic vs Inorganic (WC) | Predators | Pesticide | 1 | 1,0485 | 0,4393 | | |
| | | Field | 1 | 0,95499 | 0,4603 | | |
| Organic vs Inorganic (WC) | Predators | Field | 1 | 0,9818 | 0,4219 | | |
| | | Pesticide | 1 | 0,62506 | 0,6468 | | |
| Organic vs Inorganic (WC) | Predators | Field | 1 | 1,3636 | 0,345 | | |
| | | Pesticide | 1 | 1,3636 | 0,345 | | |

| Table 8 (cont.): Effect of farm, field type (open vs. closed) and pesticide usage (organic vs. inorganic) and their interaction on beta diversity of various feeding guilds on blueberry crops collected using vacuum method and a PERMANOVA pairwise test between farms | | | | | | |
|--|--------------|-----------------|----|----------|---------|---------------------|
| | | Variables | df | Pseudo-F | P(perm) | Post hoc |
| Organic vs Inorganic (WC) | Parasitoids | Field | 1 | 1,0184 | 0,4302 | |
| | | Pesticide | 1 | 1,0006 | 0,4457 | |
| | | FieldxPesticide | 1 | 0,93932 | 0,4884 | |
| Organic vs Inorganic (WC) | Pollinators | Field | 1 | 0,63828 | 0,7 | |
| | | Pesticide | 1 | 1,0758 | 0,3941 | |
| | | FieldxPesticide | 1 | 0,80902 | 0,542 | |
| Organic vs Inorganic (WC) | Detritivore | Field | 1 | 0,70549 | 0,6652 | |
| | | Pesticide | 1 | 1,284 | 0,2848 | |
| | | FieldxPesticide | 1 | 0,59912 | 0,6164 | |
| MP vacuum | | | | | | |
| | All guilds | Farm | 2 | 5.6145 | 0.0001 | AD≠LB;AD≠DS; LB≠DS |
| | Phytophagous | Farm | 2 | 8.779 | 0.001 | AD≠LB;AD≠DS; LB≠DS; |
| | Predators | Farm | 2 | 1.3449 | 0.133 | |
| | Parasitoids | Farm | 2 | 1.6679 | 0.0326 | AD=LB;AD≠DS;LB=DS |
| | Pollinators | Farm | 2 | 1.5356 | 0.052 | |
| | Detritivores | Farm | 2 | 3.4533 | 0.0004 | AD≠LB;AD=DS; LB≠DS |
| | Formicidae | Farm | 1 | 6.5396 | 0.0048 | AD≠LB |

DISCUSSION

In this study I set out to examine the main factors that influence the diversity and variance in arthropod communities on blueberries in South Africa. As expected, there was large seasonal variation in richness and abundance of arthropods, with highest numbers recorded in the warmer summer months. There were strong contrasts in numbers of arthropods between different production areas (different farms and different provinces) which may be related not only to specific production methods used, but also due to influx of arthropods from the immediate surrounds of the blueberry fields. Whether blueberries were produced in open fields or in tunnels also significantly affected arthropod numbers. Organic production had an overall positive effect on arthropod numbers as opposed to inorganic production methods, especially in terms of increased predator numbers. This study therefore highlights the significant effects that production method and location could have on pests and predators in fields, which would have significant impacts on production costs, profits and the environment.

Effect of different variables on arthropods associated with blueberries

Location effect

There were often significant differences in arthropod species richness, abundance and community assemblage composition between the different farms evaluated in this study. This variation is highly likely due to biogeographical location and/or agricultural practices employed at different farms (Alvarez et al. 2001). The overwhelming effect of location on arthropod taxa present in fields is exemplified by the very low numbers of taxa shared between farms and the high numbers of unique species collected at each location, irrespective of the province where farms were located. The actual species present within fields may therefore be drastically different on each farm purely due to location and demonstrates that farming practices need to be particularly pliable to adapt to each unique situation. Although not empirically tested, this likely signifies that the numbers of pests and beneficial arthropods within fields will be highly linked to the surrounding vegetation, regardless of agricultural practices (Lee et al. 2001; Lewinsohn et al. 2005; Rocca and Greco 2011; Schellhorn et al. 2014). This follows other studies that indicate that arthropod composition associated with crop production is often according to the regional pool of species (Jeanneret et al. 2003; Rocca and Greco 2011). When planning integrated management of pests and promotion of beneficial arthropods, there should therefore be a thorough understanding of the factors that influence the movement of these organisms into fields. For example, to increase numbers of beneficial arthropods such as predators and pollinators in blueberry fields one would need to optimise surrounding fields for their benefit, as has been suggested for many other crops (Walton and Isaacs 2011; Penca et al. 2017; Whitehouse et al. 2018).

Specific farming practices employed within fields will also influence the numbers and identity of arthropods present. For example, in the Western Cape Province, numbers of arthropod individuals were significantly higher at Sonop farm than at both Gelukstroom and Lushof. Overall numbers of arthropod species were generally similar between the latter two farms. Generally higher numbers of arthropods at Sonop farm may be due to specific farming practices employed at this farm, which differs by having a less stringent pesticide regime. In the Mpumalanga province, differences in arthropod abundance between Amsterdam farm and the other two farms could be ascribed to management practices as the former relied heavily on use of pesticides. Interestingly, in the two provinces pesticide use had an opposite effect on arthropod numbers. Pesticide use in the Western Cape decreased overall arthropod abundance, while in Mpumalanga it was increased. This may be due to different pesticides

being in use and / or arthropod identities, some might be more susceptible to pesticide, however this was not investigated. Specific agricultural practices within fields that could influence arthropod numbers seem to not be as important as pesticide usage. For example, in the Mpumalanga Province Lydenburg farm had lots of weeds growing amongst blueberry crops and the Dullstroom farm had a well-maintained field with short grass. They therefore differ in this important management aspect. However, the reliance on chemicals at these farms were similar, and they had similar abundances of all groups of arthropods evaluated in this study.

There was an interplay between the numbers of phytophagous arthropods and herbivores, which was especially evident at farms in the Western Cape Province. When the abundance of predators was low at a particular farm, there was a high number of plant feeding species (e.g. at Sonop and Gelukstroom farms). In addition to high number of predators, Sonop also had high numbers of parasitoids. This provides some evidence that it may be beneficial to farmers to manage fields in a way to optimise the numbers of predatory and parasitic arthropods to help manage herbivore numbers. This is in contrast to earlier studies that indicate that phytophagous species numbers are more reliant on resources than top-down control (Root 1973), but in line with more recent studies that highlight the importance of predators in managing herbivore numbers (Isaacs et al. 2009; Letourneau and Goldstein 2001).

Seasonal effect

A strong seasonal effect was detected for arthropods associated with blueberries in the Western Cape Province. A large number of arthropods species and individuals were observed between the warmer months of August and December using both sampling methods. This highlights the well-known link between arthropod developmental rates and warmer periods of time (Leach and Isaacs 2018) and is a good indication of when to focus control efforts. This is not surprising for blueberry production in this area, as plants would be in various optimal developmental stages for both phytophagous and predatory arthropods. Crops would have new leaves, flowers and fruit that would be attractive to pollinators, pests and predators. The remains of pruned branches in between the crop rows will also promote litter-consuming arthropods. Different guilds did, however, peak at different times. For example, pollinating species started increasing in numbers in August and were most abundant in October. Phytophagous numbers were highest in December. The parasitoids and predator numbers peaked in December and February, likely due to higher availability of food sources following the increase of herbivore numbers. However, the month's delay in the peak of natural enemies after that of phytophagous taxa may be too late to effectively control their numbers, as was found for flower thrips (Arévalo et al. 2009). Therefore, in terms of pest management, I advocate an integrated pest management system where the use of insecticides should augment other control strategies

(Feber et al. 1997; Cárdenas et al. 2015). For example, pesticides that target the pests themselves but are not harmful to predators and parasitoids would reduce pest numbers to a level where predators can effectively control pest populations (Wightman et al. 1982; Gaigher and Samways 2010; Dasonville et al. 2013).

Field type (open vs. closed production) effect

Altered micro environments (unique microclimates) are achieved by the use of high tunnels covered in plastic which interfere with light and other physical parameters (Lamont 2009). The use of various physical barriers in this protected cultivation method in the Western Cape Province is aimed at inhibiting pest invasions (Cane and Payne 1993; Antignus et al. 1996; Antignus 2000. Costa et al. 2002). The advantages are profit driven, both in terms of improved production and pesticide management. Exclusion of rain in such tunnels also promotes crop quality and shelf life and reduces the chances of fungal disease attack (Wells 1998; Jiang et al. 2004; Demchak 2009; Lamont 2009). Whether the production of blueberries in tunnels is beneficial in terms of arthropod numbers has not yet been investigated in South Africa.

I found that there was generally higher species richness and abundance of arthropods in open versus closed fields when using the vacuum sampler. This was largely driven by high numbers of plant feeding species in the open fields. Natural enemies were significantly more species-rich in the closed fields, but with higher abundance in open fields. Therefore, overall there seems to be a benefit to planting blueberries in tunnels, as herbivore numbers are reduced.

However, these patterns were reversed when investigating only the smaller taxa that were sampled using the clippings method. Here, numbers of herbivorous species and individuals were significantly higher in closed systems than in open fields. This is likely because some smaller arthropods prefer enclosed warm, humid environments where they are less exposed to high winds etc. that would negatively influence their behavioural ecology (Milholland and Meyer 1984; Scherm and Krewer 2008; Leach and Isaacs 2018).

Thus the decision to plant blueberries in open vs. closed fields, at least in terms of pests, will be dictated by which taxa are likely to cause the most damage. For example, *Bemisia* spp. was the most abundant species shared between the two fields, but had significantly more individuals in open than in closed fields. This is in line with previous studies conducted which showed a reduction of infestation levels of *Bemisia* spp. and other pests, such as aphids and thrips, on different crops when plastic cover was used (Antignus et al. 1996; Antignus 2000; Costa et al. 2002; Chyzik et al. 2003; Chiel 2006). Thus, if the most damaging pest in the demchakea is whitefly species, then production in an open system is preferable. Conversely, if larger

arthropods, such as Coleoptera, are more damaging, production in more closed systems may prove to be best.

Inorganic vs. organic farming practices

In this study I found that whether blueberries were produced in organic vs. inorganic fields had little effect on arthropod numbers, especially in closed production systems. Phytophagous arthropods and predators were however noticeably higher in open-organic systems. This is in line with a study by Whitehouse et al. (2018) in the main blueberry producing state of Georgia in the USA that used the same sampling method (vacuum) employed here. They observed higher abundance of parasitoids and predators in organic fields than in conventional fields. However, the effect of open vs. closed production systems in our study seemed to be more important for arthropods than pesticide usage, as significant effects between organic and inorganic farms generally disappeared when including this interacting factor. Therefore, in terms of pest of blueberries, organic production may be favoured as it does not lead to a large increase in pest species, while possibly promoting the biodiversity of predators. It should therefore be more profitable due to higher prices obtained in markets and less investments in terms of pesticides. However, it should be noted that blueberry production is fairly new in South Africa (Meyer and Prinsloo 2003; Sikuka 2017) and future pest outbreaks could be more severe than those currently experienced (new pest associations or novel genotypes of known pests) which will then likely necessitate the use of pesticides for control (Elsner and Whalon 1985; Hancock and Draper 1989; Bolda et al. 2010; Hulbert et al. 2011; Roubos et al. 2014; Carroll et al. 2015).

CONCLUSIONS

This study represents the first comprehensive study on arthropod diversity on blueberries in South Africa. The arthropod composition of various guilds was highly influenced by geographical location and agricultural practice, making control of pests for this emerging crop quite challenging as each farm would need to develop their own pest management strategy. Closed fields seem to be able to suppress the numbers of some pest taxa, but this effect is taxon dependent. Farmers would need to weigh up the extra production costs associated with production in tunnels, with the risk of damage by particular groups of herbivores. Organic farming promoted slightly higher numbers of natural enemies, but again this was dependent on production in the open or in tunnels. Organic production is however advocated as it compared favourably in terms of pest numbers to inorganic production systems, it would have less significant environmental impact, and would be linked to higher profitability.

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Chapter 3

Supplementary description of the blueberry bud mite, *Acalitus vaccinii* (Keifer, 1939) (Acari: Trombidiformes: Eriophyidae), based on specimens from South Africa

ABSTRACT

In 2014 the most devastating eriophyoid pest of blueberry, the blueberry bud mite, *Acalitus vaccinii* (Acari: Trombidiformes: Eriophyidae) was detected in the Mpumalanga province of South Africa for the first time. This is the first detection of this pest outside of North America, to which it is native. It was noted that the current description of this species does not meet modern minimum standards and there is therefore a strong need to review the description. In this chapter I re-describe *A. vaccinii* from South African specimens and include counts, measurements, photographs and line drawings of all morphologically important characters of females, males and immatures. Features are presented and compared using line drawings, phase contrast light microscopy and scanning electron microscopy. Information regarding biology and morphological variation is discussed and a key to Eriophyoidea species known on *Vaccinium* worldwide is provided.

INTRODUCTION

Blueberries, *Vaccinium* spp. (Ericaceae), are an internationally cultivated crop native to North America. In South Africa (SA) the blueberry industry was established approximately 32 years ago (Meyer and Prinsloo 2003) to cater for growing international trade demand from North America during their winter season and due to the health benefits of the fruits (Lazarus and Schmitz 2000; Staff 2000). Today, blueberries are planted in all provinces in South Africa: Free State, Gauteng, Northern Province, Mpumalanga, KwaZulu Natal, Western Cape (Meyer and Prinsloo 2003), Eastern Cape, North West (Sikuka 2017) and Limpopo (Erasmus 2013). About

80% of South African blueberries are exported to the Northern hemisphere (Erasmus 2013; Sikuka 2017).

The blueberry bud mite (BBM), *Acalitus vaccinii* (Keifer 1939) (Acari: Trombidiformes: Eriophyidae) is considered the most devastating eriophyoid pest of blueberries (Keifer 1939; de Lillo and Duso 1996). It occupies the scales of buds and blossoms of wild and cultivated blueberries. The mite feeds on developing plant tissues (Keifer 1941), causing poor growth and low yield. Symptoms of infestation include red blistering on buds, production of small leaves and fruits and malformed flowers (Garcia-Salazar 2002). Infestations by *A. vaccinii* in commercial blueberry plantations can lead to substantial yield reductions, as affected buds do not produce fruit. These symptoms are very similar to those of winter stress (Figure 1), and it is therefore important to confirm the presence of the mite using a microscope (Garcia-Salazar 2002; Weibelzahl and Liburd 2010) before applying mitigatory actions. Species identification of Eriophyidae mites is frequently difficult, and for accurate identifications, high quality descriptions and keys are important. Accurate identification of specimens is important for optimal management and control programmes as different species may have different tolerances to control measures. Accurate identification is also essential for accurate decision making by biosecurity organizations at borders.

In 2012 damage due to *Acalitus vaccinii* was observed on a single farm in the Mpumalanga province and the mite was identified for the first time in South Africa in 2014 (Craemer 2018). This was the first time that this pest has been recorded outside of North America. Within two years of its detection, it had caused an estimated 80% reduction in fruit production, resulting in substantial losses for the farmer (Craemer 2018). Further surveys by the South African Department of Agriculture, Forestry and Fisheries (DAFF) also confirmed blueberry bud mite infestations in other locations within Mpumalanga, but showed that it had not yet spread to other provinces (DAFF, pers. comm.). This was a relief as the mite was still absent from the Western Cape, which is the biggest blueberry production area. The restricted distribution of *A. vaccinii* in South Africa necessitates continued monitoring, management and containment programs to prevent further spread.

In an effort to accurately identify this newly discovered pest in SA, it became clear that a supplementary description of *A. vaccinii* will be useful (Craemer 2018). Accurate identification of eriophyoids requires a high level of expertise and this difficulty in making a proper identification is compounded by incomplete species descriptions and lack of identification keys. In this particular case, no comprehensive key to the more than 90 valid *Acalitus* species worldwide or to eriophyoids on *Vaccinium* spp. existed. South African individuals of *A. vaccinii* were initially identified (Craemer 2018) by using the key to eriophyoid world genera by Amrine et al. (2003)

and comparison with the original and subsequent descriptions of *A. vaccinii* known on *Vaccinium* (Keifer 1939). The identification was confirmed by comparing the morphology of the specimens to descriptions of the 17 *Acalitus* species known in Africa (Meyer 1990), and to the two *Acalitus* species known on Ericaceae (Keifer 1965; 1966).

Acalitus vaccinii was first described by Keifer (1939) as *Eriophyes vaccinii* from specimens collected in North Carolina, USA. It was then transferred to *Aceria* (Keifer 1946) and later to *Acalitus* (Baker and Neunzig 1970) based on the lack of a foretibial seta. Keifer's description included measurements and drawings of some characteristics of the protogyne female, with brief mention of the male. Later, Baker and Neunzig (1970) described immatures of *A. vaccinii*. Although these descriptions are adequate for positive identification of the species, they do not meet modern standards for eriophyoid descriptions (Amrine and Manson 1996; de Lillo et al. 2010). Additionally, some morphological features were not measured or described, some features were over-looked, and some life stages (most notably the male) were not adequately described.

With recent improvements in visualisation equipment, the original description can now be reviewed and supplemented with additional information for accurate identification and taxonomic comparisons. According to these new standards an adequate description should have at least seven drawings in the style of Keifer, depicting: 1) dorso-ventral view of the whole mite; 2) lateral view showing gnathosomal details and setae; 3) annuli and microtubercles; 4) prodorsal shield showing ornamentation; 5) coxal area, showing coxae, genital coverflap and internal genitalia of both sexes; 6) lateral view of legs; and 7) empodium. In addition to line drawings, measurements of different characters and counts of annuli should be included. These characteristics should be included for all life-stages where possible (Amrine and Manson 1996; de Lillo et al. 2010).

In this chapter I review and augment the existing morphological descriptions of *A. vaccinii* females and immatures and describe the male fully for the first time. I present more detailed drawings, photographs, counts and measurements of all morphologically important characters of females, males and immatures. Additional detailed information is provided using modern techniques, including digital images of slide-mounted mites from phase contrast light microscopy (PCLM) and Scanning Electron Microscopy (SEM) for clearer understanding of the structure of tiny features. To aid in identification and future comparison of Eriophyoidea found on blueberries, a key to known eriophyoid species on *Vaccinium* is provided.

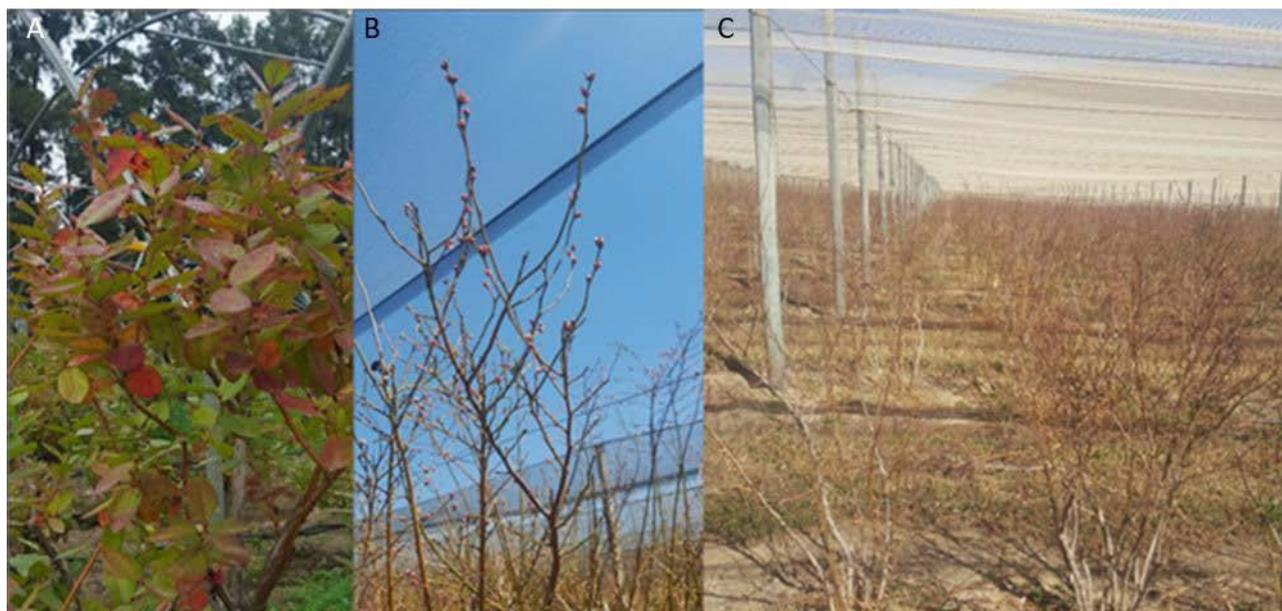


Figure 1: Winter stress a) red leaves of jewel cultivar at Lushof farm in Western Cape b) elliot plants with red flower buds and red stems and c) without leaves at Dullstroom farm in Mpumalanga Province

MATERIAL AND METHODS

Mite collection and preparation

Blueberries were sampled and eriophyoid mites collected between March 2015 and November 2016 from cultivated blueberries under hail net at farms in the Mpumalanga Province of South Africa. Farms were near the towns of Dullstroom, Lydenburg and Amsterdam. Due to the fact that this is a quarantine pest, information is sensitive and the individual farm names cannot be provided. Amsterdam farm had 10 hectares of various cultivars, namely *V. corymbosum* 'Bluecrop' (6 years old), 'Berkley', 'Elliott' and 'Spartan' and *V. virgatum* 'Centurion' (all 14 years old). Dullstroom farm had four hectares of four and eight year old 'Elliott' plants. Lydenburg had three hectares of 25 year old *V. virgatum* 'Climax' and 'Delite' plants. Samples were collected by the first author or the particular farmer. Thirty samples of 30cm long shoots were taken at random per variety and per block. To prevent possible cross contamination, secateurs were sterilized between collections in different blocks. Shoot samples were wrapped in damp paper towels and placed into separate re-sealable plastic bags labelled with the collection information. These were kept in a cooler box in the field and at 4°C in the laboratory until examination. Mites stayed alive at this temperature for about three months after sample collection. The collected material was examined by the first author for the presence of mites using a stereo microscope at a minimum of 30X magnification. Each bud was examined by first removing the outer scale layer and repeating this until the innermost parts were exposed. Eriophyid mites were collected with a

fine needle and placed into a drop of sorbitol-isopropyl alcohol medium until mounting (de Lillo et al. 2010).

Morphological description

Phase contrast light microscopy (PCLM)

Mites were slide-mounted using F-medium according to Keifer (1975) and de Lillo et al. (2010). Specimens were mounted both dorso-ventrally and laterally in order to study different characters. Slide-mounted specimens were air dried in an oven at 40°C for a minimum of 2 weeks or until completely dry before sealing the coverslip and morphological examination and identification with phase contrast at 1000X magnification. The mites were identified to the genus *Acalitus* using the key to the world eriophyoid genera by Amrine et al. (2003) and to species using the description of Keifer (1939). Mounted specimens were studied using different compound microscopes, depending on specific features examined. A Zeiss Axioskop Imager M2 microscope (Zeiss, New York, NY), equipped with a drawing tube and Zeiss AxioCam Cc5 digital camera and ZEN 2012 software, was used for line drawings and capturing of phase contrast images. Measurements were done with a Leica DM 2500 microscope (Leica Weitzlar, Germany) connected to a Leica digital camera and Leica application suite v 3.1.0 software. Measurements and counts were done on live digital images allowing live focus to ensure the accurate viewing of very small and fine features.

The morphological terminology in this chapter follows Lindquist & Amrine (1996). Characters chosen for drawing follow the recommendations of Amrine and Manson (1996) and de Lillo et al. (2010). Measurements and counts were taken from 18 specimens, comprising 12 adult females, 2 adult males, 2 nymphs and 2 larvae. Measurements are in micrometers (μm) and refer to length of the morphological characters unless specified otherwise. Measurement ranges (minimum to maximum), rounded off to one decimal place, are presented in Table 1, and means are rounded off to the nearest integer with the exception of point four to point six (0.4 - 0.6), which are presented in the descriptive text. Ink drawings were digitized using a Bizhub C 558 copier/scanner machine at the highest resolution.

Slide-mounted voucher material of all stages was deposited in the mite collections of the Department of Agriculture Forestry & Fisheries, Plant Quarantine Station in Stellenbosch, South Africa and of the Agricultural Research Council, National Collection of Arachnida —Acari in Pretoria, South Africa.

Scanning electron microscope (SEM)

To study structural details on the surface of mite individuals, a three dimensional image was obtained using a scanning electron microscope. Scanning electron microscopy was performed at the electron microbeam unit of the Central Analytical Facility of Stellenbosch University, Stellenbosch, South Africa. Low temperature scanning microscopy was not available at this university, therefore specimens were processed at ambient temperature using a Leo®1430VP SEM (Leo Electron Microscopy Ltd., Cambridge, United Kingdom) and Zeiss Merlin Field Emission SEM (Zeiss Merlin FE-SEM) (Microscopy, New York, US). Specimens were processed either untreated or after critical point drying or after conductive coating using gold to obtain usable images.

Untreated specimens were processed and imaged at ambient temperature using a Leo®1430VP SEM. Living mites were carefully mounted using a minute pin under a stereo microscope on aluminium stubs with double sided carbon tape to prevent them from moving. The specimens were observed without further manipulation under beam conditions of 7kV, ca. 1.5nA. Beam scanning was performed as quickly as possible to process the image before specimens started to shrink. The same specimen could not be scanned more than twice without shrinking. This resulted in usable whole mite images (figures 11 and 12). Images were captured digitally using SmartSEM software.

For conductive-coated specimens, living mites were stub-mounted and coated with a thin (~10nm thick) layer of gold, using an Edwards S150A Gold Sputter Coater. Gold-coated mites were processed using a Zeiss Merlin Field Emission SEM. The InLens detector was used for imaging, using beam conditions of 5kV accelerating voltage, a 250pA probe current with a working distance of approximately 4mm. This treatment was favoured for close observation of small characters, as it produced high quality images (figure 6a). However, it could not be used for whole mite images, as the mite tended to collapse under these harsh beam conditions.

Critical drying point treatment was used for some specimens. Living mites were placed into FAA solution (2 parts 37% formaldehyde: 10 parts 95% ethanol: 1 part glacial acetic acid: 7 parts deionized water) for 2 days. These samples were then dehydrated in a five-step ethanol series of 35%, 50%, 75%, 95% and 100%, for 15 minutes at each step. Dehydrated samples were critical point dried using a Quorum E3000 Series drier, then mounted on stubs and sputter coated with gold as described previously. Images of these individuals were captured using a Leo®1430VP Scanning electron microscope, with beam conditions 20kV and approximately 1.5nA, with a spot size of 300nm. The method resulted in usable whole mite images (figure 2). However, lots of mites were required for this process due to losing the specimens during the transfer in the dilution series.

Taxonomic key

A taxonomic key to all Eriophyoidea species known from *Vaccinium* spp. world-wide was compiled. This key is host-specific (as opposed to country- or region-specific), as eriophyoids are mostly host-specific. The key was adapted from the *Revised keys to the World Genera of Eriophyoidea (Acari: Prostigmata)* by Amrine et al. (2003), *The World Crop Pests* by Lindquist and Amrine (2003) and original species descriptions.

RESULTS

Supplementary description

Superfamily: ERIOPHYOIDEA Nalepa, 1898

Family: Eriophyidae Nalepa, 1898

Subfamily: Eriophyinae Nalepa, 1898

Tribe: Aceriini Amrine and Stasny, 1994

Eriophyes vaccinii Keifer, 1939

Aceria vaccinii (Keifer, 1939)

Acalitus vaccinii (Keifer, 1965)

FEMALE (n = 12), (figures 2 – 10), **Idiosoma**–Whitish, wormlike body 210 including pedipalp, 192 excluding gnathosoma, 56 wide (at the level of c2 setae). **Gnathosoma**–20, directed forward and slightly downward, basal part covered by small, pointed frontal lobe, chelicerae 19, palp coxal seta ep 4, apico-ventral setae v 2, palp genual setae d absent.

Prodorsal shield (figure 3)–Prodorsal shield oval, 25.5 long, 45 wide; frontal lobe small, thin, triangular, anteriorly pointed or slightly rounded. Prodorsal shield with pair of usually obscure admedian lines on posterior ¼ of shield between scapular setae, more or less curving outwards from rear, then curving inwards, few granules on the outer side of scapular tubercles, with eye-like structures on their outer side partly margined with single rounded (figure 4), shallow ridge, band of granules on outer margins of shield and on epicoxal area (sensu Chetverikov and Craemer 2015). Scapular setae sc 22.4, 23 apart, projecting posteriad.

Legs (figure 5)–Legs with all usual segments. *Leg I*: 20, trochanter 4.5, femur 5, basiventral femoral seta bv absent, genu 3, antaxial genual seta I'' 19; tibia 4, paraxial tibial seta I' absent; tarsus 5, paraxial unguinal tarsal seta u' 2, paraxial fastigial tarsal seta ft' 6, antaxial fastigial tarsal seta ft'' 15. Tarsal solenidion ω 6, slightly curved, sometimes straight and slightly knobbed, tarsal empodium em 4.6, simple, symmetrical, 6-rayed (figure 6). *Leg II*: 19, trochanter 3.6, femur 4.6, basiventral femoral seta bv 5, genu 3, antaxial genual seta I'' 18; tibia 3.5, tarsus

4.5, paraxial unguinal tarsal seta *u'* 3.5, paraxial fastigial tarsal seta *ft'* 7, antaxial fastigial tarsal seta *ft''* 16. Tarsal solenidion ω 7, slightly curved, sometimes straight, and slightly knobbed. Empodium *em* 8.4, simple, symmetrical, 6-rayed.

Coxisternal area (figure 7)–Suboral plate rounded, with few granules and three slight longitudinal elevations medially (only visible with SEM). Coxisternal plates I and II ornamented with rounded to elongated granules, granules arranged in single row, and parallel to and close to margin between coxisternal plates and leg trochanters. Anterolateral setae on coxisternal plate I *1b* 6, 10 apart, proximal setae on coxisternal plate I *1a* 19, 13 apart, proximal setae on coxisternal plate II *2a* 28.5, 24 apart. Inverted Y-shaped prosternal apodeme. 2 complete and 2 incomplete microtuberculate annuli between external genitalia and coxae.

External genitalia (figure 7)–Genital coverflap 12, 19 wide, with 8-12 longitudinal ridges, usually in two uneven transverse ranks, some ridges are longer stretching over both ranks. Moderate distance behind coxae, not appressed to coxae. Pregenital plate (sensu Flechtmann et al. 2015) present, with elongated tubercles in about four transverse rows arranged in more or less two transverse areas with the basal two rows slightly rounded. Proximal setae of coxisternal plate III *3a* 26, 19 apart.

Opisthosoma (figures 8 & 9)–Opisthosoma dorsally arched with 76 dorsal and 62 ventral microtuberculate annuli (from first annulus posterior to coxae II). Dorsally and ventrally with round to oval microtubercles, ventrally gradually elongated towards the rear, dorsally becoming more elongated and vague (probably subsurface) towards the rear until spiny microtubercles protruding from the posterior annulus margins of the telosoma. Opisthosomal seta *c2* 27 on ventral annulus 11, 50 apart; opisthosomal seta *d* 39.5 on ventral annulus 22, 39 apart; opisthosomal seta *e* 39.5 on ventral annulus 37, 26 apart; opisthosomal seta *f* 13, on annulus 6 from the rear, 16 apart, fine at apex. Opisthosomal setae *h1*, minute, less than 0.5. Opisthosomal setae *h2* 53, finely tapered.

Internal genitalia–(figure 10).

Deutogynes–deutogynes were not observed during this study.

MALE (n = 2), (figure 11) morphology similar to female except for genitalia. **Idiosoma**–Whitish, wormlike body 182 including pedipalp, 164 excluding gnathosoma, 52 wide (at the level of *c2* setae). **Gnathosoma**–21, directed forward and slightly downward, basal part covered by a small pointed frontal lobe, chelicerae 18, pedipalpi coxal setae *ep* 5, apico-ventral setae *v* 2, pedipalp genual setae *d* absent.

Prodorsal shield–Prodorsal shield oval 23 long, 42 wide; frontal lobe small, thin, triangular, anteriorly pointed or slightly rounded. Ornamentation similar to female. Scapular setae *sc* 19, 23 apart, projecting posteriorly.

Legs–with all usual segments. Granules are arranged in a horizontal line at the base of the trochanter. *Leg I*: 17, trochanter 3, femur 4, basiventral femoral seta *bv* absent, genu 3, antaxial genual setae *l''* 15; tibia 3, paraxial tibial setae *l'* absent; tarsus 4.6, paraxial unguinal tarsal seta

u' 2, paraxial fastigial tarsal setae ft' 7.6, antaxial fastigial tarsal setae ft'' 13. Tarsal solenidion ω 6, slightly curved, sometimes straight, and slightly knobbed, tarsal empodium em 5, simple, symmetrical, 6-rayed. *Leg II*: 18, trochanter 3, femur 5, basiventral femoral seta bv 3.5; genu 3, antaxial genual setae l'' broken could not be measured; tibia 2.6, paraxial tibial setae l' absent; tarsus 4, paraxial unguinal tarsal seta u' 2, paraxial fastigial tarsal setae ft' 3, antaxial, fastigial tarsal setae ft'' 16. Tarsal solenidion ω 7.6, slightly curved, sometimes straight, and slightly knobbed. Empodium em 4, simple, symmetrical, 6-rayed.

Coxisternal area—Suboral plate rounded, with few granules and three slight longitudinal elevations medially (only visible with SEM). Coxisternal plates I and II ornamented with rounded to elongated granules, granules arranged in single row, and parallel to and close to margin between coxisternal plates and leg trochanters. Anterolateral setae on coxisternal plate I $1b$ 4, 7 apart, proximal setae on coxisternal plate I $1a$ 20, 11 apart, proximal setae on coxisternal plate II $2a$ 25, 20 apart. Inverted Y-shaped prosternal apodeme. 2 complete and 2 incomplete microtuberculate annuli between external genitalia and coxae.

Opisthosoma—Opisthosoma similar to female, dorsally arched with 63 dorsal and 59 ventral microtuberculate annuli (from first annulus posterior to coxae II). Dorsally and ventrally with round to oval microtubercles, ventrally, gradually elongated towards the rear, telosome dorsally with spiny microtubercles protruding from the posterior margin of the annuli. Opisthosomal setae $c2$ 23 on ventral annulus 9, 50 apart; opisthosomal setae d 23 on ventral annulus 20, 39 apart; opisthosomal setae e 23 on ventral annulus 29, 39 apart; opisthosomal setae f 13, on annulus 5 from the rear, 17 apart, fine at apex. Opisthosomal setae $h1$, minute, less than 0.5.

Opisthosomal setae $h2$ 40, relatively long and finely tapered.

External genitalia—Genitalia 12.5 long, 15 wide, moderate distance behind coxae, not appressed to coxae. Proximal setae on coxisternal plate III $3a$ 7 and 14 apart, with dense irregularly arranged granules posterior to $3a$.

NYMPH ($n = 2$), (figure 12) **Idiosoma**—chunky and shorter than adults, translucent to whitish, wormlike body, 156 long, 51 wide (at the level of $c2$ setae). Gnathosoma—20, directed forward and slightly downward, chelicerae 17, pedipalp coxal seta ep 2.5, apico-ventral setae v 1.8, pedipalp genual setae d absent.

Prodorsal shield—unlike adult female and male, granules are not visible, faint admedian lines.

Scapular setae sc 16, 22 apart, projecting posteriorly.

Legs— with all usual segments. *Leg I*: 14, trochanter 3, femur 4.4, basiventral femoral setae bv absent, genu 2, antaxial genual setae l' 13; tibia 2, paraxial tibial setae l' absent; tarsus 4.6, paraxial unguinal tarsal seta u' 1.4, paraxial fastigial tarsal setae ft' 3, antaxial fastigial tarsal setae ft'' 10. Tarsal solenidion ω 4, slightly curved, blunt to slightly knobbed. Empodium em 3.5 simple, 4-rayed. *Leg II*: 13, trochanter 2, femur 3.5, basiventral femoral bv setae 2, genu 2, antaxial genual setae l'' 14; tibia 2, paraxial tibial setae l' absent; tarsus 3.5, paraxial unguinal tarsal seta u' 1.6, paraxial fastigial tarsal setae ft' 4.5, antaxial fastigial tarsal setae ft'' 13. Tarsal solenidion 7, slightly curved, blunt to slightly knobbed. Empodium em 4 simple, 4-rayed.

Coxisternal area—suboral plate rounded, sometimes with faint curved lines, fewer granules than female adult. Prosternal apodeme not visible. Coxisternal plates I and II ornamented with very few granules. Anterolateral setae on coxisternal plate I *1b* 3, 7 apart, proximal setae on coxisternal plate I *1a* 11, 10 apart, proximal setae on coxisternal plate II *2a* 21, 19 apart.

Opisthosoma—Opisthosoma with 51 dorsally arched and 45 ventral semiannuli. Ventrally, few, scattered oval to round microtubercles arranged medially in a band about the width of the distance between setae *3a*, and approximately 10 micrometers on the inside of setae *d*, up to a short distance posterior to *d*. Dorsally, oval to round microtubercles spreading over a wider area compared to the ventral side, present medially in a band about the width of the distance between setae *sc* arranged in an hourglass shape. Opisthosomal setae *c2* 14.5, 43 apart on annulus 11, opisthosomal setae *d* 28, 32 apart on annulus 16; opisthosomal setae *e* 25, 19 apart on annulus 25; opisthosomal setae *f* 9, 15 apart on annulus 41, or on annulus 5 from the rear. Seta *h1* minute, seta *h2* 40.

Genitalia—External genitalia absent. Proximal setae of coxisternal plate III *3a* 4, 9 apart.

LARVA (n= 2), (figure 13), **Idiosoma**—translucent, wormlike body 105 (including pedipalp), 53 wide. **Gnathosoma**—14.5, slightly bent. Chelicerae 14, pedipalp coxal setae *ep* 3, apico-ventral setae *v*, not visible for measurements. Pedipalp genual setae *d* absent.

Prodorsal shield—Prodorsal shield, smooth, 20 long, 35 wide, admedian lines and granules not visible. Scapular setae *sc* 9, 20 apart, projecting posteriorly.

Legs—Legs with all usual segments. *Leg I*: 12, trochanter 3, femur 3, genu 3, antaxial genual setae *l''* 13; tibia 2, tibial setae *l'* absent; tarsus 3, paraxial unguinal setae *u'* 2, paraxial fastigial tarsal setae *ft'* 5, antaxial fastigial tarsal setae *f''* 10. Tarsal solenidion ω 4, slightly curved, blunt to slightly knobbed. Empodium *em* 3, simple, 3-rayed. *Leg II*: 11, trochanter 2, femur 3, basiventral femoral seta *bv* 3, genu 2, antaxial genual setae *l''* 14; tibia 1, tarsus 3, paraxial unguinal tarsal seta *u'* 1, paraxial fastigial tarsal setae *ft'* 4, antaxial fastigial tarsal setae *ft''* 9. Tarsal solenidion ω 5, slightly curved, blunt to slightly knobbed. Empodium *em* 3, simple, 3-rayed.

Coxisternal area— suboral plate rounded, sometimes with faint curved lines, fewer granules than female adult. prosternal apodeme not visible. Coxisternal plates I and II ornamented with very few granules. Anterolateral setae on coxisternal plate I *1b* 2, 7 apart, proximal setae on coxisternal plate I *1a* 4, 9 apart, proximal setae on coxisternal plate II *2a* 9, 17 apart.

Opisthosoma—Opisthosoma dorsally arched with 30 dorsal and 30 ventral annuli. Opisthosomal microtubercles were absent or present on the one or both the ventral and dorsal sides. In dorsal view, irregular shaped to pointed microtubercles scattered towards the rear end. In ventral view, few oval to rounded microtubercles between *3a* and *d* setae-area and no microtubercles present beyond setae *d*. On both sides, the microtubercles are along setae *3a* and on the dorsal rear end. Opisthosomal setae *c2* 60, 50 apart on annulus 3 or 4, opisthosomal setae *d* 6, 28 apart, on annulus 11; setae *e* 3 long, 19 apart on annulus 16; setae *f* 8 long, 16 apart on annulus 27, or annulus 4 from the rear. Setae *h2* 23 long, Setae *h1* minute.

External genitalia—Genital coverflap absent. Proximal setae of coxisternal plate III 3a 2, 6 apart.

Table 1: Measurements (in μm) and counts of morphological characteristics of *Acalitus vaccinii*.
Abbreviations: L = length, W = width, n = number

| | Female Mean \pm SD (n=12) | Female Min–Max | Male Min–Max (n=2) | Nymph Min–Max (n=2) | Larva Min–Max (n=2) |
|-----------------------------------|-----------------------------------|-------------------|--------------------------|---------------------------|--|
| BODY SIZE | | | | | |
| body L (pedipalpi included) | 210.4 \pm 36.0 | 138.5–261.2 | 172–191.1 | 142.6–169.7 | 111.8–128.3 |
| idiosomal L (gnathosoma excluded) | 192.3 \pm 34.0 | 118.3–233.1 | 151.8–176.1 | 126.4–150.1 | 96.8–113.5 |
| body W | 56.0 \pm 4.7 | 47.9–63 | 49.7–55.1 | 47.1–51.8 | 51.8–54.8 |
| GNATHOSOMA | | | | | |
| L | 20.4 \pm 1.7 | 16.9–23.1 | 20.7–21.0 | 17.6–22.9 | 14.4–14.6 |
| setae <i>ep</i> (basal setae) L | 4.3 \pm 0.7 | 3.5–5.8 | 4.3–5.1 | 1.6–3.4 | 3.0–3.0 not visible for measurement |
| setae <i>v</i> L | 1.7 \pm 0.1 | 1.5–1.8 | 1.7–2.7 | 1.4–2.1 | |
| chelicerae L | 18.9 \pm 4.4 | 12.3–24.69 | 17.1–19.1 | 17.0–17.3 | 10.5–13.3 |

Table 1 (cont.): Measurements and counts of morphological characteristics of *Acalitus vaccinii*.
Abbreviations: L = length, W = width, n = number

| | Female Mean \pm SD (n=12) | Female Min–Max | Male Min–Max (n=2) | Nymph Min–Max (n=2) | Larva Min–Max (n=2) |
|--|-----------------------------------|-------------------|--------------------------|---------------------------|---------------------------|
| PRODORSAL SHIELD | | | | | |
| L | 25.5 \pm 1.3 | 23.4–27.5 | 22.8–24.1 | 24.6–25.8 | 19.2–20.6 |
| W | 44.8 \pm 4.9 | 30.8–50.5 | 39.2–44.4 | 39.4–43.4 | 32.4–36.7 |
| setae sc L | 22.4 \pm 1.2 | 20.0–24.2 | 18.2–20.7 | 15.5–17.4 | 8.2–9.4 |
| OPISTHOSOMA | | | | | |
| annuli before c2 | 10.6 \pm 0.5 | 10.0–11.0 | 9.0–9.0 | 6.0–7.0 | 3.0–4.0 |
| L setae c2 | 26.6 \pm 4.2 | 19.0–33.0 | 21.7–23.3 | 13.8–15.3 | 6.3–6.3 |
| distance between setae c2 | 49.6 \pm 5.8 | 36.5–59.48 | 48.2–50.9 | 42.4–42.8 | 50.5–50.5 |
| n of annuli between c2 and d | 11.2 \pm 0.6 | 10.0–12.0 | 9.0–9.0 | 8.0–8.0 | 6.0–6.0 |
| position setae d | 22.0 \pm 0.9 | 21.0–24.0 | 18.0–20.0 | 16.0–16.0 | 1.0–10.1 |
| setae d L | 39.5 \pm 9.0 | 27.9–51.5 | 17.6–28.4 | 26.9–28.4 | 6.0–6.0 |
| distance between setae d | 39.2 \pm 5.5 | 28.38–48.67 | 37.1–40.2 | 31.5–31.8 | 28.2–28.2 |
| position setae e | 36.9 \pm 2.0 | 35.0–40.0 | 21.0–28.0 | 25.0–25.0 | 16.0–16.0 |
| n of annuli between d and e | 13.8 \pm 1.3 | 12.0–16.0 | 12.0–12.0 | 7.0–8.0 | 5.0–6.0 |
| setae e L | 39.5 \pm 7.9 | 24.2–50.3 | 31.2–34.2 | 24.6–25.8 | 3.1–3.1 |
| distance between setae e | 25.7 \pm 3.2 | 18.9–30.6 | 24.0–25.4 | 18.6–18.9 | 18.7–18.7 |
| position setae f | 57.3 \pm 5.9 | 51.0–65.0 | 39.0–53.0 | 41.0–41.0 | 26.0–27.0 |
| position setae f from rear | 6.0 \pm 0.0 | 6.0–6.0 | 4.0–5.0 | 4.0–5.0 | 4.0–4.0 |
| n of annuli between f & h | 6.5 \pm 0.5 | 6.0–7.0 | 7.0–8.0 | 4.0–4.0 | 3.0–3.0 |
| n of annuli between e and f | 20.1 \pm 3.0 | 11.3–23.0 | 17.0–19.0 | 15.0–15.0 | 8.0–10.0 |
| setae f L | 12.8 \pm 1.7 | 11.0–17.3 | 13.9–17.9 | 8.6–9.6 | 8.0–8.0 |
| distance between setae f | 16.3 \pm 1.1 | 15.0–18.6 | 16.7–17.0 | 15.3–15.5 | 15.0–15.5 |
| setae h2 (caudal setae) L | 53.1 \pm 7.8 | 35.0–59.0 | 38.0–41.2 | 37.2–43.0 | 23.1–23.1 |
| setae h1 (accessory setae) L | <0.05 \pm 0.0 | <0.05 \pm 0.0 | <0.05 \pm 0.0 | minute | minute |
| n of dorsal annuli lateral to shield | 3.0 \pm 1.0 | 2.0–5.0 | 2.0–2.0 | 3.0–3.0 | 0.0–1.0 |
| n of dorsal annuli(start beneath the shield) | 73.4 \pm 9.1 | 59.0–86.0 | 60.0–61.0 | 47.0–48.0 | 32.0–33.0 |
| total n of dorsal annuli | 76.5 \pm 8.4 | 65.0–88.0 | 62.0–63.0 | 50.0–52.0 | 29.0–31.0 |
| total n of ventral annuli | 64.8 \pm 4.8 | 57.0–72.0 | 50.0–54.0 | 44.0–45.0 | 30.0–30.0 |
| COXAL AREA | | | | | |
| setae 1a L | 19.4 \pm 4.1 | 11.9–24.9 | 19.1–21.8 | 10.7–12.2 | 3.8–4.4 |
| setae 1a distance apart | 12.6 \pm 2.7 | 4.6–15.8 | 10.6–11.1 | 10.0–10.3 | 7.9–9.5 |
| setae 1b (1st coxal setae) L | 5.8 \pm 1.0 | 4.5–7.9 | 4.0–4.6 | 2.9–3.1 | 1.7–1.9 |
| setae 1b distance apart | 9.8 \pm 4.7 | 7.3–24.5 | 7.3–7.4 | 6.9–8.1 | 6.4–7.3 |
| setae 2a (3rd coxal setae) L | 28.5 \pm 4.8 | 22.6–36.4 | 15.8–19.3 | 19.5–23.0 | 8.3–9.5 |
| coxal setae 2a distance apart | 22.3 \pm 4.7 | 8.2–27.1 | 19.9–21.5 | 18.7–18.8 | 15.8–18.7 |
| distance setae 1a to 2a | 7.14 \pm 0.6 | 6.4–8.2 | 7.4–7.6 | 5.7–6.4 | 4.5–5.3 |
| complete annuli in coxigenital region | 0.0 | 0.0 | 2.0–2.0 | 0.0 | 0.0 |
| half annuli in coxigenital region | 2.4 \pm 0.9 | 2.0–3.0 | 2.0–2.0 | 0.0 | 0.0 |
| total annuli in coxigenital region | 2.4 \pm 0.9 | 2.0–3.0 | 4.0–4.0 | 0.0 | 0.0 |

Table 1 (cont.): Measurements and counts of morphological characteristics of *Acalitus vaccinii*.
Abbreviations: L = length, W = width, n = number

| | Female Mean \pm SD (n=12) | Female Min–Max | Male Min–Max (n=2) | Nymph Min–Max (n=2) | Larva Min–Max (n=2) |
|---|-----------------------------------|-------------------|--------------------------|---------------------------|---------------------------|
| LEG I | | | | | |
| L (from base of trochanter) | 20.3 \pm 5.4 | 3.7–24.7 | 16.1–17.9 | 13.8–15.0 | 11.9–12.7 |
| trochanter L | 4.5 \pm 0.6 | 3.6–5.5 | 2.7–3.6 | 2.7–2.9 | 2.7–4.1 |
| femur L | 4.9 \pm 0.8 | 3.3–6.5 | 3.8–4.5 | 4.3–4.5 | 2.6–3.5 |
| genu L | 3.1 \pm 0.2 | 2.8–3.3 | 3.1–3.4 | 1.9–2.5 | 2.7–2.9 |
| tibia L | 3.8 \pm 0.6 | 3.1–5.4 | 2.6–3.2 | 2.1–2.5 | 2.1–2.7 |
| tarsus L (excluding extremities) | 5.1 \pm 0.6 | 4.4–6.3 | 4.1–5.1 | 3.7–3.8 | 2.9–3.2 |
| em(tarsal empodium) L | 4.7 \pm 0.4 | 4.3–5.9 | 4.3–4.8 | 3.4–3.6 | 3.0–3.5 |
| ω (tarsal solenidion) L | 5.8 \pm 0.5 | 5.2–6.8 | 5.9–6.0 | 3.8–4.8 | 3.7–3.8 |
| setae <i>u'</i> L | 1.9 \pm 0.4 | 1.4–2.9 | 2.4–2.4 | 1.4–1.5 | 1.5–1.9 |
| setae <i>ft'</i> L | 15.1 \pm 1.6 | 12.4–17.8 | 12.5–13.6 | 9.9–10.1 | 9.0–10.2 |
| setae <i>ft''</i> L | 6.4 \pm 1.6 | 2.9–9.3 | 7.1–8.09 | 2.9–3.1 | 4.7–5.9 |
| setae <i>l''</i> L | 19.2 \pm 2.1 | 16.7–23.5 | 14.7–15.3 | 9.5–16.3 | 6.6–10.4 |
| LEG II | | | | | |
| L (from base of trochanter) | 18.9 \pm 4.8 | 3.8–21.4 | 17.4–18.3 | 12.9–13.7 | 3.1–11.3 |
| trochanter L (dorsal) | 3.6 \pm 0.5 | 3.0–4.8 | 2.9–3.2 | 1.8–2.2 | 1.9–11.2 |
| femur L (ventral) | 4.6 \pm 0.9 | 3.0–6.2 | 4.3–5.4 | 3.4–3.6 | 2.9–2.9 |
| genu L (dorsal/ventral) | 3.2 \pm 0.8 | 2.4–5.6 | 2.7–2.9 | 2.1–2.5 | 1.6–2.3 |
| tibia L (dorsally) | 3.5 \pm 0.7 | 2.9–5.5 | 2.3–2.8 | 1.7–1.8 | 1.3–1.3 |
| tarsus L (excluding extremities) ventrally | 4.5 \pm 0.5 | 3.7–5.3 | 4.4–4.5 | 3.4–3.7 | 2.6–2.9 |
| em (tarsal empodium) L | 8.4 \pm 10.9 | 4.3–4.3 | 4.2–4.5 | 3.5–4.2 | 2.8–2.9 |
| solenidion ω (tarsal solenidion) L | 6.7 \pm 1.8 | 1.5–8.0 | 7.6–7.6 | 6.6–6.8 | 5.1–5.6 |
| setae <i>u'</i> (mesal setae) L | 3.5 \pm 4.2 | 1.9–17.0 | 1.8–2.1 | 1.5–1.8 | 1.2–1.4 |
| setae <i>ft'</i> (dorsal tarsal setae) L | 16.2 \pm 4.2 | 4.0–19.9 | 14.7–16.8 | 11.6–14.4 | 9.1–10.7 |
| setae <i>ft''</i> (lateral tarsal setae) L | 6.7 \pm 4.4 | 3.9–20.0 | 3.1–3.7 | 3.3–5.6 | 3.9–4.4 |
| setae <i>l''</i> (genual setae) L | 18.5 \pm 4.5 | 4.5–21.0 | broken | 14.1–14.1 | broken |
| setae <i>bv</i> (femoral setae) L | 4.7 \pm 0.9 | 3.5–6.3 | 3.4–3.6 | 2.2–2.3 | 2.5–2.8 |
| EXTERNAL GENITALIA | | | | | |
| genital coverflap W | 19.4 \pm 1.9 | 13.8–20.8 | absent | absent | absent |
| genital coverflap L | 12.2 \pm 2.0 | 7.4–14.5 | absent | absent | absent |
| male genitalia W | absent | absent | 14.7–16.3 | absent | absent |
| male genital area L | absent | absent | 10.7–10.7 | absent | absent |
| setae <i>3a</i> (genital setae) L | 9.1 \pm 1.6 | 7.0–12.8 | 7.0–7.0 | 3.1–4.4 | 2.2–2.8 |
| distance between setae <i>3a</i> | 15.6 \pm 1.4 | 13.4–18.0 | 14.1–14.6 | 8.0–9.1 | 6.0–6.8 |
| annuli between <i>3a</i> and <i>c2</i> | 3.4 \pm 4.6 | 3.0–6.0 | 3.0–3.0 | 0.0–1.0 | 0.0–0.1 |

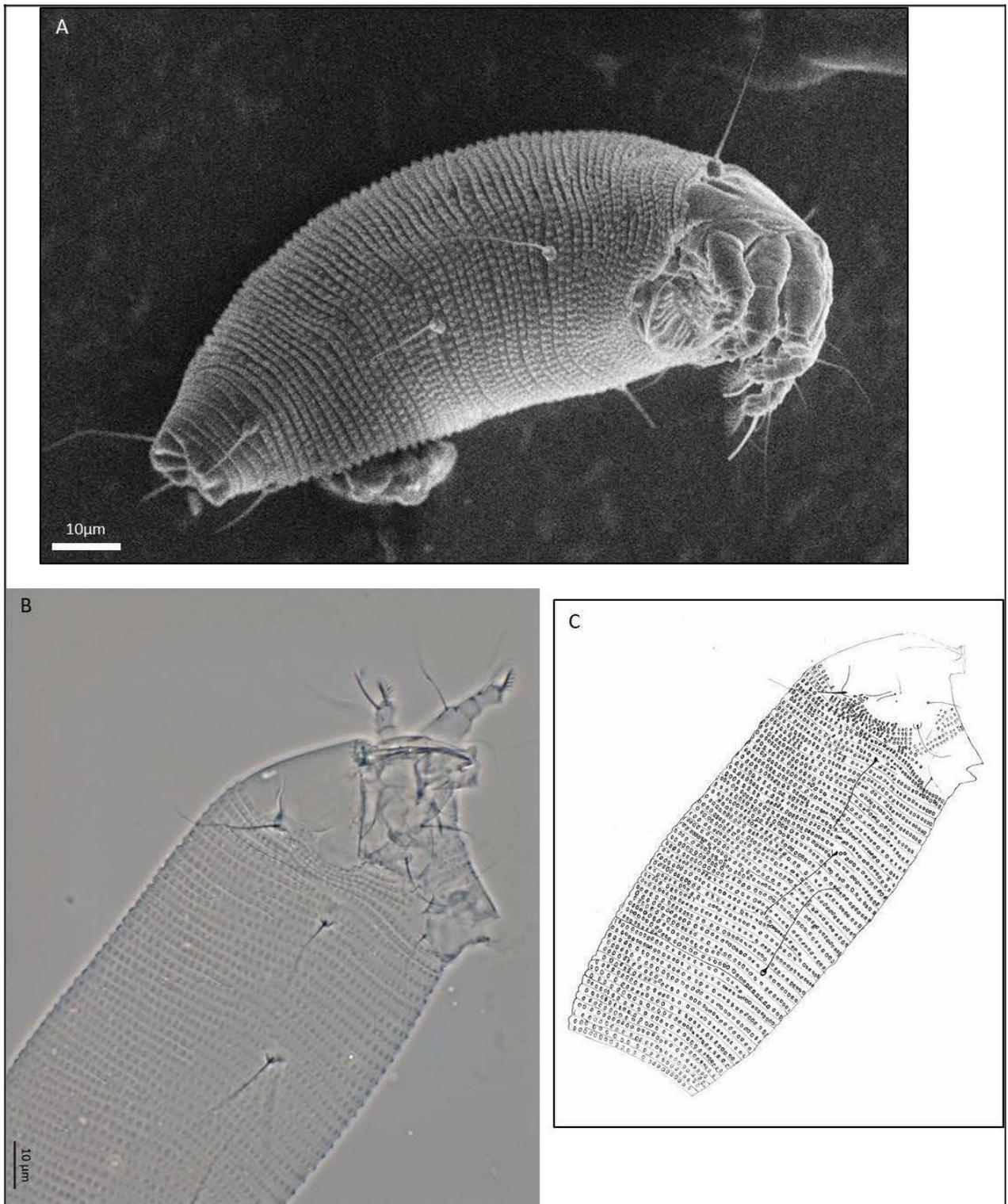


Figure 2: SEM (a), PCLM (b) and line drawing (c) of lateral view of the opisthosoma of *Acalitus vaccinii*.

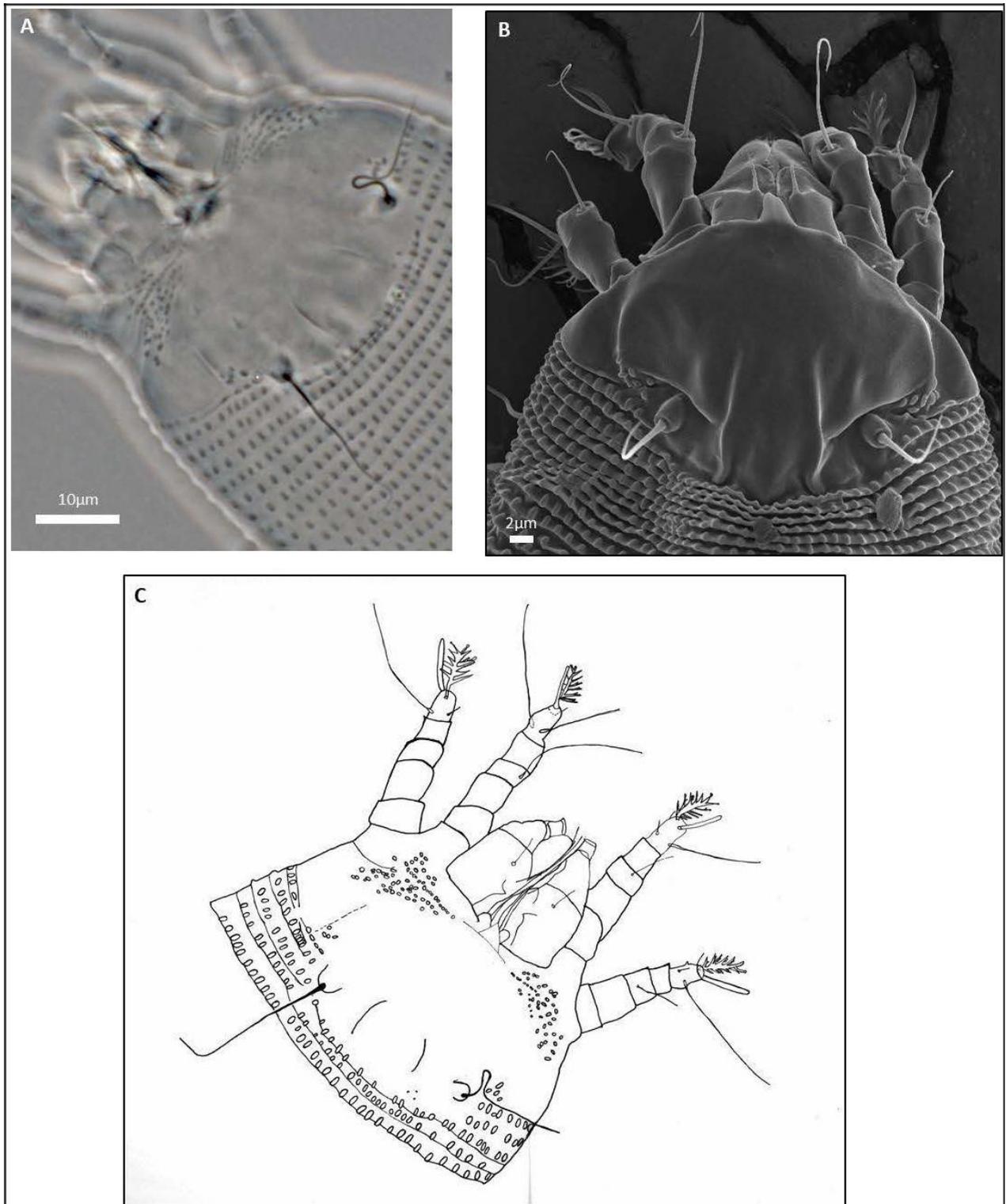
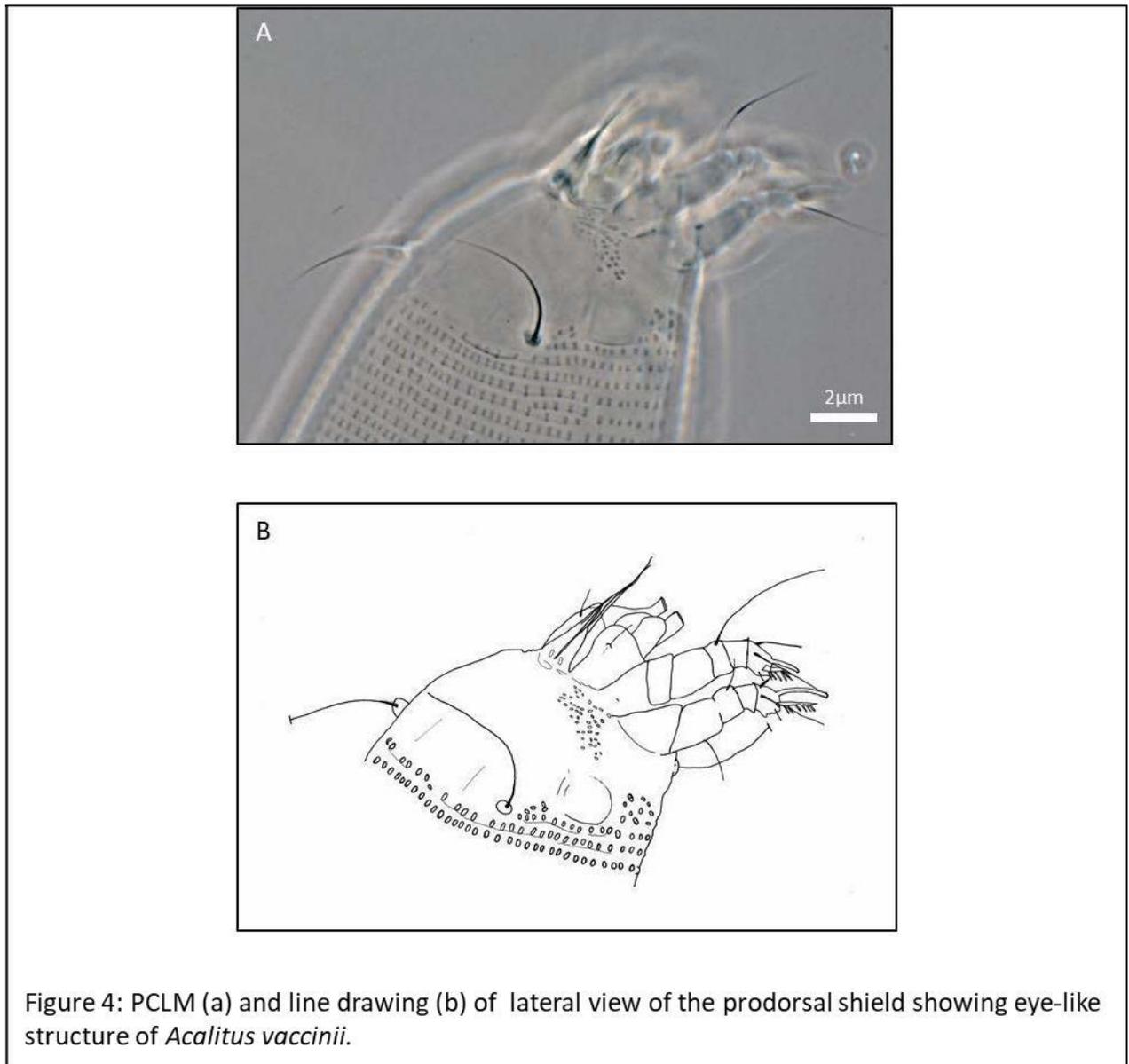


Figure 3: PCLM (a), SEM (b) and line drawing (c) of the dorsal view of the prodorsal shield of *Acalitus vaccinii*.



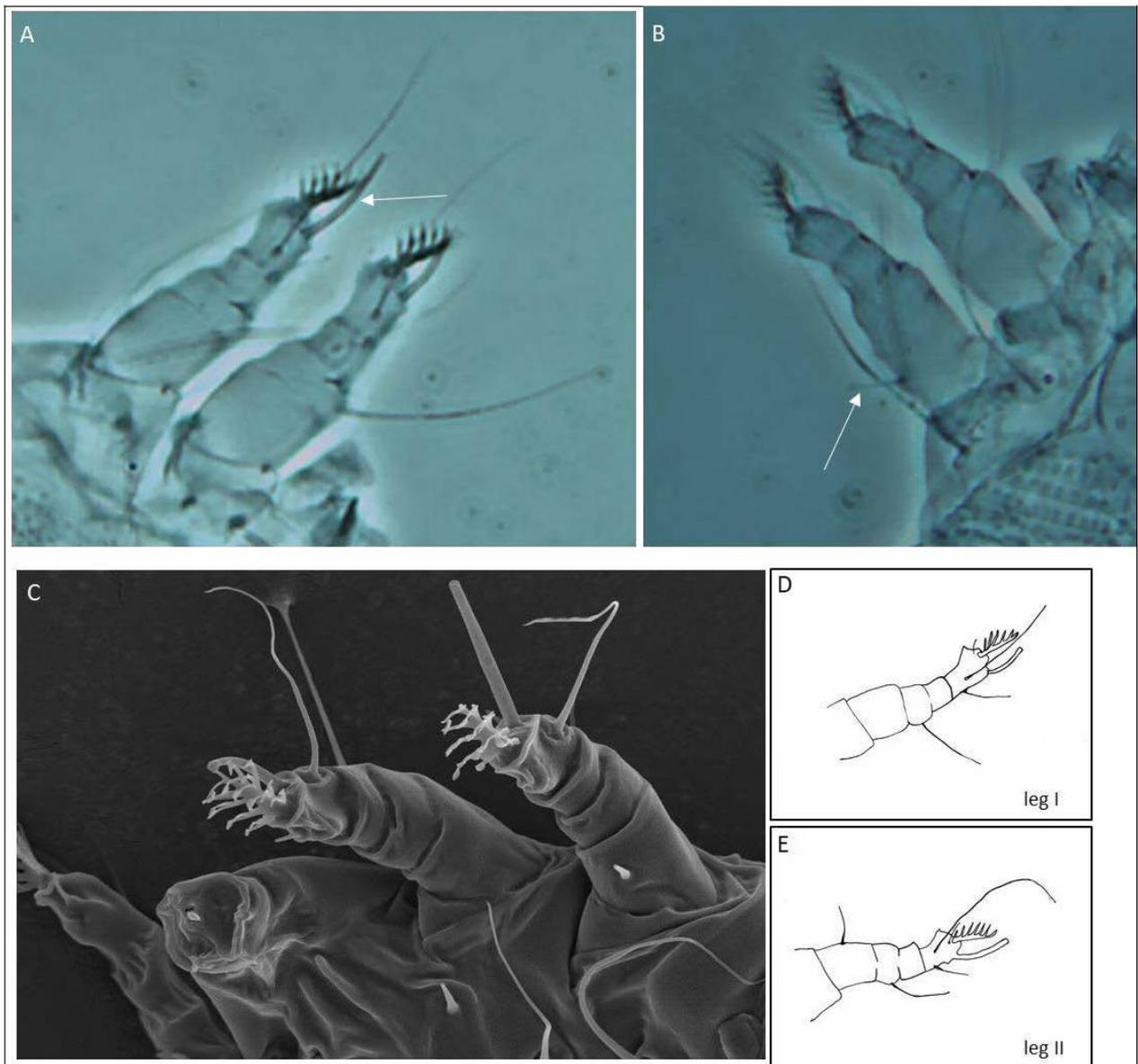


Figure 5: PCLM (a,b), SEM (c) and line drawing (d,e) of legs of *Acalitus vaccinii*. Arrows show the tarsus II solenidion (a) and femur II seta (b).

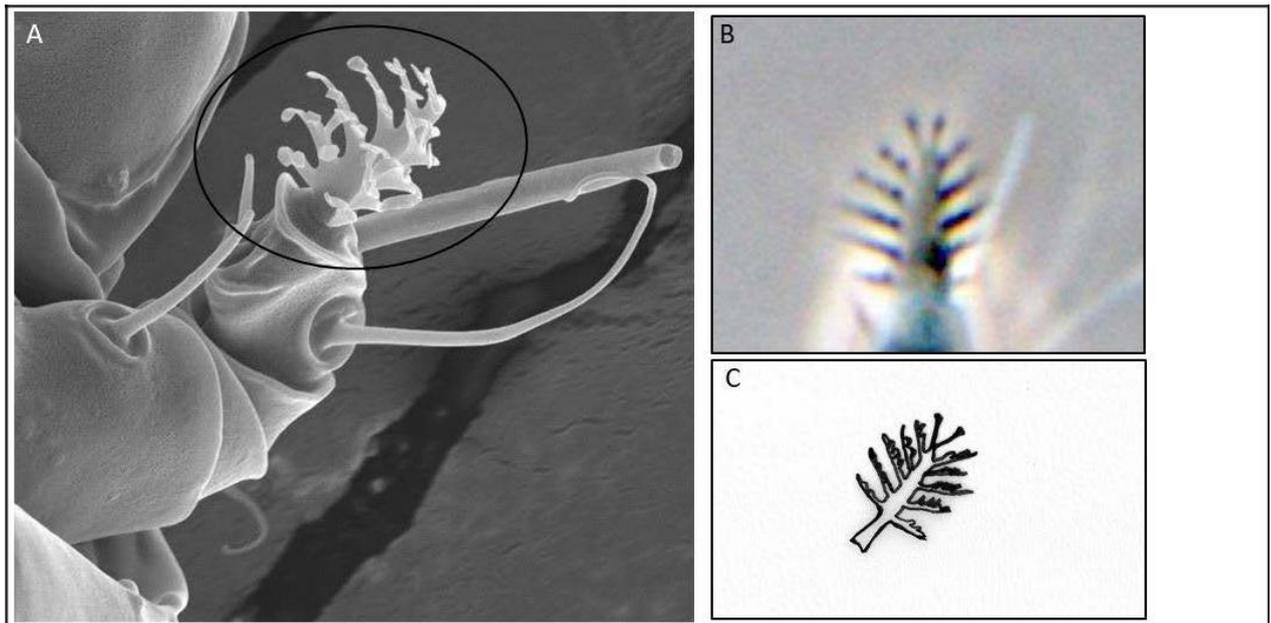


Figure 6: SEM (a), PCLM (b) and line drawing (c) of empodia of *Acalitus vaccinii*.

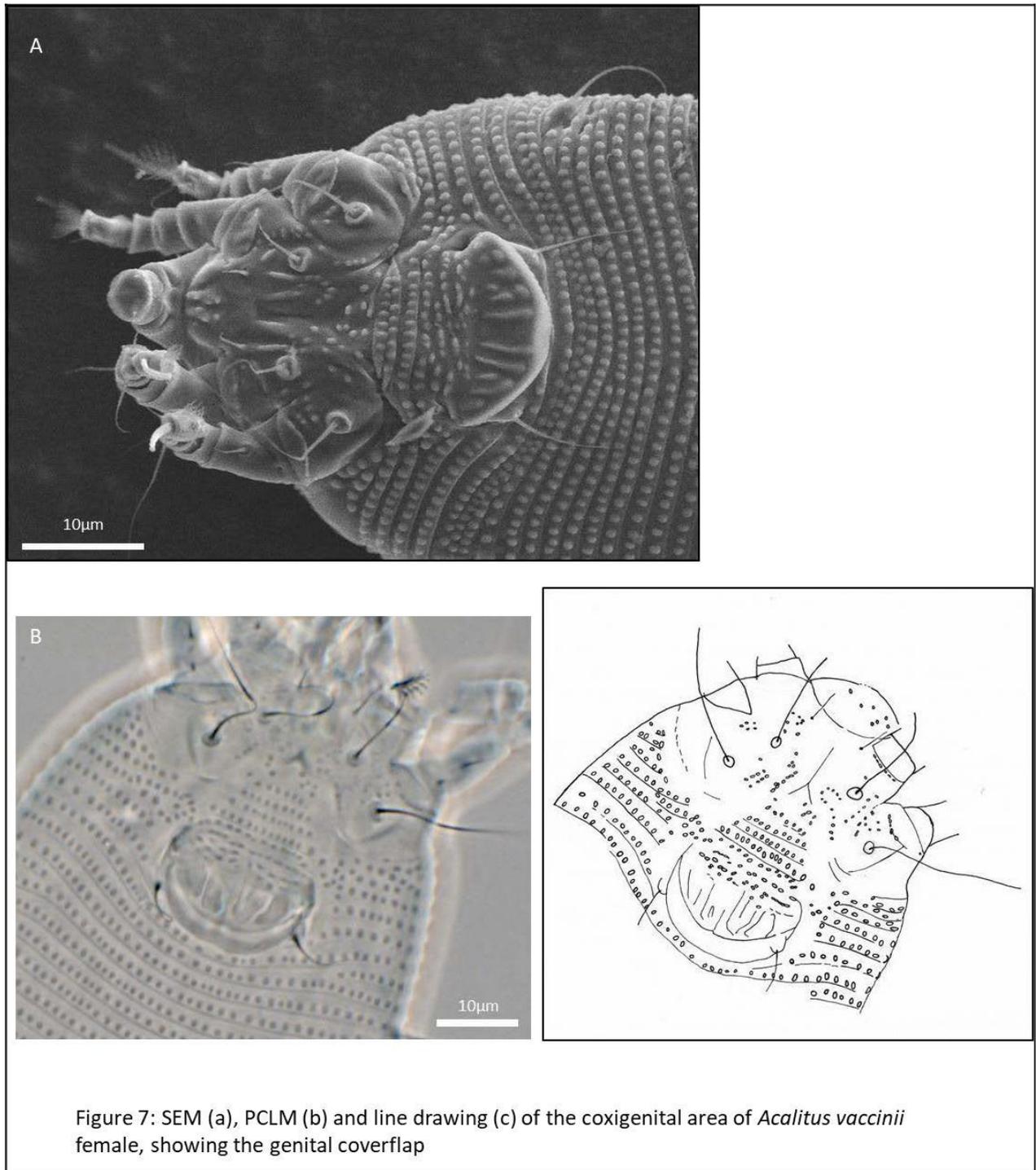
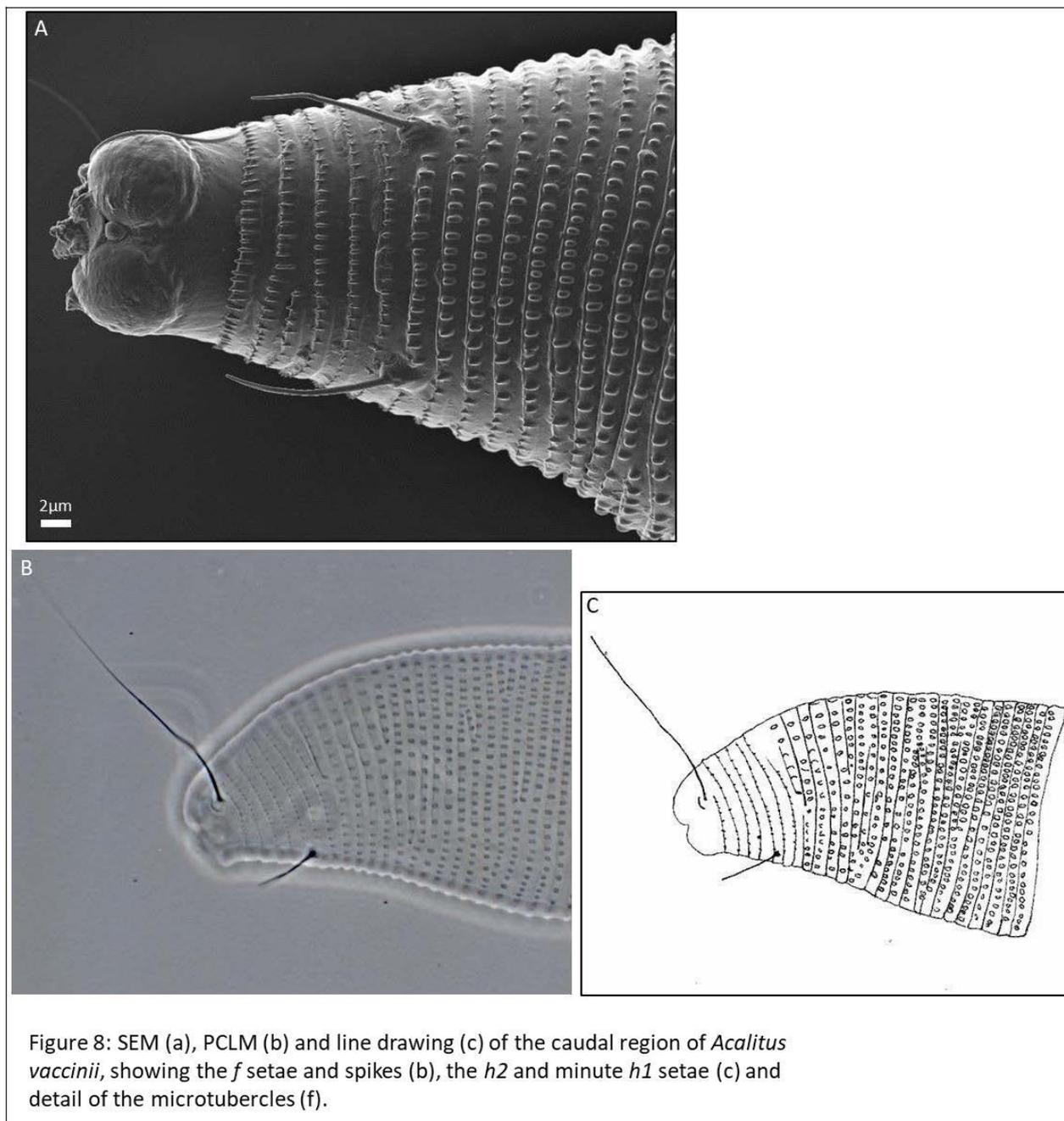


Figure 7: SEM (a), PCLM (b) and line drawing (c) of the coxigenital area of *Acalitus vaccinii* female, showing the genital coverflap



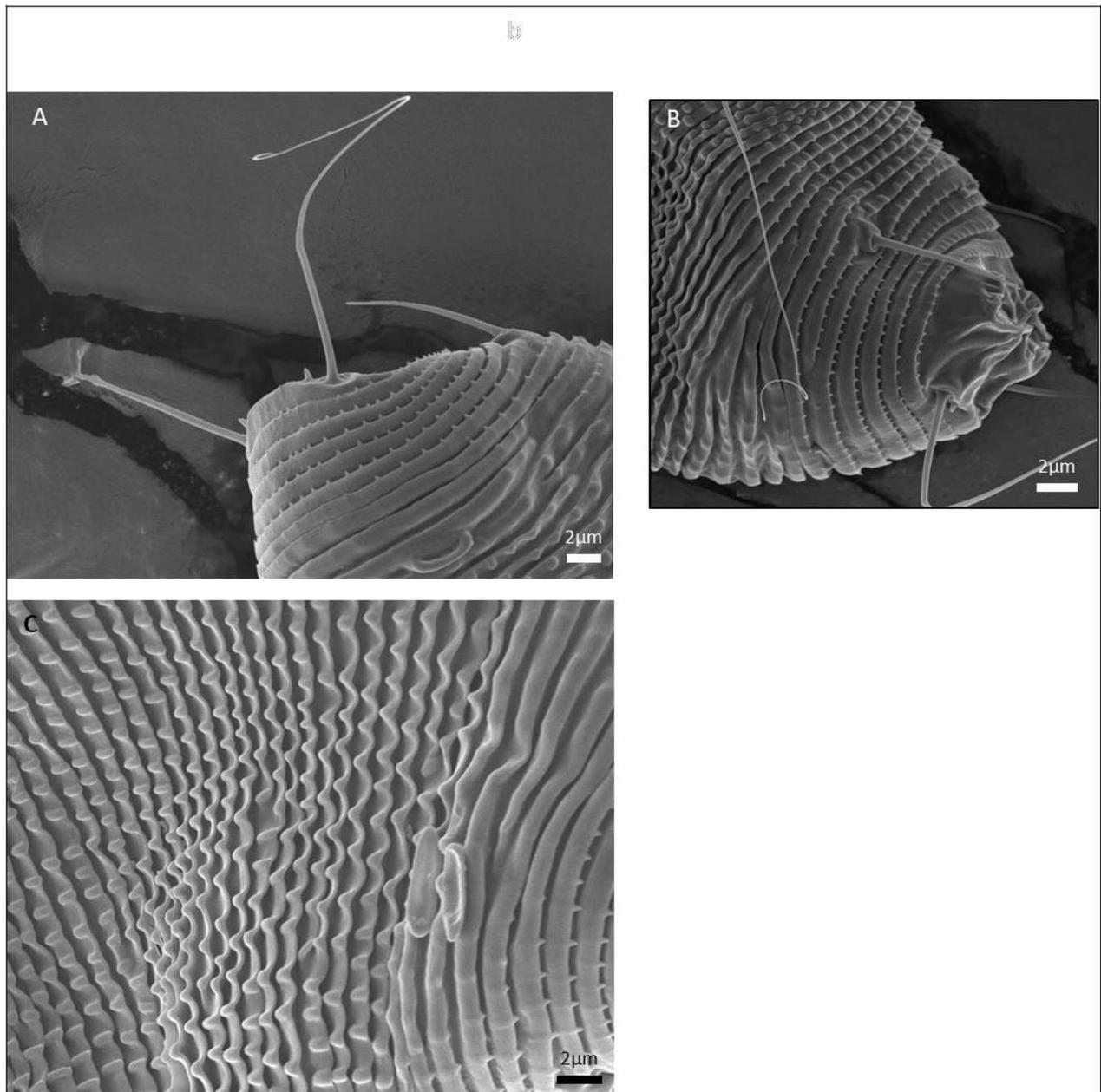


Figure 9: SEM showing the long *h2* and minute *h1* setae (a), *f* setae and spikes (b), and detail of the microtubercles (c).

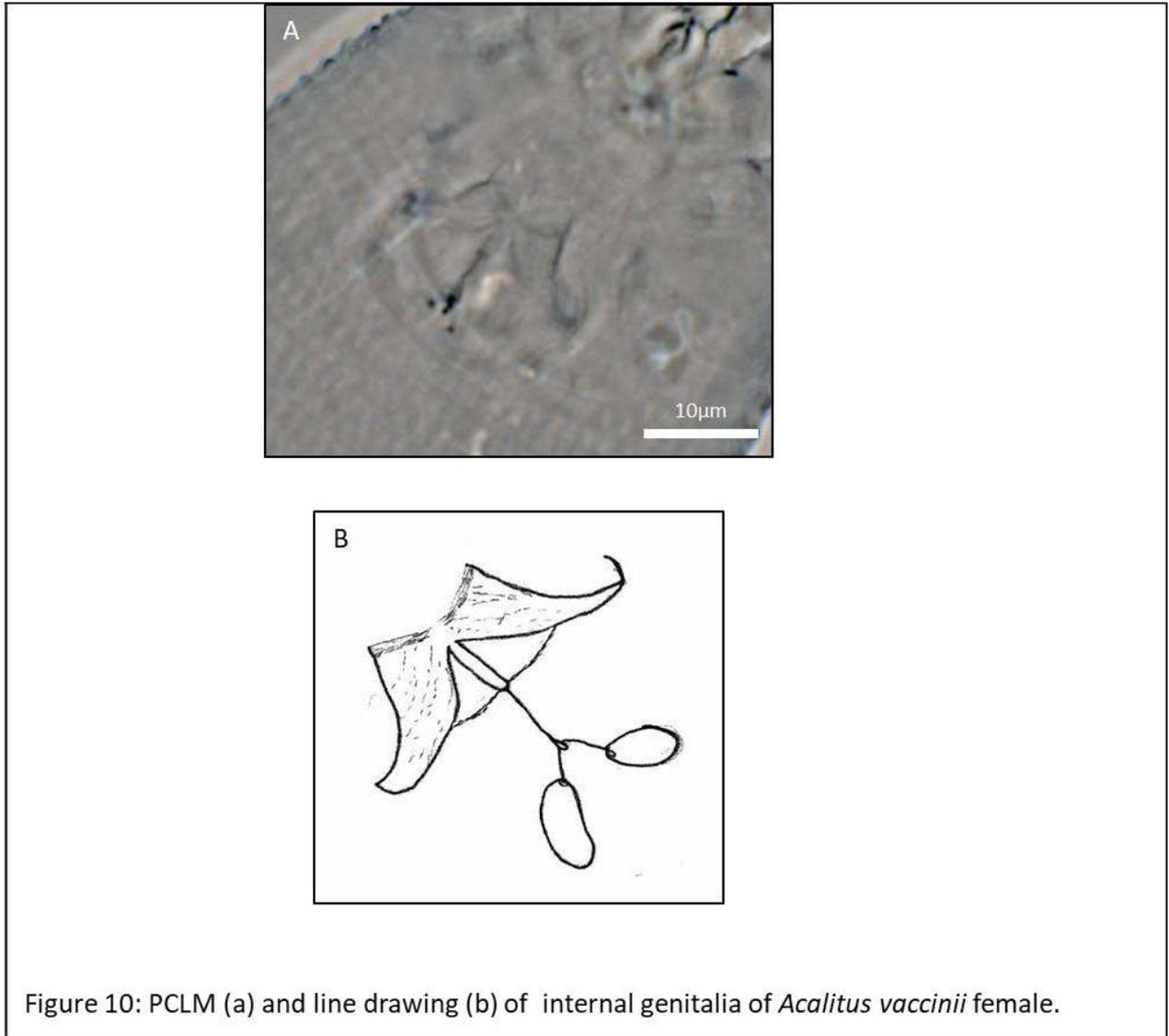


Figure 10: PCLM (a) and line drawing (b) of internal genitalia of *Acalitus vaccinii* female.

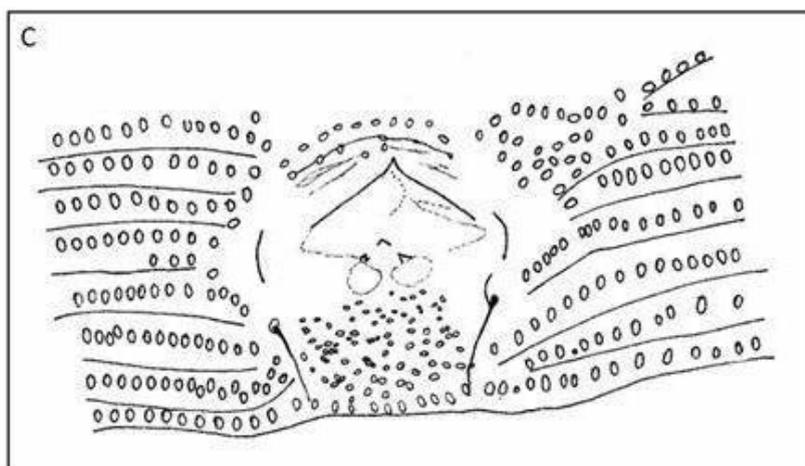
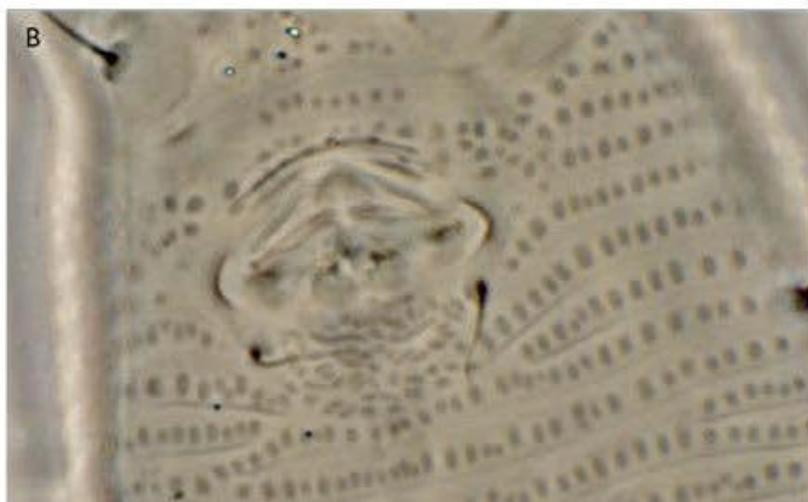
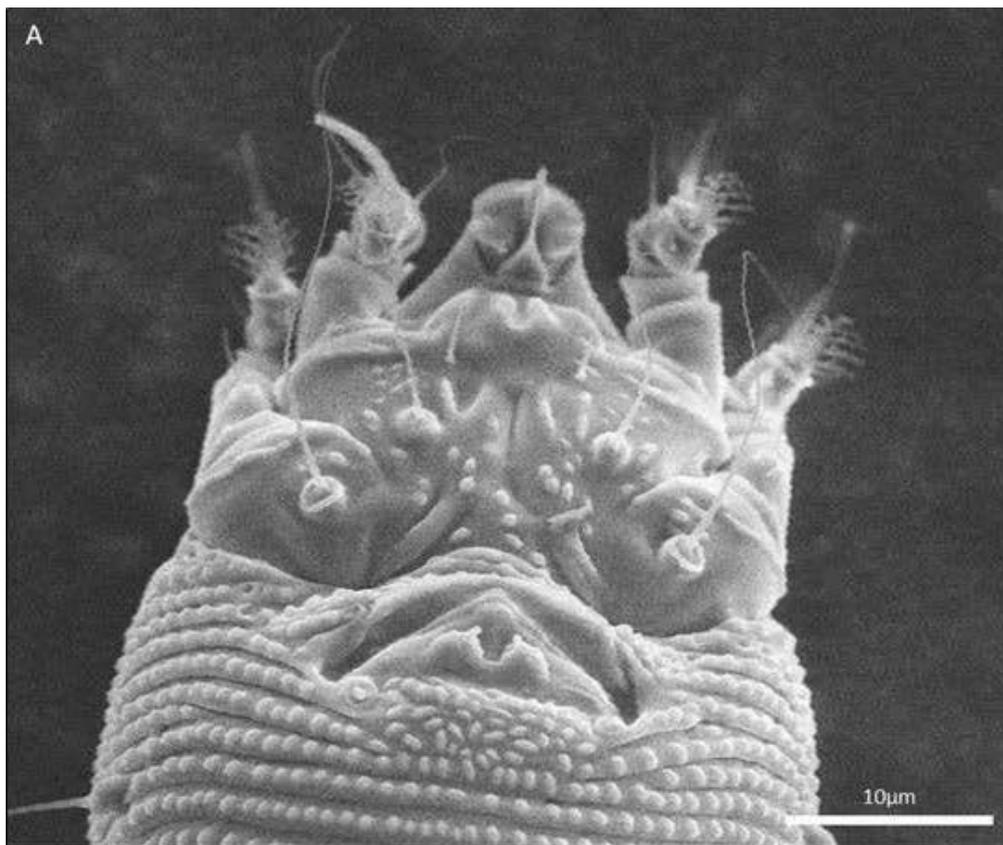
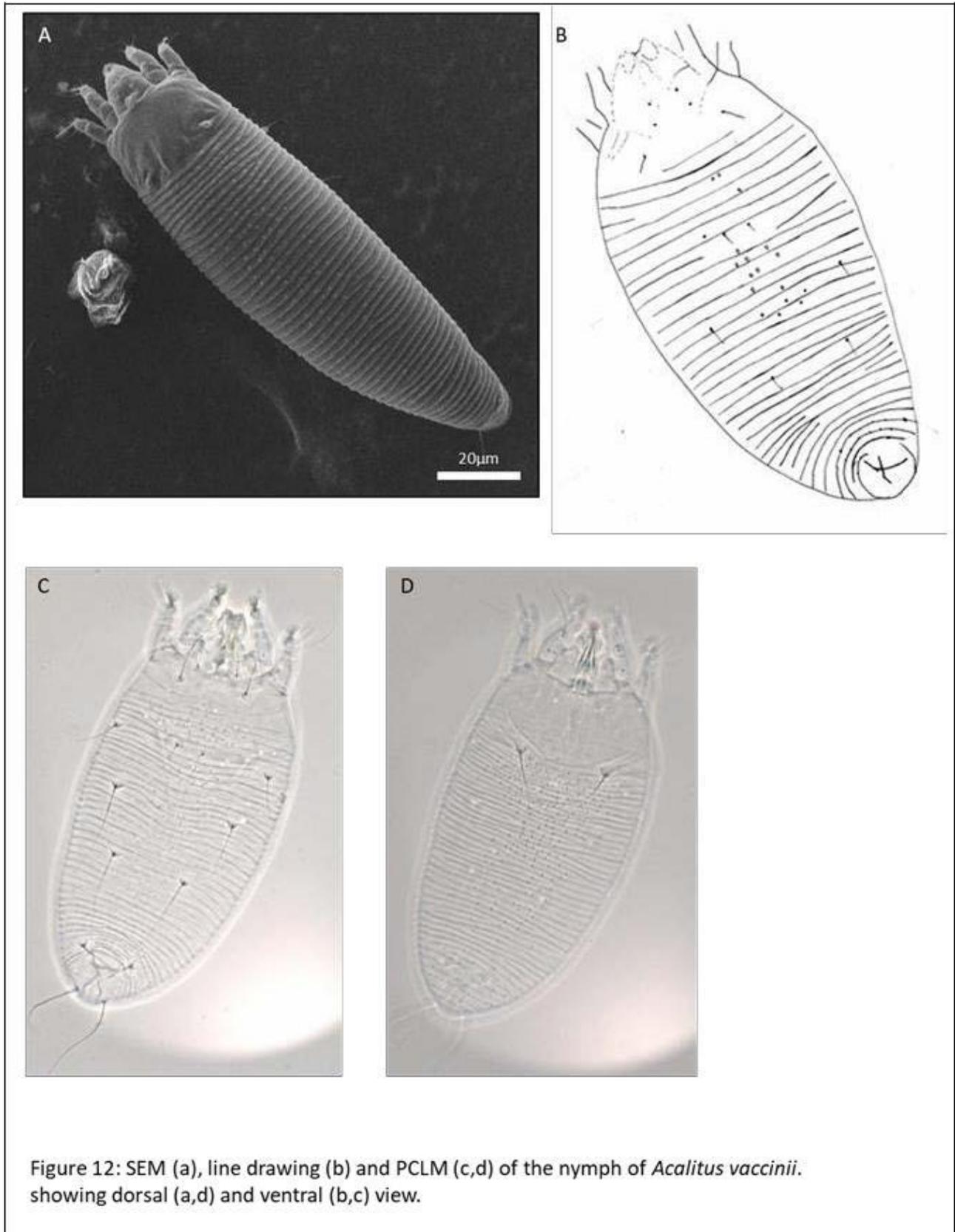
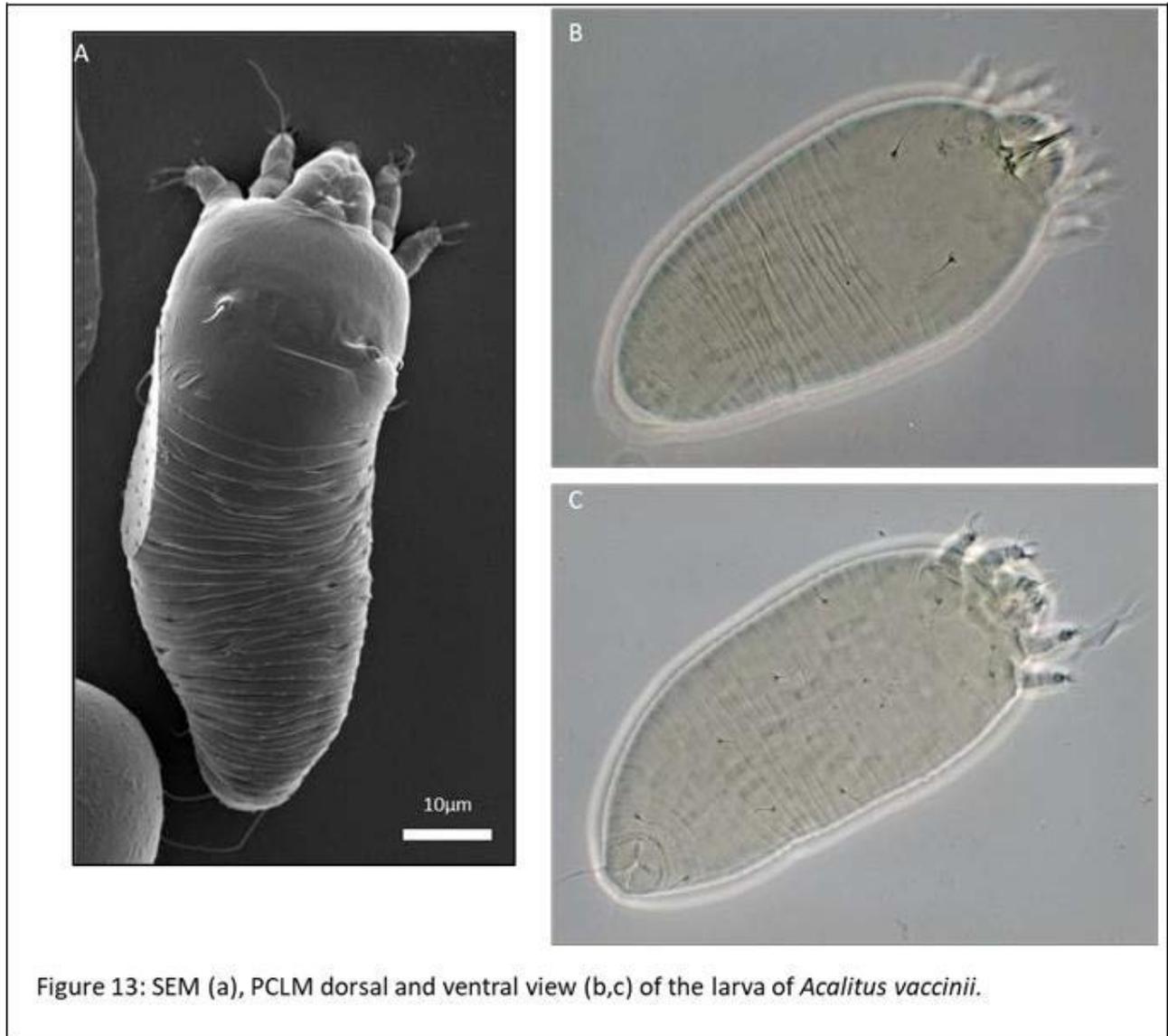


Figure 11: SEM (a), PCLM (b) and line drawing (c) of the coxigenital region of *Acalitus vaccinii* male.





Key to Eriophyoidea known on *Vaccinium* species

Adapted from Amrine *et al.* (2003), Lindquist and Amrine (1996) and individual species descriptions. It must be noted that the key presented here (as with all identification keys) is not conclusive and final. Only the currently known species on *Vaccinium* are included.

1 Gnathosoma large in comparison to body; cheliceral stylets relatively long, abruptly bent down near base, pedipalps attenuate, enclosing the long-form oral stylet, empodia often large, entire or divided, female coverflap usually smooth, female genital apodeme of moderate length, often narrowed anteriorly.....Diptilomipidae Keifer 1944 (one species known on *Vaccinium*)

..... Prodorsal shield wide with ridges, complete median and admedian lines, submedian lines incomplete, four cells on each side of anterior shield, empodium 5-rayed divided, coverflap with basal granules and 14 distal ridges, smooth dorsal annuli, ventral annuli with rounded microtubercles, coxal area sculpted with granules, prosternal apodeme present, occur as vagrants on the underside of the leaves of *Vaccinium bracteatum* ***Diptacus bracteatus***
Li, Wei and Qin 2009

1' Gnathosoma of various sizes, often large, with straight or slightly and evenly curved chelicerae. Pedipalps enclose a short-form oral stylet. Prodorsal shield always with anterior setae, legs often with solenidion ϕ on tibia of leg I, spermathecal tubes often long (3–5 times longer than that of Eriophyidae and Diptilomiodae) and extending diagonally forward then recurving caudadPhytoptidae Murray 1877..... (no species known on *Vaccinium*)

1'' Gnathosoma usually small in comparison to body, with short straight or slightly curved chelicerae, pedipalps with terminal segments short and truncate enclosing the short-form oral stylet. Empodia usually not divided and simple, female genital coverflap usually with ridges, genital apodeme usually of moderate anterior length....Eriophyidae Nalepa 1898 (8 species known on *Vaccinium*) 2

2 Tibiae absent or reduced, leg I paraxial tibial setae *l'* absent (no species known on *Vaccinium*)

2' Tibiae always of normal size and distinct from tarsi, leg I paraxial tibial setae *l'* usually (but not always) present 3

3 Prodorsal shield small and without scapular tubercles, *sc* very small on lateral margin and directed laterally; coxae widely separated anteriorly, opisthosoma lacking setae *d* and *e*; female genitalia located between coxae II; coverflap without ridges Ashieldophyinae Mohanasundaram 1984 (no species known on *Vaccinium*)

3' Prodorsal shield and opisthosomal projections not as above4

4 Female genital apodeme bent up and shortened, female genitalia appressed to coxae spreading coxae apart more than normal, and, in lateral view, usually noticeably projecting from venter; genital coverflap with ridges typically arranged in 2 uneven ranks Cecidophyinae Keifer 1966 (no species known on *Vaccinium*)

4' Female apodeme usually extending moderate distance forward. Female genitalia usually not appressed to coxae and not spreading them further apart than normal, and, in lateral view, lying more on level with venter. Genital coverflap variably ornamented, ridges typically occur in one (rarely 2) ranks 5

5 Vermiform shaped mites, annuli subequal dorsoventrally, frontal lobe typically absent, or with a light projection over gnathosoma base; if frontal lobe present, then it is narrow, basally flexible, and combine with narrow annuli Eriophyinae Nalepa 1898 (one species known on *Vaccinium*)

.....No opisthosomal ridges; leg I with both basiventral femoral seta and paraxial tibial setae absent; forecoxae often confluent, coxal setae *2a*, *1a* & *1b* present, coverflap with 8–10 ridges in a single row, sometimes in two uneven transverse ranks and ridges stretching over both ranks; 6-rayed empodium; tarsal solenidion slightly knobbed; prodorsal shield without strong central lines, inverted Y-shaped prosternal apodeme; rounded, granulate suboral plate; occurs in buds of *Gaylussacia baccata* and *Vaccinium* species ***Acalitus vaccinii*** (Keifer 1939)

5' More fusiform shaped mites, annuli typically dorsoventrally differentiated (broad dorsal subannuli and narrow ventral subannuli), frontal lobe usually broad-based and rigid Phyllocoptinae Nalepa 1892 6

6 Scapular setal tubercles usually set ahead of prodorsal shield rear margin, directing setae *sc* anteriorly, dorsally or convergently. Opisthosoma with a single middorsal ridge or with 3 or more longitudinal ridges with prominent middorsal ridge. Middorsal ridge ending in a broad furrow before termination of suboral ridges. Opisthosomal dorsum flattened in cross section. All leg and opisthosomal setae present. Prodorsal shield without projections *Calepitrimerus* Keifer, 1938 (3 species known on *Vaccinium*) 7

6' Scapular setal tubercles set ahead or near prodorsal shield rear margin, directing setae *sc* forward or dorsally, medially or convergently posteriad. Opisthosoma evenly arched, round in cross section, and less sharply tapered posteriorly. Opisthosomal shape variable: some species with broad dorsal semi-annuli and narrow ventral semi-annuli, while others with little dorsoventral differentiation. All leg and opisthosomal setae present. Prodorsal shield with frontal

- lobe *Phyllocoptes* Nalepa 1887 (4 species known on *Vaccinium*)
 8
- 7** Pinkish, wax stripes along the ridges. Prodorsal shield with a central ridge extending back and ending just beyond the dorsal tubercles setting. Broad and blunt frontal lobe, setae *sc* projecting up and ahead, setae *h1* absent, 3-rayed empodium, smooth annuli, genital coverflap with 8–9 ridges and weak horizontal markings at the top, occur around the lateral buds of fresh succulent twigs of *Vaccinium ovatum* ***Calepitrimerus gilsoni*** Keifer 1953
- 7'** Light amber yellow colour. Prodorsal shield pattern obscure, or virtually absent, frontal lobe with spines, *sc* setae projecting up and forward, prosternal apodeme moderately long, setae *h1* present, 6–rayed empodium, coverflap with 6–8 ridges, vagrants on both sides of the leaf of *Vaccinium atrococcum* ***Calepitrimerus darrowi*** Keifer, 1940
- 7''** Prodorsal shield with lateral lines and granules, median line absent, admedian lines curving back, submedian lines curving back from side of anterior shield lobe and joining with *sc* tubercles, annuli with fine and elongate microtubercles, weak middorsal opisthosomal ridge extends back to 25th–30th dorsal annuli, coxae ornamented with curved lines and granules, prosternal apodeme divided and short, coverflap with two ranks of faint parallel markings at the top and 8 weak longitudinal ridges at the bottom, vagrants on both sides of the leaves of *Vaccinium parvifolium*..... ***Calepitrimerus olympici*** Keifer, 1971
- 8** Amber, flattened wedge shaped body, 4-rayed empodium on leg I and 6-rayed empodium on leg II, genital coverflap with 6 ridges, sparse upper surface leaf vagrant of *Vaccinium amoenum*..... ***Phyllocoptes vandinei*** Keifer, 1939
- 8'** Same number of empodial rays on leg I and leg II, empodium 4 or 5-rayed, genital coverflap with 8–10 ridges 9
- 9** Empodium 5-rayed, genital coverflap with 8 ridges, *h1* setae 5µm. Light brown, with flattened body, unforked sternal line, legs with knobbed solenidion. Vagrant on the underside of *Vaccinium oxycocci* leaves..... ***Phyllocoptes oxycocci*** Roivainen, 1947
- 9'** Empodium with 4 rays, genital coverflap with 10 ridges, *h1* setae is tiny10
- 10** Female broader and shorter, 65-70 µm wide, 170-180 µm long. Yellowish, spindle form body shape, prosternal apodeme indistinctly forked, legs with curved and knobbed solenidion, genital coverflap with 10 ridges, vagrant on the underside of the leaves of *Vaccinium vitis idaea*
 ***Phyllocoptes vitisidaee*** Roivainen 1951
- 10'** Female is narrower and longer, 42-46 µm wide, 185-220 µm long. Empodium 4-rayed, genital coverflap with 10 longitudinal ridges, occur on *Myrtillus uliginosa* and *M. nigra* causing

dry and leathery leaves, also occurs on young leaves of *Vaccinium myrtillus* making them thin and withered ***Phyllocoptes vaccinii*** (Flögel and Goosmann, 1933)

DISCUSSION

When identifying the newly detected eriophyoid pest in South Africa, it became clear that the description of *Acalitus vaccinii* needed revision. Although identification of *A. vaccinii* was done without ambiguity when compared to other *Acalitus* species in Africa and known species on Ericaceae in North America, identification was a tedious process as the currently published descriptions are insufficient and no comprehensive key to species was available.

Morphology

The original description of *A. vaccinii* by Keifer (1939), although adequate for identification, no longer meets the current standards for a species description. Keifer included excellent line drawings, the style of which is still used as the standard in current guidelines (Amrine and Manson 1996; de Lillo 2010). Keifer's (1939) line drawings of *A. vaccinii* depict the main diagnostic characters, including the longitudinal ridges on the female genital coverflap, ornamentation of the prodorsal shield with two admedian lines between the scapular setae, and the aggregation of granules around a possible eye-like structure on the side of the shield. In the current study, these structures were clearly observed. However, Keifer's description is lacking in that he did not include measurements of all features, and there are some structures that he did not include in either the drawing or text description. This makes it difficult to use his description when comparing to unknown or closely related species.

Keifer's measurements of *A. vaccinii* are within the range of the minimum and maximum values found in this study and are therefore accurate. Keifer measured 33 female and 5 male characteristics, as compared to the 71 characters measured for females, 69 for males and 68 for immatures in this study. He did not measure the idiosoma, gnathosomal setae (*ep*, *d*, and *v*), distance between opisthosomal setae, leg segments or setae, leg II empodium, genital setae or distance between setae. The position of setae *f*, number of annuli between setae, number of dorsal and ventral annuli, and annuli between setae were not recorded. Additionally, Keifer's description did not describe male morphology and included very few male measurements. I suspect Keifer neglected to measure every character, considering them unnecessary for identification or redundant. He did not describe or measure males in detail, reasoning that: 1)

description is only based on the adult female; and 2) since eriophyoids are host specific, it will be unlikely to find males of one species together with females of another, concluding that identification of females will correspond to males found on the same host. Moreover, Keifer did not include some key features in his original description. Most notably, he did not record the presence of the *h1* (accessory) setae, whereas this study observed its presence in all life stages. He also did not include the leg *u'* (mesal) setae. It is highly possible that the observation of these setae in the current study is due to advancements in microscopy.

In addition to Keifer (1939), Baker and Neunzig (1970) described the males and immatures of *A. vaccinii*. Most features observed in the present study correspond to those previously observed, with the exception of differences in the presence and arrangement of the opisthosomal microtubercles in immatures. The original description by Baker and Neunzig (1970) presented the larva without microtubercles and the nymph with microtubercles covering the entire opisthosoma. In the present study, the irregular-shaped to pointed microtubercles on the gnathosoma of the larvae were either absent or present on just one or on both the ventral and dorsal sides of the mite. The nymph had microtubercles that were more widely spread on the dorsal side as compared to the ventral side and were arranged medially in a band about the width of the distance between setae *sc* arranged in an hourglass shape.

Many measurements that are standard for modern descriptions were not presented by Baker and Neunzig (1970) for the immature life stages. The few measurements presented cannot be used for comparison with current standards and procedures, as the way in which measurements were taken is not stipulated. For example, it is not clear if the idiosoma measurement included the gnathosoma; and the method for counting the annuli is not known. Measurements were also not taken for a number of setae. For example the gnathosomal setae (*ep*, *v*), opisthosomal setae (*e*, *f*), coxal setae (*1a*) and leg setae (*u'* and *l''*). The length of a number of characters (e.g. chelicerae, leg segments) and the distances and number of annuli between some setae were not presented.

Scanning Electron Microscopy (SEM)

To further enhance the quality of the description, Scanning Electron Microscopy (SEM) was used. This is an improvement on the traditional method of examining mite morphology using phase contrast microscopy only. It generates a highly magnified three-dimensional image showing the mite in its natural state. This allows one to study minute characteristics and complex shapes clearly and without distortion, unlike the uncertainty in observing slide mounted specimens with light microscopy. SEM images show increased detail compared to phase

contrast light microscopy (PCLM). Although it is an expensive and sometimes time-consuming technique, the additional information it contributes to a taxonomic study is invaluable.

In addition, Low Temperature SEM (LTSEM) has the ability to further preserve turgidity of structures such as leg walls and joints and it remains the best option for studying soft-bodied eriophyoid mites (Echlin 1970; 1978; Achor et al. 2001) However, the technique is expensive and very few LTSEM laboratories are available for use in South Africa.

In the absence of LTSEM, ambient temperature scanning electron microscopy (ATSEM) may be used. However, specimen preparation for ATSEM is time-consuming and allows room for errors, and results might have artefacts in them, especially shrinking. Generally, the use of SEM in routine/diagnostic analysis is limited by the cost of the equipment, the number of specimens available for processing, inability to recover the processed specimens, scarcity of trained personnel and the lack of microscopes to mobilize the use of low temperature technique (Fisher and Dowling 2010). I encourage more frequent use of SEM images in order to increase popularity and consequently its availability and use.

Biology

The presence of all life stages (females, males, immatures and eggs) of *A. vaccinii* on cultivated blueberries confirmed the crop as its host. Many eriophyoid species have two female forms: a summer form (protogyne) and a winter form (deutogyne) (Baker et al. 1996; Manson and Oldfield 1996). The protogyne is the active form, feeding and breeding, and is the form on which the taxonomic description is based. The deutogyne is produced during adverse conditions, usually during winter. It is inactive, and does not eat or breed. Protogynes and deutogynes are morphologically distinct, with the deutogyne usually showing reduced ornamentation and other features, but increased shielding (Baker et al. 1996; Manson and Oldfield 1996). It is important to establish whether both forms of a species occur in a particular area in order to avoid future misidentification of the deutogyne as a separate species (or even genus) because of morphological differences (Zhao 2000; Smith et al. 2010; Guo et al. 2015). In this study, specimens of *A. vaccinii* were collected throughout the year, but no deutogynes were observed. In North America, deutogynes were noted only in colder areas (Cromroy and Kuitert 2001). The absence of deutogynes in South Africa might be explained by the mild winter conditions of Mpumalanga (8–19°C) (South African Weather Services, 2018) in the past 12 years, in comparison to the mite's natural range (-1–7°C) (www.usclimatedata.com/climate/united-states/us). The moderate climate in South Africa may not induce a temperature-related change in morphology, and therefore deutogynes were not found here. All life stages remained viable and feeding in the winter season which may result in increased population size and more injury

due to feeding. Consequently the mite would become a more serious pest in warm regions than in colder regions.

CONCLUSION

With the lack of comprehensive key to species, continuous discovery of new species and advancement in eriophyoid studies, it is important to re-look at older species descriptions for standardisation. These old species descriptions can then be used efficiently for identification. It is also important to provide high quality and complete descriptions of economically important species to facilitate accurate identification. Standards set by Amrine and Manson (1996) and de Lillo et al. (2010) promote high quality species descriptions. Studying all life stages, including female, male and immatures, allows for additional characters that may not be visible on females. These characters might help to better understand intraspecific morphometric variability. The standard allows for additional information to be incorporated and correction of morphological observations which are not due to intra-specific variation but due to artefacts. This can be achieved through improved microscopy techniques such as Scanning Electron Microscopy. SEM is also important to improve morphological data which might provide more insight for further phylogenetic studies. It allows comparison of closely related specimens for phylogenetic studies and to study variations in species. SEM is not readily available worldwide, thus to facilitate taxonomic differentiation in other studies, line drawing from slide mounted specimens remains the core for species description.

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Chapter 4

General Conclusion

This study was aimed at understanding factors that may influence arthropod richness, abundance and communities focussing on detrimental and beneficial taxa associated with the blueberry production in South Africa. It also became clear that one of the limitations with a study like this is the identification of pest species, and mites in particular. I therefore also set out to improve the currently incomplete identification procedure of the blueberry bud mite.

In chapter 2, the diversity of arthropods associated with blueberry crops was assessed in the Mpumalanga and Western Cape Provinces. The effect of different production systems (open versus tunnel production; organic versus inorganic regimes) and the influence of season was also investigated. Arthropods were grouped according to general feeding guilds in order to understand multiple different effects of these variables on community assemblages (Alvarez et al. 2001). In Chapter 3, information on the ecology and taxonomy of the newly-introduced eriophyoid pest, the blueberry bud mite, *Acalitus vaccinii* Keifer was studied. This species was initially described before modern taxonomic procedures became the norm and I therefore provided additional morphological information for its re-description. Original counts and measurements of all of the important morphological characters of females, males and immatures originating from material in the USA, were compared to those of the South African specimens. A detailed up-to-date morphological description, including clear illustrations and micrographs of all important morphological characters is therefore presented. In addition, a key to Eriophyoidea species known on *Vaccinium* worldwide was developed.

Study outcomes

The main focus of this study was to assess arthropod numbers in blueberry fields in two provinces, the Mpumalanga Province, a minor production area, and in the Western Cape Province, the largest production area of blueberries in South Africa (Sikuka 2017). It represents the first comprehensive study on the arthropods associated with this crop in South Africa. Arthropods were collected using two different methods, each aimed at assessing the diversity of different sets (or size-classes) of arthropods. Vacuum sampling was used to collect larger

foliage-associated arthropods that closely connects with the blueberry crop (Dietrick et al. 1959). The clipping method focussed on the collection of smaller arthropods found in cryptic spaces on plant material such as mites, thrips and whiteflies. All arthropods in agricultural fields are ecologically important and may also be of economic importance as they contain beneficial and pest species (Alvarez et al. 2001; Olfert et al. 2002). To gain a more complete picture of arthropod communities, individuals collected using abovementioned methods were grouped into general feeding guilds (Letourneau and Goldstein 2001) as phytophages, predators, pollinators, parasitoids, detritivores and scavengers.

All variables assessed in this thesis had an influence on the numbers of arthropods in production areas of blueberries in South Africa. The two most prominent influencing variables were seasonal changes in numbers and communities, and the effect of location (Jeanneret et al. 2003). As expected, numbers of species and individuals of different guilds peaked at different times of the year, but generally high numbers of phytophagous species and predators were recorded during hottest months of the year. Farms in the Western Cape Province had different communities and species of arthropods than farms in Mpumalanga. However, large variations were also observed between farms in each production area. These results indicate that most arthropods found on the blueberry crops, including the pest taxa, likely invade this crop from the immediate surrounding areas (Berry 1996; Altieri 1999; Feber et al. 1997; 1998; Clark 1999). Location likely also influenced arthropod communities due to different agricultural practices on the different farms (Lee et al. 2001; Lewinsohn et al. 2005; Rocca and Greco 2011; Schellhorn et al. 2014). Each farm therefore has to build its own unique pest monitoring and control programme in order to best control the specific pests on that property, but also to promote the numbers of beneficial species. This is because my results also showed that farms with higher predator and parasitoid numbers also generally had reduced numbers of phytophagous species.

The effect of field type (open vs. closed) was assessed by comparison of arthropod communities collected from open fields to communities from blueberry plants grown in covered tunnels. The advantages of production in closed fields are profit driven both in terms of improved production and pesticide management. Exclusion of rain promotes crop quality and shelf life and reduces the chances of fungal disease attack (Wells 1998; Jiang et al. 2004; Demchak 2009; Lamont 2009). Protected cultivation allows for an extension of the production season, the season maybe extended into the rainy season or into spring depending on the geographical location (Jensen and Malter 1995; Leach and Isaacs 2018). It is believed that covered tunnels provide altered micro environments for arthropods and limited access to the crop (Wells 1998; Jiang et al. 2004; Demchak 2009; Lamont 2009). Based on the vacuum sampling method, open fields housed greater diversity and higher numbers of phytophagous arthropods confirming the efficacy of this method. However, this pattern differed when

assessing arthropod numbers using the clipping method. Using this method, plants under cover generally had higher numbers of phytophagous arthropods. Therefore, pest management programmes should also document the amount of crop destruction by specific taxa in order to best control their numbers when deciding on the use of open or closed production systems for this purpose.

The influence of pesticide usage on arthropod numbers was assessed in the Mpumalanga Province by comparisons between two organic farms (no pesticides) vs. a farm that used pesticides. Here, the farm that used pesticide generally had higher numbers of pest taxa and individuals than the two organic farms, indicating that organic production may be much more cost effective in this region than inorganic production. The influence of pesticide usage was also assessed in organic versus inorganic blocks on a single farm in the Western Cape Province. In this case, even blocks that were grown as organic may sometimes be treated with organic pesticides. Results indicated that organic production promoted higher diversity and numbers of beneficial taxa such as pollinators (Mordellidae) (Wilson et al. 1999). Interestingly, whether blueberries were produced organically or inorganically had little effect on the numbers of phytophagous species. It is therefore likely much more profitable to produce blueberries organically as this method would reduce costs related to pest control (pesticide application etc.) and market prices for organically grown produce is usually much higher than for those produced inorganically. Organic production practices would also promote a much healthier environment and increase the numbers of beneficial species in production areas.

In Chapter 3 I set out to augment the outdated morphological species description of the blueberry bud mite pest, *Acalitus vaccinii* using new microscopy techniques. I reviewed and upgraded the original morphological descriptions of *A. vaccinii* female and immature stages and described the male for the first time. All morphologically important characters were re-measured for all developmental stages including the male. Newly observed characters were also measured and are included in the emended description to further improve identification. Corrections in the presentation of various morphological characters were also made. The new description (re-description) therefore now conform to the current standards of presenting a species description as set out by Amrine and Manson (1996) and de Lillo (2010). All of the important morphological characters are therefore still presented as line drawings in the style of Keifer HH and form the core of the species description. However, as microscopy techniques have improved considerably in the recent past, good quality and clarity micrographs of morphological characters was also added. These were produced using phase contrast light microscopy (PCLM) and scanning electron microscopy (SEM) (Echlin 1970; 1978; Achor et al 2001). PCLM micrographs presented detailed views of morphological characters on slide mounted specimens, while SEM presented three dimensional images of mites. SEM therefore

provided clear details of complex and tiny morphological structures that other method could not produce. All images (line drawings, PCLM images, SEM images) are presented side by side to give a clear overview of the morphological characters under these different presentation methods (de Lillo et al. 2010). The first key to Eriophyoid species known from *Vaccinium* was also developed. This key will aid future species identification and comparisons of Eriophyoidea fauna found on blueberries across the globe.

Limitations and future studies

This study was limited by the availability of literature and expertise on the pests of arthropods on blueberries. This is a fairly new crop in South Africa and it is still accumulating pest species, however, apart from the blueberry bud mite, very few pests known from other parts of the world affect this crop locally. Numerous arthropods collected in this study are not well known on crops and their relationship with the crop could not be established. Identification of arthropods to species level was therefore not achievable in the confines of the current study which may present challenges for their control in future. In addition, species estimations showed that more species would likely have been sampled with increased sampling effort, which could have provided additional important information for analyses. As both farm location and production methods had large impacts on arthropod numbers, it was not always possible to discern which one of these was the dominant factor. When investigating other factors it would therefore be advisable to limit studies to smaller scales where possible. Other variables that may influence pest and other arthropod numbers may therefore be masked when surveying at this large scale. Variations may also be limited by survey of similar cultivars and same age cultivars. Factors that were excluded here and that should be investigated in future studies include fertilizer regimes, differences in the susceptibility between different cultivars, differences in the impact of different pesticides (organic and inorganic) and factors that change with changes in microclimatic conditions (e.g. temperature, moisture, radiation etc.). The present study could also have been improved by including sampling of the surrounding vegetation to assess the influx of arthropods to blueberry fields from surrounding land uses, as this factor was identified as particularly important in the present study.

With regards to the description of *Acalitus vaccinii*, the quality of this description could be much improved using a molecular marker. However, after numerous attempts, I was not able to obtain a good quality CO1 sequence of this species. DNA extraction and sequencing is particularly challenging for minute organisms such as these Eriophyoid mites. These sequences could also be used to build a molecular phylogeny of Eriophyoid mites on *Vaccinium* species to uncover true identities, general evolution and even movement across the globe (in population genetic studies).

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Appendix

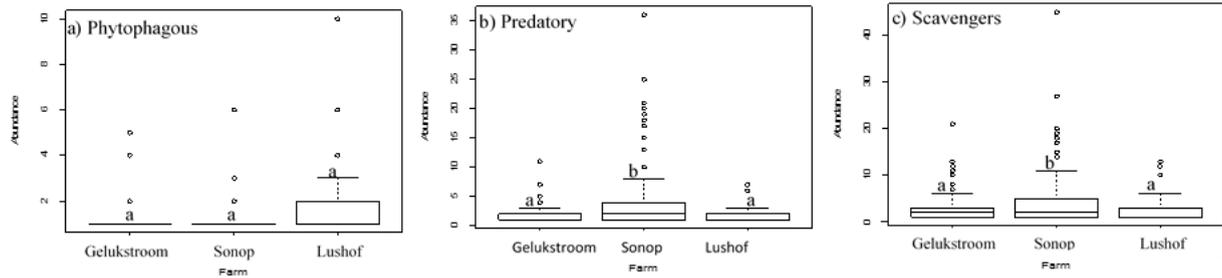
Appendix 1: Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage (inorganic vs. organic) on alpha diversity of arthropods associated with blueberries collected using the clipping sampling method. Posthoc tests are not shown.

| Richness | df | chi-square | p- | Dispersion (rdev/rdf) |
|-------------------------------------|----|------------|--------|--------------------------|
| All guilds (WC) | | | | 0,9 |
| farm | 2 | 86,662 | <0,001 | |
| month | 5 | 148,3 | <0,001 | |
| field | 1 | 33,736 | <0,001 | |
| farm*month*field | 27 | 124,58 | <0,001 | |
| Phytophagous (WC) | | | | 0,05 |
| farm | 2 | 0,042 | 0,979 | |
| month | 4 | 0,9261 | 0,921 | |
| field | 1 | 0,0172 | 0,896 | |
| farm*month*field | 9 | 0,1908 | 1.000 | |
| Predators (WC) | | | | 0,07 |
| farm | 2 | 0,848 | 0,848 | |
| month | 5 | 0,9858 | 0,9858 | |
| field | 1 | 0,0536 | 0,8169 | |
| farm*month*field | 21 | 1,9435 | 1.000 | |
| Scavengers (WC) | | | | 0,05 |
| farm | 2 | 0.2671 | 0.875 | |
| month | 5 | 0.4625 | 0.9934 | |
| field | 1 | 0.2578 | 0.6117 | |
| farm*month*field | 23 | 1.2284 | 1.000 | |
| Organic vs Inorganic all guild (WC) | | | | 0,8 |
| pesticide | 1 | 0,7051 | 0,4011 | |
| field | 1 | 12,119 | <0,001 | |
| pesticide*field | 1 | 10,173 | <0,05 | |
| Scavenger (WC) | | | | 0.04 |
| pesticide | 1 | 0.0088 | 0.9254 | |
| field | 1 | <0.0001 | 0.9902 | |
| pesticide*field | 1 | 0.0718 | 0.7887 | |
| Phytophagous (WC) | | | | 0.13 |
| pesticide | 1 | 0.1409 | 0.707 | |
| field | 1 | 0.0229 | 0.8798 | |
| pesticide*field | 1 | 0.0067 | 0.9349 | |
| Predators (WC) | | | | 0.04 |
| pesticide | 1 | 0.0476 | 0.8272 | |
| field | 1 | 0.0121 | 0.9126 | |
| pesticide*field | 1 | 0.0112 | 0.9156 | |

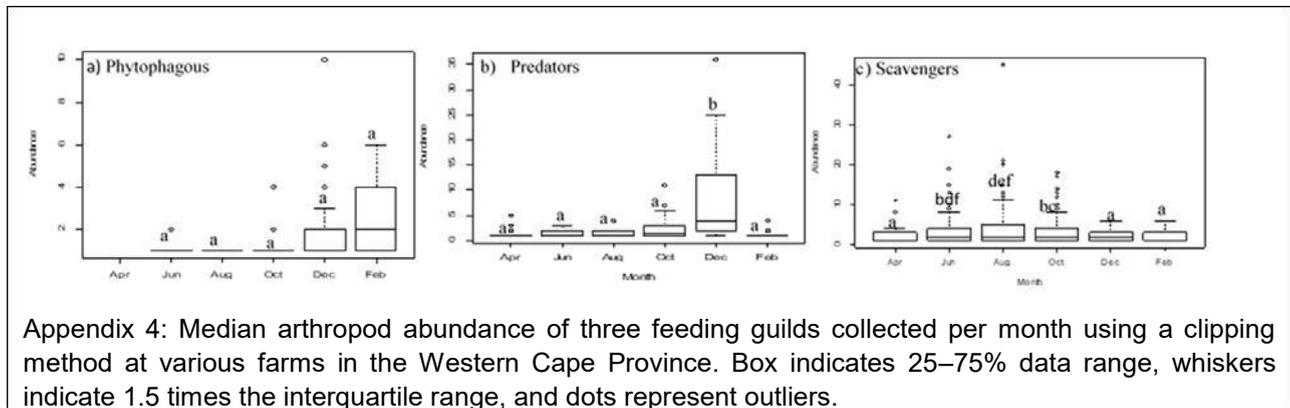
Appendix 2: Results of generalized linear models to investigate the effect of locality (farm), season (month), field type (open vs. closed) and pesticide usage (inorganic vs. organic) on abundance of arthropods associated with blueberries collected using the clipping sampling method.

| | df | chi-square | p- | Posthoc | Dispersion (rdev/rdf) |
|-------------------------------------|----|------------|---------|-----------|-----------------------|
| Overall guilds (WC) | | | | | 3,33 |
| farm | 2 | 523,11 | <0,001 | fig. 14b | |
| month | 5 | 751,6 | <0,001 | fig. 17b | |
| field | 1 | 85,089 | <0,001 | fig. 19b | |
| farm*month*field | 27 | 779,18 | <0,001 | not shown | |
| Phytophagous | | | | | 0,9 |
| farm | 2 | 2,4905 | 0,288 | appx. 3a | |
| month | 4 | 6,3419 | 0,175 | appx. 4a | |
| field | 1 | 0,0348 | 0,852 | appx. 5a | |
| farm*month*field | 9 | 4,6471 | 0,8639 | not shown | |
| Predators | | | | | 2 |
| farm | 2 | 40,349 | <0,001 | appx. 3b | |
| month | 5 | 257,66 | <0,001 | appx. 4b | |
| field | 1 | 5,8699 | <0,05 | appx. 5b | |
| farm*month*field | 21 | 98,424 | <0,001 | not shown | |
| Scavengers | | | | | 3,02 |
| farm | 2 | 75,316 | <0,001 | appx. 3c | |
| month | 5 | 47,202 | <0,001 | appx. 4c | |
| field | 1 | 2,6733 | 0,102 | appx. 5c | |
| farm*month*field | 23 | 139,06 | <0,001 | not shown | |
| Organic vs inorganic systems | | | | | 1,6 |
| pesticide | 1 | 43,122 | <0,001 | not shown | |
| field | 1 | 83,225 | <0,001 | not shown | |
| pesticide*field | 1 | 27,485 | <0,001 | appx. 6 | |
| Phytophagous | | | | | 1.12 |
| pesticide | 1 | 2.576 | 0.1085 | not shown | |
| field | 1 | 2.073 | 0.1499 | not shown | |
| pesticide*field | 1 | | | not shown | |
| Predators | | | | | 0.88 |
| pesticide | 1 | 4.7046 | 0.0301 | not shown | |
| field | 1 | 0.7748 | 0.3787 | not shown | |
| pesticide*field | 1 | 0.1921 | 0.6612 | not shown | |
| Scavengers | | | | | 1.26 |
| pesticide | 1 | 2.3513 | 0.1252 | not shown | |
| field | 1 | 17.339 | <0.0001 | not shown | |
| pesticide*field | 1 | 6.3714 | 0.0414 | appx. 7 | |

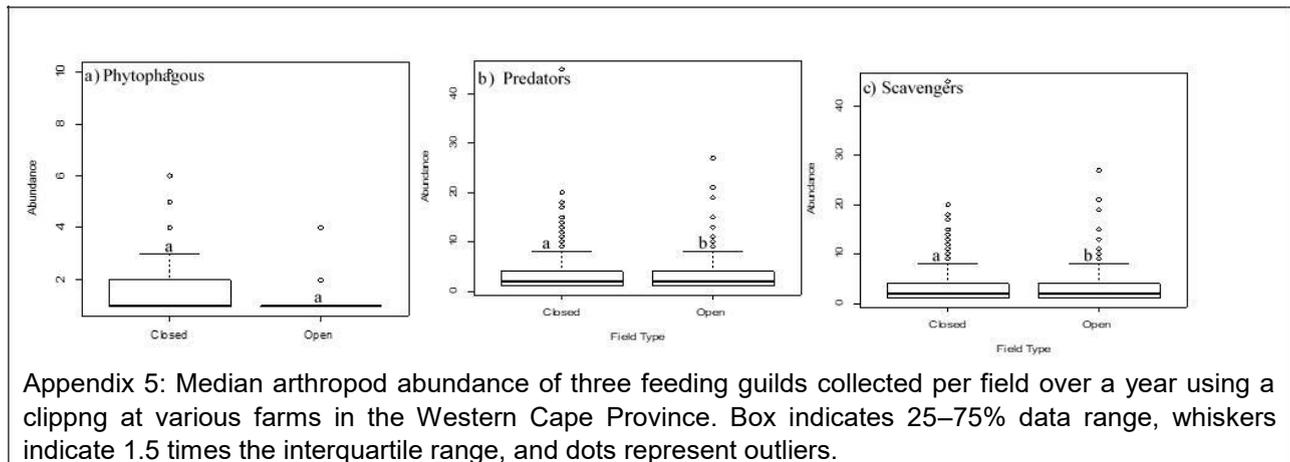
* analysis to test the effect of farm was done using Kruskal-Wallis ANOVA



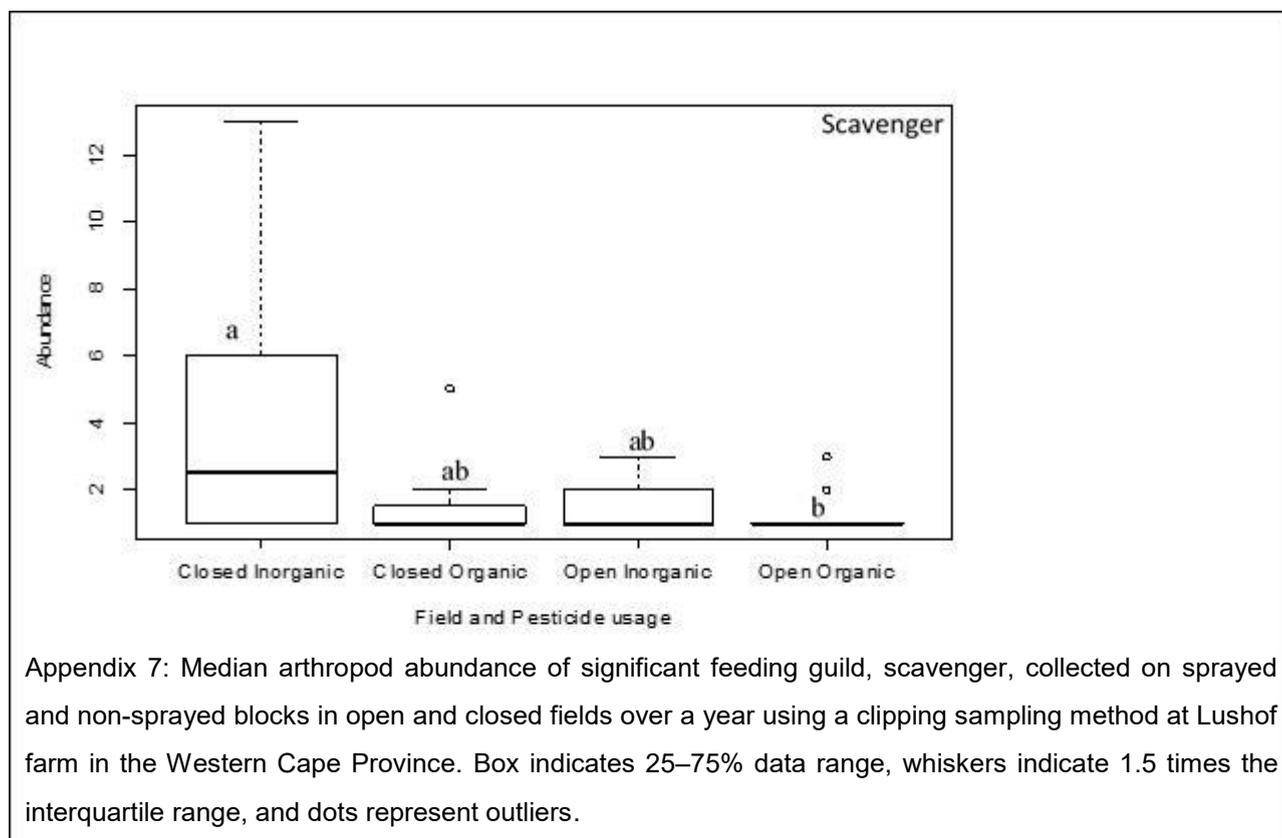
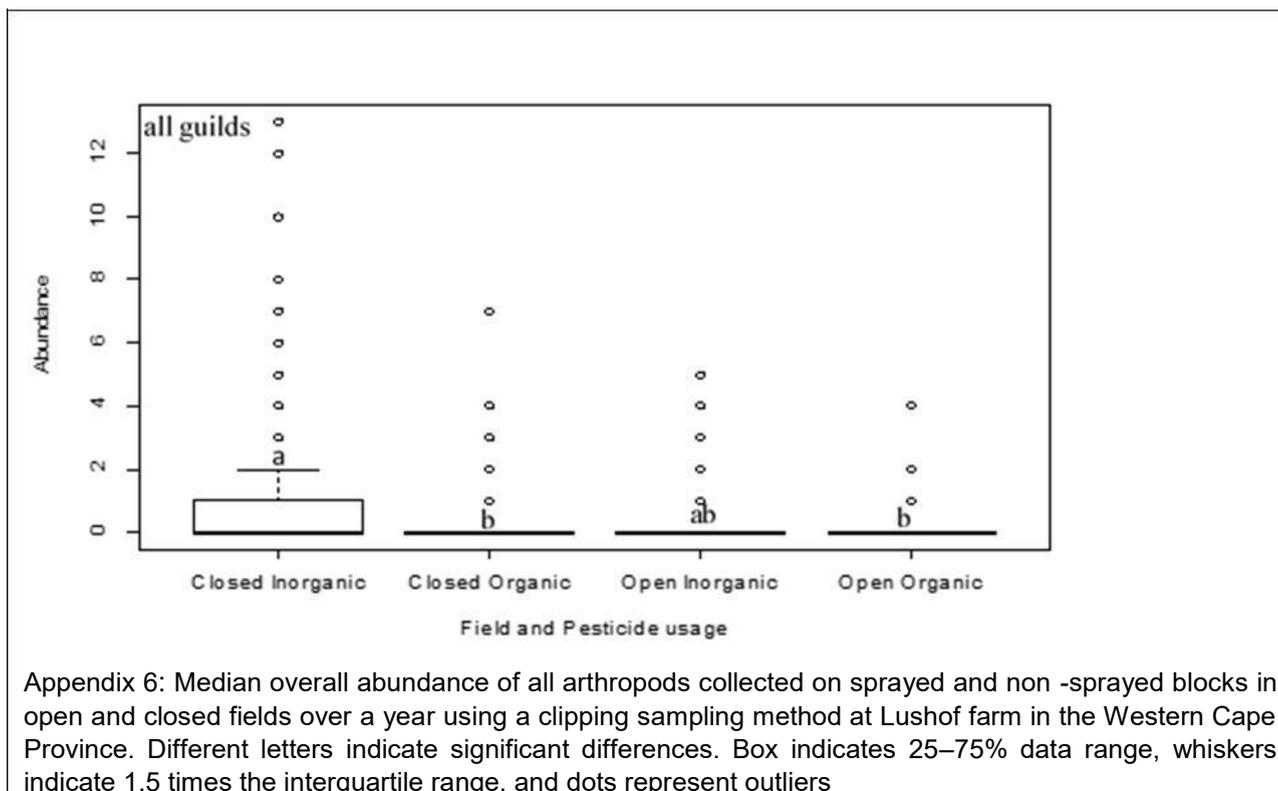
Appendix 3: Median arthropod abundance of three feeding guilds collected over one year using a clipping sampling method at various farms in the Western Cape Province. Different letters indicate significant differences. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

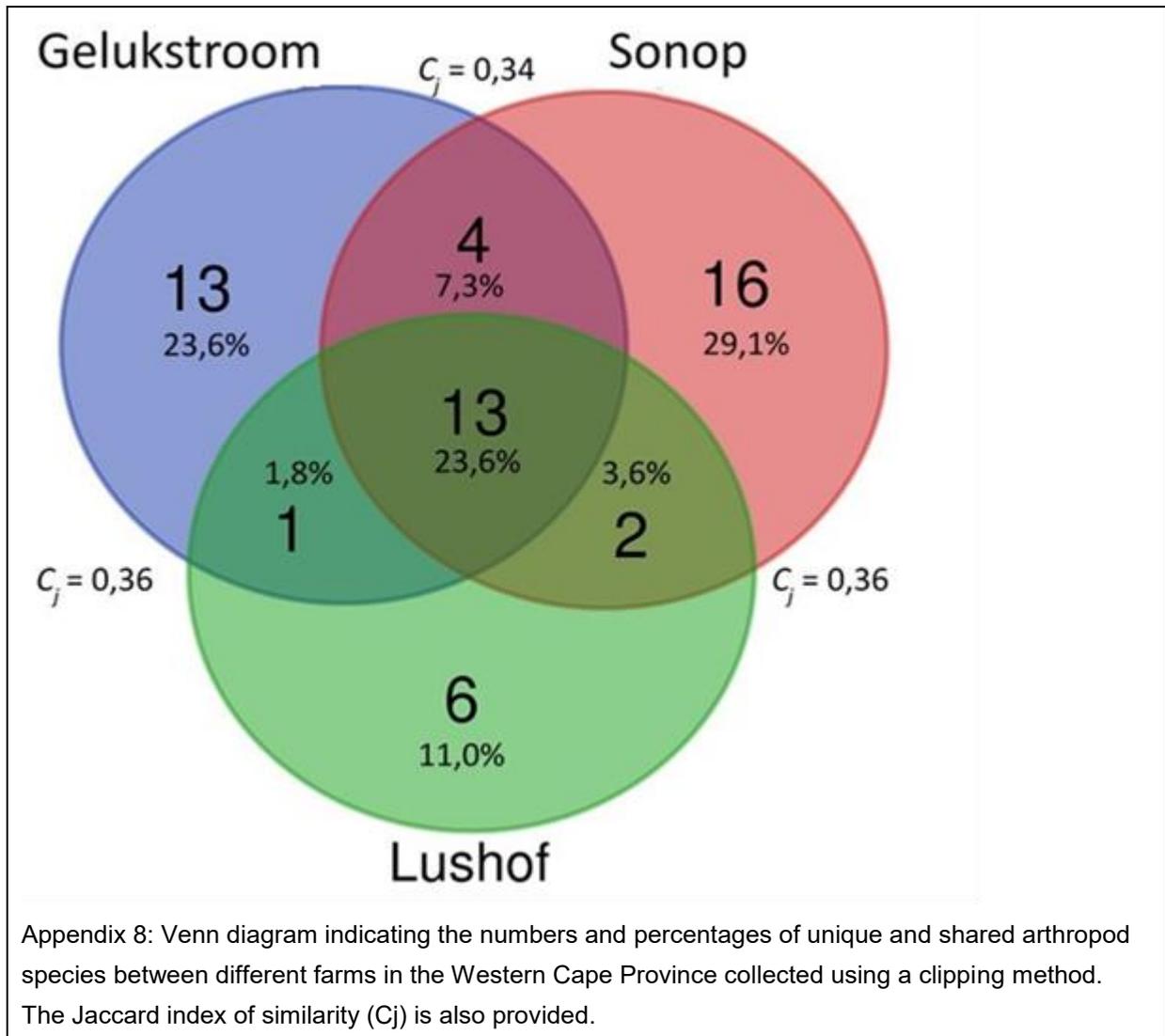


Appendix 4: Median arthropod abundance of three feeding guilds collected per month using a clipping method at various farms in the Western Cape Province. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

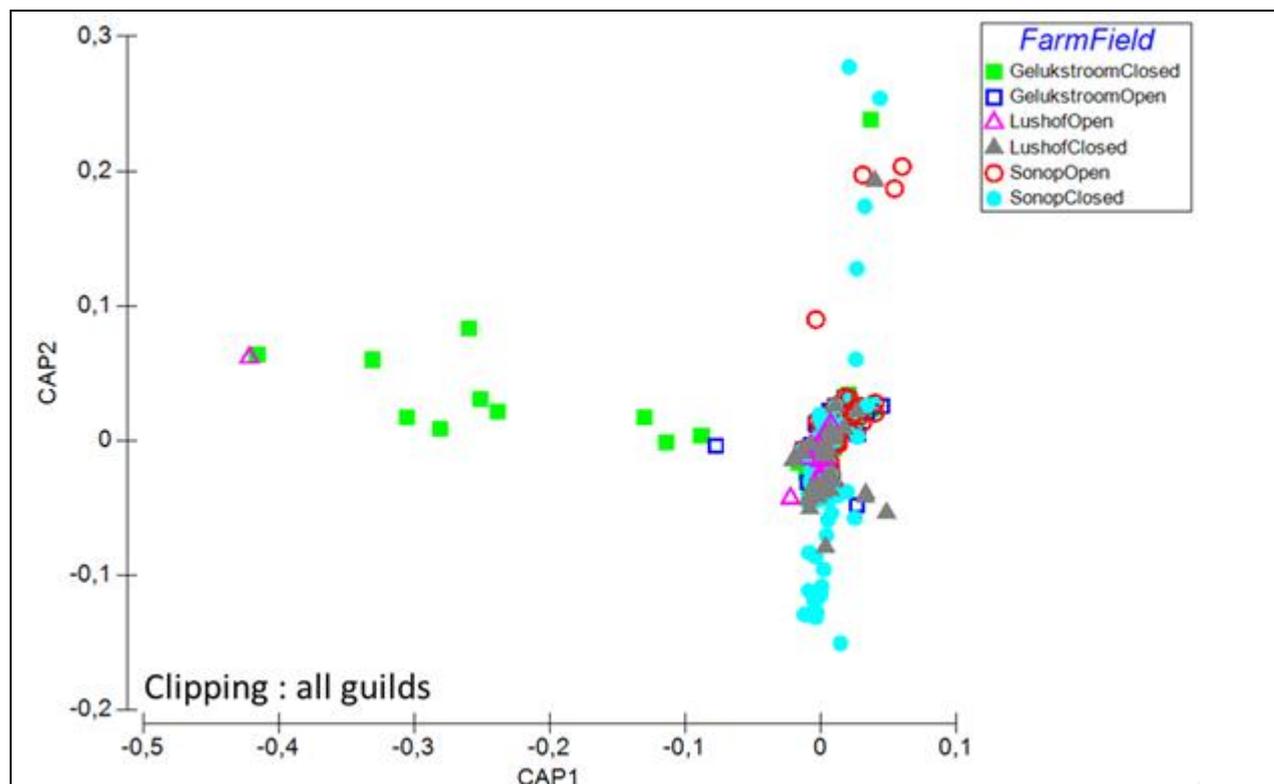


Appendix 5: Median arthropod abundance of three feeding guilds collected per field over a year using a clipping at various farms in the Western Cape Province. Box indicates 25–75% data range, whiskers indicate 1.5 times the interquartile range, and dots represent outliers.

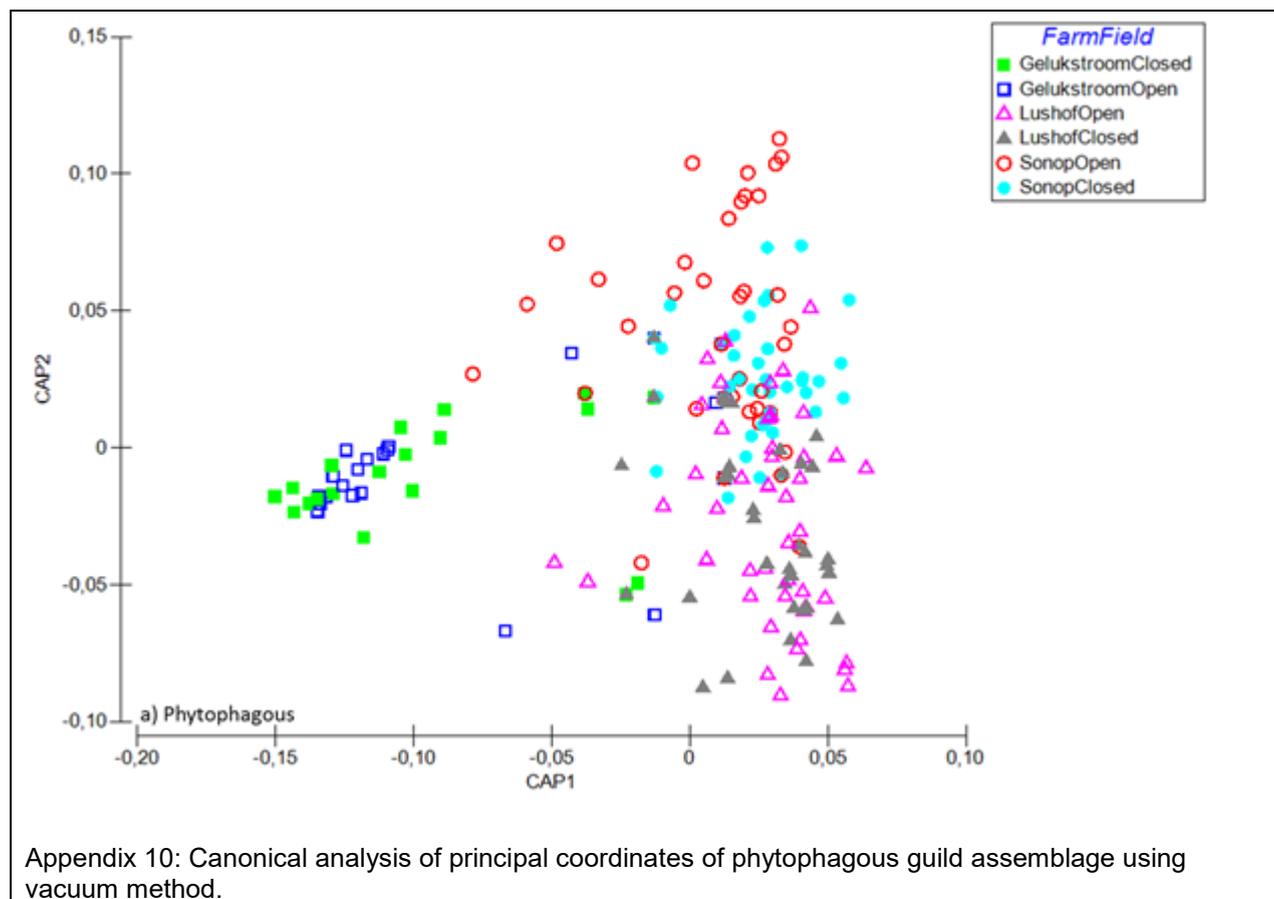




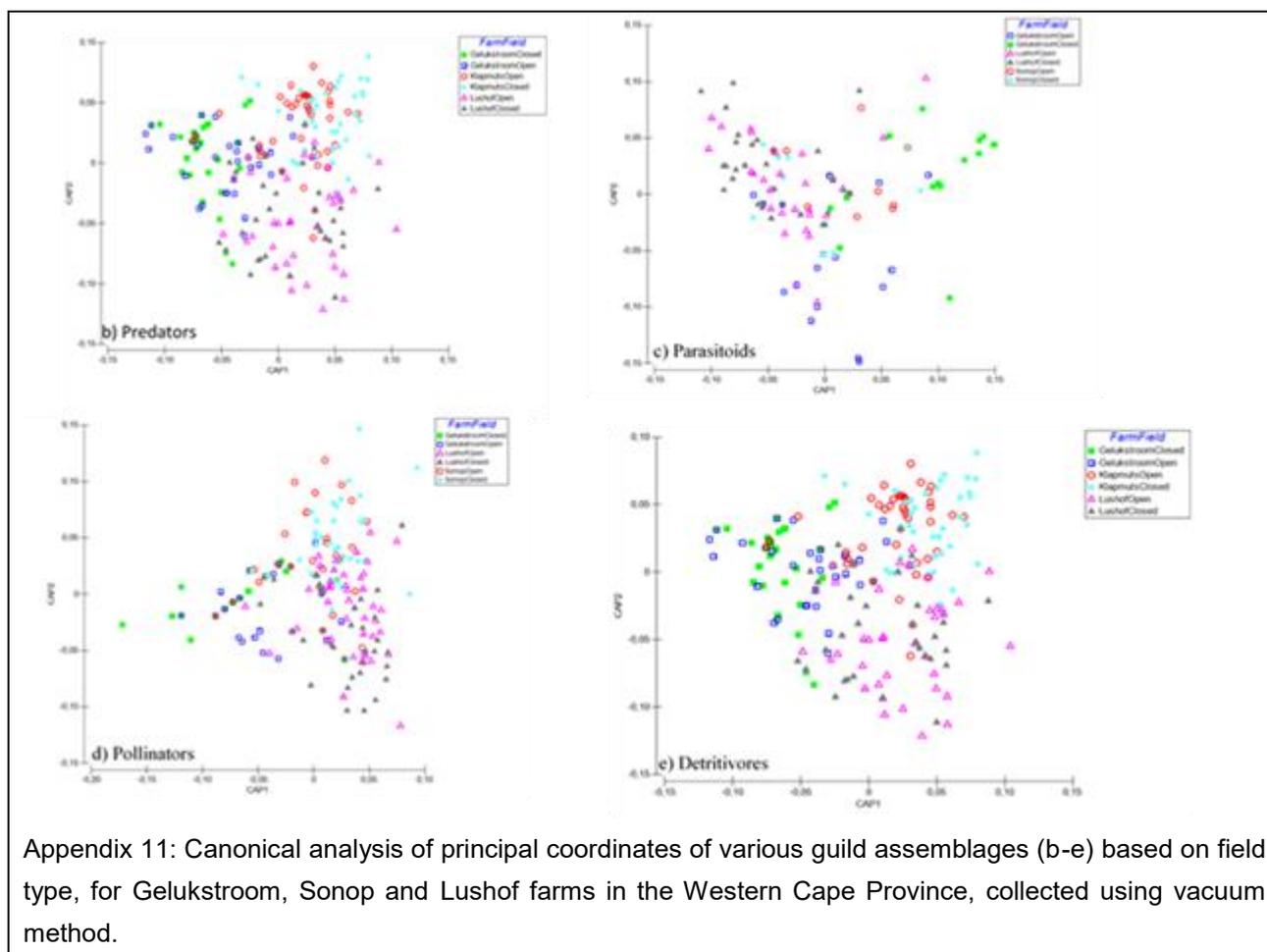
Appendix 8: Venn diagram indicating the numbers and percentages of unique and shared arthropod species between different farms in the Western Cape Province collected using a clipping method. The Jaccard index of similarity (C_j) is also provided.



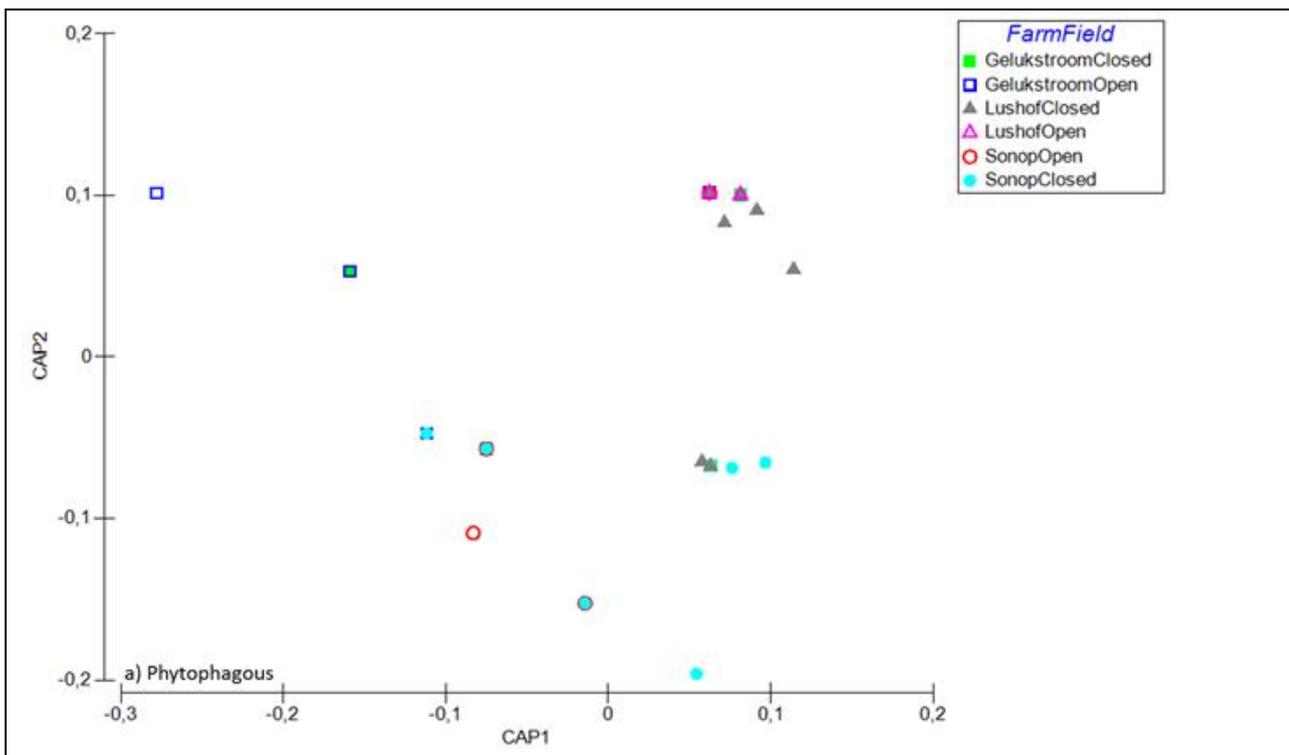
Appendix 9: Canonical analysis of principal coordinates of overall guild assemblages based on field type, for Gelukstroom, Klapmuts and Lushof farms in the Western Cape Province, collected using clipping sampling method.



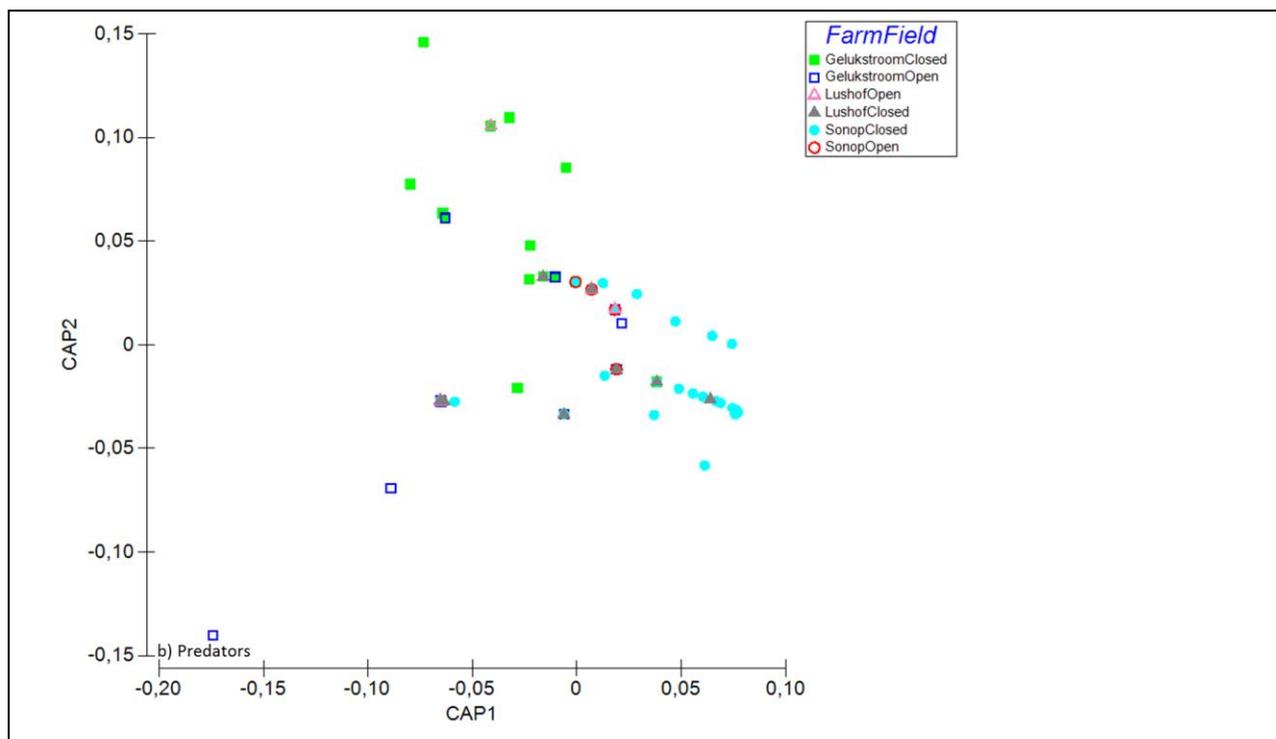
Appendix 10: Canonical analysis of principal coordinates of phytophagous guild assemblage using vacuum method.



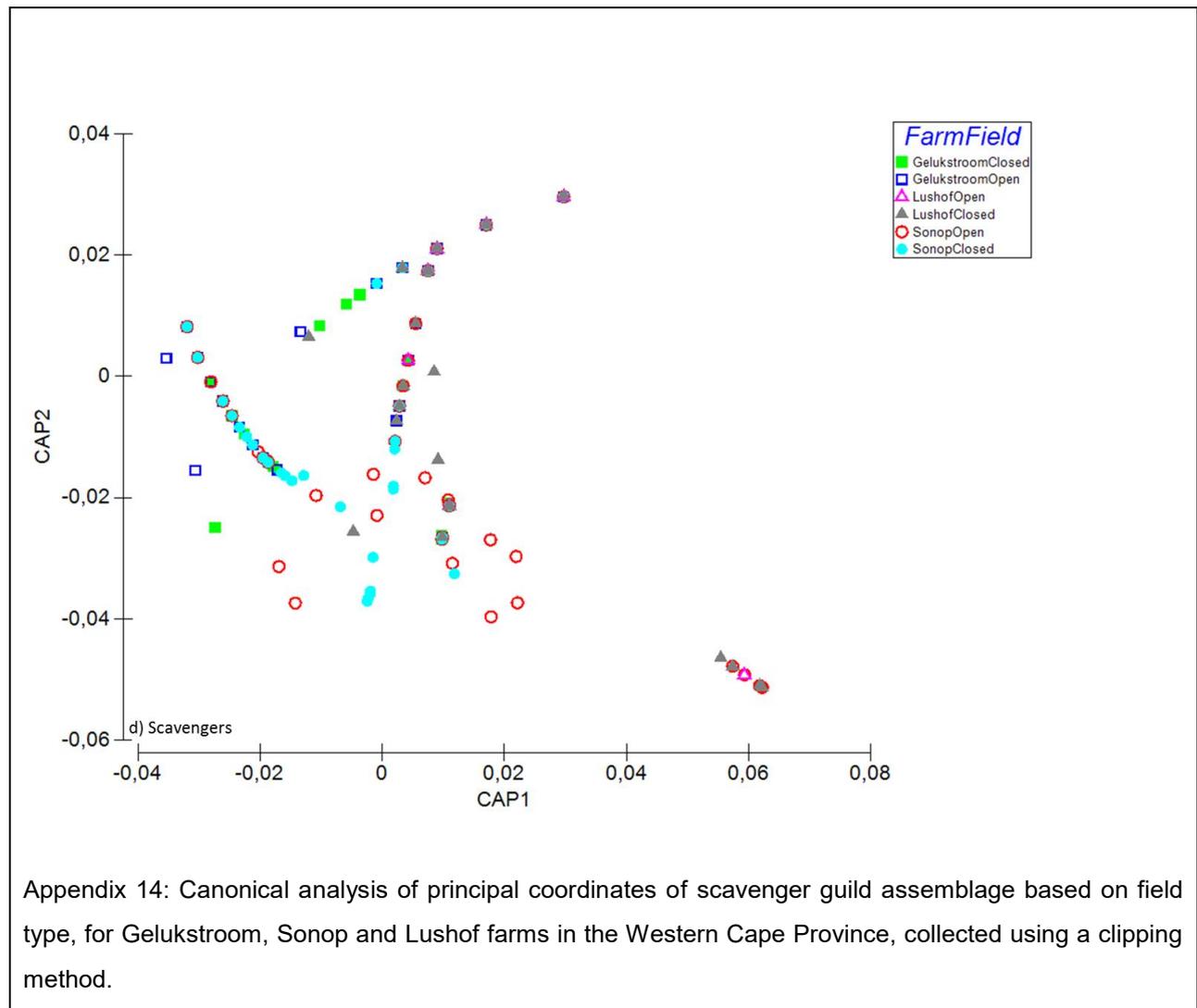
Appendix 11: Canonical analysis of principal coordinates of various guild assemblages (b-e) based on field type, for Gelukstroom, Sonop and Lushof farms in the Western Cape Province, collected using vacuum method.



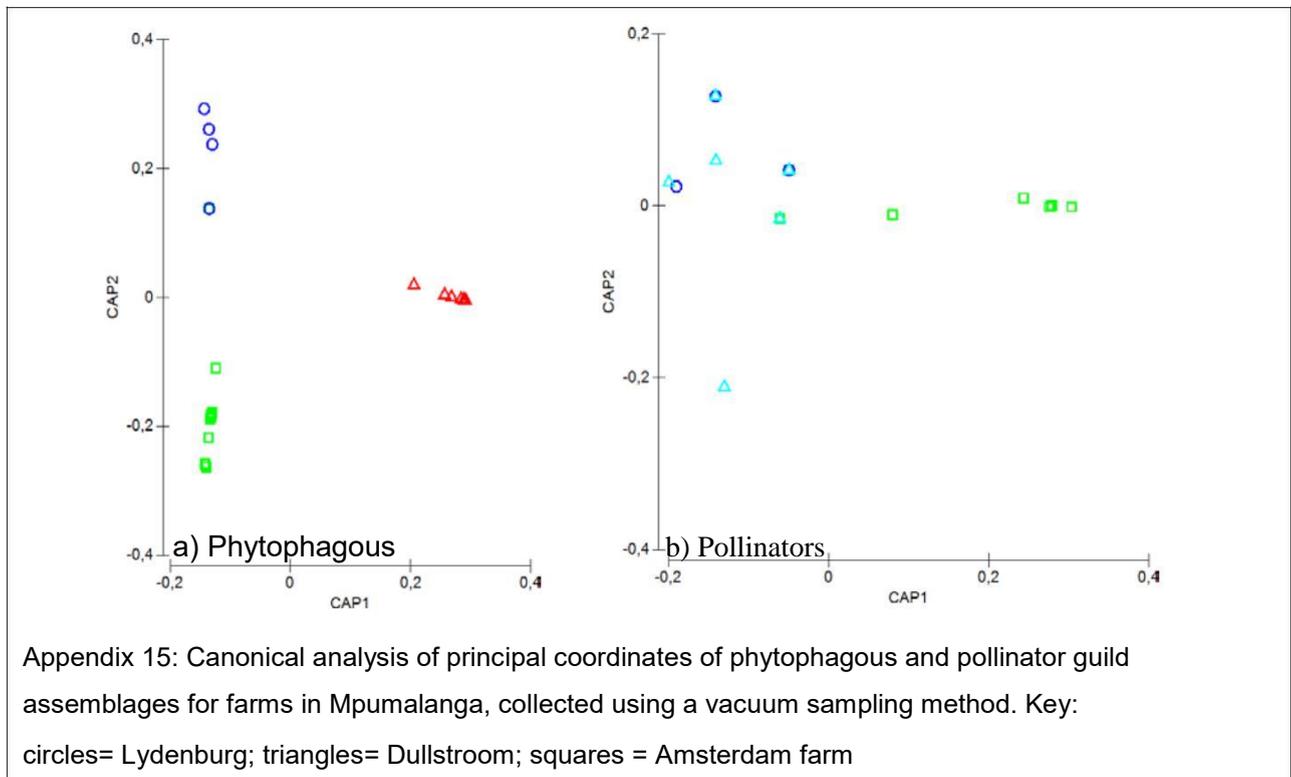
Appendix 12: Canonical analysis of principal coordinates of phytophagous guild assemblage based on field type, for Gelukstroom, Sonop and Lushof farms in the Western Cape Province, collected using a clipping method.

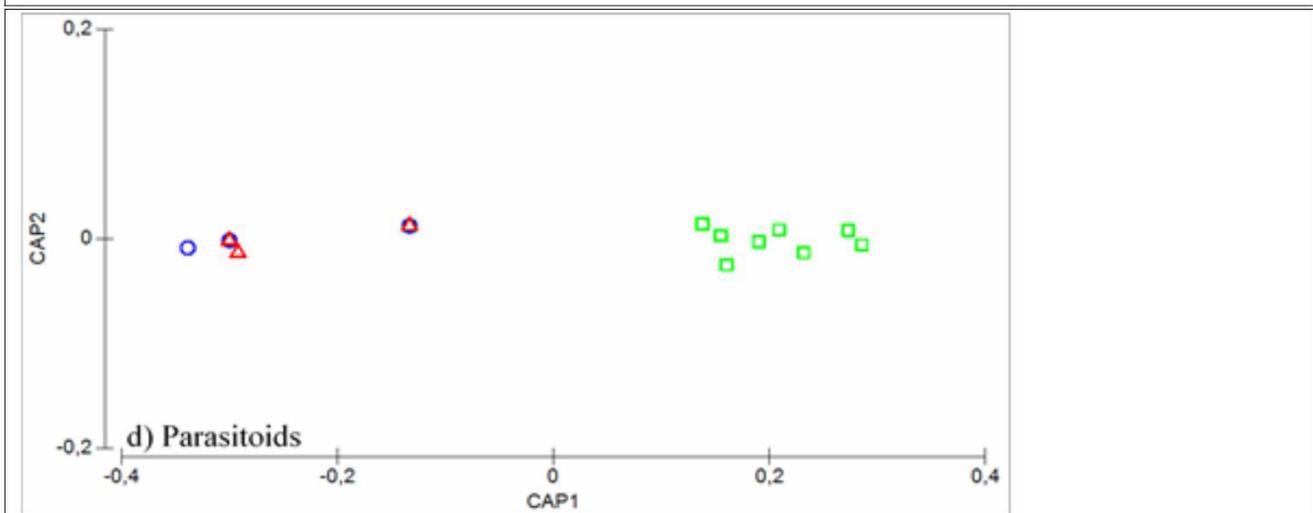
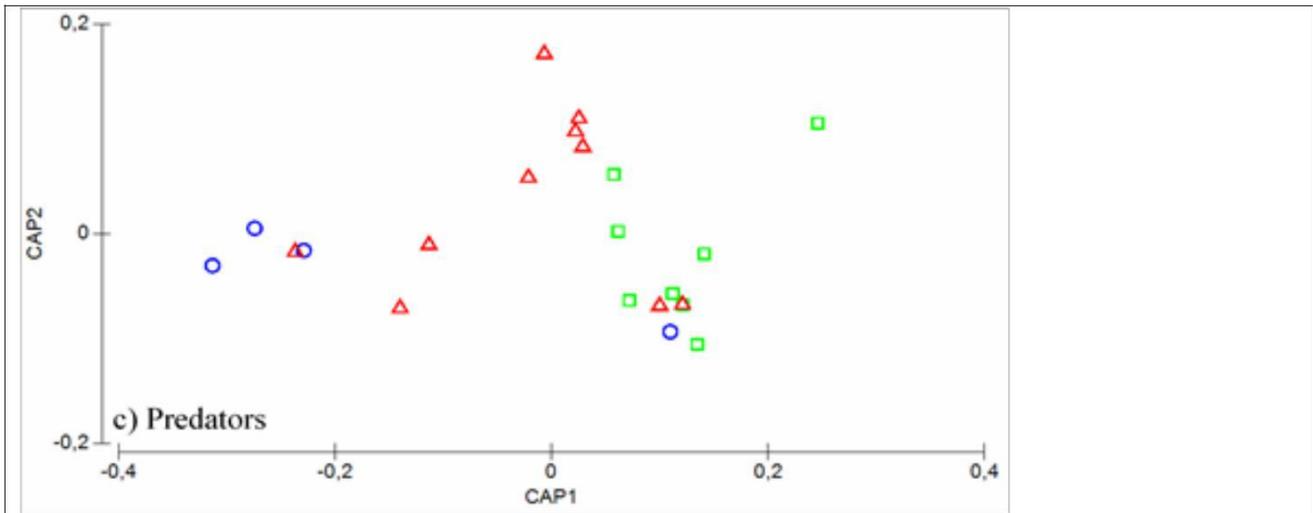


Appendix 12: Canonical analysis of principal coordinates of phytophagous guild assemblage based on field type, for Gelukstroom, Sonop and Lushof farms in the Western Cape Province, collected using a Clipping method.

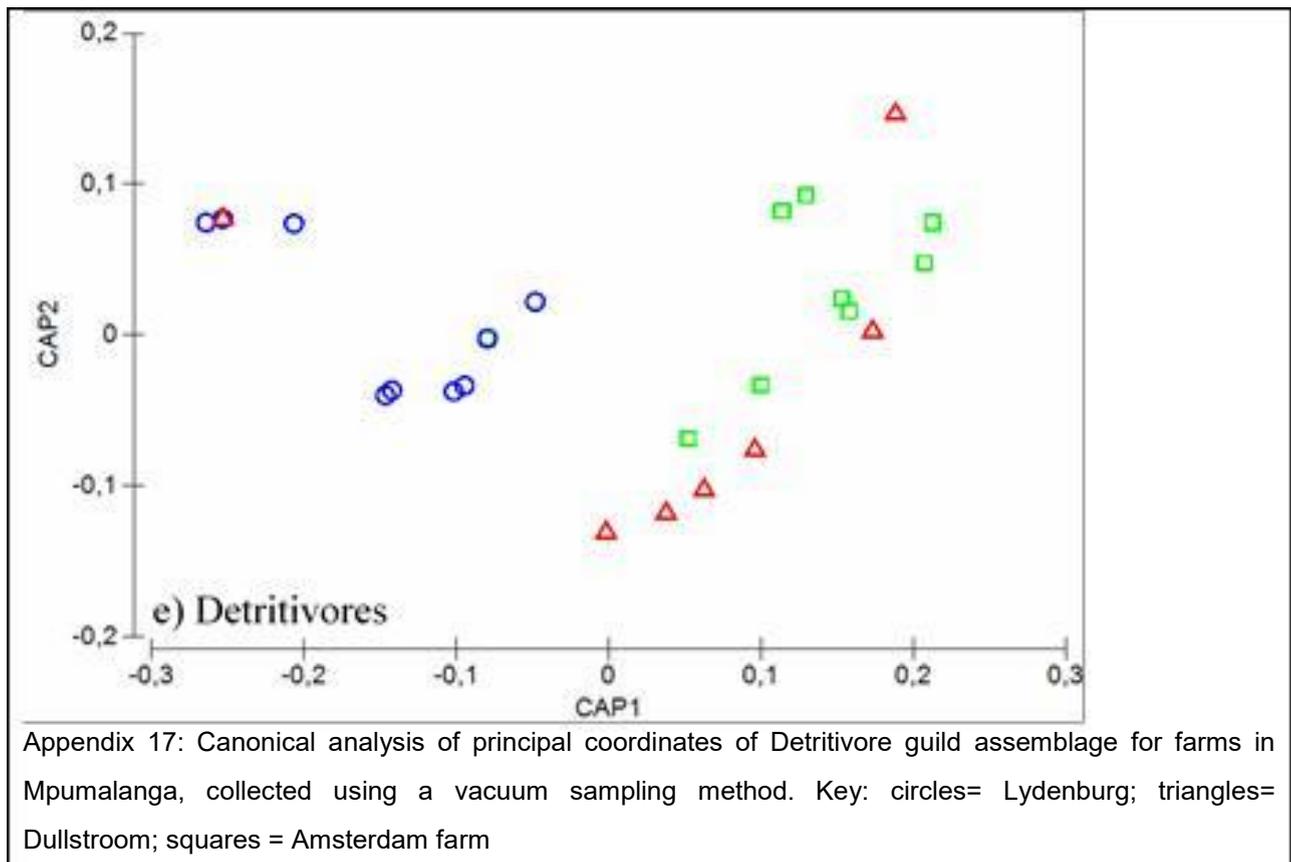


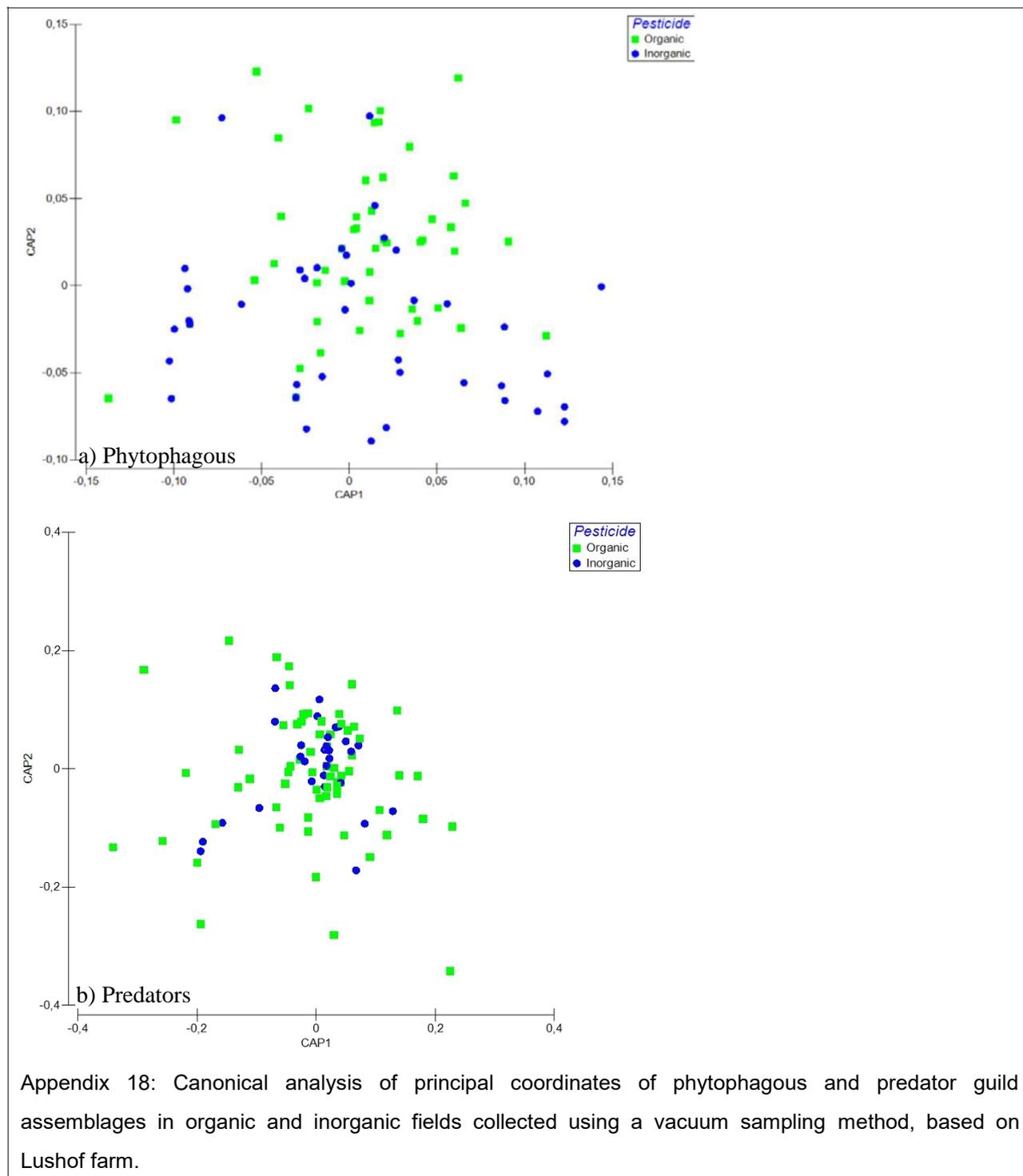
Appendix 14: Canonical analysis of principal coordinates of scavenger guild assemblage based on field type, for Gelukstroom, Sonop and Lushof farms in the Western Cape Province, collected using a clipping method.

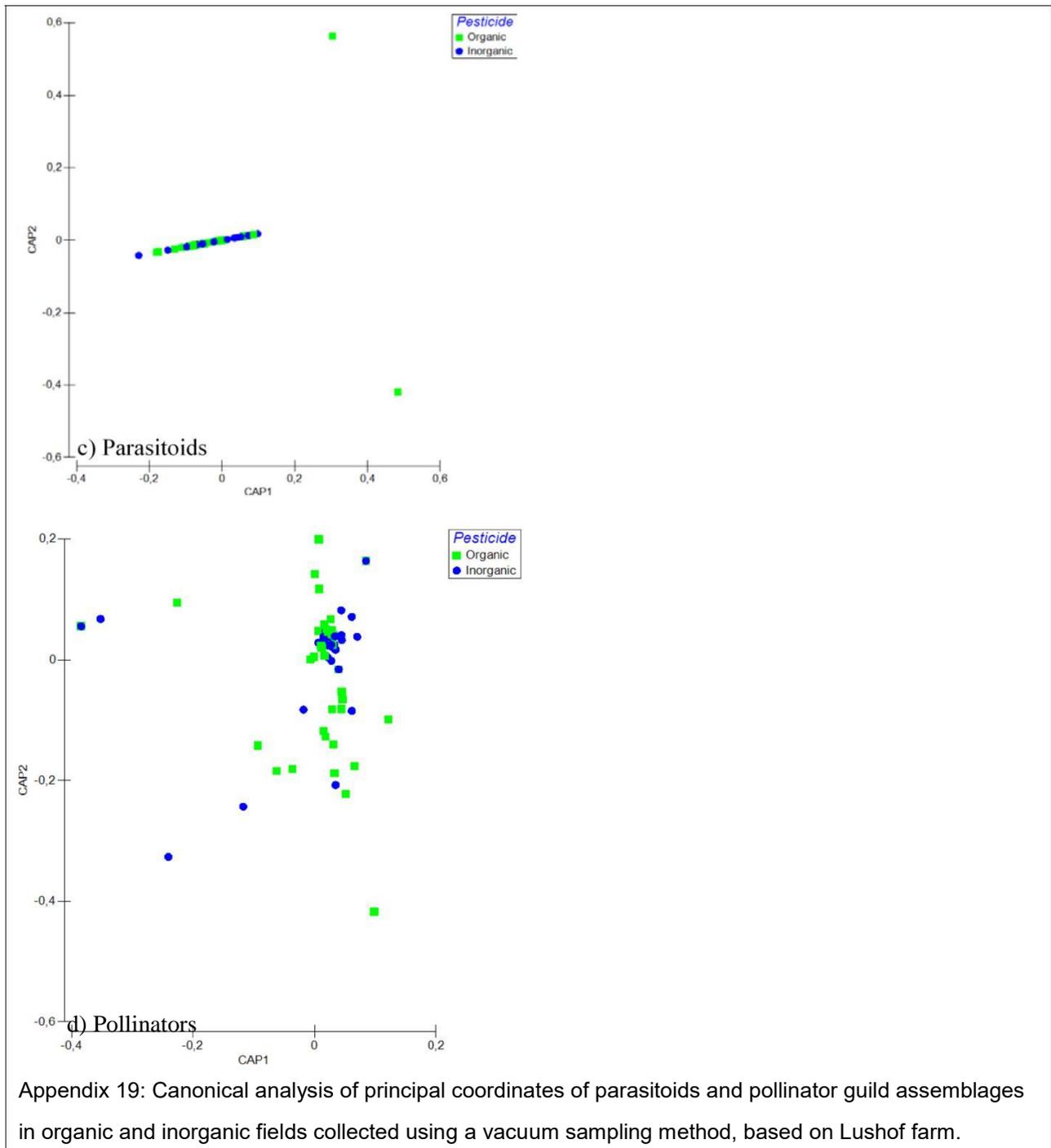




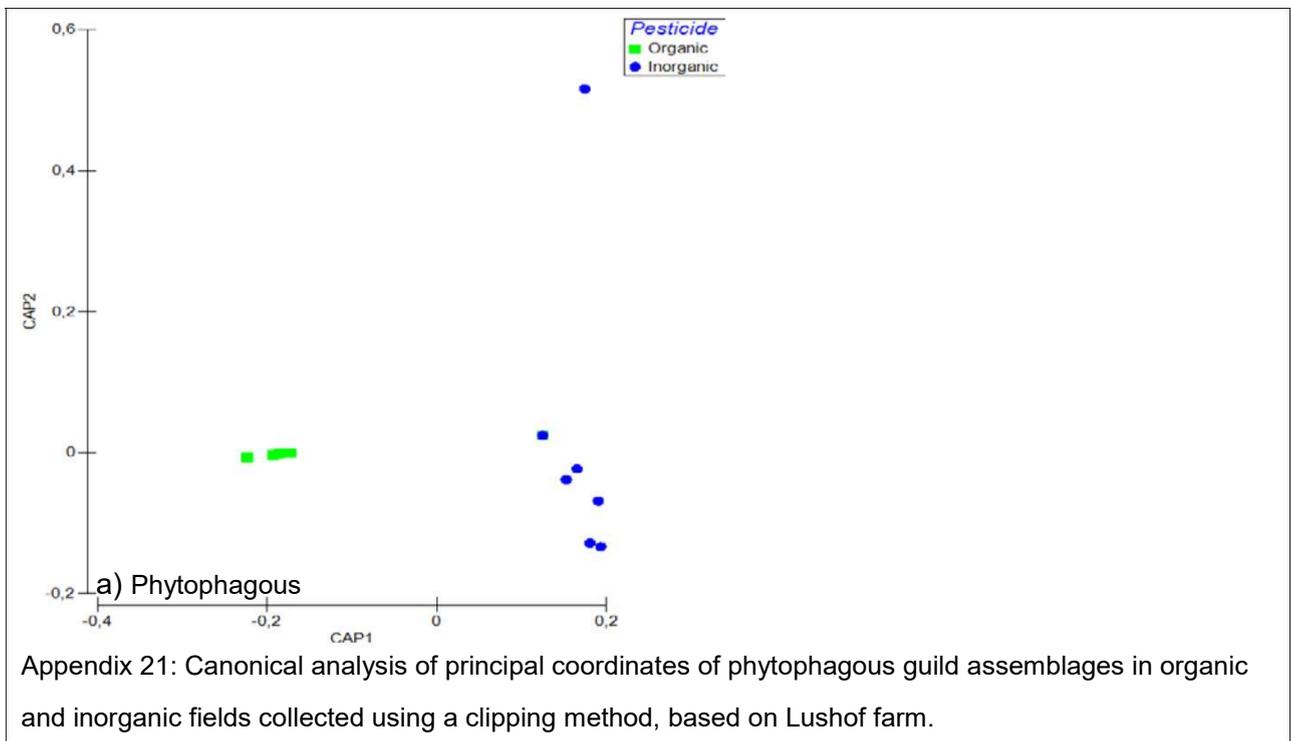
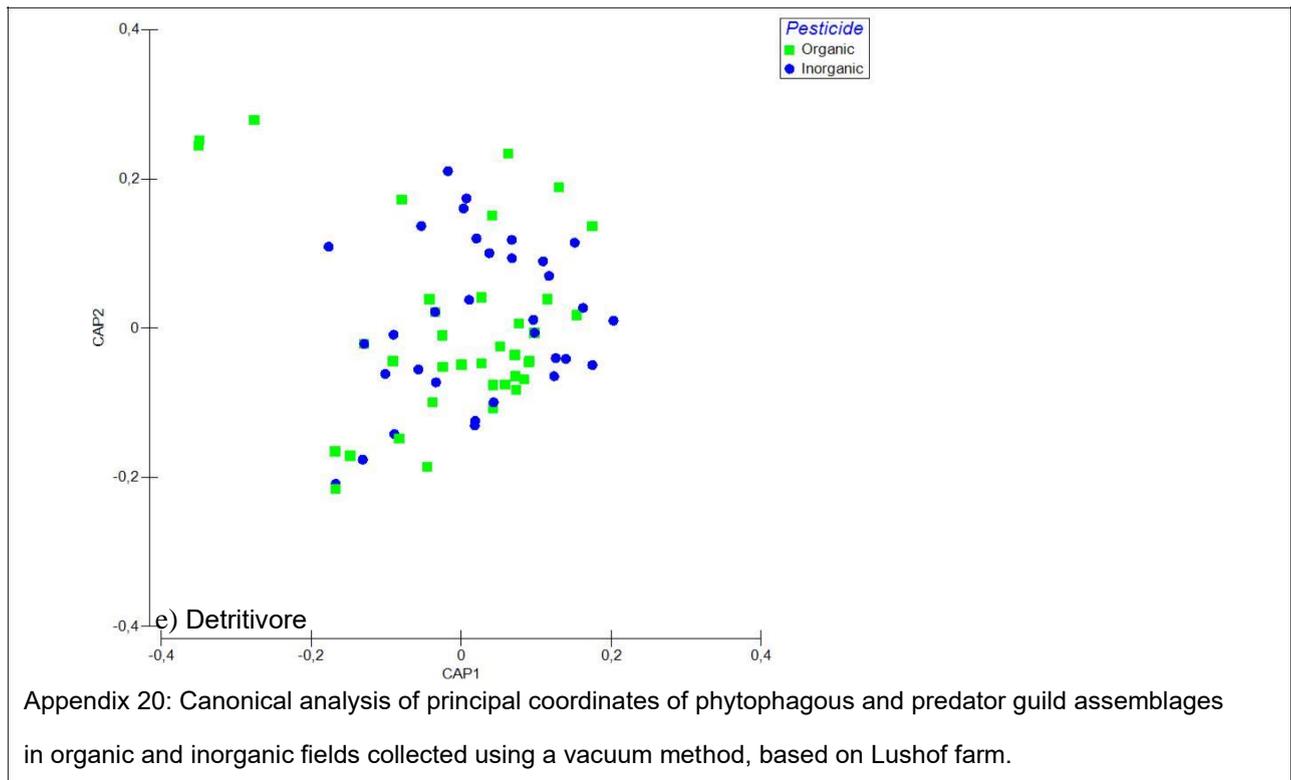
Appendix 16: Canonical analysis of principal coordinates of predators and parasitoids guild assemblages assemblage for farms in Mpumalanga, collected using a vacuum sampling method. Key: circles= Lydenburg; triangles= Dullstroom; squares = Amsterdam farm







Appendix 19: Canonical analysis of principal coordinates of parasitoids and pollinator guild assemblages in organic and inorganic fields collected using a vacuum sampling method, based on Lushof farm.



Appendix 22: Effect of farm, field type (open vs. closed) and pesticide usage (organic vs. inorganic) and their interaction on beta diversity of various feeding guilds on blueberry crops collected using a clipping method and a PERMANOVA pairwise test between farms.

| | | Variables | df | Pseudo-F | P(perm) | Post hoc | |
|----------------------------------|--------------|------------------|------------|-----------------|----------------|---------------------|---------------------|
| WC Clipping | All guilds | Farm | 2 | 2.1169 | 0.0135 | GS≠LH; KM≠LH; GS=KM | |
| | | Field | 1 | 1.3657 | 0.2498 | | |
| | | FarmxField | 2 | 1.5624 | 0.074 | | |
| | | Phytophagous | | | | | |
| | | | Farm | 2 | 1.409 | 0.1445 | KM≠LH; GS=LH; GS=KM |
| | | | Field | 1 | 0.51 | 0.7269 | |
| | | | FarmxField | 2 | 0.5375 | 0.9307 | |
| | | Predators | | | | | |
| | | | Farm | 2 | 1.544 | 0.1463 | |
| | | | Field | 1 | 2.628 | 0.0375 | |
| | | | FarmxField | 2 | 3.0775 | 0.0369 | |
| | | Scavengers | | | | | |
| | | Farm | 2 | 3.1031 | 0.0089 | KM≠LH; GS≠LH; GS=KM | |
| | | Field | 1 | 0.79316 | 0.5377 | | |
| | | FarmxField | 2 | 1.9515 | 0.062 | | |
| Field type | | | | | | | |
| Open field (WC) | All guilds | Farm | 2 | 1,7861 | 0,027 | KM≠LH; GS=LH; GS=KM | |
| | | Month | 5 | 3,346 | 0.0001 | | |
| Closed field (WC) | All guilds | Farm | 2 | 0,852 | 0,3535 | | |
| | | Month | 4 | 1,4104 | 0,1624 | | |
| Pesticide usage | | | | | | | |
| Organic vs Inorganic (WC) | All guilds | Field | 1 | 1,4245 | 0,2902 | | |
| | | Pesticide | 1 | 1,3653 | 0,2227 | | |
| | | FieldxPesticide | 1 | 2,4418 | 0,2093 | | |
| Organic vs Inorganic (WC) | Phytophagous | Field | 1 | No test | | | |
| | | Pesticide | 1 | No test | | | |
| | | FieldxPesticide | 1 | No test | | | |
| Organic vs Inorganic (WC) | Predators | Field | 1 | 1,887 | 0,0927 | | |
| | | Pesticide | 1 | 0,7016 | 0,6766 | | |
| | | FieldxPesticide | 1 | No test | | | |
| Organic vs Inorganic (WC) | Scavengers | Field | 1 | 10,18 | 0,2091 | | |
| | | Pesticide | 1 | 0,86713 | 0,4762 | | |
| | | FieldxPesticide | 1 | no test | | | |

Appendix 23: Species list of various feeding guilds collected using poking method at six farms

| <i>Farm</i> | Order | Family | Species | Individuals collected |
|--------------------|---------------------|----------------|-----------------------|------------------------------|
| | <i>Detritivores</i> | | | |
| <i>Amsterdam</i> | Coleoptera | unknown | COL D | 59 |
| | Diptera | Sciaridae | SCIARIDAE 1 | 2 |
| | | unknown | DIP 2 | 6 |
| | | | DIP 3 | 1 |
| | | | DIP 4 | 3 |
| | | | DIP 8 | 2 |
| | | | DIP 9 | 1 |
| | Psocoptera | unknown | PSOC A | 1 |
| | <i>Dullstroom</i> | Coleoptera | unknown | COL D |
| Diptera | | unknown | DIP 13 | 1 |
| | | | DIP 2 | 5 |
| <i>Gelukstroom</i> | Coleoptera | Cryptophagidae | Cryptophagid 1 | 5 |
| | | | Micrambe sp. | 5 |
| | Diptera | Anisopodidae | DIP O | 10 |
| | | | DIP T2 | 2 |
| | | Chamaemyiidae | DIP C | 10 |
| | | Mycetophilidae | DIP T | 6 |
| | | Sciaridae | SCIARID 2 | 8 |
| | SCIARID 3 | | 2 | |
| | | | SCIARID 4 | 2 |
| | | | SCIARIDAE | 35 |
| | | unknown | DIP 12 | 2 |
| | DIP CCC | | 1 | |
| | | | DIP CD | 12 |
| | | | DIP D1 | 1 |
| | | | DIP KK3 | 1 |
| | | | DIP TT | 1 |
| | | | DIP U | 7 |
| | Myriapoda | Julidae | Ommattoiulus moreleti | 8 |
| | | unknown | MILLI 2 | 10 |
| | Prostigmata | Tydeidae | Tydeus grabouwi | 3 |
| | | Tydoidea | Tydoidea | 1 |
| Psocoptera | Ectopsocidae | PSOC 2 | 5 | |
| | | Peripsocidae | PSOC 1 | 51 |
| | | PSOC 1B | 4 | |
| | | PSOC 2 | 2 | |
| | | PSOC 3 | 5 | |
| <i>Sonop</i> | Coleoptera | Cryptophagidae | Cryptophagid 1 | 112 |
| | | Curculionidae | Cryptophagid 1 | 5 |
| | | Tenebrionidae | COL 8 | 23 |
| | | unknown | COL 10 | 1 |
| | | | COL 27 | 2 |
| | Collembola | Entomobryoidea | ENTO 1 | 7 |

| | | | |
|-------------|----------------|-----------------------|-----|
| | Poduroidea | POD 1 | 1 |
| Diptera | Anisopodidae | DIP O | 70 |
| | Chamaemyiidae | DIP C | 53 |
| | Lonchopleridae | DIP Q | 18 |
| | Mycetophilidae | DIP T | 155 |
| | | MYCETOPHIL 1 | 16 |
| | | MYCETOPHIL 2 | 35 |
| | Sciaridae | DIP H | 35 |
| | | SCIARID 2 | 16 |
| | | SCIARIDAE | 92 |
| | unknown | DIP B | 5 |
| | | DIP D | 11 |
| | | DIP E | 1 |
| | | DIP GG | 5 |
| | | DIP HH | 2 |
| | | DIP I | 92 |
| | | DIP J | 7 |
| | | DIP JJ | 7 |
| | | DIP K | 10 |
| | | DIP MM | 1 |
| | | DIP N | 18 |
| | | DIP NN | 2 |
| | | DIP P | 1 |
| | | DIP QQ | 1 |
| | | DIP U | 1 |
| Myriapoda | Julidae | Ommattoiulus moreleti | 144 |
| Prostigmata | Tydeidae | Tydeus sp. | 1 |
| Psocoptera | Ectopsocidae | PSOC 2 | 1 |
| | Peripsocidae | PSOC 1 | 41 |
| | | PSOC 1B | 1 |
| | | PSOC 3 | 1 |
| BLATTODEA | Blattidae | Blattid 1 | 1 |
| Coleoptera | Cryptophagidae | Cryptophagid 1 | 147 |
| | | Micrambe sp. | 2 |
| | Cryptophagidae | Cryptophagid 1 | 16 |
| | | Micrambe sp. | 5 |
| | Cryptophagidae | Micrambe sp. | 1 |
| | Tenebrionidae | COL 28 | 1 |
| | | COL 29 | 1 |
| | | COL 8 | 1 |
| | unknown | COL 10 | 18 |
| | | COL 27 | 3 |
| Collembola | Entomobryoidea | ENTO 1 | 3 |
| | | ENTO 2 | 2 |
| | | ENTO 3 | 3 |
| | unknown | COLLE 1 | 1 |
| Diptera | Anisopodidae | DIP O | 15 |
| | | DIP T2 | 18 |

Lushof

| | | | | |
|------------------|-------------|-------------------|-----------------------|-----|
| | | Chamaemyiidae | DIP C | 138 |
| | | Lonchopleridae | DIP Q | 9 |
| | | Mycetophilidae | DIP T | 85 |
| | | | MYCETOPHIL 1 | 7 |
| | | | MYCETOPHIL 2 | 7 |
| | | Psychodidae | DIP T4 | 14 |
| | | Sciaridae | DIP H | 42 |
| | | | SCIARID 2 | 140 |
| | | | SCIARID 3 | 29 |
| | | | SCIARID 4 | 24 |
| | | | SCIARID 5 | 2 |
| | | | SCIARIDAE | 53 |
| | | unknown | DIP AAA2 | 1 |
| | | | DIP B | 1 |
| | | | DIP BB3 | 4 |
| | | | DIP CD | 4 |
| | | | DIP D | 35 |
| | | | DIP D1 | 21 |
| | | | DIP GG | 17 |
| | | | DIP HH | 2 |
| | | | DIP I | 5 |
| | | | DIP JJ | 88 |
| | | | DIP K | 1 |
| | | | DIP K1 | 10 |
| | | | DIP KK3 | 5 |
| | | | DIP MM | 5 |
| | | | DIP N | 22 |
| | | | DIP NN | 24 |
| | | | DIP QQ | 1 |
| | | | DIP RR2 | 1 |
| | | | DIP T3 | 6 |
| | | | DIP TT | 2 |
| | | | DIP U | 11 |
| | | | DIP WW2 | 1 |
| | | | DIP X3B | 1 |
| | Myriapoda | Julidae | Ommattoiulus moreleti | 15 |
| <i>Lydenburg</i> | Blattodea | Blattidae | BLAT 1 | 1 |
| | Coleoptera | unknown | COL D | 6 |
| | Diptera | Sciaridae | SCIARIDAE 1 | 1 |
| | | | SCIARIDAE 2 | 1 |
| | | unknown | DIP 13 | 1 |
| | | | DIP 9 | 1 |
| | Psocoptera | unknown | PSOC B | 1 |
| | | Formicidae | | |
| <i>Amsterdam</i> | Hymenoptera | Formicidae | FORM A | 3 |
| | | | FORM B | 3 |
| | | | FORM C | 2 |
| | | | FORM D | 4 |

| | | | | |
|--------------------|--------------------|-----------------|------------------|----|
| | | | FORM E | 1 |
| | | | FORM F | 2 |
| | | | FORM G | 1 |
| <i>Gelukstroom</i> | Hymenoptera | Formicidae | Formicid 1 | 2 |
| | | | Formicid 2 | 12 |
| | | | Formicid 3 | 18 |
| | | | Formicid 4 | 1 |
| | | | Formicid 5 | 2 |
| | | | Formicid 6 | 1 |
| <i>Sonop</i> | Hymenoptera | Formicidae | Formicid 6 | 21 |
| | | | Formicid 8 | 18 |
| <i>Lushof</i> | Hymenoptera | Formicidae | Formicid 2 | 3 |
| | | | Formicid 8 | 1 |
| | | | Formicid 9 | 2 |
| <i>Lydenburg</i> | Hymenoptera | Formicidae | FORM B | 1 |
| | | | FORM F | 1 |
| | | | FORM H | 42 |
| | | | FORM I | 1 |
| | Fung vores | | | |
| <i>Amsterdam</i> | Coleoptera | Coccinellidae | COCCI B | 1 |
| | | unknown | COL E | 2 |
| | Sarcoptiformes | Acaridae | Rhizoglyphus sp. | 2 |
| <i>Gelukstroom</i> | Thysanoptera | Phlaeothripidae | Phlaeothrips sp. | 1 |
| <i>Sonop</i> | Thysanoptera | Phlaeothripidae | Phlaeothrips sp. | 3 |
| <i>Lushof</i> | Thysanoptera | Phlaeothripidae | Phlaeothrips sp. | 98 |
| | Parasitoids | | | |
| <i>Amsterdam</i> | Diptera | Sciaridae | SCIARIDAE 1 | 1 |
| | | unknown | DIP 3 | 5 |
| | Hemiptera | Aphididae | Aphid 1 | 1 |
| | Hymenoptera | Vespidae | VESP 1 | 1 |
| | | unknown | HYM 1 | 1 |
| | | | HYM 2 | 9 |
| | | | HYM 4 | 6 |
| | | | HYM 5 | 4 |
| | | | HYM 7 | 2 |
| | | | ICH 1 | 1 |
| | | | ICH 2 | 1 |
| | | | ICH 3 | 2 |
| | | | ICH 4 | 1 |
| <i>Dullstroom</i> | Hymenoptera | Braconidae | HYM 9 | 1 |
| | | unknown | HYM 8 | 1 |
| | | | ICH 1 | 1 |
| | | | ICH 4 | 1 |
| <i>Gelukstroom</i> | Diptera | Milichiidae | DIP 27 | 1 |
| | | Pyrgotidae | Pyrgotid 3 | 1 |
| | | Simuliidae | DIP 15 | 2 |
| | | Tethinidae | DIP M | 2 |
| | | unknown | DIP B1 | 1 |

| | | | | |
|--------------|-------------|---------------|------------|----|
| | Hymenoptera | Braconidae | Braconid 1 | 2 |
| | | unknown | HYM B | 27 |
| | | | HYM C | 17 |
| | | | HYM D | 1 |
| | | | HYM E | 5 |
| | | | HYM G | 6 |
| | | | HYM G3 | 2 |
| | | | HYM H | 3 |
| | | | HYM I | 5 |
| | | | HYM J | 3 |
| | | | HYM K | 2 |
| | | | HYM L | 3 |
| | | | HYM M | 3 |
| | | | HYM N | 2 |
| | | | HYM O | 4 |
| | | | HYM Q | 2 |
| | | | HYM R | 2 |
| | | | HYM S | 1 |
| | | | HYM T | 1 |
| | | | HYM U | 1 |
| | | | HYM V | 1 |
| <i>Sonop</i> | Diptera | Drosophilidae | DROS 5 | 1 |
| | | Pyrgotidae | Pyrgotid 1 | 6 |
| | | | Pyrgotid 3 | 1 |
| | | Tethinidae | DIP M | 4 |
| | Hymenoptera | Vespidae | VESP 2 | 2 |
| | | unknown | HYM AA2 | 3 |
| | | | HYM B | 9 |
| | | | HYM BB | 1 |
| | | | HYM BB1 | 1 |
| | | | HYM BB2 | 2 |
| | | | HYM CC | 1 |
| | | | HYM E | 3 |
| | | | HYM E2 | 1 |
| | | | HYM FF | 1 |
| | | | HYM G1 | 1 |
| | | | HYM G3 | 1 |
| | | | HYM GG | 1 |
| | | | HYM I | 3 |
| | | | HYM J | 2 |
| | | | HYM JJ | 1 |
| | | | HYM L | 1 |
| | | | HYM M | 1 |
| | | | HYM O | 2 |
| | | | HYM Q | 10 |
| | | | HYM Q2 | 1 |
| | | | HYM R | 2 |
| | | | HYM S | 3 |

Lushof

| | | | |
|-------------|------------|------------|----|
| | | HYM Z | 3 |
| Diptera | Pyrgotidae | Pyrgotid 1 | 51 |
| | | Pyrgotid 3 | 2 |
| | Tethinidae | DIP M | 2 |
| | unknown | DIP B1 | 13 |
| | | DIP CC2 | 1 |
| | | DIP F2 | 1 |
| | | DIP YY2 | 2 |
| Hymenoptera | Vespidae | VESP 3 | 2 |
| | unknown | HYM A | 1 |
| | | HYM AA | 1 |
| | | HYM B | 6 |
| | | HYM BB | 12 |
| | | HYM C | 3 |
| | | HYM E | 24 |
| | | HYM E2 | 22 |
| | | HYM E4 | 1 |
| | | HYM EU | 1 |
| | | HYM F | 18 |
| | | HYM G1 | 20 |
| | | HYM G2 | 1 |
| | | HYM G3 | 18 |
| | | HYM H | 1 |
| | | HYM HH | 2 |
| | | HYM I | 17 |
| | | HYM II | 3 |
| | | HYM J | 2 |
| | | HYM K | 2 |
| | | HYM L | 12 |
| | | HYM L2 | 13 |
| | | HYM LL | 3 |
| | | HYM M | 10 |
| | | HYM N | 3 |
| | | HYM O | 11 |
| | | HYM O2 | 6 |
| | | HYM OO | 4 |
| | | HYM P | 8 |
| | | HYM PP | 6 |
| | | HYM Q | 2 |
| | | HYM Q2 | 6 |
| | | HYM R | 8 |
| | | HYM S | 1 |
| | | HYM S2 | 2 |
| | | HYM UU | 2 |
| | | HYM V | 7 |
| | | HYM X | 1 |
| | | HYM X1 | 1 |
| | | HYM Y | 2 |

| | | | | |
|--------------------|---------------------|---------------|----------------------------|------|
| | | | HYM Z | 18 |
| | | | HYM ZZ | 2 |
| <i>Lydenburg</i> | Diptera | Simuliidae | DIP 15 | 1 |
| | | unknown | DIP 14 | 1 |
| | Hymenoptera | unknown | HYM 8 | 1 |
| | | | ICH 1 | 1 |
| | Phytophagous | | | |
| <i>Amsterdam</i> | Coleoptera | Chrysomelidae | COL A | 32 |
| | Diptera | Tephritidae | TEPH 2 | 1 |
| | | | TEPH A | 1 |
| | | unknown | DIP 6 | 5 |
| | Hemiptera | unknown | HEM B | 1 |
| | | | HEM E | 1 |
| | | | HEM F | 1 |
| | Hymenoptera | unknown | HEM C | 4 |
| | | | HEM D | 4 |
| | Thysanoptera | Thripidae | Thrip 1 | 6 |
| <i>Dullstroom</i> | Coleoptera | Chrysomelidae | COL G | 1 |
| <i>Gelukstroom</i> | Coleoptera | Coccinellidae | COL 3 | 3 |
| | | Curculionidae | Phlyctinus colossus type 1 | 6 |
| | | Silvanidae | Silvanid 1 | 3 |
| | Diptera | Tephritidae | TEPHRITID 6C | 1 |
| | | | TEPHRITID 9 | 1 |
| | Hemiptera | Aleyrodidae | Bemisia afer | 2577 |
| | | Aphididae | Aphid 1 | 2 |
| | | | Aphid 2 | 1 |
| | | Cicadellidae | Cicadellid 3 | 9 |
| | | | Cicadellid 4 | 11 |
| | | Lygaeidae | Lygaeid 1 | 1 |
| | | Margarodidae | MARGARODID 1 | 1 |
| | | Miridae | Mirid 1 | 37 |
| | | | Mirid 2 | 13 |
| | | | Mirid 3 | 9 |
| | | | Mirid 4 | 2 |
| | | | Miridjuv 1 | 10 |
| | | Pentatomidae | Pentatomid 1 | 1 |
| | | Psyllidae | Psyllidjuv 1 | 1 |
| | Thysanoptera | Thripidae | Thrip | 7 |
| | | | Thripjuv | 1 |
| | | Thripidae | Thrip | 1 |
| | | unknown | THYjuv | 2 |
| <i>Sonop</i> | Coleoptera | Chrysomelidae | Chrysomelid | 10 |
| | | | Chrysomelid 25 | 1 |
| | | Coccinellidae | COL 3 | 11 |
| | | Curculionidae | Curculionid 4 | 9 |
| | | | Curculionid 6 | 1 |
| | | | Curculionid 9 | 1 |
| | | Elateridae | Elaterid | 1 |

| | | | |
|------------|---------------|---------------|-----|
| | Tenebrionidae | COL 26 | 4 |
| | unknown | COL 12 | 1 |
| | | COL 16 | 4 |
| | | COL 24 | 1 |
| | | COL 9 | 13 |
| Dermaptera | unknown | Derma 1 | 1 |
| Diptera | Agromyzidae | DIP CC | 7 |
| Hemiptera | Aleyrodidae | Bemisia afer | 15 |
| | Aphididae | Aphid 1 | 2 |
| | | Aphid 2 | 56 |
| | | Aphid 3 | 1 |
| | | Aphid 4 | 1 |
| | Cicadellidae | Cicadellid 1 | 1 |
| | | Cicadellid 2 | 1 |
| | | Cicadellid 3 | 4 |
| | | Cicadellid 6 | 1 |
| | Coreidae | Coreid 1 | 1 |
| | Diaspididae | Scale 1 | 1 |
| | Lygaeidae | Lygaeid 1 | 14 |
| | Miridae | Mirid 1 | 585 |
| | | Mirid 10 | 1 |
| | | Mirid 11 | 1 |
| | | Mirid 12 | 3 |
| | | Mirid 13 | 2 |
| | | Mirid 14 | 28 |
| | | Mirid 15 | 1 |
| | | Mirid 2 | 68 |
| | | Mirid 3 | 5 |
| | | Mirid 4 | 16 |
| | | Mirid 7 | 1 |
| | | Mirid 8 | 3 |
| | | Mirid 9 | 4 |
| | | Miridjuv 2 | 4 |
| | | Miridjuv 3 | 28 |
| | | Miridjuv 4 | 2 |
| | | Miridjuv 5 | 7 |
| | Pentatomidae | Pentatomid 2 | 1 |
| | | Pentatomid 3 | 2 |
| | | Pentatomid 4 | 1 |
| | | Pentatomid 5 | 1 |
| | Psyllidae | Psyllid 2 | 1 |
| | | Psyllid 3 | 2 |
| | | Psyllid 6 | 2 |
| | Pyrrhocoridae | Pyrrhocorid 1 | 2 |
| | unknown | HEM 02 | 1 |
| | | HEM 03 | 1 |
| | | HEM 04 | 1 |
| | | HEM 05 | 2 |

Lushof

| | | | |
|--------------|---------------|----------------------------|-----|
| | | Hemjuv 1 | 2 |
| Lepidoptera | unknown | LEPjuv 2 | 1 |
| Orthoptera | Acrididae | ACRI 1 | 1 |
| | | ACRI 2 | 1 |
| | | ACRI 3 | 2 |
| | | ACRI LARVA | 1 |
| | Caelifera | CAEL 1 | 2 |
| | Tettigonidae | TET 1 | 1 |
| Thysanoptera | Thripidae | Thrip | 3 |
| | unknown | THY 01 | 2 |
| | | THY 02 | 1 |
| Coleoptera | Cantharidae | Cantharid | 1 |
| | Chrysomelidae | Chrysomelid | 102 |
| | | COL 33 | 1 |
| | | COL 34 | 1 |
| | | COL 37 | 1 |
| | Coccinellidae | COL 3 | 34 |
| | Curculionidae | Curculionid 11 | 1 |
| | | Curculionid 12 | 1 |
| | | Curculionid 4 | 2 |
| | | Curculionid 5 | 1 |
| | | Curculionid 6 | 1 |
| | | Curculionid 7 | 8 |
| | | Phlyctinus callosus | 4 |
| | | Phlyctinus colossus type 1 | 2 |
| | | Phlyctinus colossus type 2 | 3 |
| | Elateridae | Elaterid | 47 |
| | Melyridae | COL 35 | 53 |
| | Tenebrionidae | COL 26 | 2 |
| Dermaptera | unknown | Derma 2 | 1 |
| Diptera | Agromyzidae | DIP CC | 43 |
| | | DIP U2 | 55 |
| | | DIP X4 | 3 |
| | Tephritidae | TEPHRITID 4 | 2 |
| | | TEPHRITID 6 | 1 |
| | | TEPHRITID 6A | 1 |
| | | TEPHRITID 6B | 1 |
| | | TEPHRITID 6C | 1 |
| | | TEPHRITID 7 | 1 |
| Hemiptera | Aleyrodidae | Bemisia afer | 1 |
| | Aphididae | Aphid 1 | 1 |
| | | Aphid 2 | 20 |
| | Aphididae | Aphid 1 | 6 |
| | | Aphid 2 | 113 |
| | | Aphid 3 | 16 |
| | Cicadellidae | Cicadellid 2 | 22 |
| | | Cicadellid 3 | 23 |
| | | Cicadellid 4 | 26 |

| | | |
|---------------|-----------------|-----|
| | Cicadellid 5 | 3 |
| Cicadellidae | Cicadellid 1 | 1 |
| Coreidae | Coreid 1 | 19 |
| Coreidae | Coreid 1 | 10 |
| | Coreid 2 | 1 |
| Lygaeidae | Lygaeid 1 | 19 |
| | Lygaeid 2 | 7 |
| Miridae | HEM 06 | 1 |
| | Mirid 1 | 462 |
| | Mirid 11 | 1 |
| | Mirid 14 | 8 |
| | Mirid 16 | 1 |
| | Mirid 17 | 1 |
| | Mirid 18 | 1 |
| | Mirid 19 | 1 |
| | Mirid 2 | 40 |
| | Mirid 20 | 1 |
| | Mirid 3 | 54 |
| | Mirid 4 | 40 |
| | Mirid 8 | 3 |
| | Mirid 9 | 4 |
| | Miridjuv 3 | 1 |
| | Miridjuv 5 | 8 |
| Miridae | Mirid 1 | 38 |
| | Mirid 14 | 8 |
| | Mirid 2 | 6 |
| | Mirid 21 | 1 |
| | Mirid 22 | 1 |
| | Mirid 23 | 1 |
| | Mirid 3 | 58 |
| | Mirid 8 | 4 |
| | Mirid 9 | 1 |
| | Miridjuv 2 | 1 |
| | Miridjuv 6 | 13 |
| | Miridjuv 7 | 4 |
| Monophlebidae | Icerya purchasi | 6 |
| Pentatomidae | Pentatomid 2 | 2 |
| | Pentatomid 3 | 38 |
| | Pentatomid 4 | 1 |
| | Pentatomid 6 | 1 |
| Pentatomidae | Pentatomid 1 | 1 |
| | Pentatomid 2 | 1 |
| | Pentatomid 3 | 1 |
| Psyllidae | Psyllid 3 | 1 |
| Pyrrhocoridae | Pyrrhocorid 1 | 1 |
| unknown | HEM 06 | 1 |
| | Hemjuv 1 | 1 |
| | HEMjuv 2 | 2 |

| | | | | |
|--------------------|--------------|--------------|-----------|----|
| <i>Lydenburg</i> | Lepidoptera | unknown | LEPjuv 10 | 1 |
| | | | LEPjuv 12 | 1 |
| | | | LEPjuv 13 | 1 |
| | | | LEPjuv 14 | 1 |
| | | | LEPjuv 15 | 1 |
| | | | LEPjuv 17 | 1 |
| | | | LEPjuv 18 | 2 |
| | | | LEPjuv 2 | 9 |
| | | | LEPjuv 3 | 5 |
| | | | LEPjuv 6 | 1 |
| | | | LEPjuv 7 | 6 |
| | | | LEPjuv 9 | 4 |
| | Orthoptera | Acrididae | ACRI 3 | 1 |
| | | Gryllidae | GRY 1 | 8 |
| | | | GRY 2 | 2 |
| | | Tettigonidae | ENSJUV 1 | 1 |
| | | | TET 2 | 1 |
| | Thysanoptera | Thripidae | Thrip | 1 |
| | | unknown | THY 03 | 1 |
| | | | THY 04 | 1 |
| Hemiptera | Aphididae | Aphid 1 | 17 | |
| | Cicadellidae | CICAD 1 | 2 | |
| | | CICAD 2 | 1 | |
| | Psyllidae | PSY 1 | 1 | |
| | unknown | HEM G | 2 | |
| | | HEM H | 1 | |
| | | Hemjuv 1 | 4 | |
| Orthoptera | Gryllidae | GRY 1 | 2 | |
| Pollinators | | | | |
| <i>Amsterdam</i> | Coleoptera | unknown | COL F | 2 |
| | Diptera | unknown | DIP 10 | 4 |
| | | | DIP 5 | 1 |
| | | | DIP 7 | 14 |
| | | | TIP 1 | 1 |
| | | | TIP 2 | 1 |
| | | | TIP 3 | 1 |
| | Hymenoptera | unknown | HYM 3 | 2 |
| | | | HYM 6 | 1 |
| | Lepidoptera | unknown | LEP 1 | 2 |
| <i>Dullstroom</i> | | | LEP 2 | 1 |
| | | | LEP 3 | 1 |
| | Coleoptera | Nitidulidae | COL H | 1 |
| | Diptera | unknown | DIP 16 | 2 |
| | | | TIP 1 | 1 |
| | Hymenoptera | unknown | HYM 6 | 2 |
| | Lepidoptera | unknown | LEP 4 | 2 |
| | | LEP 5 | 1 | |
| | | LEP 6 | 2 | |

| | | | | | |
|--------------------|---------------|---------------|--------------|-----------|---|
| | | LEP 7 | 1 | | |
| <i>Gelukstroom</i> | Diptera | Calliphoridae | DIP F | 1 | |
| | | Chironomidae | CHIRON 2 | 102 | |
| | | Drosophilidae | DROS 1 | 8 | |
| | | | DROS 2 | 1 | |
| | | | DROS 3B | 1 | |
| | | Tipulidae | DIP AA | 13 | |
| | | | DIP LL | 4 | |
| | | unknown | DIP CC3 | 9 | |
| | | | DIP DD | 1 | |
| | | | DIP II | 1 | |
| | | | DIP LLL | 2 | |
| | | | DIP S | 7 | |
| | Hymenoptera | Apidae | APID 1 | 3 | |
| | | unknown | HYM W | 1 | |
| Lepidoptera | unknown | LEP 01 | 1 | | |
| | | LEP 03 | 1 | | |
| | | LEP 05 | 3 | | |
| | | LEP 07 | 1 | | |
| | | LEP 08 | 1 | | |
| | | LEP 09 | 1 | | |
| | | LEP 10 | 1 | | |
| | <i>Sonop</i> | Coleoptera | Nitidulidae | COL 22 | 4 |
| | | | | Nitidulid | 1 |
| | | | unknown | COL 6 | 1 |
| Diptera | | Chironomidae | CHIRON 1 | 3 | |
| | | | CHIRON 2 | 199 | |
| | | | CHIRON 2 | 12 | |
| | | | Chironomidae | 11 | |
| | | Culicidae | DIP G | 7 | |
| | | | DIP V | 6 | |
| | | Curtonidae | DIP W | 2 | |
| | Drosophilidae | DIP L | 6 | | |
| | | DROS 1 | 4 | | |
| | | DROS 2 | 6 | | |
| | | DROS 3 | 2 | | |
| | | DROS 4 | 124 | | |
| | Milichiidae | DIP Z | 3 | | |
| | Tipulidae | DIP AA | 15 | | |
| | | DIP LL | 4 | | |
| | unknown | DIP BB1 | 3 | | |
| | | DIP BB2 | 3 | | |
| | | DIP DD | 24 | | |
| | | DIP E1 | 1 | | |
| | | DIP EE | 10 | | |
| | | DIP II | 1 | | |
| | | DIP KK | 1 | | |
| | | DIP OO | 71 | | |

Lushof

| | | | |
|-------------|---------------|-----------|-----|
| | | DIP PP | 1 |
| | | DIP R | 2 |
| | | DIP RR | 1 |
| | | DIP S | 18 |
| | | DIP Y | 30 |
| Hymenoptera | unknown | HYM DD | 1 |
| | | HYM W1 | 1 |
| Lepidoptera | unknown | LEP 01 | 1 |
| | | LEP 03 | 7 |
| | | LEP 04 | 3 |
| | | LEP 05 | 5 |
| | | LEP 06 | 1 |
| | | LEPjuv 2 | 1 |
| Coleoptera | Mordellidae | COL 15 | 18 |
| | Nitidulidae | COL 22 | 9 |
| | | Nitidulid | 5 |
| Diptera | Calliphoridae | DIP F | 1 |
| | Chironomidae | CHIRON 1 | 12 |
| | | CHIRON 2 | 206 |
| | | CHIRON 3 | 14 |
| | | CHIRON 4 | 8 |
| | Conopidae | DIP D2 | 41 |
| | Culicidae | DIP G | 13 |
| | | DIP V | 1 |
| | Curtonidae | DIP W | 17 |
| | Drosophilidae | DIP L | 3 |
| | | DROS 1 | 4 |
| | | DROS 2 | 2 |
| | | DROS 3 | 5 |
| | | DROS 4 | 138 |
| | Tabanidae | Tabanid 5 | 2 |
| | Tipulidae | DIP AA | 38 |
| | | DIP AA1 | 1 |
| | | DIP LL | 28 |
| | unknown | DIP AAA | 9 |
| | | DIP BB1 | 1 |
| | | DIP DD | 63 |
| | | DIP DD2 | 7 |
| | | DIP F3 | 1 |
| | | DIP FFF | 4 |
| | | DIP II | 18 |
| | | DIP II2 | 1 |
| | | DIP K2 | 22 |
| | | DIP KK | 5 |
| | | DIP KK1 | 12 |
| | | DIP KK2 | 4 |
| | | DIP LLL | 4 |
| | | DIP OO | 31 |

Gelukstroom

| | | | |
|--------------|-----------------|------------------|----|
| | | SP 12 | 1 |
| | | SP 13 | 4 |
| | | SP 2 | 1 |
| | | SP 3 | 2 |
| | | SP 4 | 3 |
| | | SP 5 | 2 |
| | | SP 6 | 1 |
| | | SP 7 | 2 |
| | | SP 8 | 5 |
| | | SP 9 | 4 |
| Coleoptera | Coccinellidae | COCCI C | 1 |
| Hemiptera | Reduviidae | HEM I | 47 |
| Araneae | unknown | SP A | 4 |
| | | SP B | 22 |
| | | SP C | 4 |
| | | SP D | 2 |
| | | SP E | 1 |
| | | SP F | 2 |
| | | SP G | 7 |
| | | SP H | 1 |
| | | SP I | 1 |
| | | SP J | 3 |
| | | SP L | 2 |
| | | SP M | 1 |
| | | SP N | 1 |
| | | SP O | 1 |
| | | SP P | 1 |
| Coleoptera | Anobidae | Anobid 1 | 1 |
| | Anthicidae | Formicomus sp. 1 | 4 |
| | Coccinellidae | Coccinelid 1 | 3 |
| | | Coccinelid 2 | 4 |
| | | Coccinelid 3 | 1 |
| | | Coccinelid 4 | 1 |
| | unknown | COL 2 | 2 |
| | | COL 4 | 1 |
| | | COL 5 | 1 |
| | | COLjuv 1 | 2 |
| Diptera | Phoridae | DIP X | 5 |
| Hemiptera | Reduviidae | REDUVIID 1 | 6 |
| Mesostigmata | Phytoseiidae | Phytoseiidae | 13 |
| | unknown | Mesostigmata | 12 |
| Neuroptera | Chrysopidae | Chrysopid 1 | 37 |
| | | Chrysopidjuv | 2 |
| | Coniopterygidae | Coniopterygidjuv | 7 |
| | Hemorobiidae | Hemorobid 1 | 1 |
| | unknown | Neurojuv 1 | 1 |
| Prostigmata | Anystidae | Anystidae | 12 |
| | Bdellidae | Bdellidae | 1 |

Sonop

| | | | |
|--------------|----------------|-------------------|-----|
| | Stigmaeidae | Stigmaeidae | 1 |
| Thysanoptera | Aeolothripidae | Aeolothrip | 3 |
| Araneae | unknown | SP C1 | 1 |
| | | SP A | 56 |
| | | SP B | 29 |
| | | SP B1 | 3 |
| | | SP C | 28 |
| | | SP C1 | 1 |
| | | SP E | 1 |
| | | SP EE | 1 |
| | | SP FF | 59 |
| | | SP G | 58 |
| | | SP GG | 1 |
| | | SP J1 | 5 |
| | | SP J2 | 7 |
| | | SP JJ | 2 |
| | | SP KK | 5 |
| | | SP L | 2 |
| | | SP M | 105 |
| | | SP M1 | 14 |
| | | SP M2 | 1 |
| | | SP N | 1 |
| | | SP NN | 1 |
| | | SP P | 67 |
| | | SP P1 | 3 |
| | | SP P3 | 2 |
| | | SP PP | 4 |
| | | SP Q | 3 |
| | | SP Q1 | 3 |
| | | SP U | 1 |
| | | SP W | 3 |
| | | SP X | 29 |
| | | SP X | 3 |
| | | SP X1 | 16 |
| | | SP X2 | 3 |
| | | SP Y | 1 |
| Coleoptera | Anthicidae | Anthicid 3 | 6 |
| | | Formicomus sp. 1 | 9 |
| | | Formicomus sp. 2 | 16 |
| | Cantharidae | Cantharid | 1 |
| | Coccinellidae | Coccinellid 1 | 1 |
| | | Coccinellid 10 | 2 |
| | | Coccinellid 2 | 45 |
| | | Coccinellid 3 | 40 |
| | | Coccinellid 6 | 1 |
| | | Coccinellid 7 | 4 |
| | | Coccinellid larva | 7 |
| | | COL 11 | 110 |

Lushof

| | | | |
|--------------|----------------|---------------|-----|
| | Staphylinidae | Staphylinid | 1 |
| | unknown | COL 2 | 1 |
| Diptera | Phoridae | DIP X | 26 |
| | unknown | DIP BB | 16 |
| Hemiptera | Reduviidae | Reduviid 2 | 4 |
| | | Reduviid 3 | 6 |
| | | Reduviid 4 | 1 |
| Mantodea | Mantidae | MAN 01 | 24 |
| | | MAN 07 | 1 |
| | unknown | MAN 06 | 2 |
| Mesostigmata | Macrochelidae | Macrochelidae | 1 |
| | Phytoseiidae | Phytoseiidae | 1 |
| Neuroptera | Chrysopidae | Chrysopid 1 | 16 |
| | Hemorobiidae | BEROTH 1 | 3 |
| | unknown | Neurojuv 2 | 1 |
| Prostigmata | Anystidae | Anystidae | 24 |
| | Erythraeidae | Erythraeidae | 4 |
| Thysanoptera | Aeolothripidae | Aeolothrip | 4 |
| Araneae | unknown | SP B1 | 1 |
| | | SP A | 10 |
| | | SP AA | 5 |
| | | SP B | 14 |
| | | SP C | 135 |
| | | SP C1 | 18 |
| | | SP C2 | 161 |
| | | SP C3 | 11 |
| | | SP C7 | 1 |
| | | SP CC | 4 |
| | | SP D | 1 |
| | | SP E | 25 |
| | | SP F | 1 |
| | | SP FF | 2 |
| | | SP G | 19 |
| | | SP GG | 1 |
| | | SP I | 8 |
| | | SP I1 | 2 |
| | | SP J1 | 3 |
| | | SP J2 | 7 |
| | | SP JJ | 12 |
| | | SP K | 1 |
| | | SP KK | 12 |
| | | SP L | 6 |
| | | SP L1 | 1 |
| | | SP M | 118 |
| | | SP MM | 3 |
| | | SP OO | 1 |
| | | SP P | 5 |
| | | SP P3 | 6 |

| | | | |
|--------------|----------------|--------------------|----|
| | | SP Q | 3 |
| | | SP Q1 | 3 |
| | | SP RR | 1 |
| | | SP S | 8 |
| | | SP SS | 2 |
| | | SP TT | 1 |
| | | SP W | 3 |
| | | SP X | 3 |
| | | SP X1 | 18 |
| Coleoptera | Anthicidae | Anthicid 3 | 2 |
| | | Notoxus cucullatus | 1 |
| | | Formicomus sp. 1 | 2 |
| | | Formicomus sp. 2 | 17 |
| | Anthicidae | Notoxus cucullatus | 3 |
| | Carabidae | COL 36 | 2 |
| | Claridae | COL 32 | 1 |
| | Coccinellidae | Coccinellid 10 | 7 |
| | | Coccinellid 12 | 55 |
| | | Coccinellid 5 | 60 |
| | | Coccinellid 6 | 2 |
| | | Coccinellid 7 | 3 |
| | | Coccinellid 9 | 6 |
| | | Coccinellid larva | 6 |
| | Staphylinidae | Staphylinid | 14 |
| Diptera | Empididae | DIP X2 | 22 |
| | Mycetophilidae | DIP BFG | 19 |
| | Phoridae | DIP X | 12 |
| Hemiptera | Reduviidae | Reduviid 1B | 2 |
| | | Reduviid 2 | 1 |
| | | Reduviid 3 | 3 |
| | Reduviidae | Reduviid 2 | 1 |
| | | Reduviid 4 | 1 |
| | | Reduviid 5 | 1 |
| | Scutelleridae | Scutellerid 1 | 12 |
| | Tingidae | Tingid 2 | 1 |
| | Tingidae | Tingid 2 | 1 |
| Mantodea | Mantidae | MAN 01 | 32 |
| | | MAN 04 | 13 |
| | | MAN 07 | 2 |
| | unknown | MAN 06 | 8 |
| Mesostigmata | Phytoseiidae | Phytoseiidae | 4 |
| Neuroptera | Chrysopidae | Chrysopid 1 | 50 |
| | Hemerobiidae | BEROTH 2 | 1 |
| | | BEROTH 3 | 1 |
| | Myrmeleontidae | myrm larvae | 1 |
| | unknown | Neurojuv 3 | 1 |
| Prostigmata | Anystidae | Anystidae | 28 |
| | Bdellidae | Bdellidae | 1 |

| | | | | |
|-------------------|--------------|----------------|----------------|----|
| | | Caligonellidae | Caligonellidae | 1 |
| | | Erythraeidae | Erythraeidae | 3 |
| Lydenburg | Thysanoptera | Aeolothripidae | Aeolothrip | 1 |
| | Araneae | unknown | SP 1 | 11 |
| | | | SP 10 | 1 |
| | | | SP 12 | 2 |
| | | | SP 6 | 1 |
| Scavengers | | | | |
| Amsterdam | Coleoptera | Latridiidae | COL B | 5 |
| | | | COL C | 3 |
| | | | COL C | 9 |
| | Diptera | unknown | DIP 11 | 3 |
| | | DIP 12 | 3 | |
| Dullstroom | Coleoptera | Latridiidae | COL C | 1 |
| Lydenburg | Coleoptera | Latridiidae | COL C | 12 |

Appendix 24: Species list of various feeding guilds collected using clipping method at six farms

| Farm | Order | Family | Species | Individuals collected | |
|---------------------|----------------|----------------|-------------------|------------------------------|---|
| Fungivores | | | | | |
| Gelukstroom | Prostigmata | Tarsonemidae | Tarsonemidae | 29 | |
| | | | Tarsonemus sp. | 5 | |
| | | | Tarsonemus waitei | 8 | |
| | | Tarsonemidae | Tarsonemidae | 1 | |
| | | Thysanoptera | Phlaeothripidae | Phlaeothrips sp. | 1 |
| Sonop | Prostigmata | Tarsonemidae | Tarsonemidae | 5 | |
| | | | Tarsonemus waitei | 4 | |
| | Sarcoptiformes | Acaridae | Acaridae | 1 | |
| | | | Caloglyphus sp. | 1 | |
| Lushof | Prostigmata | Tarsonemidae | Tarsonemidae | 9 | |
| | | | Tarsonemus waitei | 7 | |
| | | | Tarsonemidae | Tarsonemidae | 2 |
| Phytophagous | | | | | |
| Gelukstroom | Hemiptera | Aleyrodidae | Bemisia afer | 5 | |
| | | Coccidae | Coccidae | 2 | |
| | | Diaspididae | Parlatonia sp. | 2 | |
| | | Monophlebidae | Icerya purchasi | 1 | |
| | | | Icerya sp. | 4 | |
| | | Pseudococcidae | Pseudococcidae | 2 | |
| | | Prostigmata | Tenuipalpidae | Tenuipalpidae | 1 |
| | | Thysanoptera | Thripidae | Scirtothrips aurantii | 1 |
| | | | | Thrips fumosus | 1 |
| | | | unknown | THYjuv | 4 |
| Sonop | Hemiptera | Diaspididae | Diaspididae | 2 | |

| | | | | |
|--------------------|--------------------|-----------------|------------------------------|-----|
| | | Monophlebidae | Icerya sp. | 1 |
| | Prostigmata | Tenuipalpidae | Brevipalpus californus sp. B | 2 |
| | | | Brevipalpus lewisi | 1 |
| | | | Brevipalpus sp. | 1 |
| | | | Tenuipalpidae | 1 |
| | | Tetranychidae | Tetranychidae | 2 |
| | Thysanoptera | Thripidae | Frankliniella occidentalis | 1 |
| | | | Scirtothrips aurantii | 6 |
| | | | Thrip | 4 |
| | | | Thrips tabaci | 3 |
| | | Thripidae | Scirtothrips aurantii | 4 |
| | | unknown | THYjuv | 1 |
| <i>Lushof</i> | Hemiptera | Aphididae | Aphid | 3 |
| | | | Aphis sp. | 4 |
| | | Diaspididae | Diaspididae | 6 |
| | | | Parlatonia sp. | 1 |
| | | Pseudococcidae | Hemiberlesia sp. | 1 |
| | | | Pseudococcidae | 2 |
| | Prostigmata | Tetranychidae | Tetranychus sp. | 1 |
| | Thysanoptera | Thripidae | Heliothrips haemorrhoidalis | 2 |
| | | | Scirtothrips aurantii | 13 |
| | | | Scirtothrips sp. | 1 |
| | | | Thrips tabaci | 1 |
| | | unknown | THYjuv | 32 |
| | | | | |
| | Poll nators | | | |
| <i>Lushof</i> | Lepidoptera | Arctiidae | LEP 14 | 1 |
| | | | | |
| | Predators | | | |
| <i>Gelukstroom</i> | Mesostigmata | Phytoseiidae | Phytoseiidae | 59 |
| | | unknown | Mesostigmata | 23 |
| | Neuroptera | Coniopterygidae | Coniopterygidjuv | 10 |
| | Prostigmata | Anystidae | Anystidae | 4 |
| | | Bdellidae | Bdellidae | 1 |
| | | Iolinidae | Iolinidae | 13 |
| | | Stigmaeidae | Stigmaeidae | 10 |
| | | Trombiculoidea | Trombiculoidea | 1 |
| <i>Sonop</i> | Araneae | Aranaeidae | Aranaeidae | 1 |
| | | Theridiidae | Theridiidae | 2 |
| | | unknown | Araneae | 3 |
| | Coleoptera | Coccinellidae | Coccinellid larva | 1 |
| | Mesostigmata | Phytoseiidae | Phyto immature | 1 |
| | | | Phytoseiidae | 51 |
| | | unknown | Mesostigmata | 5 |
| | Prostigmata | Iolinidae | Iolinidae | 327 |
| | | | Agistemus collyerae | 1 |
| <i>Lushof</i> | Araneae | Eutichiridae | Cheiracanthium sp. | 2 |
| | | Linyphiidae | Linyphiidae | 1 |
| | | unknown | Araneae | 1 |
| | Coleoptera | Coccinellidae | Coccinellid larva | 2 |

| | | | | |
|--------------------|----------------|---------------------|------------------|-----|
| | | Hippodama variegata | 1 | |
| Mesostigmata | Phytoseiidae | Phytoseiidae | 19 | |
| | unknown | Mesostigmata | 20 | |
| Prostigmata | Iolinidae | Iolinidae | 16 | |
| | Stigmaeidae | Stigmaeidae | 1 | |
| Sca engers | | | | |
| <i>Gelukstroom</i> | Prostigmata | Triophtydeidae | Triophtydeidae | 2 |
| | | Tydeidae | Brachytydeus sp. | 1 |
| | | | Tydeidae | 89 |
| | | | Tydeus grabouwi | 267 |
| | | | Tydeus sp. | 86 |
| | | Tydoidea | Tydoidea | 8 |
| | Sarcoptiformes | Oribatida | Oribatida | 1 |
| <i>Sonop</i> | Prostigmata | Triophtydeidae | Triophtydeidae | 3 |
| | | Tydeidae | Brachytydeus sp. | 13 |
| | | | Tydeidae | 61 |
| | | | Tydeus grabouwi | 346 |
| | | | Tydeus sp. | 123 |
| | | | Tydeus sp. A | 39 |
| | | Tydeidae | Tydeidae | 2 |
| | | | Tydeus grabouwi | 14 |
| | | | Tydeus sp. | 1 |
| | | Tydoidea | Tydoidea | 17 |
| Sarcoptiformes | Oribatida | Oribatida | 2 | |
| <i>Lushof</i> | Prostigmata | Tydeidae | Brachytydeus sp. | 1 |
| | | | Tydeidae | 70 |
| | | | Tydeus sp. | 40 |
| | | | Tydeus sp. A | 32 |
| | | | Tydeus sp. B | 15 |
| | | Tydoidea | Tydoidea | 6 |
| | | Tydoidea | Tydeidae | 2 |