

Department of Environmental Biology

**Oil mallee plantings and arthropod biodiversity in the Western
Australian wheatbelt: effects of host species, nutrition, and leaf
chemistry**

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Doctor of Philosophy

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Declaration

To the best of my knowledge and belief this thesis contains no material previously published by any other person except where due acknowledgment has been made.

This thesis contains no material which has been accepted for the award of any other degree or diploma in any university.

Signature:

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Figure 52: Bubble plot of leaf essential oils. Essential oil levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of leaf essential oils are represented by coloured bubbles a) cineole, b) pinene. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined. _____ 127

Figure 53: Bubble plot of sideroxylonals. Sideroxylonal levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of sideroxylonals are represented by coloured bubbles. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined. _____ 129

ABSTRACT

Since European settlement, around 93% of the Western Australian wheatbelt has been cleared for agriculture, leading to a range of environmental problems, including erosion, salinity, and loss of biodiversity. Recently, oil mallees have been developed for use in the reduction and prevention of salinity, and in the production of oil, activated carbon, and electricity. While some work has been done on aspects of mallee ecology in order to maximize productivity, little is known about their usefulness as a source of biodiversity in natural and agricultural systems.

This study concentrates on the canopy arthropod fauna of the mallees. While canopy research involving arthropods is common in tropical systems, there is limited information on temperate systems and still less relating to agro-forestry or conservation plantings in agricultural areas.

Sampling was conducted in alleys of oil mallee vegetation and in remnant vegetation in the wheatbelt of Western Australia. Three mallee species: *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*, and two native remnant species: *Eucalyptus wandoo* subsp. *wandoo* and *E. astringens* were used in the study. Trees were sampled for arthropods by canopy knockdown spraying in October 2005 and May 2006. Samples were sorted to the ordinal level in the laboratory. Coleoptera (beetle) specimens were identified to the species level. Leaf and soil samples were taken at each site and from each tree species in order to determine the levels of nutrition available to herbivorous arthropods. Leaves were also collected for terpenoid (essential oil) extraction and formylated phloroglucinol (sideroxydonal) analysis to determine the influence of leaf chemistry. Data were tested for homogeneity of variance and transformations were done where necessary. A range of statistical analyses including, analyses of variance, LSDs, coefficients of correlation, MDS ordinations, ANOSIM analysis, and the BEST procedure, were conducted on the data collected.

It was determined that, for this study, chemical knockdown would be an ideal method for sampling such arthropods. A preliminary study examined the effect of repeated sampling of the same tree on canopy arthropod assemblages and found there was no effect of re-sampling on the ordinal level richness or total abundance of arthropods collected at the second sampling, six months later. As a consequence, we can be reasonably confident that the results in other sections of the thesis have not been confounded by the need to re-sample the same trees.

The effect of oil mallee host species on canopy arthropod assemblages was examined. It was found that while there were minor differences in the presence or absence of some of the leaf blemishes recorded between species, with leaf folding being more prevalent on *E. kochii* and psyllids more common on *E. loxophleba*, there was no significant difference between the species in terms of ordinal richness or total abundance of arthropods. As there were very few differences observed between the three mallee species, it made it relatively simple to compare mallees generally with remnant vegetation. This was important, as mallees were treated as a single entity being compared to the two remnant species in other parts of the thesis.

The ordinal richness and total abundance of canopy arthropods in two types of eucalypt vegetation, woodland eucalypts in remnant vegetation and mallee eucalypts in farm alley plantings was compared. Intuitively, we would expect native remnant vegetation to support a greater diversity of arthropods than any planted vegetation, simply by virtue of the native vegetation being in place for a longer period of time. The results of this study however, did not support this view. Arthropod richness and abundance were not significantly different between the tree species. Leaf blemishes and their associated sedentary arthropods also showed no significant differences between the species and overall very few differences between the mallee and remnant vegetation types were evident.

The influence of major soil and leaf nutrients on arthropod assemblages, both in natural and planted eucalypt stands, were explored. It appears that arthropod abundance, in particular, is related to soil and leaf nutrient levels. There was, however, only limited

evidence of increased arthropod ordinal richness in response to greater nutrient levels. Generally, high nutrient levels tended, instead, to reduce arthropod ordinal richness. Of the soil nutrients, phosphorus was the most influential, with high levels of phosphorus tending to relate to higher arthropod abundances. For leaf nutrients, phosphorus and nitrogen were important, with high levels of phosphorus being associated with lower ordinal richness, while high levels of leaf nitrogen were related to higher arthropod abundances. As high soil phosphorus and high leaf nitrogen tended to occur together, it is difficult to say whether one or the other is responsible for increases in arthropod abundance, though intuitively one would suggest that good soil nutrition led to enhanced plant quality, which in turn increased arthropod abundance.

The role of leaf essential oils and other secondary plant compounds in determining arthropod abundance and ordinal richness was also examined. A number of secondary plant compounds were present in the host tree species tested, and there were wide variations in the levels of these compounds between the species tested. A range of relationships, including deterrent and attractant effects, were observed between arthropod assemblages and the various secondary plant compounds. Generally speaking, mallee species had high cineole and low pinene levels and remnant species had the opposite. Sideroxylonals showed no such pattern with vegetation type, being high in *E. loxophleba* and absent in the other mallee species. In terms of their influence on arthropods, the compounds varied in their effects. Pinene had a generally negative effect, while cineole had a generally positive one. Sideroxylonal, however, was more complicated in its effects as it had a negative effect on ordinal richness, but was positively correlated with Hemiptera numbers. This suggests that it is highly unlikely that any one of these compounds can explain the pattern of arthropod assemblages observed in isolation. It seems more probable that complex interactions between these chemicals cause changes in nutritional quality and palatability of foliage, influencing the feeding behaviour, development, distribution and abundance of herbivores, in turn affecting predator densities and feeding behaviour.

The ways in which arthropod biodiversity may be influenced by the factors of host tree species, soil and plant nutrition and leaf chemistry were examined in more detail using

the order Coleoptera as an example. Season of sampling was found to have an influence, with both beetle richness and abundance being higher at the first than the second sampling. Site, on the other hand, had very little influence. Of all the tree species, *E. polybractea* had both the highest beetle species richness, and high levels of beetle abundance. Strong similarities were apparent between beetle assemblages resident on the same tree species. No influence of soil or leaf nutrients on either beetle richness or abundance was observed. However, beetle abundance was found to have a negative relationship with leaf pinene. Cineole levels were lower in the remnant species (which tended to have similar assemblages), while pinene was high in *E. loxophleba* subsp. *lissophloia* and the remnant species. The other major leaf chemical examined, sideroxylonal had no significant impact on beetle richness or abundance. Statistical analysis selected cineole as the single factor best explaining the pattern of beetle assemblages observed, though this result should be treated with caution due to possible confounding of the results as a consequence of interaction between the factors.

The broad aim of this research was to determine whether oil mallee plantings enhance arthropod biodiversity in agricultural landscapes. The results of this thesis show that oil mallees do support high levels of arthropod biodiversity. Overall, the mallees had a level of diversity, not dissimilar to that of high quality remnant vegetation. When planted in alleys across agricultural fields, they represent a significant change in the vegetative and architectural diversity of the landscape, and can have a positive influence on the environment by supporting beneficial arthropods and other native animals, reducing dryland salinity, and improving the aesthetics of the wheatbelt.

Aside from their environmental credentials, oil mallees also provide the potential for farmers to make an income from something designed to benefit the environment. If oil mallee farming can be developed appropriately, it has the potential to benefit not only farmers and the environment, but the community of Western Australia as a whole.

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CHAPTER 1

GENERAL INTRODUCTION

BACKGROUND

Introduction

Since European settlement in 1829, around 93% of the Western Australian wheatbelt has been cleared for agriculture (Saunders and Curry 1990). This has led to a range of environmental problems including erosion, salinity, and loss of biodiversity. According to the Commonwealth government's (1996) *Australia: State of the Environment Report* "two of the six major environmental problems affecting Australia are loss of biodiversity and land degradation resulting from over clearing of native vegetation".

Biodiversity

According to the Commonwealth Biological Diversity Advisory Committee (1993), biological diversity is the sum of all genetic, species and ecosystem diversity in a given area. It is generally agreed by researchers, government and the public alike, that some form of biodiversity conservation is needed, though how to achieve this is the subject of much debate. Bangert *et al.* (2005) challenges traditional notions of conservation, suggesting that the preservation of selected dominant or keystone species is inadequate. To truly conserve diversity, Bangert *et al.* (2005) propose that we need to protect habitat and genetic diversity, including that of hybrid species.

Wilson (1988) suggests that "each species is unique and intrinsically valuable" and that while we need to preserve biodiversity, it is also important that we view it as a "global resource". The Commonwealth Biological Diversity Advisory Committee (1993) agrees in its report on biodiversity and its value when it stated that "five thousand plant species have been used as food by humans, but less than twenty now feed the world's population

and just three or four carbohydrate crops are staples for the vast majority”. Wilson (1988) also points out that while we currently exploit less than 1% of living species to provide our food, there are over 75,000 edible plants in the world.

Apart from problems relating to how we use diversity, one of the major difficulties with the conservation of biodiversity is the fact that we do not really know the extent of biodiversity. Wilson (1988) says that currently there are approximately 1.4 million species of all kinds of living organisms that have been identified and that this is far short of his estimate of a total of 5 to 30 million species on the earth. This suggests that it is impossible to know the numbers of species facing extinction since we have no real idea how many species currently exist (Wilson 1988).

Value of Biodiversity

Biodiversity conservation is often seen as ‘a nice thing to do’ but is perceived to lack the urgency and importance to gain significant public support. However, the value of biodiversity should not be dismissed. The Commonwealth Biological Diversity Advisory Committee (1993) advises that biodiversity is important in the provision of a range of benefits including:

- ecosystem services, such as the protection of water resources, soil formation and nutrient cycling;
- biological resources, such as food, medicinal resources and wood products; and
- social benefits, such as recreation, tourism and cultural resources (e.g. for aboriginal people).

According to Hobbs *et al.* (2003), one of the basic principles of biodiversity is the idea that all diversity is good and that a variety of species and an array of habitats would create a greater overall diversity than just a single ecosystem, regardless of how valuable that ecosystem might appear. For example, Wilson (1988) points out that the species diversity of tropical rainforests ‘borders on the legendary’ but does this mean that they should be preserved at the expense of all other ecosystems?

In Western Australia, the majority of threatened plant taxa (214 out of 351) occur in the mid-west, wheatbelt and south coastal regions, the same regions that have been extensively cleared for agriculture (Atkins 2003). While in itself this could have major implications for plant biodiversity, we must also consider that most insect herbivores are relatively specific feeders which are restricted to particular species, genera or families of plants (Bernays and Graham 1988). Arthropods are also important contributors to ecosystem function and are essential components of several vertebrate diets (Hobbs *et al.* 2003). Thus, the loss of a single plant species could affect the diversity of both invertebrate and vertebrate animals, and ultimately entire ecosystems, through a series of flow-on effects. This is supported by Majer *et al.* (1999), who claim that due to high levels of insectivory amongst native vertebrates, any changes to the composition and abundance of arthropods could impact significantly not only on the plants and arthropods they feed on, but also on the vertebrates that prey on them.

Arthropod Diversity

Australia is one of only 12 countries worldwide that together account for 75 percent of the total biodiversity on the planet (McNeely *et al.* 1992). This, combined with Erwin's (1982) estimate of 30 million arthropod species worldwide, suggests that estimates of 140000 insect species for Australia (Nielsen and West 1994) are excessively low. This is supported by higher than expected diversity collected in recent studies from eucalypts (Majer *et al.* 2000), and by a study conducted by Majer *et al.* (1994) which came to the conclusion that the 'number of Australian insect species has been grossly underestimated'.

According to Ozanne (2005), around 50 percent of forest insect species can be found in the canopy, with up to 13 percent being true canopy specialists. The current study will focus on canopy arthropods in oil mallee eucalypts and, as indicated by Majer *et al.* (1994), most canopy arthropod studies have been conducted in the tropics, with scant attention being paid to temperate ecosystems. Springett (1978) suggested that the normal insect consumption of foliage in eucalypt forests is more severe than elsewhere. Yet according to Majer *et al.* (1994), we still have a very limited knowledge of eucalypt

canopy arthropods, aside from groups considered to be forestry pests. Plantations are generally expected to have poorer insect diversity than remnants and native forests. Work by Cunningham *et al.* (2005) supports this, although they found that plantations of blue gums shared many of their species with the remnant vegetation. In fact the fauna of plantations could be regarded as a subset of that found in the natural vegetation.

Diversity is most commonly measured in terms of species richness and species abundance. The Commonwealth Biological Diversity Advisory Committee (1993) defines species richness as ‘the number of species in a defined area’ and species abundance as ‘the relative number of individuals of different species’ in a given area. Southwood *et al.* (1982) agrees, advising that the number of species present is the main measure of a fauna’s diversity.

The current study uses canopy knockdown, which samples a considerable proportion of the diversity present. According to Ozanne (2005), because of the huge diversity collected by this method, a single sampling event of this kind can yield samples that take substantial inputs of time merely to clean out debris and sort to order. Work by Majer *et al.* (1994) showed a strong correlation between ordinal and species profiles, indicating that the former provides some reflection of the species richness of a sample or a site. This provides some justification for the majority of the samples used in this study being identified only to the ordinal level.

Farm Forestry and Biodiversity

Farm forestry is rapidly expanding, especially in southern Australia (Figure 1), and is seen by many as an opportunity to develop profitable timber and other products, while at the same time addressing environmental problems (Hobbs *et al.* 2003). While biodiversity benefits are touted as stemming from farm forestry, Hobbs *et al.* (2003) assert that little research has been done to substantiate these claims and little advice or support is available for those who wish to improve the biodiversity value of their farm plantings. Dames and Moore (1999) agree, suggesting it is generally assumed that farm

forestry improves biodiversity on agricultural land merely because trees are planted where crops once stood. They also argue that some aspects of farm forestry are not beneficial to the environment or biodiversity, especially when profitability is at stake. In order to truly integrate farm forestry and biodiversity, Dames and Moore (1999) suggest that there must be some flexibility, as many of the objectives of the two activities are likely to be antagonistic. For example, chemical sprays to remove insects and increase productivity would be detrimental to biodiversity through killing non target organisms, some of which are potential food items for birds and lizards.



Figure 1: A southern Australian Tasmanian blue gum plantation.

In Western Australia, one of the major environmental issues is dryland salinity (Ferdowsian *et al.* 1996), and as this problem continues to grow, the need to take action becomes more urgent. While some work has been done to show the benefits of oil mallees and other agro-forestry operations in reducing the effects of salinity, there has been limited research into the biodiversity benefits of farm forestry. In general, the focus has been on profitability rather than environmental outcomes (Hobbs *et al.* 2003). This is understandable as, farm forestry requires the financial input of farmers, many of whom are already struggling with declining terms of trade. Simply put, for any large scale

investment by farmers to occur in environmentally based farming systems, such as agroforestry, the new system must at least match the profitability of existing systems (Bartle *et al.* 2007).

Oil Mallees

While eucalypts are commercially grown across the world, their natural distribution is almost totally restricted to Australia, with a few tropical exceptions to the north (Williams and Brooker 1997). Eucalypts take three main growth forms, ‘forest’ trees, ‘woodland’ trees and ‘mallees’. Mallees are small trees, usually less than 10 m tall, deep-rooted, multi-stemmed from the base, and are common to low rainfall, arid and semi-arid regions of Australia (Brooker and Kleinig 1990).

One of the commercial attractions of oil mallees is their ability to quickly develop a substantial lignotuber, allowing trees to resprout repeatedly after canopies are harvested or damaged by fire (Noble 2001, Wildy and Pate 2002). This feature makes oil mallees ideal for short-rotation coppice cropping; where trees are established, entire canopies are mechanically harvested, the trees are left to resprout and within only a few years the trees are ready to be harvested again (Davis 2002).

Historically, oil mallees have been grown for oil and biomass (Bartle and Shea 2002). The uncommonly high concentrations of oil, in particular cineole, which could be extracted from oil mallee leaves, have been one of their main attractions (Bartle *et al.* 1996). Eucalyptus oil with high cineole content already has markets in the pharmaceutical trade (Barton *et al.* 1989), while other uses for cineole continue to be found. For example, it has been discovered that the addition of small quantities of cineole can have a stabilizing effect on petrol-ethanol fuel mixes (Tjandra 1986).

In Western Australian, however, salinity has been one of the major drivers for the development of the oil mallee industry. Bartle and Shea (2002) suggest that, in the long term, commercial perennial crops, whether pastures, cereals or trees, will be the only effective way to reduce groundwater recharge and prevent salinity impacts on a wide

scale. The lack of suitable options for use in low rainfall regions, combined with the salinity control potential of deep-rooted mallee species, plus its long history of cultivation, made mallees the obvious choice for farm forestry development in the wheatbelt of Western Australia (Bartle and Shea 2002). For these reasons, the use of oil mallees in short-cycle coppice crops, which can be harvested regularly without the need for replanting, is showing promise as one of the best options for profitable salinity control in the Western Australian wheatbelt (Bartle *et al.* 2007).

The timing and location of this study is particularly opportune, as the oil mallee industry, while still in its infancy, is growing rapidly. As of 2003, ten thousand hectares of the Western Australian wheatbelt had been planted to oil mallees, with most mallee plantings configured as narrow belts of trees through wider alleys of annual crops or pastures (Figure 2), and this area continues to expand (Enecon Pty Ltd 2001).



Figure 2: Oil mallee alley in WA wheatbelt.

While salinity is one of the driving factors behind the growth of the oil mallee industry in Western Australia, profitability remains the key. The potential to produce bioenergy, eucalyptus oil and activated carbon from oil mallees in a single process is one

development which has been eagerly anticipated (Fung *et al.* 2002, Enecon Pty Ltd 2001). As interest from energy companies, investors and farmers alike begins to grow, and with testing of a demonstration processing facility at Narrogin designed to produce oil, activated carbon and electricity concurrently underway (Figure 3) (Bartle and Shea 2002), this is an ideal time to further explore the potential benefits of these plants.



Figure 3: Demonstration processing facility at Narrogin.

AIM AND OBJECTIVES

The broad aim of this research was to determine the habitat value of oil mallee plantings in terms of canopy arthropod biodiversity in agricultural landscapes. In order to achieve this aim, a series of supporting objectives were formulated. These objectives are stated below as a set of questions which must be answered in order to accomplish the stated aim:

- Does the canopy-dwelling arthropod assemblage vary between oil mallee species?

- Are canopy-dwelling arthropod assemblages that are present on planted oil mallees similar to those found on remnant eucalypt vegetation?
- Do soil and leaf nutrient levels influence canopy-dwelling arthropod assemblages?
- Do leaf essential oils and secondary plant compounds influence canopy-dwelling arthropod assemblages?

The ways in which the answers to these questions might influence the arthropod biodiversity of planted and remnant mallee trees are shown schematically in Figure 4.

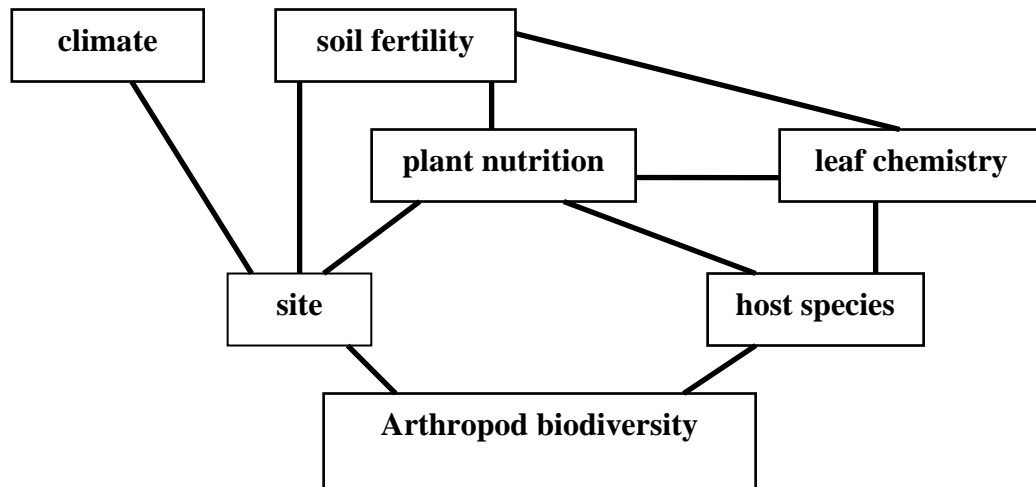


Figure 4: Schematic diagram showing how the various components of the research interact to contribute to arthropod biodiversity.

This research complements a wider study of biodiversity in the wheatbelt currently being conducted by CSIRO Sustainable Ecosystems. If it can be shown that biodiversity in agricultural landscapes is improved through planting oil mallees, it will provide further impetus for their establishment in farming systems. This could prove important in the fight against dryland salinity as, while oil mallees are already known to help reduce salinity in farming systems, information on their environmental and financial benefits is limited.

SIGNIFICANCE

The use of oil mallees in the reduction and prevention of salinity, and in the production of oil, activated carbon, and electricity, is a relatively new practice. While some work has been done on aspects of mallee ecology in order to maximize productivity, little is known about their usefulness as a source of biodiversity in natural and agricultural systems (Bartle and Shea 2002). For this reason, biodiversity studies involving mallees will add significant data in what is a new area of study.

This study will concentrate on the canopy arthropod fauna of the mallees. While canopy research involving arthropods is common in tropical systems, there is limited information on temperate systems and still less relating to agro-forestry or conservation plantings in agricultural areas. Hence, this work will fill a void in the scientific literature by using commonly used techniques in a different environment.

This study comes at an opportune time for the Western Australian oil mallee industry as, while growing rapidly, it is still in its early development. This means that results from this work will be available early enough to assist in the decision making processes of a large proportion of growers and investors, and will provide baseline data to support future work on oil mallees by other researchers.

In summary, this project is ideally situated to explore a gap in the literature created by the lack of research on arthropods associated with eucalypt species, as well as the limited data on oil mallee and other farm based forestry effects on biodiversity. This work also addresses the general lack of knowledge about how nutrition, plant host species and leaf chemistry interact to determine the abundance and richness of arthropod assemblages.

THESIS STRUCTURE

The arrangement of chapters is discussed here and the ways in which they interconnect are shown diagrammatically in Figure 5. Chapter 2 gives a general description of the study areas and overall experimental design used. Chapters 3 to 8 describe the results of field studies and present the core of new findings for this thesis. In Chapter 3, a review of arthropod sampling methods is presented and the advantages and disadvantages of the major method selected, chemical knockdown, are explained. This chapter also describes a preliminary study conducted to determine whether repeated sampling has an effect on canopy arthropods. An in-depth comparison of three oil mallee species in terms of their canopy-dwelling arthropod assemblages is conducted in Chapter 4. Chapter 5 compares the arthropod biodiversity in planted and remnant eucalypt vegetation. An exploration of the effects of soil and leaf nutrition on canopy-dwelling arthropod assemblages is detailed in Chapter 6. Chapter 7 examines the influence of leaf essential oils and secondary plant compounds on canopy-dwelling arthropod assemblages. Chapter 8 describes a case study involving the order Coleoptera, which brings together the various aspects influencing arthropod diversity in tree canopies, plus species specific data. The final section of the thesis provides some general discussion, conclusions and recommendations for the future uses of oil mallees in biodiversity programs.

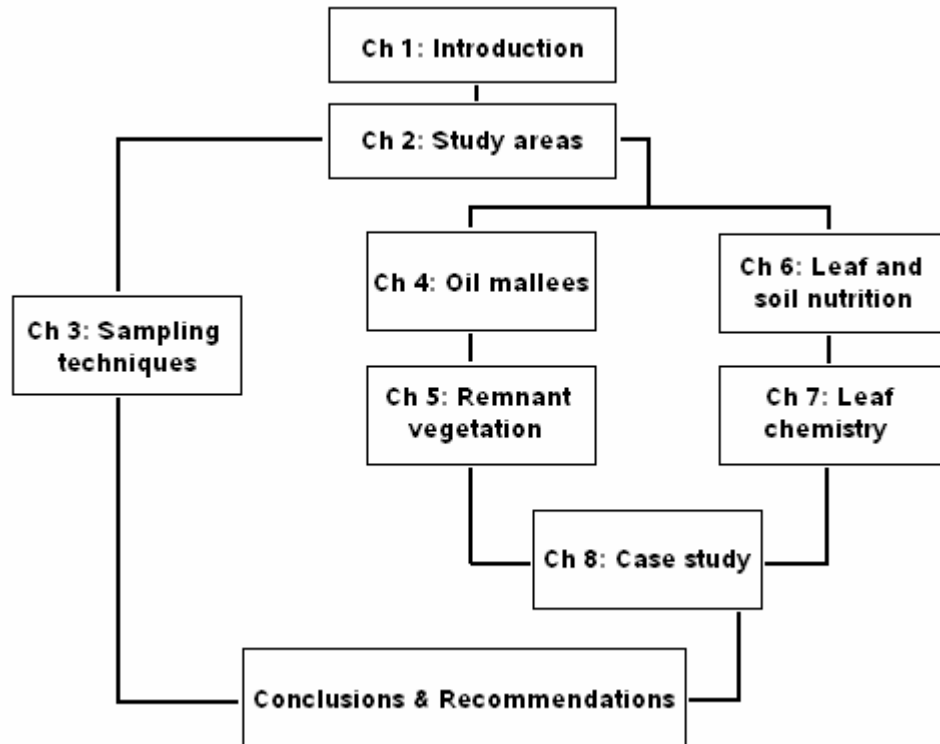


Figure 5: Flow diagram of the thesis structure showing how each of the chapters relate and build upon each other to lead to the conclusions and recommendations.

CHAPTER 2

DESCRIPTION OF STUDY AREAS

LOCATION OF STUDY AREA

The study area was located south-east of Perth in the southern central wheatbelt of Western Australia. Sites were spread between Narrogin and Harrismith in the south and Pingelly and the Tutanning Nature Reserve in the north (Figure 6).

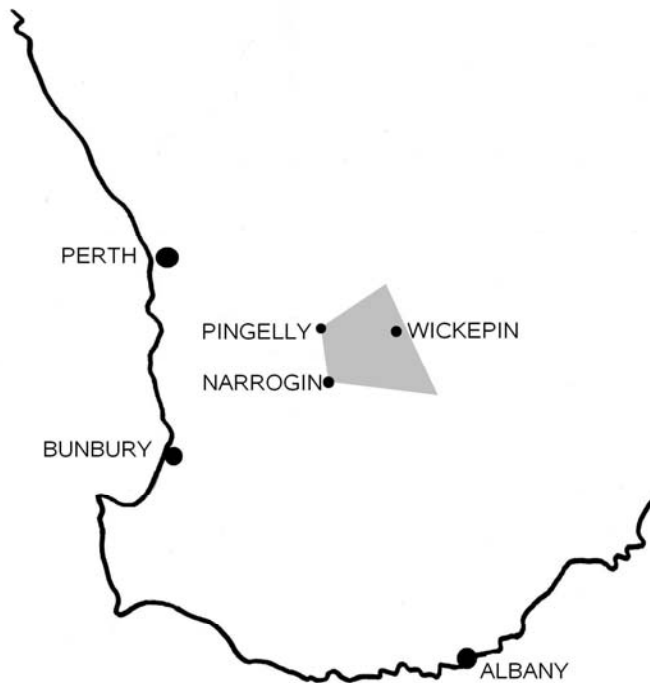


Figure 6: Map of the south-west of Western Australia, showing the study area shaded grey.

Seven sampling sites were selected within the study area (Figure 7). Four ‘mallee sites’ were selected, consisting of planted rows or alleys of oil mallee vegetation. These sites incorporated each of three oil mallee species, located in the same or adjoining paddocks. A further three ‘remnant sites’ were selected. These sites consisted of two species of

native eucalypt in uncleared remnant vegetation located alongside a commercial oil mallee planting of one of the three previously selected oil mallee species. All sites were selected to be representative of healthy vegetation of the chosen habitat type.

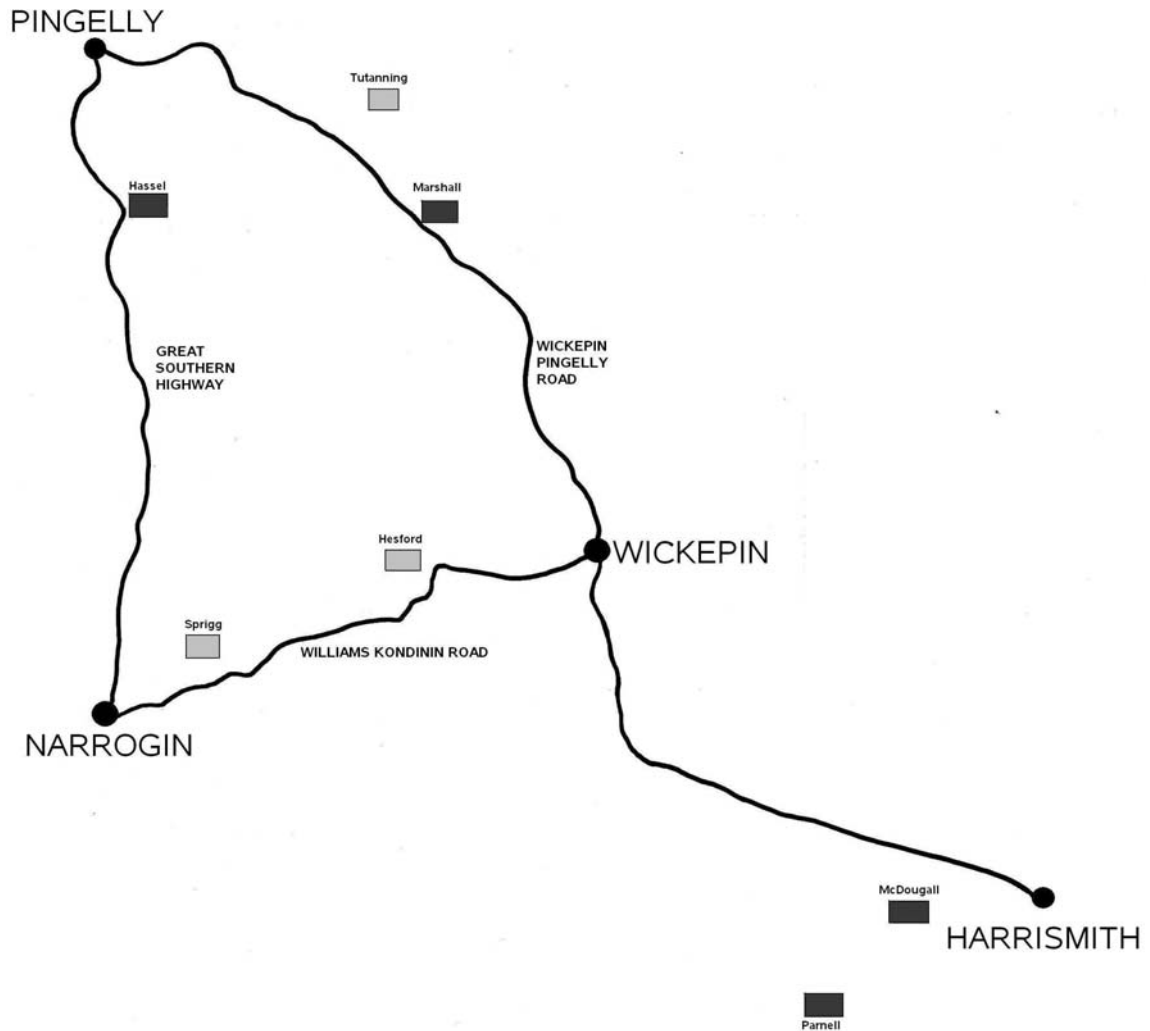


Figure 7: Map of site locations within the study area. Oil mallee sites are shaded dark grey, remnant vegetation sites are shaded light grey.

CLIMATE

The climate of the study area is categorized as Mediterranean and is typified by hot dry summers and cool, wet winters. General weather conditions were recorded on each day of sampling (Appendix A). The Narrogin townsite was used as a source of climate data

for the study area, as it is the major centre in the area and had climate data available dating back to 1891.

Rainfall

Figure 8 shows the long-term average and the recorded monthly rainfall for the study period. The long-term average annual rainfall for Narrogin is 495.9 mm. During the study period, the annual recorded rainfall was lower than the long-term average in 2005 (469.8mm) and much lower than the average in 2006 (342.6mm). The 2005 year had a similar pattern of rainfall to the long-term average, apart from a wetter than average autumn. The 2006 year, on the other hand, had very high rainfall during January and had a very dry winter, making the rainfall pattern almost reverse of that of the normal rainfall distribution for the area.

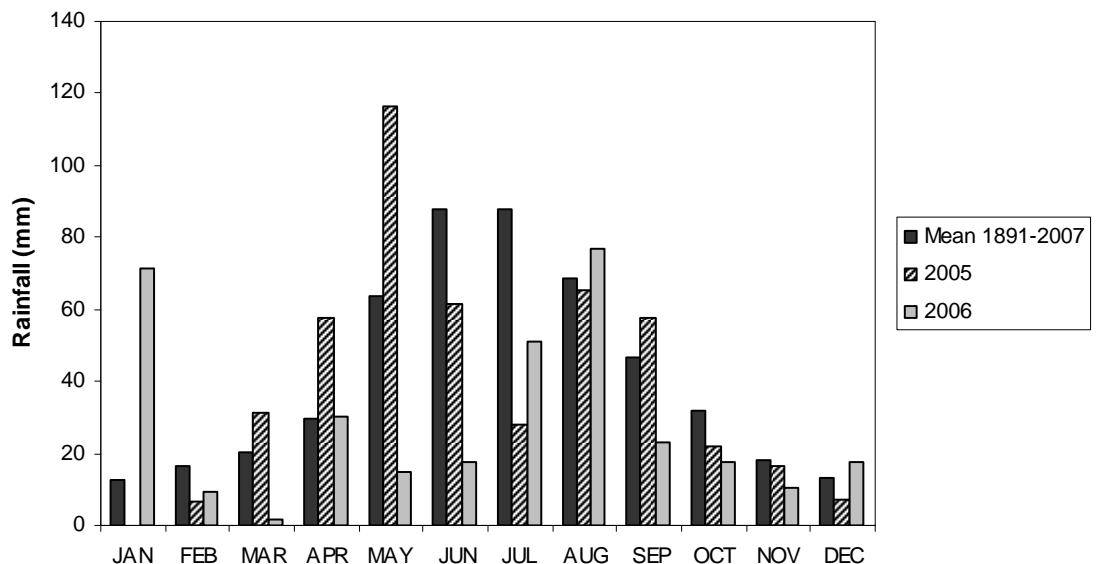


Figure 8: Long-term average monthly rainfall (1891-2007) and monthly rainfall recorded during the study period (2005- 2006) for the Narrogin townsite. (Source: Bureau of Meteorology 2007).

Temperature

Maximum and minimum temperature values for the two years of the study, as well as figures from the long-term average, are presented in Figure 9. Overall, the two years of the study experienced similar maximum and minimum temperatures to those reported in

the long-term average. However, there were a few notable differences. There were spikes in the mean minimum temperature in 2005 in both March and May. In 2006, minimum temperatures were lower than average in June and higher than average in November. The 2006 maximum temperatures were also variable, with the January figures being lower than average and the maxima from August to December consistently being above the long-term average.

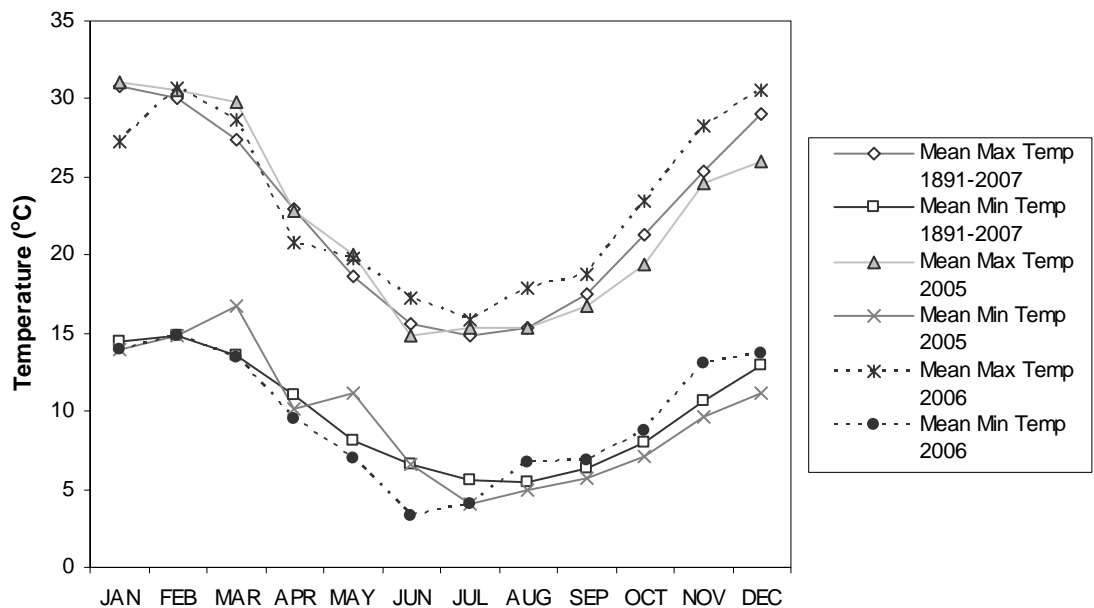


Figure 9: Long-term average maximum and minimum monthly temperatures (1891-2007) and mean monthly maximum and minimum temperatures recorded during the study period (2005-2006) for the Narrogin townsite. (Source: Bureau of Meteorology 2007).

SOIL TYPES

Geologically speaking, McArthur (1991) defines the Narrogin area as one consisting of “Archaean granites, gneisses, and migmatites, with some meta-sediments and volcanics” occurring in a few areas. Researchers from a number of different disciplines have defined approximately the same area, giving it names as diverse as the ‘Dwarda ecological region’ (Gentilli 1979), ‘the zone of younger laterites’ (Mulcahy 1967), and ‘the Narrogin semi-stripped etchplain’ (Finkl and Churchwood 1973). McArthur (1991) described the area as distinctive because of its pattern of landforms. It is easy to see why

descriptions vary so widely in such a diverse landscape, which ranges from un-weathered rock, granite outcrops and laterite-capped hills to wide valley plains and long gentle slopes.

The study site soils generally consist of sandy topsoil with varying amounts of gravel or clay deeper in the profile. The soils of the study area fall into three main types, described by McArthur (1991) as:

1) Quailing

- laterite plateau remnants divided into
 - shallow sandy and gravelly soils over lateritic duricrust
 - lateritic duricrust outcrops
 - deep yellow sands;

2) Kauring

- grey gritty sands over pale ferruginous gravels and duricrust; and

3) Malebelling

- yellow duplex soils developed over granite and divided into
 - brown gritty sands over yellowish brown gritty sandy clay
 - greyish brown gritty sand over yellowish brown and brown mottled gritty clay.

VEGETATION TYPES

The original vegetation of the region varies from eucalypt forests and open woodlands to dense shrubland. Before clearing, species commonly occurring in the area included: Wandoo, Brown Mallet, and various other woodland and mallee eucalypts, plus *Dryandra*, *Allocasuarina*, and *Acacia* (Figures 10 and 11). After clearing, only small pockets of the original vegetation (remnants) remained. The rest has been converted to agricultural land for grazing and cereal cropping. In some instances, a third stage of development has occurred where the land has become unprofitable for cropping and has been replanted to trees in order to reverse some of the damage done by the extensive clearing of the past. Within the study area, the revegetated land mostly consists of oil

mallee vegetation that has been planted for the combined benefits of profit (from oil and biomass production) and environmental outcomes (from improved soil water use and increased biodiversity).



Figure 10: Native vegetation (Wandoo woodland).



Figure 11: Native woodland vegetation.

The vegetation being studied in this research is of two types: 1) planted oil mallees; and 2) remnant eucalypts. Mallees are defined by Williams and Brooker (1997) as eucalypts which are “multistemmed from ground level, usually less than 10 m in height, often with the crown predominantly at the ends of the branchlets”. The remnant eucalypts in this study are best described as ‘woodland trees’, which are defined as around 10-25 m in height and, although single-stemmed, they may branch a short distance above the ground (Williams and Brooker 1997). In total, five species, all belonging to the eucalypt subgenus *Symphyomyrtus*, were assessed in this study; these are outlined with descriptions (Boland *et al.* 2006) below.

Oil Mallee Species

1) *Eucalyptus polybractea* (Blue-leaved Mallee)

From the Greek *poly* (much, many) and Latin *bractea* (bract), referring to the many bracts around young buds, although not unique to this species. The Blue-leaved Mallee is a mallee 5 to 10 m tall with ash-coloured or bluish foliage (Figure 12).



Figure 12: *Eucalyptus polybractea*.

It is native to small areas around the Wyalong district of New South Wales, and Bendigo region of Victoria. Although not native to Western Australia, *Eucalyptus polybractea* has been found to grow particularly well in the wetter western wheatbelt areas (including the study area). *E. polybractea* is one of the most widely used oil mallee species due to its high concentration of cineole in the leaf, making it a valuable oil producer.

2) *Eucalyptus kochii* subspecies *borealis* (Oil Mallee)

The species was previously known in some areas as *Eucalyptus horistes*, but is now known as *Eucalyptus kochii borealis*. Named after farmer and sawmiller, Max Koch (1854-1925), who made numerous plant and seed collections; Latin *borealis* (northern) probably relating to its northern distribution in WA. *Eucalyptus kochii borealis* is a rough-barked mallee growing to 8 m tall with multiple stems arising from lignotubers below the ground (Figure 13).



Figure 13: *Eucalyptus kochii* subspecies *borealis*.

Its canopy is made up of slightly glossy leaves, which may be green or bluish green and which vary in their amount of glossiness. This species is native to Western Australia, although in the current study it is well south of its natural range and has been known to perform poorly in areas above 600mm annual rainfall.

3) *Eucalyptus loxophleba* subspecies *lissophloia* (York Gum - mallee form)

From the Greek *loxos* (crooked, slanting) and *phleps* (veins), presumably referring to the habit and leaf venation of the species. This species is a low, straggly smooth-barked mallee native to the eastern wheatbelt and goldfields of Western Australia (Figure 14).



Figure 14: *Eucalyptus loxophleba* subspecies *lissophloia*.

The juvenile leaves tend to be bluish green, becoming glossy when mature. *E. loxophleba* subspecies *lissophloia* is on the western margin of its natural range and has been shown to grow well in the study area.

Remnant Eucalypt Species

1) *Eucalyptus wandoo* subspecies *wandoo* (Wandoo)

The name Wandoo comes from the Aboriginal name used for the ‘gum’ or sap obtained from this species. Wandoo is a medium sized, white-barked gum tree up to 25 or even 30 m tall (Figure 15). It is native to the south-west of Western Australia, occurring north to Gin Gin and Bindi Bindi. Wandoo is known to occur in open forests, woodlands and shrublands.



Figure 15: *Eucalyptus wandoo* subspecies *wandoo*.

2) *Eucalyptus astringens* (Brown Mallet)

From the Latin *astringens* (binding, constrictive), this refers to the astringent properties of the bark. Brown Mallet is a small to medium sized, non-lignotuberous tree 10-25 m in height (Figure 16). This species has a straight trunk, often more than half the height of the tree, and a dome shaped canopy of moderately dense foliage. Its bark is constantly shed and, in the 1800s, collecting and selling the bark to the tanning industry was an important source of income in the Narrogin area (McArthur 1991). The Brown Mallet is native to the wheatbelt of Western Australia, inland of the areas typically dominated by jarrah forest.



Figure 16: *Eucalyptus astringens*.

EXPERIMENTAL DESIGN

Seven experimental sampling sites were set up in the localities of Narrogin, Wickiepin, and Pingelly in the wheatbelt of Western Australia on the design shown in Figure 17.

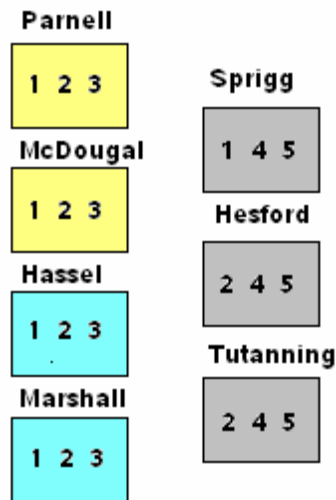


Figure 17: Diagrammatic representation of the experimental design. Colours represent habitat types and soil types: yellow = mallees on sand, blue = mallees on loamy sand, grey = remnant on shallow gravelly soils. Numbers represent tree species 1 = *Eucalyptus polybractea*, 2 = *E. loxophleba* subsp. *lissophloia*, 3 = *E. kochii* subsp. *borealis*, 4 = *E. wandoo* subsp. *wandoo*, 5 = *E. astringens*.

Four of these sites; Parnell, McDougal, Hassel, and Marshall; consisted of alleys of oil mallee vegetation from three different species: *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*. Of these four sites, two were on a sandy soil, while the other two were on a slightly heavier loamy, sand soil. This allowed not only for the comparison of the three oil mallee species over four sites, but also gave the opportunity to determine if there are differences in the performance of these species on different soil types. The reason the soil types are not more dramatically different is that oil mallees tend not to be grown on true heavy soils. Mallees generally ranged in age from 5 to 8 years, with many nearing an age at which harvesting for oil might be considered. The spacing of the mallees within rows was uniform, as they had been planted by machine. However, the space between mallee rows, known as the ‘alley’, ranged from 5 to 40 metres depending on if the growers planned to run sheep or grow crops between the rows or if the mallees were a stand-alone enterprise.

A further three sites; Sprigg, Hesford, and Tutanning; were selected in native vegetation remnants, either on farmers' properties, or in reserves. The remnants were typically located on shallow gravelly soils on, or adjacent to, lateritic outcrops. The age and spacing of the remnant species could not be controlled, as these trees were selected from those trees remaining after agricultural clearing, because of the poor soil and locations on which they were situated. The remnant vegetation at each of these sites contained two native species: *Eucalyptus wandoo* subsp. *wandoo* and *E. astringens*. Each of these sites also had planted oil mallee vegetation, in the same format as in the mallee sites, containing either *Eucalyptus polybractea*, or *E. loxophleba* subsp. *lissophloia* in an adjoining paddock. This permitted the two native species to be compared not only with each other but also with at least one oil mallee species. If oil mallees were found not to be significantly different from each other, then general comparisons between oil mallees and remnant vegetation could readily be made. This design also allows for the comparison of individual mallee species occurring alongside remnants against the same species growing with no remnants nearby. Unfortunately, not all of the desired site and tree species combinations were available, resulting in an unbalanced experimental design. Care has been taken for this not to adversely influence the statistical analyses used in later chapters.

SELECTION OF STUDY TREES

For the mallee sites, the three species selected at the same site were located, wherever possible, in the same paddock or in adjacent paddocks to limit within-site soil type variation. For each mallee species, five trees were selected from adjacent alleys of the same species. Where adjacent alleys were of a different species, trees were selected from one alley only. Trees were selected to represent the range in size and form of the mallees. Selected trees had crowns as separate as possible from adjacent trees, in order to prevent sampling error due to contamination from adjacent trees. At the remnant sites, five trees from each of the two native species, plus another five trees of the closest

occurring mallee species, were selected. As in the alleys, isolation of the crowns of selected trees from adjacent trees was important. However, in contrast to the alley sampling, trees in the remnant vegetation were selected on the basis of their practicality of sampling, as trees taller than 15 m could not be effectively sampled using the intended techniques. Hence, rather than selecting representative age and size trees, trees from the remnant sites were selected for manageability of size.

A review of sampling techniques and an examination of the effect of repeated sampling on canopy arthropod assemblages follows in Chapter 3.

CHAPTER 3

THE EFFECT OF RE-SAMPLING ON CANOPY ARTHROPOD ASSEMBLAGES

INTRODUCTION

In Chapter 2 the study sites used in this thesis were described. The aim of this chapter is to answer the question:

- Does re-sampling of vegetation influence the arthropod assemblages collected?

It is intended to achieve this by exploring the various sampling techniques available and conducting an experiment to determine the influence of re-sampling on the arthropod community. For this reason the chapter is divided into two sections:

- 1) *Background* - which reviews the various sampling techniques available and conducts an analysis of the benefits and disadvantages of chemical knockdown as a canopy arthropod collection method; and
- 2) *Effects of knockdown re-sampling* - which details an experiment conducted to determine if re-sampling of the same trees using chemical knockdown would affect the abundance and richness of arthropods collected at the second sampling.

BACKGROUND

Review of Sampling Techniques

A wide range of sampling methods has been used in canopy science. Before the use of chemical knockdown, the most common methods of sampling arboreal arthropods measured activity (of those arthropods which were large and mobile enough to be observed), rather than the actual abundance of arthropods (Stork 1988). The pros and

cons of some of the commonly used alternatives to chemical knockdown are described below.

Direct observations of arthropods in the canopy have the advantage of knowing where specimens came from, but low population densities make the work time-consuming and difficult (Paarmann and Kerck 1997). Hand collection also allows the origin of specimens in the canopy to be recorded accurately, and collections may be returned to the laboratory alive for further study. However, depending on the expertise of the worker, this method may collect very few specimens per unit of time and tiny arthropods can easily be missed (Basset *et al.* 1997).

While Lowman *et al.* (1996) found that sweeping was useful for collecting lepidopteran specimens, Basset *et al.* (1997) claim in a review of collection methods that the technique tends to be biased towards active rather than sedentary arthropods. They add that one of the difficulties with sweep netting is the considerable variation in sample size. Although sweep netting has been popular in studies of the “field layer” it is not commonly used in canopy studies.

The beating method involves using a stick to strike the vegetation and a collection tray to gather the arthropods as they become dislodged. Beating is non-destructive and easy to carry out (Lowman *et al.* 1996), as well as being very effective for collecting lepidopteran larvae (White 1975). Both Lowman *et al.* (1996) and Basset *et al.* (1997) agree that beating does not compare favourably with chemical knockdown in terms of the completeness of diversity collected. Basset *et al.* (1997) also found that the size of the catch using the beating method tended to be highly variable.

Branch clipping is a useful method for determining the exact density of foliage sampled (Basset *et al.* 1997.) The method has previously been used by Abbott *et al.* (1992) and Majer and Recher (1988) in studies of the arthropod faunas of eucalypts. However, the method is destructive of arthropod habitat and is heavily biased towards sedentary arthropods and web-spinners (Majer and Recher 1988, Basset *et al.* 1997).

Branchlet shaking (Majer *et al.* 1996) involves grabbing a small branch, about 30 cm long, and vigorously shaking it into a calico net. The major advantage of branchlet shaking is that it can be conducted in windy conditions when chemical knockdowns could not be conducted. However, in a comparison of branchlet shaking and chemical knockdowns, Majer *et al.* (1996) found that samples collected by branchlet shaking tended to contain less of the smallest and largest sized groups of arthropods than samples collected using chemical knockdown. Chemical knockdown, hence, is still the more effective of the two methods. However, in very windy weather conditions, branchlet shaking should be considered as an alternative to not sampling at all.

Malaise traps are designed to collect arthropods which fly upwards in response to a vertical obstacle and are particularly useful in collecting Diptera and Hymenoptera. Flight interception traps on the other hand target arthropods such as Coleoptera, which tend to fall when they encounter a vertical surface. The descriptions of Malaise and flight-interception traps by Basset *et al.* (1997) indicate that these traps target only flying arthropods, and that each is biased towards particular orders. In 1988, Basset described a composite interception trap which was designed to overcome some of the limitations of Malaise and flight interception traps. The resulting trap constituted a major improvement in trap design although, as acknowledged by Basset (1988), the composite interception trap would be best used in combination with chemical knockdown rather than in place of it.

Sticky traps allow for extensive replication due to their low cost (Basset *et al.* 1997). One disadvantage of sticky traps, however, as with Malaise and flight interception traps, is that they are biased towards the collection of flying arthropods. Also, the task of identifying specimens while stuck can be onerous, and once trapped in the glue, arthropods are very difficult to remove without damage.

Light traps and baited traps are useful for collecting certain taxa (Basset *et al.* 1997). The disadvantage is that only very specific taxa are collected; those that are collected are

nocturnal (light traps) or those which are attracted to certain foods or pheromones (baited traps). Light traps have the added drawback of being expensive to set up and to operate (Basset *et al.* 1997).

Vacuum samplers can be useful for sampling arthropods from a variety of habitats in the canopy, rather than just the foliage. Weaknesses in the use of vacuum samplers include their cost, weight, exhaust fumes, their potential to clog with debris, the difficulty in defining sample size and the possibility of damage to the samples (Basset *et al.* 1997).

Regardless of the type of sampling, canopy access has long been an issue, especially in larger rainforest trees. A range of measures have been developed over the years to assist in providing canopy access from aerial walkways (Wint 1983), balloon rafts (Lowman *et al.* 1993) and, recently, canopy cranes (Parker *et al.* 1992). The development of these measures has seen an explosion in canopy science as access to some of the tallest trees in the world is allowed.

There are a number of other methods available to researchers investigating arthropod biodiversity, so this is not an exhaustive list. However, it is clear from what has been discussed here, that none of these methods used alone adequately samples the diversity of arthropods in canopies.

Chemical Knockdown

One of the most important techniques used in the collection of canopy arthropods was not examined in the previous discussion. That is chemical knockdown, a method developed over time by a number of workers, including Martin (1966), Roberts (1973), Gagne (1979) and Southwood *et al.* (1982). It involves the application of an insecticide to the canopy, and subsequently the collection of arthropods falling from the treated vegetation. In the literature, chemical knockdown is variously referred to as misting, fogging, spraying or knockdown. While there are some differences in the machinery used and effectiveness under different sampling conditions, they all refer to the same basic principle. This section will first deal with the advantages and disadvantages of the

chemical knockdown technique in general, before examining the specific differences between the types of knockdown.

Natural pyrethrum and synthetic pyrethroids are the most commonly used chemicals for this sampling method, as they have a very high knockdown efficiency (Paarmann and Kerck 1997). The effect of pyrethroids on the insect nervous system results in uncontrolled movements, causing many arthropods to drop from leaves, thereby increasing their chance of collection (Stork 1991, Stork and Hammond 1997). Basset *et al.* (1997) found that chemical knockdown has the advantages of being relatively quick to carry out, and producing numerous, relatively clean samples which were easily processed. Studies by Lowman *et al.* (1996), and Majer and Recher (1988) examined various sampling techniques and agreed that, while not without problems, misting or fogging with insecticides were ultimately the most effective methods to use in collecting a large and representative sample of the canopy arthropod community. In 1987, Paarmann and Stork referred to the development of chemical knockdown as a “breakthrough” in terms of solving the problem of accessing the canopy. Many researchers (e.g., Majer and Recher 1988, Lowman *et al.* 1996, Basset *et al.* 1997, Stork and Hammond 1997, Richardson *et al.* 1999) agree that chemical knockdown collects more of the diversity of canopy arthropods, is less dependent on arthropod activity, and is not as destructive to the habitat as other sampling techniques. Chemical knockdown is also regarded by some as being the best method to use in terms of accuracy, reliability and comparability of collected material (Stork and Hammond 1997).

Some disadvantages of insecticide knockdown are that the procedure is noisy and invasive (Lowman *et al.* 1996), the equipment and chemicals can be costly, and trees can not be re-sampled until recolonization has occurred (Basset *et al.* 1997). If excess moisture is present on leaves before spraying, arthropods can become trapped on the phylloplane by the surface tension of droplets of chemical pooling on the leaves. Furthermore, as the droplets dry the arthropods then tend to adhere to the foliage rather than dropping from the canopy as intended (Ozanne 2005). Paarmann and Kerck (1997) found that some leg loss can occur in long-legged arthropods, while Basset *et al.* (1997)

suggest that having specimens which are dead when collected and the difficulty of tracing the part of the canopy from which arthropods were sampled are disadvantages of the procedure. One of the mostly frequently cited problems with chemical knockdown (Adis *et al.* 1984, Majer and Recher 1988, Lowman *et al.* 1996, Stork and Hammond 1997) is that some types of arthropods are not effectively sampled. Cryptic arthropods, such as those that inhabit nests, galleries, webs, leaf rolls or bark may not be killed by the insecticide, while sessile arthropods that are attached to the leaves, such as scale insects and psyllids, do not fall from the canopy and so are not collected. It is also not known how effectively nocturnal arthropods are sampled by this technique.

According to Stork (1988), sampling with knockdown insecticides can be divided into two types: spraying and fogging. Lowman *et al.* (1996) agrees, regarding fogging and misting as technically separate procedures, due to differences in methods of application and with fogging producing smaller droplets. Stork (1988) defines spraying as delivering “a fine spray of small droplets produced by injecting the insecticide solution into the air blast of a large motor-driven fan.” Spraying (or misting) equipment is carried on the investigator’s back and the spray can be directed up to 10 m into the canopy. Stork (1988) describes fogging as producing “much smaller droplets as the insecticide is broken up by injection into the exhaust fumes produced by a small engine”. Heat from the exhaust of the machinery produces a warm fog which rises through the canopy, making fogging much better suited to use in tall trees. However, one difficulty related to the use of fogging is that the fog is very mobile and can easily be influenced by climatic conditions (Lowman *et al.* 1996). This means that the technique is best undertaken on still days, limiting the time and numbers of days on which it can be performed. The mobility of fog can also result in knockdown over a much larger area than was originally intended, which can be a serious disadvantage when sampling closely spaced trees or sensitive habitats (Ozanne 2005). Spraying on the other hand is limited by the distance that the spray can travel, meaning that only trees of a certain height can be sampled.

EFFECTS OF KNOCKDOWN RE-SAMPLING

As chemical knockdown appeared to be the most effective way to collect a large amount of data in a small amount of time, this method was chosen for use in the current study. Spraying (or misting) was chosen in preference to fogging, as the trees being sampled were not exceptionally tall and the need to spray relatively closely spaced trees, under possibly windy conditions meant fogging would not be possible. To maximize the amount of data collected per sampling effort, it was decided to sample in the peak arthropod seasons of autumn and spring. As there were a limited number of trees available for this study, this would mean that the same trees might have to be sprayed on more than one occasion.

Since recolonization rates for these trees were not known, it could not be assumed that the first spraying had not substantially distorted or reduced the arthropod community on those trees before the second sampling occurred. The results of this study therefore have important implications for the rest of the thesis, as repeated sampling is used throughout the research. If research results are confounded as a consequence of re-sampling, it may be difficult to determine if variations between treatments are attributable to the experimental factors being tested, or if they are due to incomplete recolonization of the trees before re-sampling.

Method

Five trees of each of three species, *Eucalyptus loxophleba* subsp. *lissophloia*, *E. wandoo* subsp. *wandoo*, and *E. astringens* were selected from a site near Wickiepin, WA. Trees were selected to be as independent as possible to avoid contamination of trees which might later be a source of recolonising arthropods. In October of 2005, the trees were sampled by canopy spraying, using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] (Appendix B) was applied at a rate of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Many studies in the literature have used non-residual synthetic pyrethroids (Paarmann and Stork 1987, Stork 1991, Stork and Hammond 1997). The chemical used in this study is a contact and residual synthetic pyrethroid, with an average field half life of 90 days. It is non-toxic to

vertebrates and very slow to degrade in sunlight but breaks down rapidly in soil. This chemical was used because it was widely available and was relatively cheap compared to natural pyrethroids. Since this chemical has a residual activity, this study was needed to determine its effect on recolonization.

Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Various investigators have used different 'drop times' before collecting fallen specimens from sheets or trays (Roberts 1973, Adis *et al.* 1997, Paarmann and Kerck 1997, Azarbayjani *et al.* 1999). While some taxa drop almost immediately, time must be allowed for species which take longer to succumb to the insecticide. Most studies allow a drop time of between one and two hours; after this time the risk of contamination with taxa not from the source tree is increased (Stork and Hammond 1997). For the studies in this thesis, a short drop time of one hour was used as meat ants, *Iridomyrmex greensladei*, were observed collecting fallen arthropods from the sheets and carrying them away as a food source. The shorter drop time was used to prevent such losses from ants scavenging. Once collected, samples were stored in alcohol until sorting.

In order to prevent Occupational Health and Safety issues related to the use of pesticide chemicals, a range of measures were put in place. Personal protective equipment, including full body spray suit, gloves, boots, protective eyewear and a respirator mask, were worn during spraying. Gloves and face masks were also worn whenever handling drop sheets and spray equipment. Upon return to the laboratory arthropods were transferred to clean ethanol prior to sorting to remove any remaining chemical contamination.

In May of 2006, the same spraying and collection procedure was used to sample another 15 trees (three species x five reps), as well as the original 15 trees which had previously been sampled in October of 2005. Once collected and stored in alcohol, samples were

sorted with the aid of a stereo microscope to the ordinal level in the laboratory. Data were tested for homogeneity of variance and transformations were done where necessary. Two-way ANOVAs were conducted to determine the effect of spraying treatment on total numbers of arthropods (abundance), numbers of orders (richness) and mean individuals collected from each of the major orders. Tree species was included as a random factor in the analyses.

Results

Only the results of collections obtained in the May 2006 sampling are presented here. This is in order to perform a direct comparison of a first sampling and a second sampling conducted at the same time, during the same season. Mean arthropod numbers and frequency of arthropod orders are presented in Appendix C. Numbers of arthropod orders and total numbers of arthropods varied between the three tree species (Figure 18). Analyses of variance (Table 1) revealed that these differences were statistically significant ($P < 0.05$) (df: 2, 24), with *E. wandoo* having more individuals and orders than the other species. Differences between the two spraying treatments were much smaller, and analysis of variance (Table 1) revealed that there was no significant effect of re-spraying on arthropod abundance or ordinal richness. There was also no interaction between tree species and spraying treatment.

Table 1: Two-way ANOVA results for effect of spraying treatment and tree species on total numbers of arthropods (abundance) and numbers of orders (richness). Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right.

	Source of Variation	Differences	df	MS	F	Probability
Abundance	tree species	W ^a A ^b L ^b	2	76910.40	4.649	0.020
Abundance	spraying treatment	ns	1	9973.63	0.603	0.445
Abundance	species*sprayed	ns	2	9761.73	0.590	0.562
Abundance	error		24	16541.76		
Richness	tree species	W ^a A ^b L ^b	2	10.80	12.462	0.000
Richness	spraying treatment	ns	1	0.00	0.000	1.000
Richness	species*sprayed	ns	2	2.80	3.231	0.057
Richness	error		24	0.86		

* W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; L = *E. loxophleba* subsp. *lissophloia*.

* ns = no significant difference at $p=0.05$.

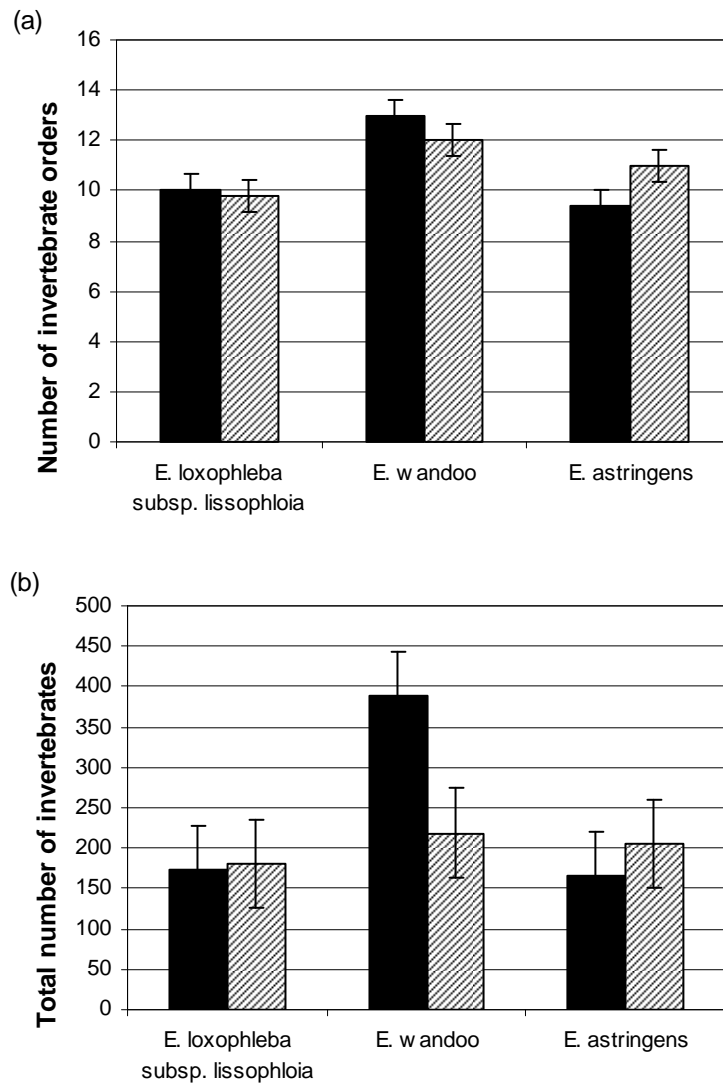


Figure 18: Variation in (a) numbers of arthropod orders and (b) total numbers of arthropods collected when sprayed once (represented by a dark bar) compared to sprayed twice (represented by shaded bar). Error bars represent standard error.

Individual orders were also examined to determine if any individual groups were influenced by re-spraying (Figure 19).

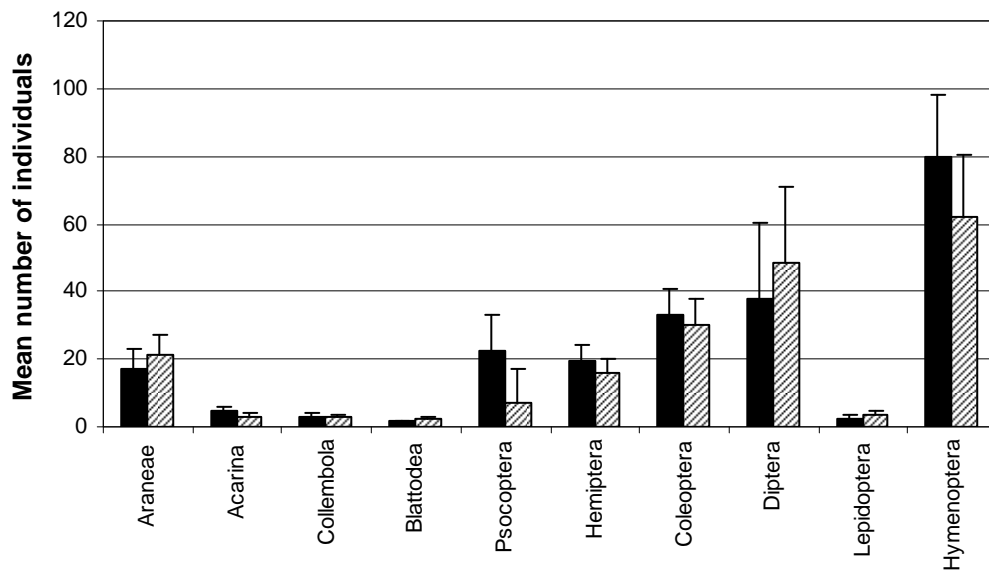


Figure 19: Mean numbers of individuals of selected arthropod orders collected from trees sprayed once (represented by a dark bar) compared to trees sprayed twice (represented by shaded bar). Error bars represent standard error.

While the graph appears to show some variation in mean numbers of individuals collected, analysis of variance (Table 2) revealed that only Lepidoptera differed significantly in the number of individuals collected from trees that had been sampled for the first or the second time. Tree species had a greater effect, with Acarina, Psocoptera, Diptera, Lepidoptera, and Hymenoptera numbers being influenced by the tree species from which they were collected. No significant interaction, however, was observed between tree species and spraying treatment.

Table 2: Two-way ANOVA results for effect of spraying treatment and tree species on mean individuals collected from each of the major orders.

Taxon	Source of Variation	df	MS	F	Probability
Araneae	spraying treatment	1	0.04940	0.49	0.490
Araneae	tree species	2	0.28360	2.83	0.079
Araneae	sprayed*species	2	0.09620	0.96	0.397
Araneae	error	24	0.10030		
Acarina	spraying treatment	1	0.00053	0.01	0.942
Acarina	tree species	2	0.50798	5.18	0.013
Acarina	sprayed*species	2	0.12456	1.27	0.299
Acarina	error	24	0.09803		
Collembola	spraying treatment	1	0.09072	1.50	0.233
Collembola	tree species	2	0.13896	2.29	0.123
Collembola	sprayed*species	2	0.07960	1.31	0.288
Collembola	error	24	0.06060		
Blattodea	spraying treatment	1	0.00605	0.13	0.725
Blattodea	tree species	2	0.12044	2.53	0.101
Blattodea	sprayed*species	2	0.00229	0.05	0.953
Blattodea	error	24	0.04767		
Psocoptera	spraying treatment	1	0.17910	0.91	0.349
Psocoptera	tree species	2	1.21240	6.19	0.007
Psocoptera	sprayed*species	2	0.59270	3.03	0.067
Psocoptera	error	24	0.1959		
Hemiptera	spraying treatment	1	0.00390	0.03	0.853
Hemiptera	tree species	2	0.07440	0.66	0.526
Hemiptera	sprayed*species	2	0.01250	0.11	0.895
Hemiptera	error	24	0.11270		
Coleoptera	spraying treatment	1	0.00430	0.04	0.850
Coleoptera	tree species	2	0.06190	0.52	0.599
Coleoptera	sprayed*species	2	0.04960	0.42	0.662
Coleoptera	error	24	0.11830		
Diptera	spraying treatment	1	0.03950	0.23	0.634
Diptera	tree species	2	1.29560	7.63	0.003
Diptera	sprayed*species	2	0.06530	0.38	0.685
Diptera	error	24	0.16980		
Lepidoptera	spraying treatment	1	0.35693	8.89	0.006
Lepidoptera	tree species	2	0.90491	22.54	0.001
Lepidoptera	sprayed*species	2	0.05060	1.26	0.302
Lepidoptera	error	24	0.04015		
Hymenoptera	spraying treatment	1	0.06789	0.87	0.361
Hymenoptera	tree species	2	0.43445	5.55	0.010
Hymenoptera	sprayed*species	2	0.04509	0.58	0.570
Hymenoptera	error	24	0.07828		

All ordinal data were log (x+1) transformed for homogeneity of variance.

Discussion

According to Stork and Hammond (1997) the proportion of a tree's fauna not killed by the insecticide and the pace at which recolonization occurs are the most important factors influencing the re-establishment of the fauna of a treated tree. Aspects such as the type of insecticide used and the proximity of other vegetation from which recolonization could occur, may also affect the rate at which arthropods return to sprayed trees.

In this study, spraying treatment was found to have no adverse influence on arthropod abundance or richness, or on the abundance of any individual order indicating that recolonization had occurred by the time of the second sampling. This was largely unexpected, as other workers (Stork 1991, Stork and Hammond 1997, Azarbayjani *et al.* 1999) have found that recolonization can be a slow process and repeated sampling may not accurately represent the fauna that would be present had the trees not previously been sampled. However, these findings can vary considerably with the time scale and taxonomic echelon being considered. Stork (1991) found that only about 20% of individuals and species had returned 10 days after the first knockdown. Stork and Hammond (1997) confirmed this in their study, in which it took three months for the faunas of sampled tree crowns to fully recover. In a study conducted by Azarbayjani *et al.* (1999), the number of species present on the trees recovered within 16 weeks of spraying and common species recolonized within a fortnight. However, the community structure was still seriously disrupted a year after being initially sampled.

Previous studies have used non-residual synthetic pyrethroids (Paarmann and Stork 1987, Stork 1991, Stork and Hammond 1997). However, the current study has used a synthetic pyrethroid chemical with residual activity, meaning that its effects could last for some time after spraying. Since this chemical has a reasonably long residual activity, it was expected that recolonization might be reduced or somewhat delayed. However, as no significant differences were detected between trees being sampled for the first time and those being resampled, it could be assumed that any residual effect had worn off before re-sampling occurred.

Despite expectations that recolonization might take over a year to complete (Azarbayjani *et al.* 1999), this has not been supported in the current work. Perhaps recolonization is rapid in this area, or conceivably six months is ample time for full recolonization to occur. Another possibility is that examination of the data at the ordinal level was not sufficient to detect some of the less apparent changes which might have occurred at a higher taxonomic resolution. Whatever the reason, resampling had no measurable effect on numbers of orders or individuals collected. This is important to note, as while re-sampling of the same trees has been used in other parts of this thesis, it appears that this has not introduced any extra experimental error to the data.

Effect of tree species will be further explored in Chapters 4 and 5.

CONCLUSION

In the background section, it was determined that for this study chemical knockdown would be ideal for sampling a large number of arthropods in a small time. In the study of effects of knockdown re-sampling, we saw that in this case there was no effect of re-sampling on the ordinal level richness or total abundance of arthropods collected at the second sampling. This means that the 6 month period between sampling was long enough to allow for a reasonable level of recolonization, or at least a return of the arthropod fauna to levels which were undetectable, at the ordinal level, from those of trees being sampled for the first time. As a consequence, we can be reasonably confident that the results in other sections of the thesis have not been confounded by the need to re-sample the same trees.

CHAPTER 4

THE EFFECT OF OIL MALLEE SPECIES ON CANOPY-DWELLING ARTHROPOD ASSEMBLAGES

INTRODUCTION

In Chapter 3 the effect of repeated sampling on canopy arthropod assemblages was explored. It was determined that repeated sampling, as conducted in this study, did not affect arthropod ordinal richness and total abundance. This chapter will attempt to answer the question:

- Does the canopy-dwelling arthropod assemblage vary between oil mallee species?

In order to do this, firstly, the concepts of diversity and host specificity will be introduced and explained in terms of how they relate to oil mallees. Then, the rest of the chapter will detail a study comparing three oil mallee species in terms of their richness, abundance and ordinal diversity.

Arthropod Diversity

What is arthropod diversity? At its most basic, diversity can be described as the variety of animal species (Southwood and Henderson 2000) and so, arthropod diversity would be the variety of arthropod species. In reality however, diversity is a far more complex concept. Whittaker (1972) divided diversity into three types:

- α diversity: the number of species within a community or habitat;
- β diversity: the rate and extent of change in species along a gradient from one habitat to others; and
- γ diversity: the number of species in a range of habitats in a geographical area, resulting from the α diversity of habitats and the β diversity between them.

In 1994 Majer *et al.* also introduced the concept of σ diversity, which refers to the diversity due to seasonal changes in species composition.

There are many components to diversity (Southwood and Henderson 2000). Here I will outline three of the most frequently used:

- richness - the number of species (or any other taxonomic classification) present in a habitat or ecosystem;
- abundance - the number of individuals in a defined area; and
- evenness - the distribution of the abundance of individuals between species (or other taxonomic levels).

In the current study richness and abundance were used as the main measures of diversity in oil mallees and remnant vegetation. Due to the volume of specimens collected during this study most samples were only identified to order, hence the use of ordinal richness.

However it is defined or measured, diversity can vary in many ways and innumerable factors can influence it. So what influences diversity? There is a huge range of variables which contribute to arthropod diversity in tree canopies. Moran and Southwood (1982) found that there were differences in the abundance of the various kinds of arthropods in tree canopies, depending on whether the leaves were broad or narrow, and that narrow-leaved species tended to have lower species richness than broad-leaved ones. Majer *et al.* (1999) found that in some instances, isolated paddock trees had a greater abundance and diversity of canopy arthropods than remnant trees and suggested that this might be due to higher nutrient levels in the paddock or to a concentrating effect on dispersing arthropods. Abbott and Wills (2001) postulate that moderate levels of disturbance may be beneficial to canopy arthropod diversity. They also claim that canopy arthropod species richness is influenced mostly by temperature, rainfall and habitat diversity.

Host Specificity

Host specificity refers in this case to the level to which the arthropod species are restricted to a particular host tree species. There is some debate about the levels of host specificity displayed by canopy arthropods and what influence specificity might have on arthropod diversity. Basset (1992) argues that while it is assumed that canopy arthropods in woodland environments are generally quite host-specific, there tends to be a decrease

in specificity in response to low host plant nutrient levels. In a study comparing the arthropod fauna of Australian forests, Recher *et al.* (1996) found that as much as 60% of the arthropod fauna collected in NSW, and 47% of that in WA forest canopies was collected only from one of the eucalypt species sampled in each of the respective forests. However, many of these species were rarely encountered, so their degree of host specificity cannot be predicted with confidence. Novotny *et al.* (2002), believes that host specificity is rare, and that most specialization is at the genus level and most often occurs in large genera (such as *Eucalyptus*). Basset (1992) also found that host specificity was reduced where plant chemical defenses and/or predator numbers were low. This agrees, in part, with work by Bernays and Graham (1988), which proposes that natural enemies are a major selection pressure for host specificity. Their work also suggests that the effect of leaf chemical composition has been overemphasized and, while this does influence behaviour, it may not be as important as first thought in the development of specialization by herbivores.

With these issues in mind, the study outlined in this chapter sought to characterize the arthropod assemblages on three oil mallee species. A resulting question is whether the three species contribute equally to the return of biodiversity when tree plantings are carried out in the agricultural landscape.

METHOD

Five trees of each of three oil mallee species, *Eucalyptus loxophleba* subsp. *lissophloea*, *E. polybractea*, and *E. kochii* subsp. *borealis* were selected at four sites; (Figure 7) Parnell, McDougall, Marshall and Hassel. At each site, the three oil mallee species were located in the same or adjoining paddocks and sampled trees were selected to be representative of the age and size range present.

The characteristics and dimensions of each tree were measured and recorded (Appendix D). For example, tree height, width (N-S and E-W), and height of canopy from base were recorded to enable the calculation of canopy volume. As the majority of trees

sampled were elliptical in shape, the formula of Thorne *et al.* (2002) was used for calculating an ellipsoid volume [$2/3\pi H(A/2 \times B/2)$], where: H is the height of the plant from the base to the top of the photosynthetically active material; A is the widest diameter of the plant measured on a North-South orientation; and B is the widest diameter of the plant measured on a East-West orientation (at right angles to A). Thorne *et al.* (2002) claim that their formula is “elastic and accurately accommodates a wide range of plant shapes and sizes”. Other factors such as alley position, and flowering status were also recorded (Appendix E). Preliminary testing with analyses of variance, however, found no significant effect of flowering or alley position on ordinal richness or total abundance of canopy arthropods. Some interaction between site and alley position was observed. However, this was determined to be an anomaly in the data produced by a single northward facing tree at one site, hence further analysis of these data was considered unnecessary.

In October 2005 (spring), and May 2006 (autumn), the trees were sampled by canopy spraying, using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] was applied at a rate of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Once collected, samples were stored in alcohol until sorting. Samples were then sorted with the aid of a stereo microscope to the ordinal level in the laboratory. Mean and frequency figures from sorted arthropod data are presented in Appendix F. With the exception of beetles (Chapter 8), arthropods were only sorted to ordinal level due to the time and labor constraints experienced during this PhD study.

For each tree, at the spring 2005 sampling, samples were also taken of each type of scale, gall, sessile arthropod and other formation of interest, by branch clipping (Appendix G). This was intended to demonstrate the diversity of arthropod fauna, and some of the leaf symptoms produced by arthropods, not collected by canopy fogging.

Tall branches were reached by use of pole mounted secateurs and clippings were taken to be representative of the diversity of leaf damage and sedentary arthropods present rather than a random sample which might not have shown the actual diversity present. After field collection, all samples were taken to Curtin University of Technology for sorting and identification.

Data were tested for homogeneity of variance and transformations were done where necessary. Abundance was corrected for canopy volume by using arthropods per cubic metre of canopy volume in the analyses. A series of analyses of variance (ANOVAs) were performed to determine if there were any significant differences between the three tree species in terms of tree height, canopy volume, richness, abundance, individuals per order, and leaf blemishes. Two or three-way ANOVAs were conducted on the data collected, with tree species as a fixed factor and site as a random factor. Season was included as a fixed factor in all analyses except the two-way analysis of leaf blemishes as the leaf blemish data were only collected in one season. LSDs were used in post hoc testing to determine where differences lie.

The results presented in this chapter fall into two main categories:

Tree physical data - which presents and compares the two physical aspects of the oil mallees which were deemed to be the most likely to impact on arthropod ordinal richness, namely tree height and canopy volume. Chemical factors such as nutritional status and leaf chemistry will be examined in later chapters.

Arthropod data - which presents the results of arthropod collections by chemical knockdown and leaf clipping. Chemical knockdown results are presented in terms of ordinal richness, abundance and individuals within orders. Collections of leaf blemish data obtained by leaf clipping are presented as presence / absence data. Seasonal impacts on arthropod collections are also examined in this section.

RESULTS

Tree Height

Tree height (Figure 20) varied greatly between the three oil mallee species, with *E. polybractea* being the tallest and *E. kochii* the shortest. It can also be seen that, on average, all species grew by an appreciable amount between sampling times.

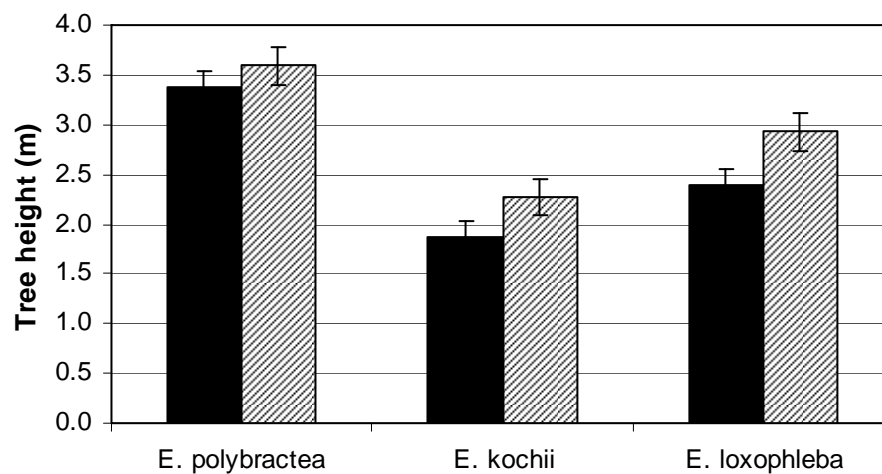


Figure 20: Mean tree height of three oil mallee species as measured in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Analysis of variance (Table 3) confirmed this, with significant ($P < 0.05$) differences in tree height between the three species observed. There was also a small difference in the effect of site, with one site (Parnell) having taller trees than the other sites. As expected, tree growth between sampling periods produced a significant difference in tree height between seasons. No interactions between factor effects were observed.

Table 3: Three-way ANOVA results for effect of mallee species, season and site on tree height, with site as a random factor. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Site	ns	3	1.191	1.453	0.319
Species	P ^a L ^b K ^c	2	20.224	25.097	0.001
Season	2 ^a 1 ^b	1	4.540	43.927	0.007
Site*Species	interaction	6	0.806	8.994	0.009
Site*Season	ns	3	0.103	1.153	0.402
Species*Season	ns	2	0.260	2.907	0.131
Site*Species*Season	ns	6	0.090	0.330	0.919

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*.

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

Canopy Volume

The three tree species examined here had varying canopy volumes (Figure 21), with *E. polybractea* having the largest and *E. kochii* the smallest canopy volume. All species also appeared to substantially increase in canopy volume over the six months between sampling.

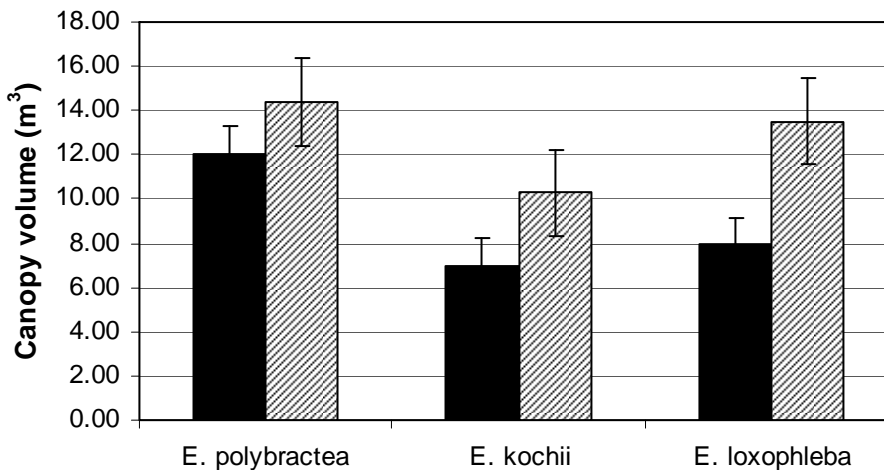


Figure 21: Mean canopy volume of three oil mallee species as determined from measurements made in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Analysis of variance (Table 4) indicated no significant influence of tree species or site on canopy volume. Season, however, had a significant influence, with the canopy volume found to be significantly greater ($P < 0.05$) at the second sampling than at the first.

Table 4: Three-way ANOVA results for effect of mallee species, season and site on canopy volume, with site as a random factor. Log canopy volume was used for this analysis. Superscripts indicate significant differences (LSD, $p = 0.05$), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	0.547	2.338	0.178
Season	2 ^a 1 ^b	1	0.629	34.882	0.010
Site	ns	3	0.191	0.769	0.548
Site*Species	interaction	6	0.234	70.405	0.000
Season*Species	interaction	2	0.050	14.956	0.005
Site*Season	interaction	3	0.018	5.429	0.038
Site*Species*Season	ns	6	0.003	0.078	0.998

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at $p = 0.05$.

* ns = no significant difference at $p = 0.05$.

Richness

Arthropod ordinal richness, as shown in Figure 22, averaged between 10 and 12 orders per tree for each of the three mallee species, with very slight differences in the numbers of orders collected between seasons.

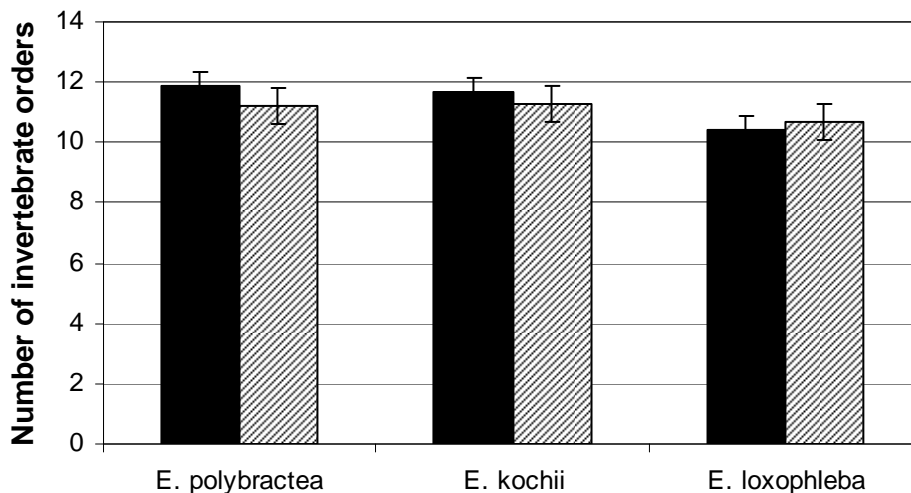


Figure 22: Mean number of arthropod orders collected on three oil mallee species in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Analysis of variance (Table 5) indicated that there were no significant differences in the numbers of orders collected from the different mallee species ($P < 0.05$). Site also had no significant effect on the number of orders. Season however did have an effect with the number of orders collected being reduced at the second sampling. There was also a significant site by species interaction.

Table 5: Three-way ANOVA results for effect of mallee species, season and site on number of orders collected per m³ of canopy volume, with site as a random factor. Superscripts indicate significant differences (LSD, $p = 0.05$), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	6.541	2.266	0.185
Season	1 ^a 2 ^b	1	5.705	23.190	0.017
Site	ns	3	2.689	0.890	0.495
Species*Season	ns	2	0.281	2.499	0.162
Season*Site	ns	3	0.246	2.186	0.191
Species*Site	interaction	6	2.887	25.651	0.000
Season*Species*Site	ns	6	0.113	0.173	0.983

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at $p = 0.05$.

* ns = no significant difference at $p = 0.05$.

Abundance

The total number of arthropods, also referred to as total abundance, is shown in Figure 23. Arthropod numbers appeared to vary, both between species and seasons. Analysis of variance (Table 6) however, showed no significant difference between species or seasons in terms of the number of arthropods collected. There was also no significant difference ($P < 0.05$) in the total number of arthropods collected at different sites. There were however some interaction effects but no consistent pattern emerged.

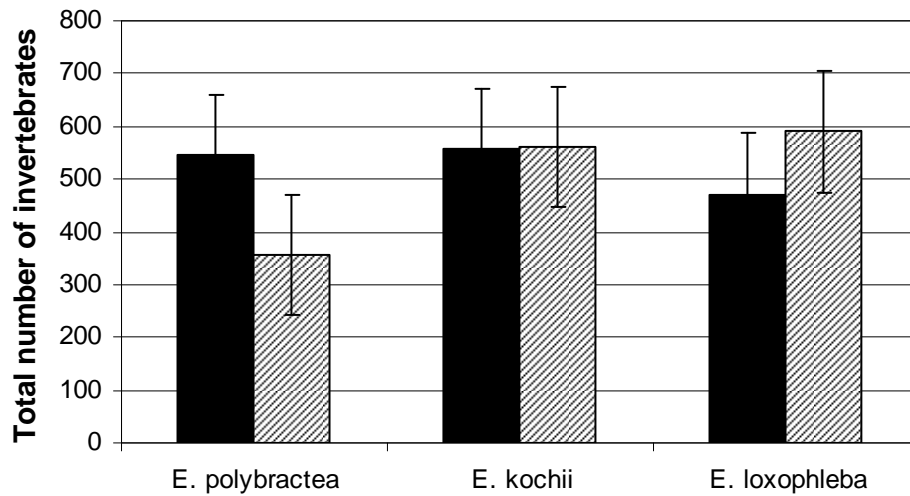


Figure 23: Mean number of arthropods collected on three oil mallee species in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Table 6: Three-way ANOVA results for effect of mallee species, season and site on total number of arthropods collected per m³ canopy volume, with site as a random factor and canopy volume values log transformed. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	1.106	1.294	0.341
Season	ns	1	0.999	1.674	0.286
Site	ns	3	0.197	0.157	0.922
Species*Season	ns	2	0.042	0.217	0.811
Season*Site	ns	3	0.597	3.091	0.111
Species*Site	interaction	6	0.854	4.425	0.047
Season*Species*Site	interaction	6	0.193	3.385	0.005

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

Individuals within Orders

There were some observable differences between mallee species in terms of the number of individuals within each order collected. Figure 24 indicates higher numbers of Psocoptera and Coleoptera on *E. polybractea*, greater levels of Hemiptera on *E. loxophleba* and more Hymenoptera on *E. kochii* than on the other mallee species.

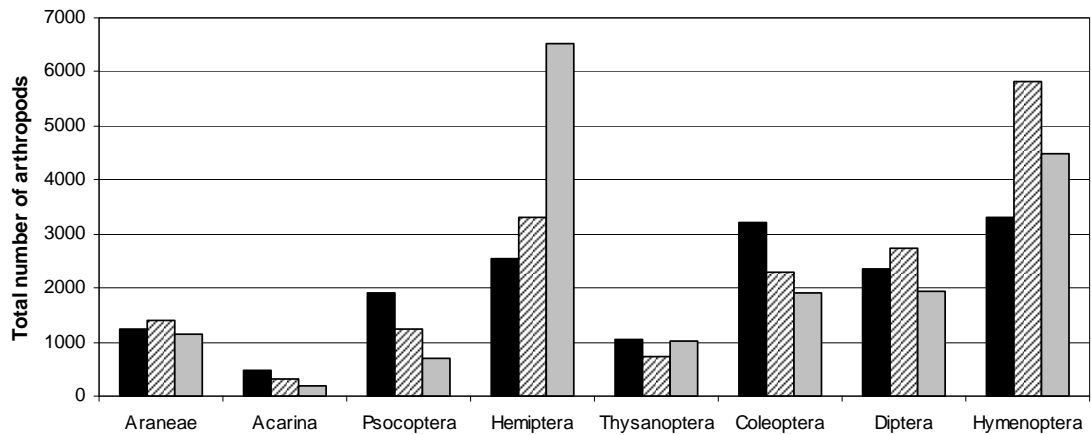


Figure 24: Total number of arthropods collected from particular orders on three oil mallee species; *E. polybractea* (represented by a dark bar), *E. kochii* subsp. *borealis* (represented by shaded bar) and *E. loxophleba* subsp. *lissophloia* (represented by a light bar).

However analysis of variance (Table 7) showed that these differences were not significant. In fact the only statistically significant differences were for differences in the numbers of Psocoptera, Hemiptera and Coleoptera between seasons, with each of these orders having lower numbers at the second time of sampling.

Table 7: Three-way ANOVA results for effect of mallee species, season and site on numbers of individuals collected from particular orders. Abundance was corrected for the canopy volume of the sampled tree. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right.

Taxon	Source of variation	Differences	df	MS	F	Probability
Araneae	Season	ns	1	0.196	0.773	0.444
Araneae	Species	ns	2	0.794	1.200	0.365
Araneae	Site	ns	3	0.199	0.256	0.855
Araneae	Season*Species	ns	2	0.001	0.007	0.993
Araneae	Season*Site	ns	3	0.253	1.821	0.244
Araneae	Species*Site	interaction	6	0.662	4.763	0.040
Araneae	Season*Species*Site	interaction	6	0.139	2.955	0.011
Acarina	Season	ns	1	6.729	1.896	0.262
Acarina	Species	ns	2	0.515	1.028	0.413
Acarina	Site	ns	3	0.738	0.188	0.899
Acarina	Season*Species	ns	2	0.043	0.362	0.710
Acarina	Season*Site	interaction	3	3.550	30.026	0.001
Acarina	Species*Site	ns	6	0.501	4.237	0.051
Acarina	Season*Species*Site	ns	6	0.118	0.656	0.685
Psocoptera	Season	1 ^a 2 ^b	1	15.833	70.087	0.004
Psocoptera	Species	ns	2	0.463	0.420	0.675

Taxon	Source of variation	Differences	df	MS	F	Probability
Psocoptera	Site	ns	3	0.451	0.370	0.778
Psocoptera	Season*Species	ns	2	0.210	1.946	0.223
Psocoptera	Season*Site	ns	3	0.226	2.091	0.203
Psocoptera	Species*Site	interaction	6	1.101	10.195	0.006
Psocoptera	Season*Species*Site	ns	6	0.108	0.823	0.555
Hemiptera	Season	1 ^a 2 ^b	1	4.300	10.333	0.049
Hemiptera	Species	ns	2	2.197	1.263	0.349
Hemiptera	Site	ns	3	2.492	1.194	0.374
Hemiptera	Season*Species	ns	2	0.209	3.032	0.123
Hemiptera	Season*Site	interaction	3	0.416	6.041	0.030
Hemiptera	Species*Site	interaction	6	1.740	25.253	0.001
Hemiptera	Season*Species*Site	ns	6	0.069	1.129	0.351
Thysanoptera	Season	ns	1	16.872	4.533	0.123
Thysanoptera	Species	ns	2	0.849	1.313	0.337
Thysanoptera	Site	ns	3	0.613	0.149	0.925
Thysanoptera	Season*Species	interaction	2	1.359	5.446	0.045
Thysanoptera	Season*Site	interaction	3	3.722	14.917	0.003
Thysanoptera	Species*Site	ns	6	0.647	2.593	0.136
Thysanoptera	Season*Species*Site	ns	6	0.250	1.400	0.223
Coleoptera	Season	1 ^a 2 ^b	1	3.912	45.524	0.007
Coleoptera	Species	ns	2	0.025	0.030	0.970
Coleoptera	Site	ns	3	0.519	1.056	0.543
Coleoptera	Season*Species	ns	2	0.415	0.953	0.437
Coleoptera	Season*Site	ns	3	0.092	0.211	0.885
Coleoptera	Species*Site	ns	6	0.835	1.916	0.224
Coleoptera	Season*Species*Site	interaction	6	0.436	6.769	0.000
Diptera	Season	ns	1	3.707	4.799	0.116
Diptera	Species	ns	2	0.958	1.140	0.381
Diptera	Site	ns	3	0.105	0.105	0.951
Diptera	Season*Species	ns	2	0.199	0.329	0.732
Diptera	Season*Site	ns	3	0.773	1.275	0.365
Diptera	Species*Site	ns	6	0.841	1.387	0.351
Diptera	Season*Species*Site	interaction	6	0.606	5.584	0.000
Hymenoptera	Season	ns	1	1.411	6.638	0.082
Hymenoptera	Species	ns	2	1.415	0.987	0.426
Hymenoptera	Site	ns	3	0.066	0.073	0.969
Hymenoptera	Season*Species	ns	2	0.149	0.202	0.823
Hymenoptera	Season*Site	ns	3	0.213	0.287	0.833
Hymenoptera	Species*Site	ns	6	1.434	1.938	0.220
Hymenoptera	Season*Species*Site	interaction	6	0.740	10.753	0.000

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

Leaf Blemish Presence / Absence

The only types of leaf blemish that showed any significant difference in their presence between the three mallee species were leaf folding and psyllids (Table 8). *E. loxophleba* subsp. *lissophloia* had significantly more psyllids than either of the other mallee species (P=0.05). In fact, only one of the *E. loxophleba* trees sampled did not have psyllids. *E. kochii* subsp. *borealis* had significantly more leaf folding than the other species. In fact, apart from on one other tree, all leaf folding was found on *E. kochii*. Site also had an influence, with leaf mining being significantly more common at site 2 (McDougall) than all the other sites. Webbing was significantly more common at site 2 than at site 1, with the other sites intermediate between the two.

Table 8: Two-way ANOVA results for effect of mallee species and site on the types of leaf symptoms observed in leaf clipping samples. Samples n/5 trees for each tree species at each site. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Symptom	Source of variation	Differences	df	MS	F	Probability
Leaf blistering	Species	ns	2	0.350	2.333	0.178
Leaf blistering	Site	ns	3	0.133	0.889	0.499
Leaf blistering	Species*Site	ns	6	0.150	0.947	0.471
Galls	Species	ns	2	1.517	3.957	0.080
Galls	Site	ns	3	1.133	2.957	0.120
Galls	Species*Site	interaction	6	0.383	3.067	0.013
Leaf folding	Species	K ^a P ^b L ^b	2	0.517	10.333	0.011
Leaf folding	Site	ns	3	0.017	0.333	0.802
Leaf folding	Species*Site	ns	6	0.050	0.500	0.805
Leaf mining	Species	ns	2	0.317	1.390	0.319
Leaf mining	Site	2 ^a 1 ^b 3 ^b 4 ^b	3	2.061	9.049	0.012
Leaf mining	Species*Site	ns	6	0.228	2.278	0.052
Psyllid	Species	L ^a P ^b K ^b	2	2.867	6.789	0.029
Psyllid	Site	ns	3	0.106	0.250	0.859
Psyllid	Species*Site	interaction	6	0.422	3.167	0.011
Scale	Species	ns	2	0.517	3.720	0.089
Scale	Site	ns	3	0.422	3.040	0.114
Scale	Species*Site	ns	6	0.139	0.575	0.748
Webbing	Species	ns	2	0.200	1.800	0.244
Webbing	Site	2 ^a 4 ^{ab} 3 ^{ab} 1 ^b	3	0.594	5.350	0.039
Webbing	Species*Site	ns	6	0.111	0.444	0.845

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*.

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel),

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

DISCUSSION

Azarbayjani *et al.* (1999) questioned whether the species found on an individual host tree are a random collection of available species or if the unique attributes of each host tree make it suitable for particular arthropod species. Each of the three mallee species used in this study vary in a number of ways, and hence might conceivably have quite different arthropod assemblages. *E. polybractea* for example, was taller than the other mallee species. Studies have shown that larger trees are considered better overwintering sites for beetles (Pollard 1968, Sotherton 1984, Dennis *et al.* 1994), and Lawton (1983) indicated that larger plants were more likely to be found and colonized than smaller ones. Kuris *et al.* (1980) also support the idea that bigger is better in terms of a host species' ability to support a variety of herbivorous arthropods. These studies suggest that being the largest of the three species present, *E. polybractea* should be expected to harbour more beetles, or have a greater diversity or abundance of arthropods than the other mallee species. This however, was not the case. Perhaps the differences in size between the mallees were not large enough to have an impact on arthropod diversity and comparisons with larger trees might produce different results.

Canopy size and structure is also a significant factor influencing the diversity of arthropods encountered according to some sources (Lawton and Price 1979, Campbell and Norman 1989). While the three species differed in their height, they all had similar canopy volumes. The fact they had similar canopy volumes, and they did not differ significantly in their arthropod assemblages, suggests that canopy volume might be a stronger indicator of the abundance and diversity of arthropod assemblages than tree height. This can not be proven in the current study however, since trees were selected to be of a similar canopy size.

While the three mallee species did not differ in their total arthropod abundance or their ordinal richness, there were some small differences in the types of leaf blemishes present

on the different species. Psyllids were more common on *E. loxophleba* than on the other mallee species and leaf folding was almost entirely restricted to *E. kochii*.

Why there were more psyllids on *E. loxophleba* than any of the other mallee species is not clear. However, Yen (2002) advises that many psyllids are specific to a single *Eucalyptus* species, suggesting that species of psyllids specific to the other two mallee species present were not located at these sites. This makes sense, as of the three mallee species, *E. loxophleba* is closest to its natural range in the study area, while *E. polybractea* is native to the eastern states and *E. kochii* has a more northern distribution in Western Australia than the study area. Since *E. loxophleba* is in its native environment, it seems entirely possible that psyllid species which have adapted to specialize on this species should be present in the study area.

E. kochii had more leaf folding than the other mallee species. The leaf folds were observed to be constructed by spiders. The main attraction to *E. kochii* for the leaf folding spiders would most likely be the size of the leaves. *E. kochii* had much more slender leaves which would suit the folding conducted by the spiders for their shelter building. The other mallees had broader leaves, which would be much harder for the small spiders to manipulate. Hence, apart from a single leaf, leaf folding was not encountered on the other mallee species. Work by Lawton (1983) indicated that some arthropod diversity might be explained by the ease with which arthropod species could exploit the different leaf shapes exhibited by host trees species. This supports the contention that the size of the leaves might be the limiting factor in the occurrence of the leaf folding spiders.

While the season of collection had no influence on the abundance of total arthropods or on any particular order, it did have some effect on ordinal richness. Overall, the ordinal richness observed was reduced at the second sampling. The possible reasons for this are varied. Possibly the difference in environmental conditions experienced in the two sampling seasons contributed to this reduction. The first sampling was conducted in spring, while the second sampling occurred during autumn. Hence, it seems likely that a

difference in flowering or some other seasonal factor may be responsible. Flowering records (Appendix G), however, do not support this. Equally as likely is the possibility that the chemical knockdown treatment was too severe for some of the more sensitive orders and hence these groups had not recolonized the area by the time of the second sampling. This is supported by Azarbayjani *et al.* (1999), who found that while common species recolonised an area within two weeks, it could take up to a year for the arthropod community to resemble that present prior to spraying. Interestingly, the fact that there was not a corresponding drop in arthropod abundance suggests that the reduced ordinal richness was a result of losing a small number of rare species or possibly, a result of remaining orders becoming more abundant in their absence. This is supported by Minor *et al.* (2004), who suggest that high levels of diversity simply increase the likelihood of redundancy and compensation. This means that the numbers and activities of species which are lost through habitat disturbance, such as insecticide spraying, could be replaced by other species with similar ecological functions.

As there were very few differences observed between the three mallee species, it will make it relatively simple to compare mallees generally with remnant vegetation. This is important to note, as mallees will be treated as a single entity being compared to two remnant species in the following chapters.

CONCLUSION

In this chapter I asked the question, does the species of oil mallee planted affect the arthropod ordinal richness encountered? The answer is no ... not significantly. While there were minor differences in the presence or absence of some of the leaf blemishes recorded between species, with leaf folding more prevalent on *E. kochii* and psyllids more common on *E. loxophleba*, there was no significant difference between the species in terms of ordinal richness or total abundance of arthropods.

While the lack of significant differences between the mallee species might appear to be a disappointing result, it is actually a benefit in the context of this study. In the next

chapter, mallee species will be treated as a single species (mallee sp.) in comparisons with remnant vegetation. Had the mallee species been significantly different this could have confounded the results in the rest of the thesis.

This chapter has examined the variability of arthropod assemblages in planted mallee vegetation. In Chapter 5 the ordinal richness of canopy arthropod assemblages in both planted mallee and native remnant eucalypt vegetation will be explored and compared.

CHAPTER 5

COMPARISON OF ARTHROPOD BIODIVERSITY IN PLANTED AND REMNANT EUCALYPT VEGETATION

INTRODUCTION

The Western Australian wheatbelt now consists of pockets of natural vegetation, often isolated and degraded, amidst a vast expanse of cleared agricultural land (Bennett 1992). The isolated and fragmented nature of the remaining natural vegetation has made the situation for local wildlife and arthropod populations tenuous. Restoring connectivity of remnants of natural vegetation has become a priority in conservation efforts, as it is believed to significantly improve the prospects of declining populations that are nearing extinction (Saunders 1991, Ralph 1991/92, Bennett 1992, Kavanagh *et al.* 2007). One of the suggested methods of restoring connectivity is to replace lost habitats through tree planting (Bennett 1992).

To date, little research has been done to determine the factors which influence biodiversity in planted eucalypts, and how they compare to the situation in remnant vegetation (Clough *et al.* 2007, Kavanagh *et al.* 2007). Intuitively, we would expect that native remnant vegetation would support a greater diversity of arthropods than any planted vegetation, simply by virtue of the native vegetation being in place for a longer period of time. As paddocks and fields are frequently ploughed and grazed, trees planted within agricultural paddocks must rely on field margin trees and nearby remnants as sources of colonizing arthropods (Clough *et al.* 2007). Many arthropods have small ranges, are poor dispersers and have specific habitat requirements (Clark and Richardson 2002), making the task of recolonisation all the more difficult without the support of within-field refugia.

Tsitsilas *et al.* (2006) and Thomas and Marshall (1999) state that arthropod biodiversity is generally low in agricultural areas, mostly as a result of the homogeneity of the landscape and the alien nature of the vegetation. They suggest that this can be rectified, however, by increasing the vegetational diversity of rural landscapes. This is supported by Clough *et al.* (2007), who found that species richness could be correlated to the amount of “non-crop area”. This can be taken a step further according to Tsitsilas *et al.* (2006), who claim that improving habitat heterogeneity can impact negatively on pest arthropods. For this reason, biodiversity plantings need not be beneficial only to the environment; they may also prove useful in the day - to - day lives of farmers.

In Chapter 4, the effect of oil mallee species on canopy arthropod assemblages was explored. It was determined that the species of oil mallee used has little impact on the arthropod assemblage observed. This chapter will attempt to answer the question:

- Are canopy-dwelling arthropod assemblages present on planted oil mallees similar to those found on remnant eucalypt vegetation?

I plan to achieve this by comparing the arthropod abundance, ordinal richness and diversity of leaf blemishes of two types of eucalypt vegetation, woodland eucalypts in remnant vegetation and mallee eucalypts in farm alley plantings.

METHOD

Five trees of each of two native woodland eucalypt species, *Eucalyptus astringens* (Brown Mallet) and *E. wandoo* subsp. *wandoo* (Wandoo), were selected at three remnant vegetation sites. Remnant trees were selected to be similar in canopy size to the mallee trees and small enough to be accessed with the spraying equipment from the ground. In the adjoining paddock at each site, one of the three oil mallee species used in Chapter 4 was also selected for sampling. These trees were located an average of 50 to 100 metres from the edge of the remnant. The same oil mallee species could not be used at all sites, as sites containing both the desired remnant species and the correct mallee species were

not available. All of the selected trees at each site were located in the same or adjoining paddocks.

The characteristics and dimensions of each tree were measured and recorded (Appendix H). Tree height, width (N-S and E-W), and height of canopy from base were recorded to enable the calculation of canopy volume. As the majority of trees sampled were elliptical in shape, the formula of Thorne *et al.* (2002) was used for calculating an ellipsoid volume [$2/3\pi H(A/2 \times B/2)$], where: H is the height of the plant from the base to the top of the photosynthetically active material; A is the widest diameter of the plant measured on a North-South orientation; and B is the widest diameter of the plant measured on a East-West orientation (at right angles to A). Thorne *et al.* (2002) claim that their formula is “elastic and accurately accommodates a wide range of plant shapes and sizes”. Other factors, such as flowering status, were recorded in case they were later deemed to have an impact on the results (Appendix I).

In October of 2005 (spring), the trees were sampled by canopy spraying, using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] was applied at a rate of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Once collected, samples were stored in 70% ethanol until sorting. In May of 2006 (autumn), the same spraying and collection procedure was repeated on the same trees. Once collected and stored in alcohol, samples were sorted with the aid of a stereo microscope to the ordinal level in the laboratory. Mean and frequency figures from sorted arthropod data are presented in Appendix F. With the exception of beetles (Chapter 8), arthropods were only sorted to ordinal level due to the time and labor constraints experienced during this study.

For each tree, branch clipping samples were also taken to obtain scales, galls, sedentary arthropods and other formations of interest (Appendix G). Samples were collected by

searching each tree for examples of leaf damage and collecting one example of each of the observed leaf damage types from each tree. This was intended to demonstrate the ordinal richness of arthropod fauna not collected by canopy fogging. After field collection, all samples were taken to Curtin University of Technology for sorting and identification.

Data were tested for homogeneity of variance and transformations were done where necessary. Abundance was corrected for canopy volume by using arthropods per cubic metre of canopy volume in the analyses. A series of analyses of variance (ANOVAs) were performed to determine if there were any significant differences between the three tree species in terms of tree height, canopy volume, richness, abundance, individuals per order, and leaf blemishes. The paddock trees were simply treated as ‘mallee’, regardless of which species was involved, since the comparison was designed to compare remnant and planted trees. Two or three-way ANOVAs were conducted on the data collected, with tree species as a fixed factor and site as a random factor. Season was included as a fixed factor in all analyses except the two-way analysis of leaf blemishes as the leaf blemish data were only collected in one season. LSDs were used in post hoc testing to determine where differences lie.

The results presented in this chapter fall into two main categories:

Tree physical data - which presents and compares the two physical aspects of the oil mallees which were deemed to be the most likely to impact on arthropod ordinal richness, namely tree height and canopy volume. Chemical factors such as nutritional status and leaf chemistry will be examined in later chapters.

Arthropod data - which presents the results of arthropod collections by chemical knockdown and leaf clipping. Chemical knockdown results are presented in terms of richness, abundance and individuals within orders. Collections of leaf blemishes caused by sedentary arthropods were obtained by leaf clipping and data are presented as presence / absence data. Seasonal impacts on arthropod collections are also examined in this section.

RESULTS

Tree Height

Tree height was measured for each of the trees sampled for arthropods during this study, at both times of sampling. Figure 25 demonstrates that this was quite variable between the three tree types compared, with *E. astringens* being the lowest and *E. wandoo* being the highest trees. These differences were not as pronounced by the second sampling in May 2006.

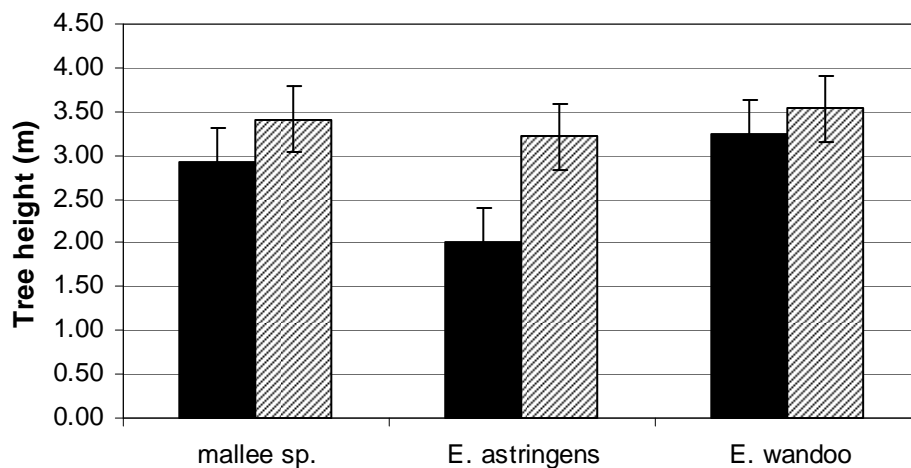


Figure 25: Mean tree height of mallee species and two species of remnant eucalypts as measured in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). The error bars represent standard errors.

Analysis of variance (Table 9) however, showed no significant differences in tree height between the three tree types measured. There was also no significant effect of site or season on tree height.

Table 9: Three-way ANOVA results for effect of tree species, season and site on tree height, with site as a random factor.

Source of variation	Significant differences	df	MS	F	Probability
Site	ns	2	1.993	1.208	0.386
Species	ns	2	0.548	0.346	0.727
Season	ns	1	2.134	9.387	0.092
Site*Species	interaction	4	1.585	9.734	0.024
Site*Season	ns	2	0.227	1.397	0.347
Species*Season	ns	2	0.240	1.472	0.332
Site*Species*Season	ns	4	0.163	0.149	0.963

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

Canopy Volume

Tree canopy volume data, as determined using Thorne *et al.*'s (2002) formula, is presented in Figure 26, with *E. wandoo* exhibiting the highest canopy volume.

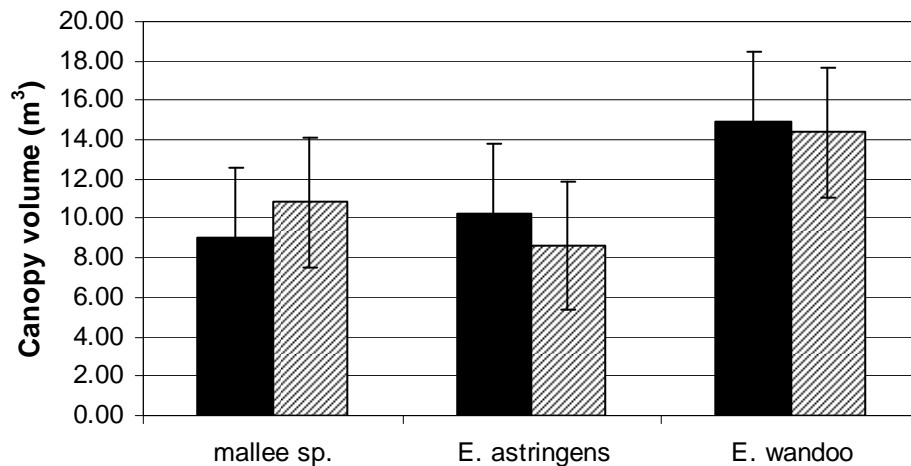


Figure 26: Mean canopy volume of oil mallee species and two remnant eucalypt species as determined from measurements made in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Analysis of variance (Table 10) however, indicated no significant differences in tree canopy volume between the three tree types. There was also no significant effect of site on canopy volume. There was, however, an effect of season, with canopy volume increasing between sampling periods.

Table 10: Three-way ANOVA results for effect of tree species, season and site on canopy volume, with site as a random factor. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right. Log canopy volume was used for this analysis.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	0.547	2.338	0.178
Season	2 ^a 1 ^b	1	0.629	34.882	0.010
Site	ns	3	0.191	0.769	0.548
Site*Species	interaction	6	0.050	70.405	0.000
Season*Species	interaction	2	0.234	14.956	0.005
Site*Season	interaction	3	0.018	5.429	0.038
Site*Species*Season	ns	6	0.003	0.078	0.998

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at $p=0.05$.

* ns = no significant difference at $p=0.05$.

Richness

For this study, richness was measured as the number of arthropod orders collected per tree. Figure 27 shows the average number of orders collected from each of the tree types. The average number of orders collected ranged between 10 and 13 orders, with slight differences in the number of orders collected between seasons.

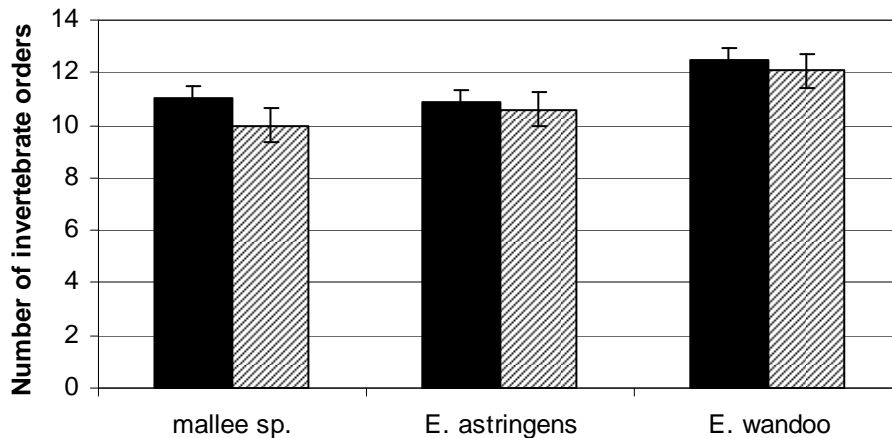


Figure 27: Mean number of arthropod orders collected on remnant eucalypts (*E. astringens*, *E. wandoo*) compared to planted mallee vegetation in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

E. wandoo appears to support the largest number of orders. Analysis of variance (Table 11) however showed this difference not to be significant. Site and season also had no significant effect on the number of orders collected.

Table 11: Three-way ANOVA results for effect of tree species, season and site on number of orders collected, with site as a random factor. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	12.700	1.498	0.297
Season	ns	1	2.133	0.184	0.697
Site	ns	3	33.644	2.038	0.235
Species*Season	ns	2	2.633	0.738	0.517
Species*Site	ns	6	8.478	2.377	0.158
Season*Site	ns	3	11.600	3.252	0.102
Species*Season*Site	interaction	6	3.567	2.942	0.011

* **interaction** = a significant interaction effect was observed at $p=0.05$.

* **ns** = no significant difference at $p=0.05$.

Abundance

The total number of arthropods, also referred to as total abundance, is shown in Figure 28.

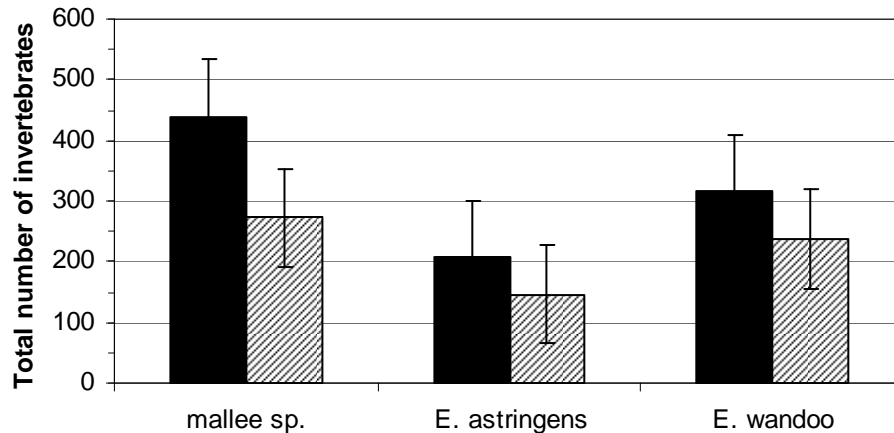


Figure 28: Mean total number of arthropods collected on remnant eucalypts (*E. astringens*, *E. wandoo*) compared to planted mallee vegetation in October 2005 (represented by a dark bar) and May 2006 (represented by shaded bar). Error bars represent standard error.

Figure 28 appears to show a clear difference between the three species in terms of the total number of arthropods collected per tree. Analysis of variance (Table 12) however revealed that there was no significant difference between the three tree species in terms of the total number of arthropods collected. There was also no effect of site or season on total arthropods and no interaction between site and species was observed.

Table 12: Three-way ANOVA results for effect of tree species, season and site on total number of arthropods collected per cubic metre of canopy volume, with site as a random factor. Canopy volume was log transformed for this analysis. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	ns	2	0.302	1.318	0.363
Season	ns	1	0.640	8.257	0.103
Site	ns	2	0.060	0.228	0.806
Species*Sample	ns	2	0.109	2.414	0.205
Species*Site	ns	4	0.230	5.085	0.072
Sample*Site	ns	2	0.078	1.718	0.289
Species*Sample*Site	ns	4	0.045	0.615	0.653

* ns = no significant difference at $p=0.05$.

Individuals within Orders

The numbers of individuals in each order varied widely between the tree species (Figure 29).

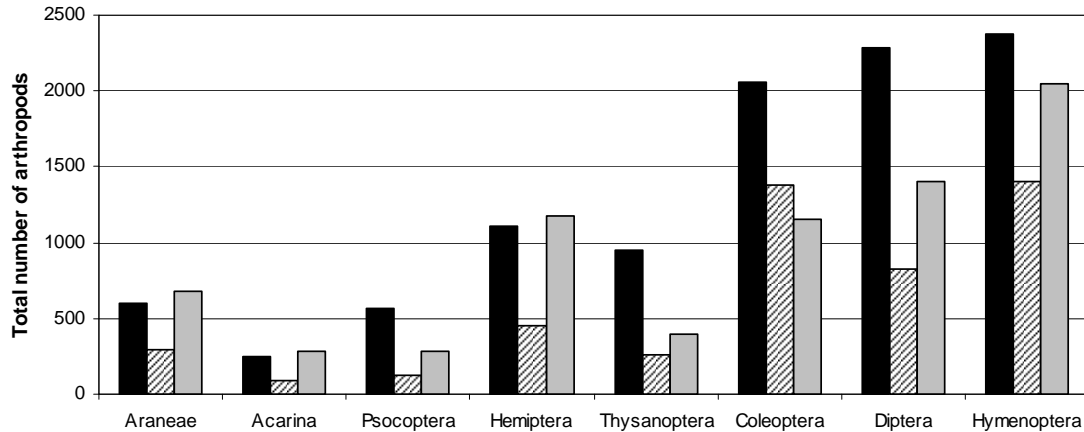


Figure 29: Total number of arthropods collected from particular orders on three tree types; mallee sp. (represented by a dark bar), *E. astringens* (represented by shaded bar) and *E. wandoo* subsp. *wandoo* (represented by a light bar).

Overall, the mallee species tended to have more individuals of each of its representative orders than the other species, while *E. astringens* consistently had the lowest numbers of individuals of the three tree types. However these observed differences were found not to be statistically significant (Table 13, $P=0.05$). There was also no significant effect of site on the abundance of any of the individual orders. Two groups however, were influenced by season. Numbers of spiders were higher at the second sampling, while Hemiptera numbers fell significantly between sampling periods.

Table 13: Three-way ANOVA results for effect of tree type, season and site on numbers of individuals collected from particular orders, with site as a random factor. Abundance was corrected for the canopy volume of the sampled tree. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right

Taxon	Source of variation	Differences	df	MS	F	Probability
Araneae	Season	2 ^a 1 ^b	1	1.407	54.128	0.018
Araneae	Species	ns	2	0.465	3.983	0.112
Araneae	Site	ns	2	0.019	0.158	0.859
Araneae	Season*Species	ns	2	0.023	1.089	0.419
Araneae	Season*Site	ns	2	0.026	1.216	0.387
Araneae	Species*Site	ns	4	0.117	5.456	0.065

Taxon	Source of variation	Differences	df	MS	F	Probability
Araneae	Season*Species*Site	ns	4	0.021	0.254	0.907
Acarina	Season	ns	1	3.310	6.423	0.127
Acarina	Species	ns	2	0.326	1.235	0.382
Acarina	Site	ns	2	0.264	0.451	0.687
Acarina	Season*Species	ns	2	0.463	2.381	0.208
Acarina	Season*Site	ns	2	0.515	2.650	0.185
Acarina	Species*Site	ns	4	0.264	1.358	0.387
Acarina	Season*Species*Site	ns	4	0.194	1.133	0.348
Psocoptera	Season	ns	1	2.509	3.073	0.222
Psocoptera	Species	ns	2	0.433	1.035	0.434
Psocoptera	Site	ns	2	0.291	0.277	0.776
Psocoptera	Season*Species	ns	2	1.037	5.581	0.070
Psocoptera	Season*Site	ns	2	0.817	4.394	0.098
Psocoptera	Species*Site	ns	4	0.418	2.252	0.226
Psocoptera	Season*Species*Site	ns	4	0.186	0.862	0.491
Hemiptera	Season	1 ^a 2 ^b	1	1.661	23.408	0.040
Hemiptera	Species	ns	2	0.543	0.793	0.513
Hemiptera	Site	ns	2	0.247	0.806	0.686
Hemiptera	Season*Species	ns	2	0.249	0.555	0.613
Hemiptera	Season*Site	ns	2	0.071	0.158	0.859
Hemiptera	Species*Site	ns	4	0.685	1.523	0.347
Hemiptera	Season*Species*Site	interaction	4	0.450	4.742	0.002
Thysanoptera	Season	ns	1	6.151	2.761	0.238
Thysanoptera	Species	ns	2	1.019	2.619	0.187
Thysanoptera	Site	ns	2	0.280	0.137	0.881
Thysanoptera	Season*Species	ns	2	0.028	0.048	0.954
Thysanoptera	Season*Site	ns	2	2.228	3.851	0.117
Thysanoptera	Species*Site	ns	4	0.389	0.672	0.645
Thysanoptera	Season*Species*Site	interaction	4	0.579	2.650	0.040
Coleoptera	Season	ns	1	0.709	3.573	0.199
Coleoptera	Species	ns	2	0.213	0.801	0.510
Coleoptera	Site	ns	2	0.202	0.562	0.618
Coleoptera	Season*Species	ns	2	0.469	4.458	0.096
Coleoptera	Season*Site	ns	2	0.199	1.886	0.265
Coleoptera	Species*Site	ns	4	0.265	2.522	0.196
Coleoptera	Season*Species*Site	ns	4	0.105	0.831	0.510
Diptera	Season	ns	1	2.856	10.436	0.084
Diptera	Species	ns	2	0.448	1.672	0.297
Diptera	Site	ns	2	0.750	2.471	0.357
Diptera	Season*Species	ns	2	0.767	3.220	0.147
Diptera	Season*Site	ns	2	0.274	1.148	0.404
Diptera	Species*Site	ns	4	0.268	1.126	0.456
Diptera	Season*Species*Site	ns	4	0.238	1.319	0.271
Hymenoptera	Season	ns	1	0.000	0.001	0.976
Hymenoptera	Species	ns	2	0.500	1.332	0.360

Taxon	Source of variation	Differences	df	MS	F	Probability
Hymenoptera	Site	ns	2	0.148	0.355	0.720
Hymenoptera	Season*Species	interaction	2	0.369	9.197	0.032
Hymenoptera	Season*Site	ns	2	0.083	2.061	0.243
Hymenoptera	Species*Site	interaction	4	0.375	9.340	0.026
Hymenoptera	Season*Species*Site	ns	4	0.040	0.479	0.751

* 1 = season 1 (October 2005); 2 = season 2 (May 2006).

* interaction = a significant interaction effect was observed at p=0.05.

* ns = no significant difference at p=0.05.

Leaf Blemish Presence / Absence

The different types of leaf blemish did not exhibit any significant difference in their occurrence on one species or another (Table 14). There was also no statistical effect of site on the presence or absence of the various leaf blemishes.

Table 14: Two-way ANOVA results for effect of tree type and site on the types of leaf symptoms observed in leaf clipping samples. Samples n/5 trees for each tree species at each site. Superscripts indicate significant differences, with values decreasing from left to right.

Symptom	Source of variation	Differences	df	MS	F	Probability
Leaf blistering	Species	ns	2	0.022	0.250	0.790
Leaf blistering	Site	ns	2	0.022	0.250	0.790
Leaf blistering	Species*Site	ns	4	0.089	1.00	0.420
Galls	Species	ns	2	0.089	1.000	0.444
Galls	Site	ns	2	0.156	1.750	0.284
Galls	Species*Site	ns	4	0.089	1.143	0.352
Leaf folding	Species	ns	2	0.022	0.182	0.840
Leaf folding	Site	ns	2	0.156	1.273	0.373
Leaf folding	Species*Site	ns	4	0.122	1.571	0.203
Leaf mining	Species	ns	2	0.200	0.545	0.617
Leaf mining	Site	ns	2	0.267	0.727	0.538
Leaf mining	Species*Site	ns	4	0.367	2.063	0.106
Psyllid	Species	ns	2	0.267	1.000	0.444
Psyllid	Site	ns	2	0.000	0.000	1.000
Psyllid	Species*Site	ns	4	0.267	1.714	0.168
Scale	Species	ns	2	0.200	0.429	0.678
Scale	Site	ns	2	0.867	1.857	0.269
Scale	Species*Site	interaction	4	0.467	5.250	0.002
Webbing	Species	ns	2	1.356	3.813	0.118
Webbing	Site	ns	2	0.089	0.250	0.790
Webbing	Species*Site	ns	4	0.356	1.882	0.135

* interaction = a significant interaction effect was observed.

* ns = no significant difference at p=0.05.

DISCUSSION

Work by Lawton (1983) indicated that, in general, larger sized trees were more complex than smaller ones, and hence had a more diverse suite of arthropods. This was mainly due to the greater variety of resource types found in larger and more complex vegetation. This however, was not supported by the current study. The tree physical data collected showed that there was no significant difference in tree height between the three vegetation types observed in this chapter, mostly because some effort was made to select trees for similarity of size. Canopy volume was also not significantly different between sites and species. It did, however, increase between seasons, indicating that the trees grew somewhat during the study period.

Arthropod ordinal richness and abundance were not significantly different between the three species or between sites or seasons either. When it came to leaf blemishes, there were also no significant differences between the species, sites or seasons. It appears that there is even less difference between the tree species examined in this chapter than there was in the previous chapter analyzing only mallee species. According to Lawton (1983), more arthropod species should be supported by host plants with greater architectural complexity. While work by Humphrey *et al.* (1999) suggested that differences in canopy structure and light interception might be important in determining arthropod species diversity. This, however, was not supported by the current study, despite *E. loxophleba* being relatively architecturally simple, with a spreading habit and open canopy, it was able to support the same richness and abundance of arthropods as the far more complex and established remnant vegetation.

Thomas and Marshall (1999) found that higher floral diversity, as present in the remnant vegetation examined here, increased arthropod diversity. This is also not supported by the current work, as no significant difference was seen between the single species planted mallee vegetation and the floristically diverse remnant. Clough *et al.* (2007) suggest that the composition of the vegetation matrix could play an important role in

colonization by arthropods, indicating that mallees located near the remnant stands could have experienced more rapid recolonization than mallees located away from remnants. Perhaps the greater diversity of arthropods supported by the remnant was able to be transferred to the mallee species, which were located close to the remnants in this study. Kavanagh *et al.* (2007) support this, claiming that efforts to reverse degradation using eucalypt plantings would be more likely to succeed if trees were planted close to remnant vegetation. In the current study, had the mallees sampled at the remnant sites been planted further from the remnants, benefits in terms of arthropod ordinal richness and abundance gained from their proximity to the remnant might have been reduced. Had this been the case, differences between the two vegetation types might have been greater.

Another possible reason for the lack of variation between the tree species examined in this chapter could be a low level of specificity of the arthropods encountered in this area. Alternatively, the assertion of Novotny *et al.* (2002) that most specialization is at the genus level, occurring most often in large genera (such as *Eucalyptus*) could prove correct in this instance. Since the current study used five *Eucalyptus* species, all from the same *Symphomyrtus* subgenus, it seems unlikely that any specialisation present in the arthropod assemblage would be detected in a species level comparison of tree species. This, combined with the low level of arthropod identification used in this study, make it difficult to detect differences in arthropod richness between the host tree species. This does not however, explain the similarities observed in arthropod abundance between the mallee and remnant vegetation types.

Season of sampling also had no statistical impact on the richness or abundance of arthropods collected. Some small impacts of season were observed, however, in numbers of individual orders. Numbers of Araneae were greater at the second sampling in autumn than at the first sampling, while there were less Hemiptera at the autumn sampling than at the first sampling in spring. It is difficult to explain the increase in Araneae, except to suggest perhaps the increase in canopy volume between seasons provided more resources for herbivores which, in turn, supported an increase in Araneae.

The reduction in Hemiptera could be a direct result of the increase in spider numbers. However, it seems superficial to suggest this is the only reason for the change. Perhaps, to some degree, the drop in Hemiptera could be attributed to seasonal changes or the influence of insecticide residues. Certainly more fresh new foliage would be available in spring than in autumn, which would support higher numbers of Hemiptera. The possibility of residues affecting Hemiptera numbers is also a valid one, as the insecticide used in this study has a field half life of 90 days (Appendix B). This means that depending on weather conditions, some residue might have remained six months later at the second sampling event. Predatory spiders would not be as likely to be influenced by pesticide residues as Hemiptera, which are predominately phytophagous. Another possibility is that Hemiptera might simply take longer to recolonise an area after knockdown sampling than Araneae. This is supported by Azarbayjani *et al.* (1999), who found that while some species could recolonise an area within a fortnight, other species could take up to a year to return following a disturbance such as insecticide knockdown.

The results of this chapter are somewhat disappointing, as expected differences between mallee and remnant vegetation did not eventuate. This can, however, be seen in a positive light, if the differences between these species in terms of richness and abundance are so small, this bodes well for the success of revegetation and conservation efforts aimed at supporting biodiversity in agricultural areas. Kavanagh *et al.* (2007) agree, deeming any eucalypt plantings useful in improving agricultural landscapes by adding to and supporting remnant vegetation and biodiversity as a whole. They believe that even the types of eucalypt plantings used in this work, which generally consist of only a single species, are still important in the conservation of birds and other fauna in rural areas. This indicates that while mallees may not be the ultimate solution, they have an important role to play in agroecosystems.

CONCLUSION

In this chapter I asked the question, does whether a host plant is a planted eucalypt or remnant eucalypt affect the arthropod ordinal richness and abundance encountered? The answer is No...not significantly. Arthropod richness and abundance were not significantly different between the three species or between sites or seasons for that matter. Leaf blemishes and their associated sedentary arthropods also showed no significant differences between the species, sites or seasons.

Even the season of sampling had no statistical impact on the richness or abundance of arthropods collected. Some small impacts of season were observed however, in numbers of individual orders with numbers of Araneae greater at sampling period 2 than at the first sampling, and numbers of Hemiptera decreasing between the first and second sampling. These are very minor differences and suggest an influence of climatic changes between seasons or chemical residues from the first sampling.

The results of this chapter are somewhat disappointing as very few differences between the mallee and remnant vegetation types were evident. However it is encouraging to note that small differences between these species in terms of richness and abundance suggest that mallees might be a good fit for revegetation programs aimed at supporting biodiversity in agricultural areas.

This chapter has compared the ordinal richness and abundance of arthropod assemblages in both planted mallee and remnant vegetation. In Chapter 6 the potential effect of soil and leaf nutrition on canopy arthropod ordinal richness and abundance will be explored.

CHAPTER 6

THE EFFECT OF SOIL AND LEAF NUTRITION ON CANOPY-DWELLING ARTHROPOD ASSEMBLAGES

INTRODUCTION

In Chapter 5, arthropod biodiversity in planted and remnant eucalypt vegetation was compared. In this chapter, some of the factors which might contribute to differences in arthropod assemblages on different eucalypt species will be explored. In 1988, Majer and Recher suggested that there were a number of possible reasons for observed differences in arthropod abundance and ordinal richness. These included the nutrient level of the foliage, secondary plant compounds, and the structure of the leaves. The levels of nitrogen and other nutrients in foliage are widely reported to be important factors contributing to arthropod nutrition, growth, and grazing patterns (Gordon 1972, Fox and Macauley 1977, Slansky and Feeny 1977, White 1978, Basset 1992). Soil nutrient levels are also considered to be important (Braithwaite *et al.* 1983, Landsberg *et al.* 1990, Blanche and Westoby 1995) due to their influence on plant nutrition. Work by Fox and Morrow (1992) found that the effects of poor soil nutrition were aggravated by the presence of insects, while Hanover (1975) linked the nutritional status of trees to their levels of insect resistance.

This chapter will attempt to answer two questions:

- Do soil nutrient levels influence canopy-dwelling arthropod assemblages?
- Do leaf nutrient levels influence canopy-dwelling arthropod assemblages?

It is intended that this will be done by comparing the canopy arthropod assemblages present in both natural and planted eucalypt stands and relating them to the levels of soil and leaf nutrients present.

METHOD

Seven experimental sampling sites were set up in the localities of Narrogin, Wickiepin, and Pingelly in the wheatbelt of Western Australia. Four of these sites, Parnell, McDougal, Hassel, and Marshall, consisted of alleys of oil mallee vegetation from three different species: *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*. Of these four sites, two were on a sandy soil, while the other two were on a slightly heavier loamy, sand soil. A further three sites, Sprigg, Hesford, and Tutanning, were selected in native vegetation remnants, either on farmers' properties, or in reserves. The remnant vegetation at each of these sites contained two native species: *Eucalyptus wandoo* subsp. *wandoo*, and *E. astringens*. Each of these sites also had planted oil mallee vegetation, containing either *E. polybractea*, or *E. loxophleba* subsp. *lissophloia* in an adjoining paddock. This was done as three sites containing the selected remnant species as well as all the same mallee species were not available.

At each site, five trees of each of the three species present were selected for arthropod sampling. In October of 2005 (spring), the trees were sampled by canopy spraying using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] was applied at a concentration of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Once collected, samples were stored in 70% ethanol until sorting. In May of 2006 (autumn), the same spraying and collection procedure was repeated on the same trees. The analysis presented in Chapter 3 indicates that sampling of the same trees after the lapse of six months is unlikely to affect the composition of arthropods at the ordinal level. Once collected and stored in alcohol, samples were sorted in the laboratory with the aid of a stereo microscope to ordinal level. Samples were only sorted to ordinal level due to the time and labor constraints of this study.

For each tree, samples were also taken of scales, galls, sedentary arthropods and other formations of interest, by branch clipping. This was intended to demonstrate the ordinal richness of arthropod fauna not collected by canopy fogging. After field collection, all samples were taken to Curtin University of Technology for sorting and identification.

Leaf and soil samples were taken at each site in order to determine the levels of nutrition available to herbivorous arthropods. At each site, 50 leaves (bulked from five trees) of each of the tree species present were collected and placed in paper bags. Leaves were collected from all over the tree canopy using pole mounted secateurs in order to obtain a representative sample of the range of leaf ages and quality. Newly emerged and senesced leaves, however, were not collected as these leaves were not suitable for the chemical analyses used. The bags of leaf material were kept in a cooler box until they were delivered to Cumming Smith British Petroleum (CSBP) for analysis. Analyses tested for levels of macro nutrients such as nitrogen, phosphorus and potassium, as well as micronutrients. Soil samples were also taken at each site. Samples were taken to 20 cm depth using a soil auger under and around each of the tree species present. Three samples were taken and bulked per tree species at each site. Samples were stored in plastic bags and delivered to CSBP for analysis. Soil samples were tested for macronutrients such as nitrogen, phosphorus and potassium as well as micronutrients, pH and conductivity. The number of samples taken was limited due to the cost of analysis.

Data were tested for homogeneity of variance and transformations were done where necessary. Analyses of variance ANOVAs were performed to determine if there were any significant differences between the seven sites in terms of soil nutrition. ANOVAs were also used to determine if there were any significant differences between the tree species with regard to soil nutrition, and leaf nutrition. One way ANOVAs were conducted because of the unbalanced nature of the experimental design. This was unavoidable due to the scarcity of suitable sites containing all of the required tree species. LSDs were used in post hoc testing to determine where differences lie.

Coefficient of correlation analyses calculated by Pearson's product-moment method were also conducted to ascertain the relationships between soil nutrients and leaf nutrition, and also between soil and leaf nutrient levels with arthropod richness and abundance. Because of the unbalanced nature of the experimental design, some of the analyses involving both site and species required the mallee and remnant sites to be examined separately to maintain reasonable degrees of freedom.

The results presented in this chapter fall into three categories:

Soil nutrition - which presents and compares the levels of the three main macronutrients present in soil: nitrogen, phosphorus and potassium. The effect of site on nutrient levels is also explored.

Leaf nutrition - which presents and compares the levels of the three main nutrients present in leaf samples: nitrogen, phosphorus and potassium. The effect of site and tree species on nutrient levels is also explored.

Arthropod data - which examines the influence of soil and leaf nutrient levels on arthropod richness and abundance.

RESULTS

Soil Nutrition

Analyses of macronutrients at the four mallee sites and three remnant sites were compared. Only macronutrients are examined in this chapter, although the full set of soil data can be seen in Appendix J. Soil nitrogen ranged between 1 and 5 mg/kg, with three of the mallee sites, but not site 4, tending to have higher levels than two of the remnant sites (Figure 30).

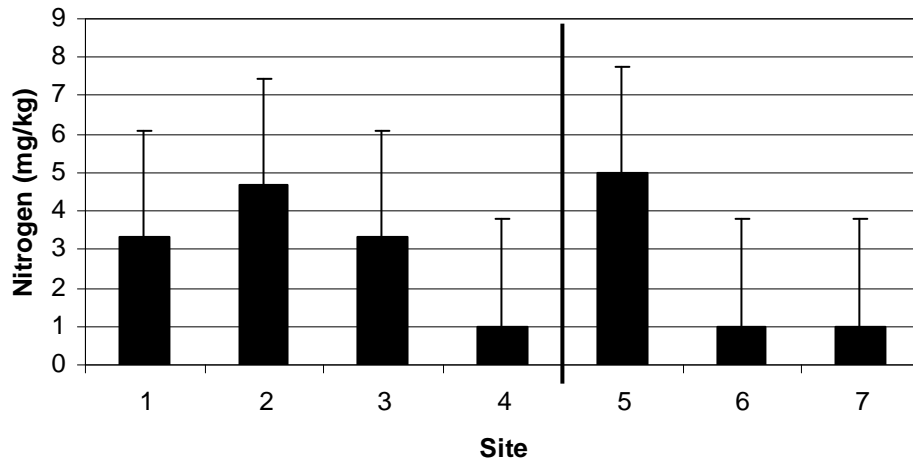


Figure 30: Soil nitrogen levels determined from bulked soil samples, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Soil phosphorus (Figure 31) ranged between 8 and 45 mg/kg, with the mallee sites consistently having numerically higher levels than the remnant sites.

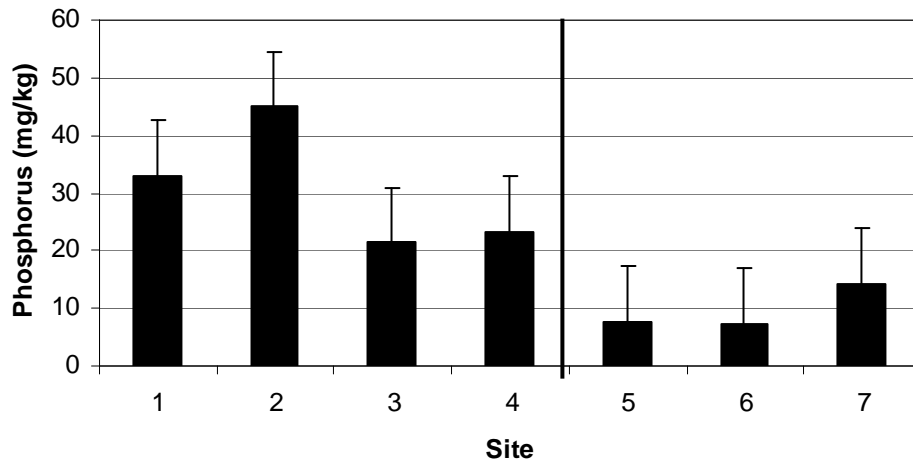


Figure 31: Soil phosphorus levels determined from bulked soil samples, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Soil potassium ranged between 40 and 160 mg/kg, with the remnant sites consistently having higher potassium levels than the mallee sites (Figure 32).

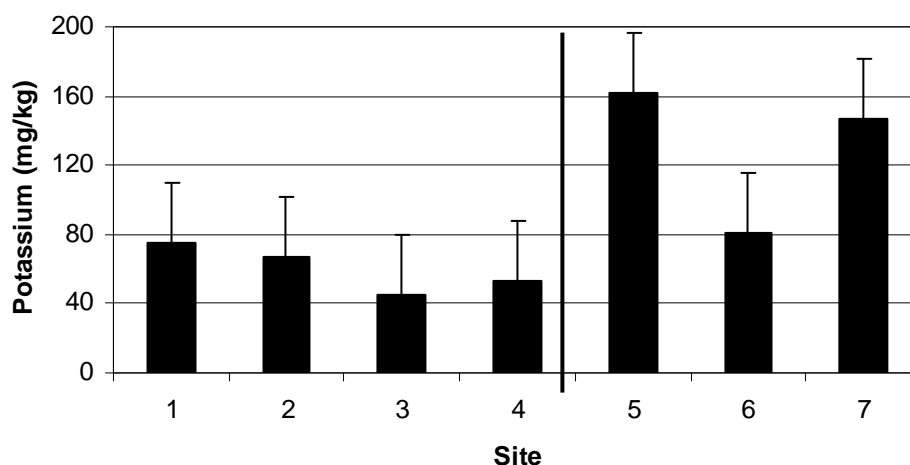


Figure 32: Soil potassium levels determined from bulked soil samples, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Analyses of variance (Table 15) examined the significance of the relationships between site and soil nutrients. The analysis found that there was no significant difference in the levels of soil nitrogen present at the seven sites. Two of the mallee sites (McDougall and Parnell) had significantly higher ($P < 0.05$) levels of phosphorus than the other mallee sites and the remnant sites. In terms of soil potassium, two of the remnant sites (Tutanning and Hesford) had significantly higher ($P < 0.05$) levels than the other remnant (Sprigg) and the mallee sites.

Table 15: One-way ANOVA results for effect of site on soil nutrients. Superscripts indicate significant differences (LSD, $p=0.05$), with values decreasing from left to right.

Soil nutrient	Significant differences	df	MS	F	Probability
Soil Nitrogen	ns	6	9.302	0.804	0.583
Soil Phosphorus	2 ^a 1 ^{ab} 4 ^{bc} 3 ^{bc} 7 ^{bc} 5 ^c 6 ^c	6	565.492	4.095	0.014
Soil Potassium	5 ^a 7 ^{ab} 6 ^{bc} 1 ^{bc} 2 ^c 4 ^c 3 ^c	6	6255.381	3.469	0.026

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

* ns = no significant difference at $p=0.05$.

Leaf Nutrition

Leaf nutrients at the four mallee sites and the three remnant sites were individually compared, as well as with all sites combined. Only macronutrients are considered in this

chapter, although the full set of data can be seen in Appendix K. There seemed to be very little variation in leaf nitrogen between sites, with values ranging between 0.7 and 1.1 percent dry weight (Figure 33).

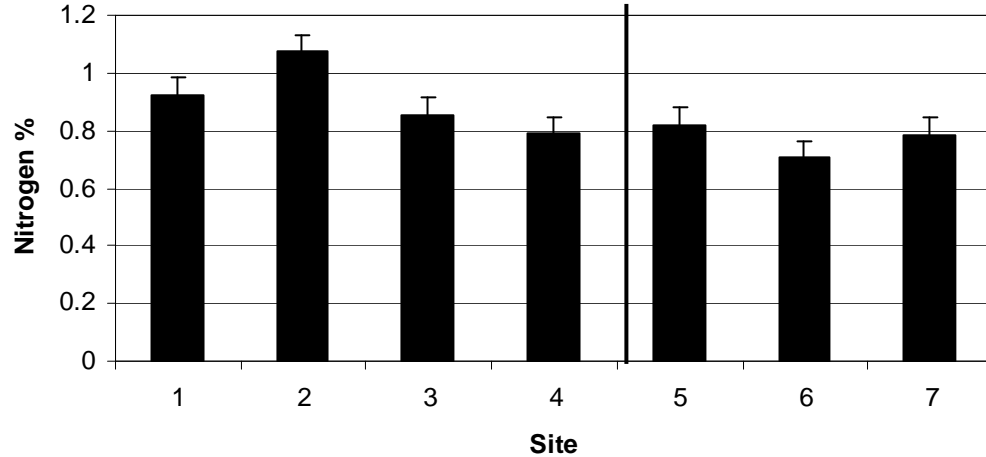


Figure 33: Leaf nitrogen levels determined from bulked leaf samples collected from throughout the canopy, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Leaf phosphorus ranged between 0.05 and 0.13% and appeared to be consistently higher in the mallee than in the remnant vegetation (Figure 34).

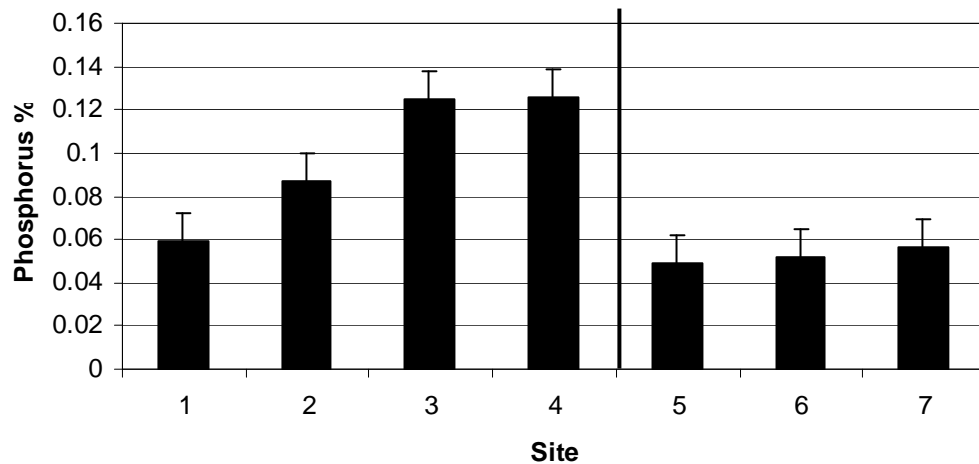


Figure 34: Leaf phosphorus levels determined from bulked leaf samples collected from throughout the canopy, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Leaf potassium ranged between 0.4 and 0.6%, with no appreciable difference between the sites being apparent (Figure 35).

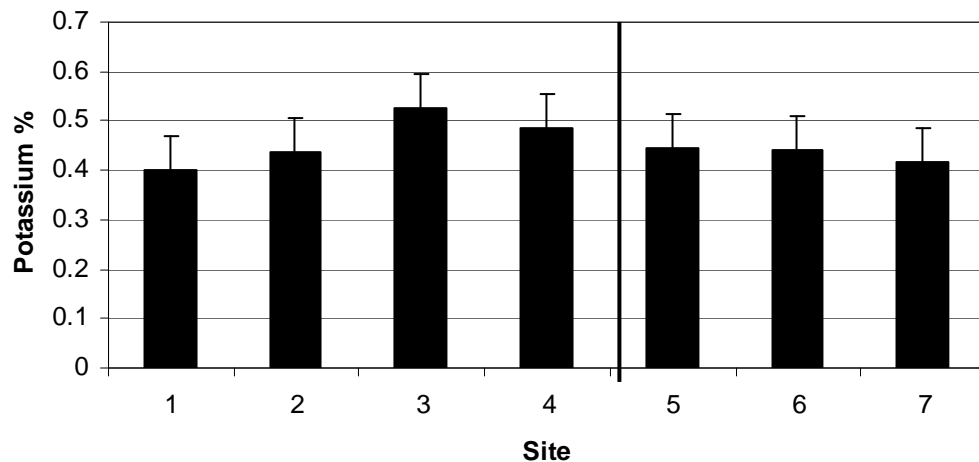


Figure 35: Leaf potassium levels determined from bulked leaf samples collected from throughout the canopy, n = 3. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

As the results could be confounded by different species at different sites, mallees and remnant species were analysed separately. No interaction terms were used due to potential confounding of the results. Analysis of variance (Table 16) showed that there was no significant difference in the levels of leaf nitrogen present at the remnant sites or between the remnant species. However, nitrogen levels at the mallee site 2 (McDougall), were higher ($P < 0.05$) than at the other mallee sites. Also *E. loxophleba* subsp. *lissophloia* was found to have significantly higher levels of leaf nitrogen than the other mallee species. Analysis of variance (Table 16) also showed that there was no significant difference in the levels of leaf phosphorus present at the remnant sites. However, at the mallee sites, two sites (Hassel and Marshall) had significantly higher ($P < 0.05$) levels of leaf phosphorus than the other sites. There was no effect of tree species on leaf phosphorus at the mallee sites, but within the remnant sites, *E. astringens* and *E. wandoo* had significantly ($P < 0.05$) lower levels of leaf phosphorus than the mallee species. Analysis of variance (Table 16) showed that there was no significant difference in the levels of leaf potassium present at the remnant or mallee sites. There was also no significant difference in leaf potassium between either the mallee or remnant tree species.

Table 16: One-way ANOVA results for effect of site and tree species on leaf nutrients. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Nutrient	Habitat	Source	Significant differences	df	MS	F	Probability
Leaf N	Mallee	Species	L ^a P ^b K ^b	2	0.024	17.720	0.003
Leaf N	Remnant	Species	ns	2	0.000	0.046	0.956
Leaf N	Mallee	Site	2 ^a 1 ^{ab} 3 ^b 4 ^b	3	0.045	32.971	0.000
Leaf N	Remnant	Site	ns	2	0.011	2.148	0.232
Leaf P	Mallee	Species	ns	2	0.000	1.111	0.389
Leaf P	Remnant	Species	M ^a W ^b A ^b	2	0.000	32.469	0.003
Leaf P	Mallee	Site	4 ^a 3 ^a 2 ^b 1 ^b	3	0.003	9.322	0.011
Leaf P	Remnant	Site	ns	2	0.000	3.875	0.116
Leaf K	Mallee	Species	ns	2	0.014	2.107	0.203
Leaf K	Remnant	Species	ns	2	0.010	6.215	0.059
Leaf K	Mallee	Site	ns	3	0.009	1.339	0.347
Leaf K	Remnant	Site	ns	2	0.001	0.405	0.692

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species.

* ns = no significant difference at p=0.05.

The relationship between soil and leaf nutrients was examined by coefficient of correlation (Table 17). At the remnant sites the only significant relationship was a strong positive correlation between soil phosphorus and leaf phosphorus. At the mallee sites there was a significant positive correlation between soil phosphorus and leaf nitrogen. When the data for all sites were combined, the only significant correlations were a strong positive one between soil phosphorus and leaf nitrogen and a negative one between soil potassium and leaf phosphorus.

Table 17: Coefficient of correlation results for the relationship between soil nutrients and leaf nutrients. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Habitat type	Soil nitrogen	Soil phosphorus	Soil potassium
Leaf nitrogen	Remnant	-0.15	0.29	0.26
Leaf phosphorus	Remnant	0.20	0.71	-0.05
Leaf potassium	Remnant	0.03	0.20	-0.06
Leaf nitrogen	Mallee	0.50	0.69	0.11
Leaf phosphorus	Mallee	-0.10	-0.29	-0.53
Leaf potassium	Mallee	-0.43	-0.47	0.12
Leaf nitrogen	Combined	0.26	0.75	-0.25
Leaf phosphorus	Combined	0.06	0.34	-0.57
Leaf potassium	Combined	-0.21	-0.13	-0.09

Arthropod Data

The influence of soil nutrients on arthropod abundance and richness was examined by coefficient of correlation (Table 18). At the remnant sites, there were no significant relationships between soil nutrients and arthropod richness or abundance. There were also no correlations between soil nutrients and arthropod richness or abundance at the mallee sites. When all the sites were examined together, the only significant correlations were a positive one between soil phosphorus and arthropod abundance and a negative one between soil potassium and arthropod abundance.

Table 18: Coefficient of correlation results for effect of soil nutrients on arthropod richness and abundance. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Habitat type	Nitrogen	Phosphorus	Potassium
Richness (orders)	Remnant	-0.63	-0.37	-0.25
Abundance	Remnant	-0.31	0.02	-0.25
Richness (orders)	Mallee	0.23	0.46	0.40
Abundance	Mallee	0.11	0.47	-0.23
Richness (orders)	Combined	-0.11	0.25	0.00
Abundance	Combined	0.04	0.60	-0.49

The influence of leaf nutrients on arthropod richness and abundance was also examined by coefficient of correlation (Table 19). At the remnant sites there were no significant relationships between leaf nutrients and arthropod richness or abundance. At the mallee sites, there was a significant positive correlation between leaf nitrogen and arthropod ordinal richness. There was also a significant negative correlation between leaf phosphorus and arthropod ordinal richness at the mallee sites. When all data were combined, the only significant relationships were a positive one between leaf nitrogen and arthropod abundance and a negative one between leaf phosphorus and arthropod ordinal richness.

Table 19: Coefficient of correlation results for effect of leaf nutrients on arthropod richness and abundance. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Habitat type	Nitrogen	Phosphorus	Potassium
Richness (orders)	Remnant	-0.08	-0.48	-0.57
Abundance	Remnant	-0.01	0.42	0.20
Richness (orders)	Mallee	0.61	-0.72	-0.28
Abundance	Mallee	0.54	-0.39	-0.35
Richness (orders)	Combined	0.42	-0.44	-0.33
Abundance	Combined	0.61	0.17	-0.12

DISCUSSION

Of the soil nutrients examined, nitrogen was not significantly different between the sites. The fact that some differences in arthropod assemblages were observed between certain sites and tree species suggests that soil nitrogen might not be the most important influence on canopy arthropod biodiversity. This is somewhat surprising as soil fertility, in general, is considered to be important to canopy arthropods. This is especially true for sedentary arthropods like galls. Work by Blanche and Westoby (1995) indicated that numbers of gall species were lower at high fertility sites and that sites with low soil fertility tended to have more complex galls than more fertile sites. Although effects on galls were not specifically examined in this work, some arthropods were influenced by soil and leaf nutrition. The ordinal method of measuring biodiversity used in this study was, however, very crude and may mask trends which could exist at the species level. This possibility will be explored further in the chapter that examines beetles down to species level (Chapter 8).

While soil nitrogen did not vary statistically between the sites, soil phosphorus and soil potassium did. Sites 1 (Parnell) and 2 (McDougall) had higher levels of soil phosphorus than the others when all sites were compared. Both sites 1 and 2 were mallee sites and these were located on a lighter sandy soil when compared to the loamy sand soil of the other mallee sites (Marshall and Hassel). Perhaps soil type can, in part, explain this difference in soil phosphorus. Soil phosphorus levels at these sites might also be associated with the use of phosphorus fertilizers on crops sown between the mallee alleys. Elevated levels of soil phosphorus were associated with higher arthropod abundance, suggesting that this nutrient may be a key influence on the variability in arthropod abundance between mallee sites. Also of interest was a positive correlation between soil phosphorus and higher levels of leaf nitrogen, which was also associated with higher arthropod abundances. It could be argued then that fertilizer use could have

improved leaf quality, either structurally and/or chemically, which in turn might have led to greater arthropod abundances (Fox and Morrow 1992).

Sites 5 (Tutanning) and 7 (Hesford) had higher levels of soil potassium than all the other sites. Both of these sites were remnant ones, suggesting further variation in soil type between these two sites and the others. Gourley (1999) suggests that potassium tends to increase with soil clay content, so perhaps higher clay content in the remnant soils is responsible for the higher levels of soil potassium observed. Another possibility is that soil potassium tends to be greater in soils which have not been cleared for agriculture. This is supported by Nye and Greenland (1964), who found that following clearing, nutrient levels in the soil depleted rapidly due to a combination of leaching and erosion. Low levels of potassium found at mallee sites could also be explained by their history as agricultural paddocks. The removal of soil nutrients in harvested crops is well recognized (Nye and Greenland 1964, Stoorvogel *et al.* 1993). This effect is exacerbated in the case of soil potassium by the limited application of fertilizers containing this element (Stoorvogel *et al.*). During the study period the managers of the mallee sites did not apply potassium fertilisers to their tree plantings. However farm records indicate that paddocks adjoining the two remnant sites which showed the highest levels of soil potassium had received applications of potash fertilizer in recent years. While the remnants themselves were not fertilized, it is possible that they were impacted by runoff from the nearby farmland (Pettit *et al.* 1998).

Leaf nutrition was more variable between both sites and tree species than was soil nutrition. Mattson (1980) regards the levels of leaf nutrients, especially nitrogen, to be important factors in the growth and development of herbivorous arthropods. One of the main findings of this chapter was that, while soil nitrogen apparently had little or no effect, high levels of leaf nitrogen were associated with greater arthropod numbers and in some instances higher arthropod ordinal richness. Nitrogen, in particular, has been implicated as a limiting factor in many plant-herbivore relationships (Soo Hoo and Fraenkel 1966a, Soo Hoo and Fraenkel 1966b, Gordon 1972, Fox and Macauley 1977, Slansky and Feeny 1977, Onuf 1978, White 1978). In fact, according to Mattson (1980),

“Some writers believe that plants with less than 1.8% nitrogen are a substandard food resource.” Site 2 (McDougall) had greater levels of leaf nitrogen than any of the other sites, perhaps related to the greater levels of soil phosphorus and nitrogen, probably resulting from fertilizers having been applied at this site. The higher arthropod numbers observed could be due to the higher nutritional quality of the leaves attracting, and being able to sustain, greater numbers of leaf-eating arthropods and, in turn, their predators. Basset (1992) found that most herbivorous arthropods were found wherever large numbers of high quality (nitrogen and water levels), young leaves were available. Moran and Hamilton (1980) claim that young foliage is more nutritious and so is more attractive to herbivores. Some researchers (Fox and Macauley 1977, White 1978) believe that nitrogen levels are an important determinant of arthropod grazing, while others claim that, apart from Ohmart’s study of chrysomelids (1991), there is little evidence to suggest that nitrogen limits arthropod numbers in eucalypts. Work by Fox and Macauley (1977) indicated that some arthropods merely ate more leaf tissue in order to compensate for low leaf nitrogen.

Leaf phosphorus, was greater at sites 3 (Marshall) and 4 (Hassel) than any of the other sites. These were mallee sites with slightly heavier soil. Overall, the mallee species, whether *E. loxophleba* subsp. *lissophloia*, *E. kochii* subsp. *borealis*, or *E. polybractea*, had higher levels of leaf phosphorus than the tree species in the remnant vegetation, regardless of whether they were at a mallee or remnant site. This suggests a possible difference in the biology or physiology of these plants, although the difference could also be a function of the different structure of the vegetation i.e. alleys versus woodland. Different leaf nutrient levels between species located in the same area might be related to differences in their root physiologies or their systems of nutrient storage (Majer *et al.* 1992). Differences in leaf nutrients between forests, on the other hand, could be due to different nutrient levels in the soils of the forests (Braithwaite *et al.* 1983, Braithwaite 1986, Landsberg *et al.* 1990). There is also the possibility, of course, that this can all be attributed to differences in fertilizer use. This suggestion is supported by work conducted by Fox and Morrow (1992) which found that levels of foliage nutrients were improved by the application of a balanced (NPK) fertilizer. In the current, study high

levels of leaf phosphorus were associated with lower arthropod ordinal richness, it is unclear why this might be the case, but it is possible that fertilizer use has enhanced conditions for a small number of common arthropod orders, causing them to dominate food resources and consequently excluding some of the rarer arthropod orders.

Despite differences between sites in terms of soil potassium levels, there were no significant differences in leaf potassium between the seven sites. Leaf potassium was also not significantly different between the tree species. This may, be due to low availability of the potassium present in the soil, which could be bound up in forms not able to be taken up by plants. Alternatively, it may simply be that potassium is not limiting to plant growth in these areas and the higher levels of potassium found at some sites are excess to plant requirements. This is supported by Gourley (1999) who claims many Australian soils are typically moderate to high in available and exchangeable potassium.

The results in this chapter are complicated by the number of variables interacting with each other. High leaf nitrogen was correlated with high soil phosphorus, which begs the question does soil phosphorus increase leaf nitrogen or is this simply a coincidence? Higher soil phosphorus was generally associated with mallee sites, indicating a possible effect of fertilizers. Higher soil potassium was associated with remnant sites, indicating that vegetation on a particular soil type tended to be left as remnant vegetation, rather than being cleared for agriculture. Leaf phosphorus tended to be high when soil potassium was low, perhaps indicating an effect of soil type. Overall, leaf nutrition was lower in the remnant species than the mallees, again perhaps as a result of fertilizer application. It is difficult to determine if soil or leaf nutrients, or the interaction of the two, are responsible for the differences observed in arthropod numbers. Further, there are other factors yet to be examined such as the effect of secondary plant compounds, which will be examined in Chapter 7.

CONCLUSION

In this chapter, I attempted to determine if soil or plant nutrition might play a role in the variation in arthropod abundance and ordinal richness described in previous chapters. It appears that arthropod abundance, in particular, is related to soil and leaf nutrient levels. There was, however, only limited evidence of increased arthropod ordinal richness in response to greater nutrient levels. Generally, high nutrient levels tended, instead, to reduce arthropod ordinal richness. Of the soil nutrients, phosphorus was the most important, with high levels of phosphorus tending to relate to higher arthropod abundances. For leaf nutrients, phosphorus and nitrogen were important, with high levels of phosphorus being associated with lower ordinal richness, while high levels of leaf nitrogen were related to higher arthropod abundances. As high soil phosphorus and high leaf nitrogen tended to occur together, it is difficult to say whether one or the other is responsible for increases in arthropod abundance, though intuitively one would suggest that good soil nutrition led to enhanced plant quality, which in turn has increased arthropod abundance. In Chapter 7, the role of leaf essential oils and other secondary plant compounds in determining arthropod abundance and ordinal richness will be examined.

CHAPTER 7

THE INFLUENCE OF LEAF ESSENTIAL OILS AND SECONDARY PLANT COMPOUNDS ON CANOPY- DWELLING ARTHROPOD ASSEMBLAGES

INTRODUCTION

In Chapter 6 the effects of soil and leaf nutrition on canopy arthropods were investigated. This chapter examines how secondary plant compounds influence this component of the fauna by asking the questions:

- Do leaf essential oils influence canopy-dwelling arthropod assemblages?
- Do secondary plant compounds, specifically sideroxylonals, influence canopy-dwelling arthropod assemblages?

It is intended that this will be done by comparing the canopy arthropod assemblages present in both natural and planted eucalypt stands and relating them to the levels of leaf essential oils and sideroxylonals present. I will begin with a discussion of the importance of leaf chemistry.

Importance of Leaf Chemistry

A number of studies have been conducted into the influence of eucalypt leaf chemistry on herbivores (Edwards 1993, Stone and Bacon 1994, Provenza *et al.* 2003). In 1959, Fraenkel suggested that the reason for the very existence of secondary plant compounds was to serve as repellants and attractants for insect herbivores. Bennett and Wallsgrave (1994) claim that many leaf chemicals have a role in defense against herbivores, pests and pathogens. However, according to Ohmart and Edwards (1991) the compounds found in eucalypts have little effect on the survival, performance and defoliation levels of arthropod herbivores.

According to Edwards (1993), eucalypts contain high levels of terpenoids, often referred to as essential oils, in their leaves but only recently has there been any evidence that they have any influence on herbivores. In their work on leaf essential oils, Wildy, Pate and Bartle (2000) found that out of 63 discernable compounds, the most ubiquitous was 1,8-cineole, with a number of other compounds, including α -pinene, present at lower concentrations. In some studies, it has been shown that high levels of 1,8-cineole can act as a feeding deterrent or anti-feedant agent against most insects, while a number of other compounds are effective against vertebrates (Edwards 1993, Stone and Bacon 1994, Provenza *et al.* 2003). Edwards' (1993) study indicated that trees with high levels of the terpenoid 1,8-cineole, in their leaves experienced very little defoliation, while trees with low cineole were heavily grazed. This is supported by work on *Eucalyptus camaldulensis* performed by Stone and Bacon (1994), who found that trees with higher levels of 1,8-cineole were less affected by insect herbivores than those with lower levels of the compound. Arthropods, and insect herbivores in particular, have developed a range of methods for dealing with these chemicals (Morrow *et al.* 1976, Ohmart and Edwards 1991, Lawler and Foley 2002). Some tolerate them by keeping consumption within certain limits, some avoid them by feeding around oil glands in the leaf, while others have more advanced methods including sequestering, metabolizing or even detoxifying chemicals which they are regularly exposed to as part of their regular diet.

The sideroxylonals are a group of formylated phloroglucinol compounds found in many species of the genus *Eucalyptus* (Wallis and Foley 2005). Close and McArthur (2002) suggest that, in general, phenolics have a negative influence on herbivores. Eschler *et al.* (2000) found that formylated phloroglucinol compounds (FPCs) in *Eucalyptus* leaves have a range of important biological functions, including influencing feeding behaviour in marsupial folivores. The most widespread of these FPCs is the sideroxylonals. Work by Moore *et al.* (2005) found that feeding by koalas on *E. melliodora* was negatively affected by sideroxylonals. To date, however, little work has been done on the effects of sideroxylonals on arthropod herbivory. This is supported by Morrow and Fox (1980), who suggest that while correlations have been found between sideroxylonal

concentrations and defoliation levels, it cannot yet be said that sideroxylonals are the cause of deterrence in insect resistant trees. Some workers even suggest that phenolics, such as sideroxylonals, can act to stimulate feeding by arthropod herbivores (Heron 1965, Bernays 1981).

Eucalyptus leaves tend to have high levels of both terpenes (essential oils) and phenolic compounds in their leaves, both of which have been implicated in determining the acceptability and nutritional quality of foliage to herbivores (Hume 1982). Provenza *et al.* (2003) highlight the fact that all plants contain some level of toxins, so complete avoidance is not practical. They suggest that interactions between nutrients and toxins may influence food and habitat selection, and thus tend to regulate plant-herbivore interactions. This is supported by Atsatt and O'Dowd (1976), who indicate that while there are many chemical defenses plants can use against herbivores, very few are lethal. Most act only to modify behaviour by reducing feeding rates or causing avoidance. They suggest that unpalatable plants or plant parts may be excluded from a herbivore's 'optimal diet'; however, this does not mean that they will never be consumed. When alternative food resources are plentiful, unpalatable and toxic plants will be avoided, however, even the most toxic species will be eaten to some extent under conditions of extreme food shortage. This chapter will compare the levels of some of the common secondary plant compounds known to occur in eucalypts and assess how these levels might influence arthropod assemblages.

METHOD

Seven experimental sampling sites were set up in the localities of Narrogin, Wickepin, and Pingelly in the wheatbelt of Western Australia. Four of these sites, Parnell, McDougal, Hassel, and Marshall, consisted of alleys of oil mallee vegetation from three different species: *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*. A further three sites, Sprigg, Hesford, and Tutanning, were selected in native vegetation remnants, either on farmers properties, or in reserves. The remnant

vegetation at each of these sites contained two native species: *Eucalyptus wandoo* subsp. *wandoo* and *E. astringens*. Each of these sites also had planted oil mallee vegetation containing either *Eucalyptus polybractea*, or *E. loxophleba* subsp. *lissophloia* nearby in an adjoining paddock. This layout of sites was selected as enough sites containing the selected remnant species, as well as, all the same mallee species were not available.

At each site, five trees of each of the three species present were selected for arthropod sampling. In October of 2005 (spring), the trees were sampled by canopy spraying, using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] was applied at a rate of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Once collected, samples were stored in 70% ethanol until sorting. In May of 2006 (autumn), the same spraying and collection procedure was repeated on the same trees. Once collected and stored in alcohol, samples were sorted with the aid of a stereo microscope to the ordinal level in the laboratory. Samples were only sorted to ordinal level due to the time and labor constraints experienced during this study.

Prior to spraying for the first invertebrate sampling in October 2005, leaves were collected for terpenoid (essential oil) analysis. The leaves were collected and analysed following the method outlined in Ammon *et al.* (1985). Ten leaves per tree were removed using pole-mounted secateurs so as not to disturb arthropods. Leaves could not be removed after spraying, as this could interfere with the chemical analysis. In the field, 3 g (wet weight) of leaf material from each of the trees was added to individual bottles with 50 ml of ethanol previously weighed into them. Bottles were left for at least 10 days to maximize the effectiveness of the solvent extraction process, before continuing with the analysis. In the laboratory, the bottles were reweighed and the mass of fresh leaf material calculated. The solution was then analysed for both water and oil to determine which essential oils were present in the samples from each tree and in what proportions.

Bottles containing no leaf material were used as controls to confirm that no significant loss of solvent occurred during sampling in the field. Samples were analysed using vapour-phase chromatographic analysis on a Varian Aerograph 2700 at the Department of Environment and Conservation (DEC) in Perth, Western Australia. All analyses were carried out three times and the data were averaged. Component oils were identified by their retention times and by co-injection with a reference compound, and confirmed by gas chromatography-mass spectrometry (Ammon *et al.* 1985).

In May 2006, prior to the second sampling for arthropods, leaves were taken for formylated phloroglucinol (sideroxylonal) analysis. Leaves were collected, processed and analysed as per the protocol described in Wallis and Foley (2005) at the Australian National University (ANU). Thirty leaves per tree were very carefully removed using pole-mounted secateurs in order not to disturb the arthropods. Leaves were not removed after spraying, as this might interfere with the chemical analysis. Foliage was freeze-dried and ground to pass through a 1 mm sieve using a Cyclotech 1093 Mill. Then 50 mg of the ground foliage was sonicated in 8 g of 7% water in acetonitrile (with 0.1% trifluoroacetic acid and 0.3000 g/L of the internal standard, 2-ethyl phenol) for 5 minutes. The extract (0.45 µm) was then filtered into an autosampler vial for analysis by reversed phase HPLC. Samples were analysed isocratically on a Waters Alliance HPLC system using 7% water in acetonitrile with 0.1% trifluoroacetic acid. The analysis produced total sideroxylonals contents of the leaves, as well as percentages of the A, B and C forms of sideroxylonal.

Data were tested for homogeneity of variance and transformations were done where necessary. Two-way analyses of variance (ANOVAs) were conducted on the data collected with site as a random factor and species as a fixed factor. ANOVAs were used to determine if there were any significant differences between the field sites and tree species in terms of leaf pinene, cineole, and sideroxylonal. LSDs were used in post hoc testing to determine where differences lie. Coefficient of correlation analyses calculated by Pearson's product-moment method were also conducted to ascertain the effects of

essential oil and sideroxylonal levels on arthropod richness and abundance, and also on selected arthropod orders.

The results presented in this chapter fall into three categories:

Essential Oils - which presents and compares the levels of the two main terpenes: cineole and pinene. The effect of site and tree species on oil levels is also explored.

Sideroxylonal - which presents and compares the levels of this major phenol group. The effect of site and tree species is also explored.

Arthropod Data - which examines the influence of essential oils and sideroxylonals on arthropod richness and abundance, as well as on selected arthropod orders.

RESULTS

Essential Oils

The levels of various essential oils were measured by gas chromatography. Gas chromatographic (GC) conditions and results are presented in Appendix L. Individual gas chromatograms are also available on CD in Appendix L. Only the two major leaf essential oils, as determined by the analysis (comprising over 55% of total leaf oils in most cases), are examined here, namely 1,8-cineole and α -pinene. Figure 36 presents the average levels of cineole in each of the tree species used in this study. This shows that, in general, mallees had greater levels of cineole than the remnant species, with *E. kochii* having the highest and *E. astringens* the lowest leaf cineole levels.

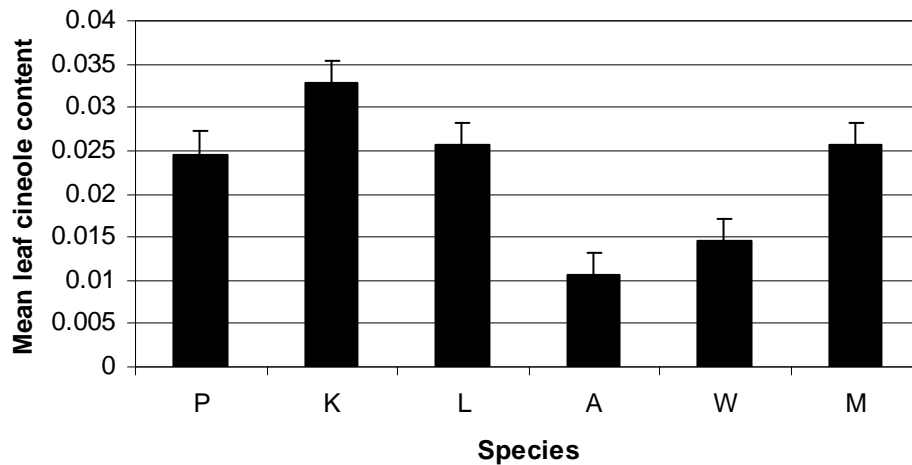


Figure 36: Mean leaf cineole content determined from bulked leaf samples collected from throughout the canopy, n = 5. Species labels: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Error bars represent standard error.

There was also considerable variability in the levels of leaf cineole between sites (Figure 37). The mallee sites (with only mallee vegetation) had higher levels of leaf cineole than remnant sites, where there were two remnant and one mallee species.

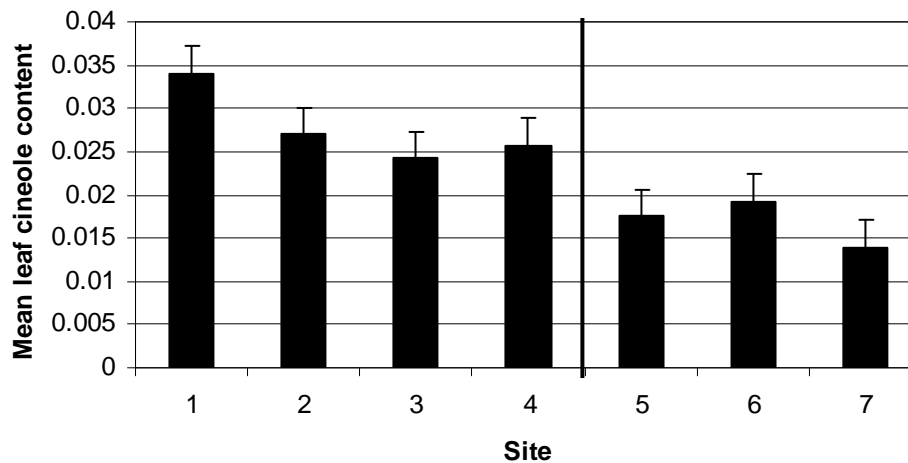


Figure 37: Mean leaf cineole content determined from bulked leaf samples collected from throughout the canopy, n = 5. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Analysis of variance (Table 20) showed that these differences were significant ($P < 0.05$). Mallee sites consistently had higher leaf cineole levels than remnant sites, with site 1 (Parnell) having the highest levels of leaf cineole, which were significantly greater than

at any other site. Regardless of the site, all of the mallee species had higher levels of leaf cineole than the two remnant species, *E. astringens* and *E. wandoo*. Within the mallees, *E. kochii* had significantly higher leaf cineole than the other mallee species, while the other mallees were not significantly different from each other in terms of their leaf cineole content. No significant interaction between site and species was observed in terms of leaf cineole.

Table 20: Two-way ANOVA results for effect of site and tree species on leaf cineole content. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Site	1 ^a 2 ^b 4 ^b 3 ^{bc} 6 ^{cd} 5 ^d 7 ^d	5	0.000	4.416	0.001
Species	K ^a L ^b M ^b P ^b W ^c A ^c	4	0.001	13.388	0.000
Site*Species	ns	10	0.000	1.091	0.379

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species.

* ns = no significant difference at p=0.05.

Figure 38 presents the average levels of pinene in each of the tree species used in this study. There is a wide variability in the levels of leaf pinene, with *E. astringens* and *E. loxophleba* having the highest levels, while *E. polybractea* and *E. kochii* had very low levels.

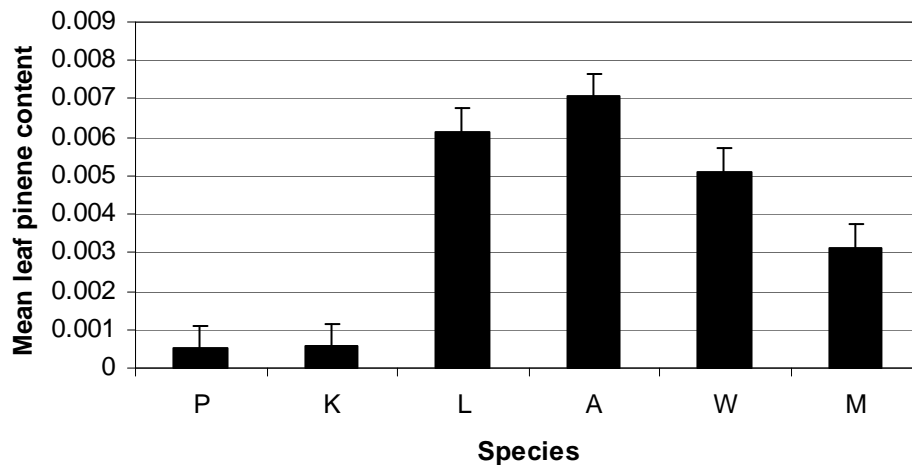


Figure 38: Mean leaf pinene content determined from bulked leaf samples collected from throughout the canopy, n = 5. Species labels: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Error bars represent standard error.

There was also a considerable difference in the levels of leaf pinene between sites (Figure 39). Mallee sites had consistently lower levels of leaf pinene than the remnant sites.

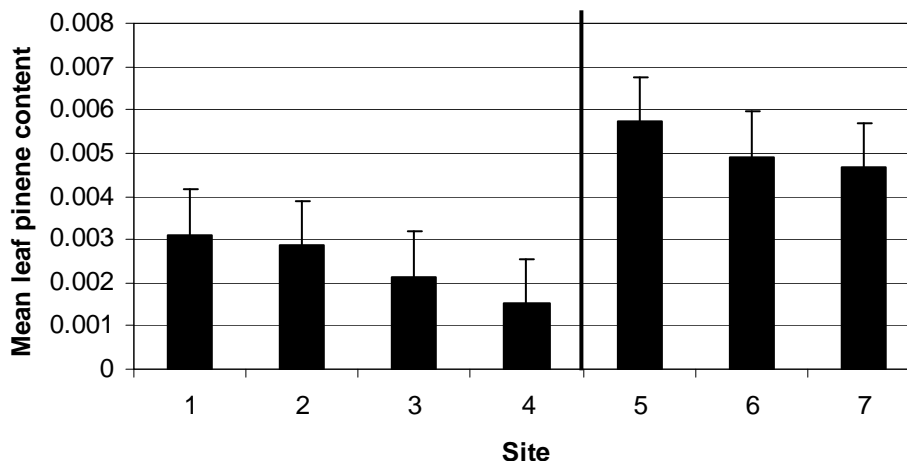


Figure 39: Mean leaf pinene content determined from bulked leaf samples collected from throughout the canopy, n = 5. Site labels 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Sites 1-4 mallee sites, 5-7 remnant sites. Error bars represent standard error.

Analysis of variance (Table 21) indicated that these differences were significant ($P < 0.05$). Mallee sites consistently had lower leaf pinene levels than remnant sites. In the case of tree species, the pattern was less clear, with one mallee, *E. loxophleba*, and one remnant species, *E. astringens*, showing the highest levels of pinene, whilst the other mallees, *E. polybractea* and *E. kochii*, had the lowest leaf pinene levels. There was also significant site by species interaction. When the data were analysed again in a one-way design, however, the relationships of the sites and species with leaf cineole levels were the same.

Table 21: Two-way ANOVA results for effect of site and tree species on leaf pinene content. Superscripts indicate significant differences (LSD, $p = 0.05$), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Site	5 ^a 6 ^{ab} 7 ^b 1 ^c 2 ^c 3 ^{cd} 4 ^d	5	0.000	3.480	0.007
Species	A ^a L ^a W ^b M ^c K ^d P ^d	4	0.000	72.520	0.000
Site*Species	interaction	10	0.000	5.934	0.000

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species.

* interaction = a significant interaction effect was observed at $p = 0.05$.

Sideroxylonals

The content of various formylated phloroglucinol compounds (FPCs) were analysed by HPLC. Only the most ubiquitous of these compounds in *Eucalyptus* species, the sideroxylonals, are examined here. Levels of sideroxylonals obtained are presented in Appendix M. There are three common types of sideroxylonals: A, B and C. However, data are presented here as total sideroxylonals. Figure 40 presents the mean levels of sideroxylonals in each of the tree species used in this study. Levels of sideroxylonals were extremely variable between species, with *E. loxophleba* having the highest levels and both *E. polybractea* and *E. kochii* containing no sideroxylonals.

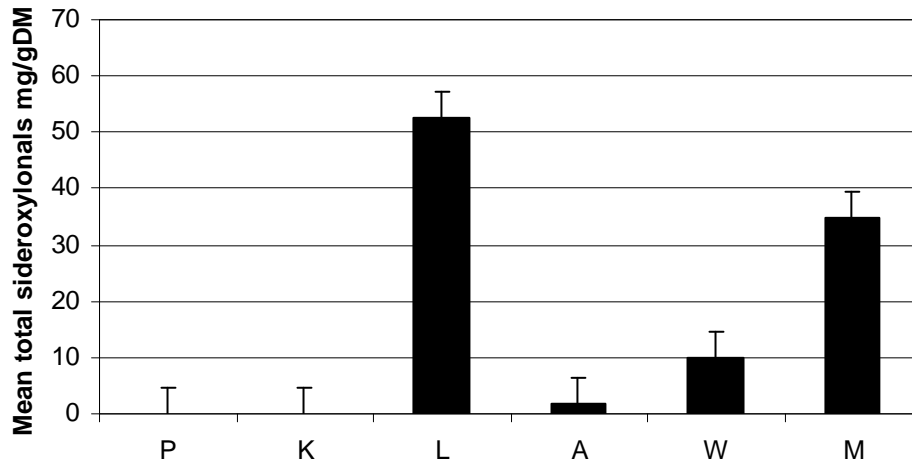


Figure 40: Mean total sideroxylonal levels determined from bulked leaf samples collected from throughout the canopy, n = 5. Species labels: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Error bars represent standard error.

Analysis of variance (Table 22) found some highly significant ($P < 0.01$) differences between the tree species in relation to their sideroxylonal levels. *E. loxophleba* had significantly higher levels of sideroxylonals than any of the other species, followed by the ‘mallee’ species (which was predominately *E. loxophleba*), while the other mallees and *E. astringens* had the lowest levels. A significant interaction between site and species was also observed. However, further testing showed that this did not significantly alter the outcome of the analysis.

Table 22: Two-way ANOVA results for effect of site and tree species on total leaf sideroxytonals. Superscripts indicate significant differences (LSD, p=0.05), with values decreasing from left to right.

Source of variation	Significant differences	df	MS	F	Probability
Species	L ^a M ^b W ^c A ^d P ^d K ^d	4	11393.711	136.754	0.000
Site*Species	interaction	10	727.489	8.732	0.000

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species.

* interaction = a significant interaction effect was observed at p=0.05.

Arthropod Data

The influence of essential oils on arthropod abundance and richness was examined by coefficient of correlation (Table 23). The essential oils that were tested did not appear to strongly affect arthropod ordinal richness in either a positive or negative way. Arthropod abundance however was negatively correlated with pinene levels and positively correlated with cineole content.

Table 23: Coefficient of correlation results for effect of essential oils on arthropod richness and abundance. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Pinene	Cineole
Richness (orders)	-0.17	0.16
Abundance	-0.20	0.40

The influence of sideroxytonals on arthropod abundance and richness was also examined by coefficient of correlation (Table 24). The sideroxytonals did not appear to have any strong relationship with arthropod abundance in either a positive or negative way. Arthropod ordinal richness, however, was negatively correlated with total sideroxytonals.

Table 24: Coefficient of correlation results for effect of sideroxytonals on arthropod richness and abundance. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Total Sideroxytonals
Richness (orders)	-0.28
Abundance	0.07

The effect of the essential oils on some of the most prevalent herbivorous and predatory arthropod orders collected was also explored. Figure 41 graphically presents the relationships between the five selected arthropod orders and the essential oils cineole and pinene.

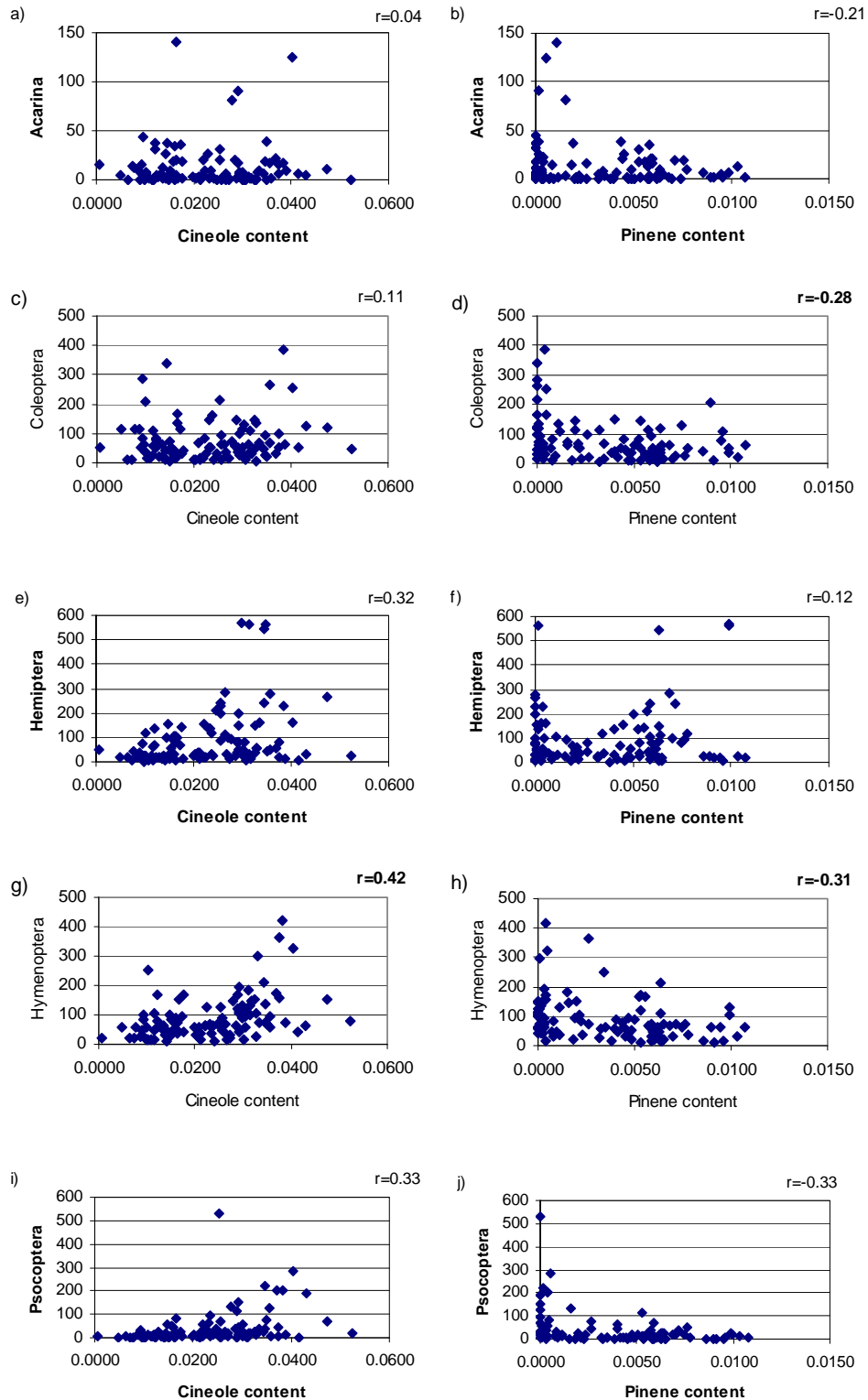


Figure 41: Effect of leaf cineole content on a) Acarina, c) Coleoptera, e) Hemiptera, g) Hymenoptera and i) Psocoptera; and effect of leaf pinene content on b) Acarina, d) Coleoptera, f) Hemiptera, h) Hymenoptera and j) Psocoptera. Values of $r > 0.195$ indicate significant relationships at the $P < 0.05$ significance level for all analyses.

The effect of the sideroxylonals on some of the most prevalent herbivorous and predatory arthropod orders collected was also explored. Figure 42 compares the relationships between the five selected arthropod orders and the sideroxylonals.

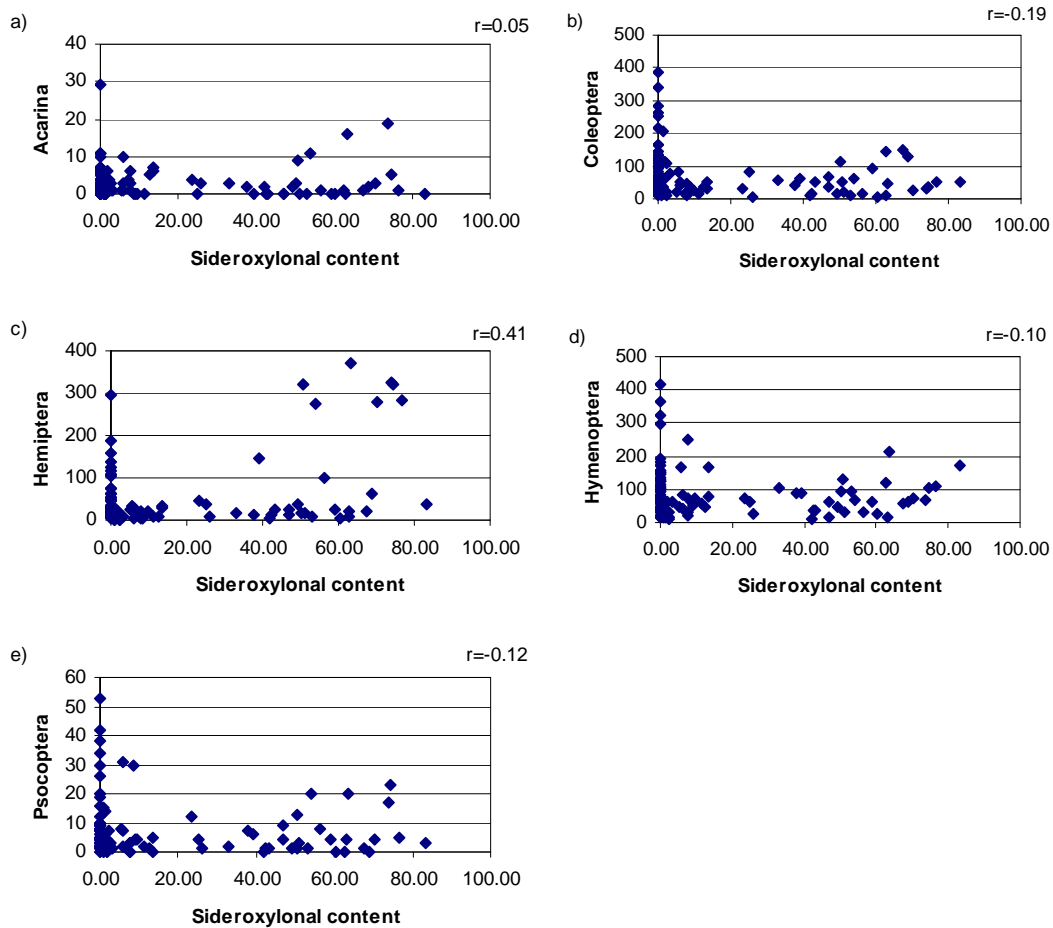


Figure 42: Effect of leaf sideroxylonal content on a) Acarina, b) Coleoptera, c) Hemiptera, d) Hymenoptera and e) Psocoptera. Values of $r > 0.195$ indicate significant relationships at the $P < 0.05$ significance level for all analyses.

The significance of the relationships observed in Figures 41 and 42 were explored using coefficient of correlation analyses. These analyses showed that cineole levels were significantly positively correlated with Hemiptera, Hymenoptera and Psocoptera. Pinene levels, on the other hand, were significantly negatively correlated with Acarina, Coleoptera, Hymenoptera and Psocoptera. The only significant correlation with sideroxylonal levels was a positive one with Hemiptera.

DISCUSSION

The attractiveness of plants to invertebrates is relative, and changes with the availability and palatability of surrounding plants to herbivores, (Atsatt and O'Dowd 1976) and with fluctuations in prey populations for predators. It is widely accepted that secondary plant compounds are one of the determining factors in the attractiveness of plants to herbivores. This is because secondary plant chemicals are known to have adverse effects on the physiology and development of many herbivores, influencing feeding behaviour, and even acting as poisons in some instances (Chapman and Blaney 1979, Wiggins *et al.* 2006). Other studies, however, suggest that these compounds can also have attractant effects on some phytophagous insect species (Bernays and Woodhead 1982, Bernays 1981, Heron 1965). Atsatt and O'Dowd (1976) assert that only a few chemical defenses are lethal, while most influence the behaviour of herbivorous arthropods, causing them to exclude certain plants or plant parts from their diet. In turn, anything that influences the abundance and behaviour of herbivores has the potential to influence predators.

In the present study, one of the major secondary plant compounds tested for was the terpenoid 1,8-cineole. The remnant species, *E. astringens* and *E. wandoo*, had lower levels of this essential oil than the mallee species. *E. kochii* had the highest levels of all the species tested, followed by *E. loxophleba*. The most obvious explanation for this is the simple genetic variation in the production of leaf essential oils between species, although it might be argued that the exposure of mallee species to fertilizers could also be responsible because of higher availability of resources required for production of secondary plant compounds (Coley *et al.* 1985). Studies by Dudt and Shure (1994), however, found that while the use of fertilizers increases plant biomass, it has no apparent influence on levels of leaf chemicals or on arthropod herbivory. Studies conducted by Edwards (1993) found that trees with high levels of the terpenoid 1,8-cineole in their leaves experienced very little defoliation by Christmas Beetles compared to those with lower cineole levels. This is not supported by the current work, however,

as there was a significant positive relationship between leaf cineole content and arthropod abundance.

The levels of the other terpenoid tested, pinene, did not follow the same pattern as that of 1,8-cineole. The mallee species, *E. polybractea* and *E. kochii*, had very low levels of leaf pinene, while the remnant species, *E. astringens*, had the highest pinene levels, followed by another mallee, *E. loxophleba*. The remnant sites had higher levels of leaf pinene compared to the mallee sites. This was due to the high levels exhibited by the remnant species, *E. astringens* and by *E. loxophleba*, the latter being present as the mallee species comparison at two of the three remnant sites. Variability in leaf pinene contents between species could help to explain their variability in arthropod abundances, as there was a significant negative relationship between leaf pinene content and arthropod abundance.

While leaf cineole and pinene contents had no impact on arthropod ordinal richness, when individual orders were examined, some significant relationships could be seen. Cineole had a generally positive influence, with high levels being correlated with high numbers of Hemiptera, Hymenoptera, and Psocoptera, while pinene had a generally negative effect, with high levels being correlated with low numbers of Acarina, Coleoptera, Hymenoptera and Psocoptera. There also appears to be a balance between the two essential oils which enhances these effects. It appears that low levels of pinene, coupled with high cineole could possibly be stimulatory to arthropods. The opposite scenario, of high levels of pinene with low cineole, may inhibit arthropods, reducing their abundance and the prevalence of certain orders. One explanation for the lack of significant impacts of essential oils on arthropod assemblages might be that anti-herbivore effects may only become apparent if a threshold in total oil concentration is exceeded (Morrow and Fox 1980). This is important to note, as tree species, and even the individual trees, in this study varied in their total oil concentrations.

The sideroxylonals, a group of formylated phloroglucinol compounds found in many species of the genus *Eucalyptus* (Wallis and Foley 2005), were the other major

secondary plant compound examined in this chapter. In this study, *E. loxophleba* had very high sideroxylonals, while the other mallee species had none. The two remnant species, *E. astringens* and *E. wandoo*, had lower levels. Wallis and Foley (2005) also found that the mallee species, *E. loxophleba*, had very high levels and claimed that its leaves could contain up to 100 mg/g DM of sideroxylonals. One reason why the remnant species might contain more sideroxylonals than two of the three mallee species is that shade-tolerant, slow growing species tend to have higher levels of phenolics, like sideroxylonals, than fast growing shade-intolerant species (Dudt and Shure 1994). The remnant environment is consistently more shaded than the open paddocks, where the mallees are planted and the open woodlands and low shrublands where they naturally occur. Despite this, the current study found no significant effect of total leaf sideroxylonals on arthropod abundance. Arthropod ordinal richness however, was significantly negatively correlated with total sideroxylonals.

Although phenols like sideroxylonals are commonly regarded as feeding deterrents for phytophagous insects, this is not supported by the current study, as the sideroxylonals did not affect arthropod abundance, except for a positive correlation with Hemiptera numbers. Furthermore, there have been instances where phenols (like sideroxylonal) have been used, rather than avoided by arthropods. Work by Bernays and Woodhead (1982) indicated that the tree locust (*Anacridium melanorhodon*) survives better and grows faster when certain phenols are present. These phenols are at high concentration in the locust's common host plants. They are retained in the insect by becoming bound in the cuticle, where it is suggested they probably stabilize proteins. According to Herms and Mattson (1992), some herbivores are able to sequester plant secondary metabolites to deter their own predators, while others use them as nutrients. Similarly, studies by Chapman and Blaney (1979) and Ohmart and Larsson (1989) found that some insects are able to sequester secondary plant compounds from the foliage on which they feed, acquiring toxic properties that provide a defense against predators and reducing their suitability as hosts for parasitoids.

What other strategies do the arthropods collected from eucalypt species containing high levels of secondary plant compounds use in order to survive, and in many cases feed, on foliage that might otherwise be considered toxic? While some herbivores are able to tolerate, utilize, or detoxify these chemicals (Chapman and Blaney 1979, Ohmart and Larsson 1989), many must resort to avoiding plants containing these compounds or use mixtures of food items to dilute their toxic effects (Bernays *et al.* 1994, Singer *et al.* 2002). This seems a likely scenario for many of the groups examined in the current study. A study by Cates (1980) found that specialized herbivores preferred young, nutritious, but highly toxic tissues, while herbivores with diets which consisted of a variety of plant taxa appeared to prefer the less nutritious, mature leaves that were lower in plant secondary compounds. Singer *et al.* (2002) found that the reaction of herbivores when encountering a new food depends heavily on its content of compounds in a manner suggestive of toxin dilution. This is one reason why mixed diets are believed to improve herbivore performance (Bernays *et al.* 1994). Alternatively, food mixing might simply be a method of selecting a range of foods to allow for a better balance of nutrients. Singer *et al.* (2002) suggest that both of these explanations may be correct. They consider that selective shifting between host plants could both balance nutritional requirements, and reduce the harmful effects of secondary plant compounds.

Why some types of vegetation contain more of these compounds than others is unclear. In the current study, mallee vegetation had more cineole than the remnant vegetation, *E. loxophleba* and *E. astringens* had higher pinene than the other species, and *E. loxophleba* had the highest levels of sideroxylonals, while the other mallee species had none. The resource availability hypothesis (Coley *et al.* 1985), suggests that species from resource-rich environments cope with herbivory by rapid growth and leaf replacement, while species from resource-poor environments use higher concentrations of secondary plant chemicals to deter arthropods (Dudt and Shure 1994). This is supported by Blanche and Westoby (1995), who found that plants growing on infertile soils tended to have higher concentrations of secondary compounds, such as oils and phenols. While this may help to explain some of the differences observed in the

concentrations of secondary plant compounds, it also seems likely that a large amount of phenotypic influence is being exerted in the current study.

The leaf chemicals examined here differed markedly in their relationships with arthropod ordinal richness, abundance and on the prevalence of particular arthropod orders. While these results are supported by a large amount of the existing literature, Fox and Macauley (1977) and Ohmart and Larsson (1989) claim that herbivore-plant relations are not influenced by variations in the leaf concentrations of tannins, essential oils, phenols and other secondary plant compounds. While the high concentrations of essential oils and phenols in eucalypt foliage might suggest that eucalypts should be relatively immune to attack from arthropods due to the adverse effects of these compounds on grazers, Fox and Macauley (1977) argue that this is far from the case. In fact eucalypts are some of the most heavily grazed tree species in the world, with species containing large amounts of terpenoids (essential oils) subject to at least as much insect herbivory as other temperate tree species which contain smaller amounts of terpenoids (Ohmart and Larsson 1989). According to these authors, secondary plant compounds do not seem to be correlated with levels of insect herbivory.

While many believe that the role of secondary compounds in plants is to deter herbivores, a number of other theories continue to be promoted. Close and McArthur (2002), claim that the main purpose of secondary compounds is to act as antioxidants, protecting leaves from photodamage, and that any effect on herbivores may be incidental. Bennett and Wallsgrave (1994) believe that secondary plant compounds serve many purposes, not only in defense against herbivore attack, but also in protection from UV, osmotic and other stresses, in the attraction of pollinators, and in allelopathic defense against other plants. Whatever their purpose, it is clear that there are a range of plant secondary compounds present in eucalypts, and some level of influence on arthropods is generally accepted. This was to some extent evident from the results presented in this chapter. The difficulty in separating herbivore responses to secondary plant compounds from those of other phytochemicals and nutrients (Singer *et al.* 2002), however, makes the results far from clear-cut. Allowances must also be made for the

fact that the analyses in this chapter were based on ordinal level arthropod data, which could have limited the sensitivity of the analyses. Chapter 8 examines one of the orders at the species level, to see if significant relationships might be clearly apparent at this taxonomic resolution.

CONCLUSION

This chapter set out to determine how secondary plant compounds influence canopy arthropods. A number of secondary plant compounds were present in the host tree species tested and there were wide variations in the concentrations of these compounds between the eucalypt species tested.

Generally speaking, mallee species had high cineole and low pinene levels and remnant species had the opposite. Sideroxylonals showed no such pattern, with vegetation type being high in *E. loxophleba* and absent in the other mallee species. In terms of their influence on arthropods, the compounds varied in their effects. Pinene had a generally negative effect, while cineole had a generally positive one. Sideroxylonal, however, was more complicated in its effects as it had a negative effect on ordinal richness, but was positively correlated with Hemiptera numbers. This suggests that it is highly unlikely that any one of these compounds can explain the pattern of arthropod assemblages observed in isolation. It seems more probable that complex interactions between these chemicals cause changes in nutritional quality and palatability of foliage, influencing the feeding behaviour, development, distribution and abundance of herbivores, in turn affecting predator densities and feeding behaviour.

This chapter explored the influence of secondary plant compounds on arthropods at the ordinal level. In Chapter 8 the combined influences of factors previously discussed in Chapters 4, 5, 6, and 7 on the Coleoptera (beetles) will be examined at the species level.

CHAPTER 8

HOW DO HOST EUCALYPT SPECIES, SOIL AND LEAF NUTRITION, LEAF ESSENTIAL OILS AND SECONDARY PLANT COMPOUNDS INFLUENCE ARTHROPOD BIODIVERSITY AT THE SPECIES LEVEL: A CASE STUDY USING THE ORDER COLEOPTERA

INTRODUCTION

In Chapters 4 and 5 the influence of host species, be it planted or remnant, was explored. In Chapter 6 the importance of nutrition was examined and in Chapter 7 the role of leaf chemistry was considered. In this chapter, the influences of each of these factors will be investigated in more depth for a single order, the Coleoptera (beetles). Probably the most pertinent quote on the importance of beetles was made by J.B.S. Haldane who, when asked what could be inferred about the Creator from the study of His creations, replied ‘an inordinate fondness for beetles’ (Evans 1975, Fisher 1988, Grove and Stork 2000). This quote reflects not only the importance of beetles in Haldane’s mind, but also refers to the sheer magnitude of the order Coleoptera in comparison with all other orders. The continuing popularity of this quote, years later, is testament to the fact that little has changed. Beetles are still one of, if not the, most abundant and speciose order, not only of arthropods, but of all life-forms. This seems cause enough for me to devote an entire chapter of this thesis to Coleoptera and the influence thereon of the previously discussed factors of host tree species, soil and plant nutrition and leaf chemistry.

This chapter draws together much of the work from previous chapters and examines the various factors influencing arthropod biodiversity at a finer scale using beetles as a case study. In order to achieve this, several questions need to be answered:

- Does host eucalypt species influence the canopy-dwelling beetle assemblage collected?
- Do soil nutrient levels influence canopy-dwelling beetle assemblages?
- Do leaf nutrient levels influence canopy-dwelling beetle assemblages?
- Do leaf essential oils influence canopy-dwelling beetle assemblages?
- Do secondary plant compounds, specifically sideroxylonals, influence canopy-dwelling beetle assemblages?
- Which factor is most important in determining canopy-dwelling beetle assemblages?

METHOD

Seven experimental sampling sites were set up in the localities of Narrogin, Wickopin, and Pingelly in the wheatbelt of Western Australia. Four of these sites, Parnell, McDougal, Hassel, and Marshall, consisted of alleys of oil mallee vegetation from three different species: *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*. A further three sites, Sprigg, Hesford, and Tutanning, were selected in native vegetation remnants, either on farmers properties, or in reserves. The remnant vegetation at each of these sites contained two native species: *Eucalyptus wandoo* subsp. *wandoo* and *E. astringens*. Each of these sites also had adjacent planted oil mallee vegetation containing either *Eucalyptus polybractea* or *E. loxophleba* subsp. *lissophloia* in an adjoining paddock. Only two of the three mallee species were directly compared to the remnant species, as no *Eucalyptus kochii* subsp. *borealis* plantings could be found adjacent to remnant vegetation, containing the selected remnant species.

At each site, five trees of each of the three species present were selected for arthropod sampling. In October of 2005 (spring), the trees were sampled by canopy spraying, using a backpack spraying machine. The synthetic pyrethroid insecticide Dominex[®] was

applied at a rate of 1 ml/L, with an average volume of 1-2 L of insecticide used on each tree. Sheets were placed beneath the trees before spraying, and 60 minutes afterwards the trees were shaken to dislodge any remaining arthropods. Specimens were removed from the sheets by shaking into trays by hand, and by use of hand held battery powered aspirators, which sucked specimens into a small collection jar. Once collected, samples were stored in 70% ethanol until sorting. In May of 2006 (autumn), the same spraying and collection procedure was repeated on the same trees. Once collected and stored in alcohol, samples were sorted with the aid of a stereo microscope to the ordinal level in the laboratory. Coleoptera (beetle) specimens were sent to Mr. Andras Szito (a professional taxonomist) for identification to the species level. Coleoptera raw data are presented in Appendix N.

Leaf and soil samples were taken at each site in order to determine the levels of nutrition available to herbivorous arthropods. At each site, 50 leaves (bulked from five trees) of each of the tree species present were collected and placed in paper bags. Randomly selected leaves were collected from all over the tree canopy, using pole mounted secateurs, in order to obtain as close to a representative sample as possible with the limited number of leaves collected. Newly emerged and senesced leaves, however, were not collected as their nutrient levels would have skewed the results. The bags of leaf material were kept in a cooler box until they were delivered to CSBP for analysis. Soil samples were also taken at each site. Samples were taken to 20 cm depth under and around each of the tree species present. Three samples were taken and bulked per tree species at each site. Samples were stored in plastic bags and delivered to CSBP for analysis.

Prior to spraying for the first invertebrate sampling in October 2005, leaves were collected for terpenoid (essential oil) analysis. The leaves were collected and analysed following the method outlined in Ammon *et al.* (1985). Ten leaves per tree were very carefully removed using pole-mounted secateurs so as not to disturb arthropods. Leaves could not be removed after spraying as this could interfere with the chemical analysis. In the field 3 g of leaf material from each of the trees was added to individual bottles with

50 ml of ethanol previously weighed into them. Bottles were left for at least 10 days to maximize the effectiveness of the solvent extraction process before continuing with the analysis. In the laboratory, the bottles were reweighed and the mass of fresh leaf material calculated. The solution was then analysed for both water and oil to determine which essential oils were present in the samples from each tree and in what proportions. Bottles containing no leaf material were used as controls to confirm that no significant loss of solvent occurred during sampling in the field. Samples were analysed using vapour-phase chromatographic analysis on a Varian Aerograph 2700 at the Department of Environment and Conservation (DEC) in Perth, Western Australia. All analyses were carried out three times and the data were averaged. Component oils were identified by their retention times and by co-injection with a reference compound, and confirmed by gas chromatography-mass spectrometry (Ammon *et al.* 1985).

In May 2006, prior to the second sampling for arthropods, leaves were taken for formylated phloroglucinol (sideroxylonal) analysis. Leaves were collected, processed and analysed as per the protocol described in Wallis and Foley (2005) at the Australian National University (ANU). Thirty leaves per tree were very carefully removed using pole-mounted secateurs in order not to disturb the arthropods. Leaves were not removed after spraying, as this might interfere with the chemical analysis. Foliage was freeze-dried and ground to pass through a 1 mm sieve using a Cyclotech 1093 Mill. Then 50 mg of the ground foliage was sonicated in 8 g of 7% water in acetonitrile (with 0.1% trifluoroacetic acid and 0.3000 g/L of the internal standard, 2-ethyl phenol) for 5 minutes. The extract (0.45 µm) was then filtered into an autosampler vial for analysis by reversed phase HPLC. Samples were analysed isocratically on a Waters Alliance HPLC system using 7% water in acetonitrile with 0.1% trifluoroacetic acid. The analysis produced total sideroxylonals contents of the leaves as well as percentages of the A, B and C forms of sideroxylonal.

Data were tested for homogeneity of variance and transformations were done where necessary. A range of statistical analyses were conducted on the data collected. To strengthen the analyses, beetle species occurring on three or less trees were excluded

from the analysis. Analyses of variance (ANOVA) were conducted to determine if significant differences in beetle species richness and abundance existed in relation to factors such as season, site and tree species. One-way ANOVAs were conducted because of the unbalanced nature of the experimental design. This was unavoidable due to the scarcity of suitable sites containing all of the required tree species. LSDs were used in post hoc testing to determine where differences lie. Coefficients of correlation calculated by Pearson's product-moment method were also used to determine if there were any significant relationships between beetle species richness and abundance with factors such as soil and leaf nutrition, leaf essential oils and sideroxylonals content.

Ordinations were conducted using the Primer 6 statistical package (Primer-E 2007) to demonstrate the degree of similarity between beetle assemblages collected from different tree species at various sites. Ordinations were constructed by multi dimensional scaling (MDS) based on Bray-Curtis similarities of fourth root transformed data. For all beetle species level analyses, species occurring on three or less trees were excluded from the analysis. ANOSIM analysis was conducted on the MDS beetle assemblage data to determine if there were any significant differences between the beetle assemblages in the main habitat types (mallee and remnant) and on each tree species. Global test results indicated whether significant differences existed and pairwise tests indicated where the main between group differences arose. Bubble plots were used to superimpose environmental data over the ordination for combined seasonal beetle assemblage data. These data were used because environmental factors, such as soil nutrients and leaf essential oils, did not correspond to any particular season. The bubble plots provided a series of visual demonstrations of the relationships between various environmental factors and the degree of beetle assemblage similarity. The significance of these relationships was determined using the BEST procedure, which uses Spearman rank correlations between the beetle assemblage (Bray-Curtis) matrix and the environmental (Euclidean distance) matrix (Clarke 1993, Clarke and Warwick 2001). The procedure compares the various environmental factors influencing the pattern of species assemblages and produces a list which ranks the strength of the correlation between the various combinations of factors and the pattern of species assemblages from the MDS.

The highest correlation represents the best match between explanatory variables and the assemblage pattern observed (Clarke 1993, Clarke and Warwick 2001).

The results presented in this chapter fall into seven categories:

Season - which presents the results of analyses to determine if the season in which beetles were collected influences beetle species richness and abundance;

Site - which presents the results of analyses designed to determine if there is any significant difference in beetle assemblages between sites;

Species - which presents the results of analyses to determine if there is any significant difference in beetle assemblages between tree species;

Soil Nutrients - which describes the results of analyses to determine the significance of any influence of soil nutrients on beetle assemblages;

Leaf Nutrients - which describes the results of analyses of the influence of leaf nutrients on beetle assemblages;

Essential Oils - which presents the results of analyses to determine the influence of the leaf essential oils cineole and pinene on beetle assemblages; and

Sideroxylonal - which presents the results of analyses to determine the influence of the secondary plant compound sideroxylonal on beetle assemblages.

RESULTS

During this study a total of 13 856 beetles were collected from 332 species, representing 40 families (Appendix N). Results of collections of beetles by chemical knockdown are presented below in terms of richness, abundance and assemblage relationships to outside factors. For this study, richness was measured as the number of beetle species collected per tree. At the first sampling in October 2005 (spring), 231 species were collected. The family with the most species was the Curculionidae. At the second sampling in May 2006 (autumn), 232 species were collected and the Curculionidae was again the most species-rich beetle family. Total number of beetles, also referred to as abundance, was also measured. At the first sampling in October 2005 (spring), a total of 8265 beetles were collected and the family with the most individuals was the Staphylinidae. At the

second time of sampling in May 2006 (autumn), a total of 5591 beetles were collected and Coccinellidae was the most abundant of the beetle families. Overall, the most abundant family was the Staphylinidae.

Season

Figure 43 presents a comparison of the beetle species richness collected during the two sampling seasons, split into the two site types ‘mallee’ and ‘remnant’. There does appear to be a difference in the number of beetle species collected between the two sampling periods, with the first time of sampling having more beetle species collected in both mallee and remnant habitats than at the second time of sampling.

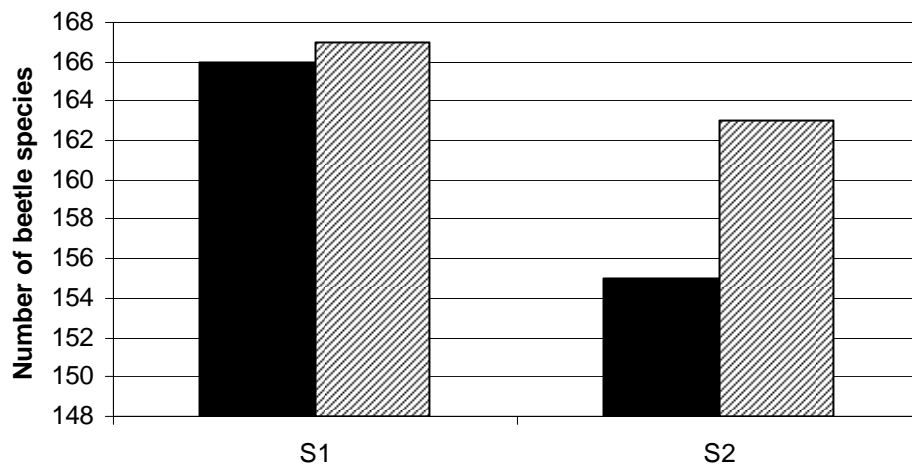


Figure 43: Total number of beetle species (richness) collected in October 2005 (S1) and May 2006 (S2) at mallee sites (represented by a dark histogram) and remnant sites (represented by shaded histogram).

Analysis of variance (Table 25), indicated that there was a significant difference ($P < 0.05$) in beetle species richness collected at the two periods of sampling from the remnant sites and when all sites were considered together. In both cases, the first period of sampling (October 2005) had greater species richness than the second period of sampling.

Table 25: One-way ANOVA results for effect of sampling period on beetle species richness. Superscripts indicate significant differences (LSD, $p=0.05$) with values decreasing from left to right.

Source of variation	Significant differences	Habitat type	MS	F	Probability
Season	1 ^a 2 ^b	Remnant	640.00	16.67	0.001
Season	ns	Mallee	58.80	1.73	0.190
Season	1 ^a 2 ^b	Combined	499.89	13.72	0.001

* 1 = Sampling period 1 (Oct 2005); 2 = Sampling period 2 (May 2006).

* ns = no significant difference at $p=0.05$.

Figure 44 presents a comparison of the total beetle numbers (abundance) collected at the two periods of sampling, split into the two site types, ‘mallee’ and ‘remnant’. There appears to be a difference between the two sampling periods in relation to beetle abundance in both site types.

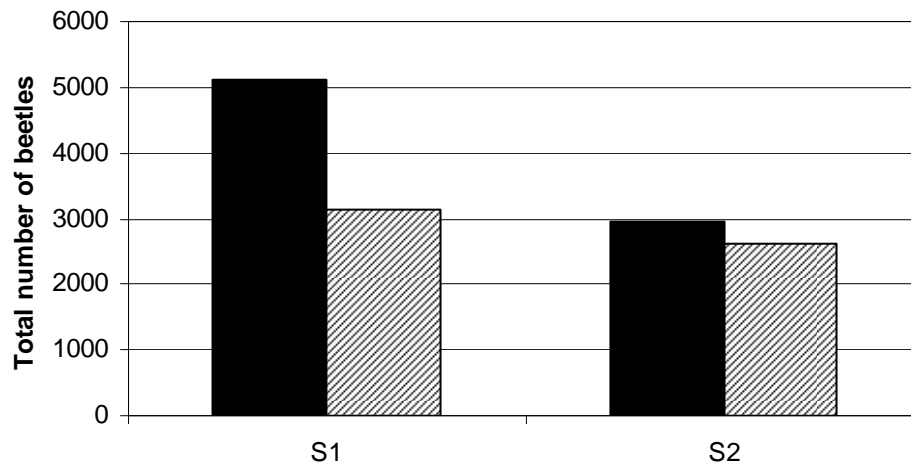


Figure 44: Total number of individual beetles collected in October 2005 (S1) and May 2006 (S2) at mallee sites (represented by a dark histogram) and remnant sites (represented by shaded histogram).

Analysis of variance (Table 26), indicated that there was a significant difference ($P<0.05$) in total beetle abundance collected at the two periods of sampling from the mallee sites and when all sites were considered together. In both cases, the first period of sampling (October 2005) had greater total beetle abundance than the second period of sampling. There was no significant difference, however, between sampling periods in terms of beetle abundance at the remnant sites.

Table 26: One-way ANOVA results for effect of sampling period on beetle abundance. Superscripts indicate significant differences (LSD, p=0.05) with values decreasing from left to right.

Source of variation	Significant differences	Habitat type	MS	F	Probability
Season	ns	Remnant	13056.0	3.46	0.066
Season	1 ^a 2 ^b	Mallee	39313.0	12.04	0.001
Season	1 ^a 2 ^b	Combined	50484.0	14.53	0.001

* 1 = Sampling period 1 (Oct 2005); 2 = Sampling period 2 (May 2006).

* ns = no significant difference at p=0.05.

Site

Seven sites were used in this study and it was important to know if site was a factor in the beetle species richness and abundance observed. Analysis of variance (Table 27) indicated that beetle species richness was not influenced by site. Beetle abundance, however, was somewhat affected by sampling site. Analysis of variance (Table 27) showed that, beetle abundance was significantly greater at site 3 (Marshall) than at sites 4 (Hassel), 7 (Hesford) and 5 (Tutanning).

Table 27: One-way ANOVA results for effect of site on beetle species richness and beetle abundance. Superscripts indicate significant differences (LSD, p=0.05) with values decreasing from left to right.

	Significant differences	df	MS	F	Probability
Richness	ns	6	149.862	2.042	0.060
Abundance	3 ^a 6 ^{ab} 1 ^{ab} 2 ^{ab} 4 ^b 7 ^b 5 ^b	6	13176.190	2.238	0.040

* 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

* ns = no significant difference at p=0.05.

Species

The effects of tree host species on beetle species richness was examined by analysis of variance (Table 28). While most species were not significantly different in terms of beetle species richness, *E. kochii* subsp. *borealis* had significantly lower (P<0.05) levels than the other species. The effect of tree host species on beetle abundance was also examined by analysis of variance (Table 28). *E. polybractea* had the highest beetle abundances, being significantly greater (P<0.05) than *E. kochii*, *E. astringens*, *E. loxophleba* and *E. wandoo*.

Table 28: One-way ANOVA results for effect of tree species on beetle species richness and beetle abundance. Superscripts indicate significant differences (LSD, p=0.05) with values decreasing from left to right.

	Significant differences	df	MS	F	Probability
Richness	P ^a L ^a W ^a M ^a A ^a K ^b	5	240.187	3.327	0.006
Abundance	P ^a M ^{ab} K ^{bc} A ^{bc} L ^c W ^c	5	21636.724	3.747	0.003

* L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astragens*; M = mallee species.

Family

Numbers of beetles collected from each of the families represented in this study are presented in Figure 45. The graph shows that the Staphylinidae, Lathridiidae, Coccinellidae, Curculionidae and Chrysomelidae were the most abundant beetle families collected.

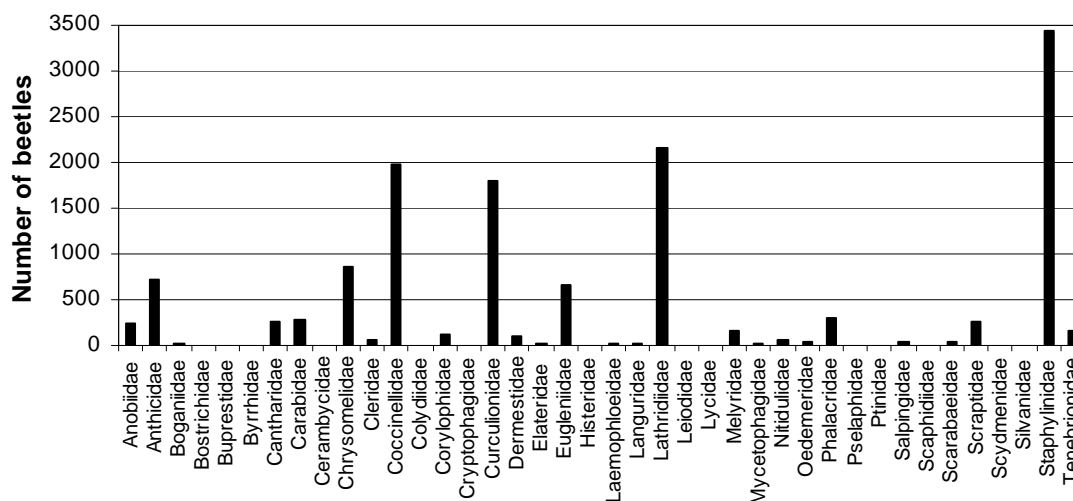


Figure 45: Total number of individual beetles collected from each of the representative families during this study.

The effects of host tree species and site on the number of beetle families collected was determined by analysis of variance (Table 29). No significant difference was found between either sites or species in relation to the number of beetle families collected.

Table 29: Two-way ANOVA results for effect of tree species and site on the number of beetle families collected. Superscripts indicate significant differences, (LSD, $p=0.05$) with values decreasing from left to right.

	Significant differences	df	MS	F	Probability
Site	ns	5	9.471	1.685	0.140
Species	ns	4	13.026	2.317	0.059
Site*Species	interaction	10	30.729	5.467	0.000

* interaction = a significant interaction effect was observed.

* ns = no significant difference at $p=0.05$.

While there was no statistical difference in the number of families collected, Figures 46 and 47 clearly indicate that certain families were more prevalent at certain sites and on certain species.

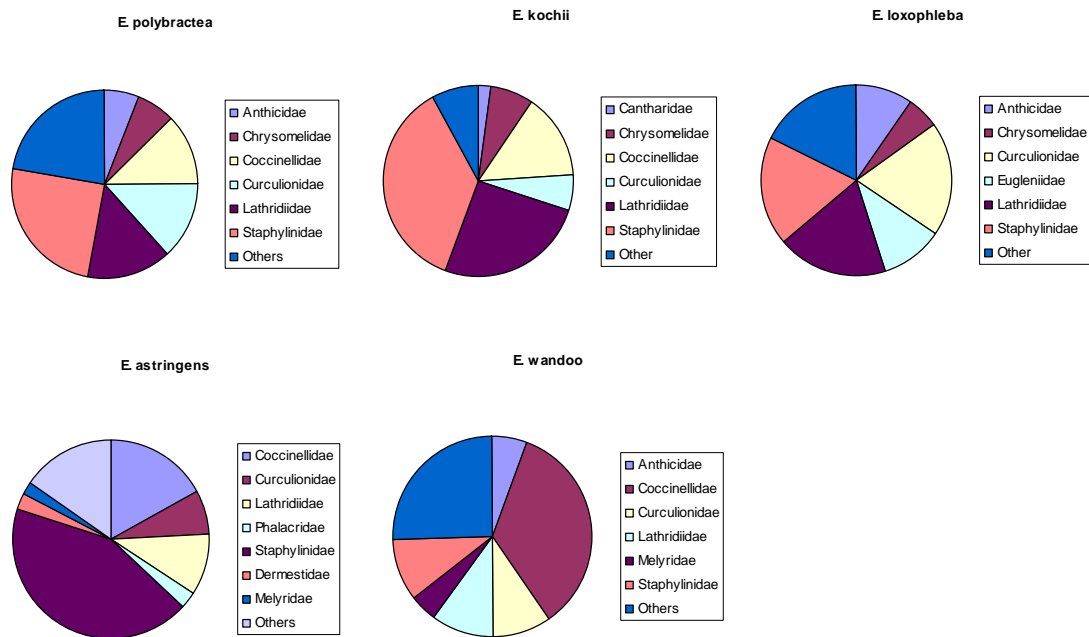


Figure 46: Pie graphs showing numbers of the major families collected from each of the tree species sampled. Species sampled were *E. polybractea*; *E. kochii* subsp. *borealis*; *E. loxophleba* subsp. *lissophloia*; *E. astringens*; and *E. wandoo* subsp. *wandoo*.

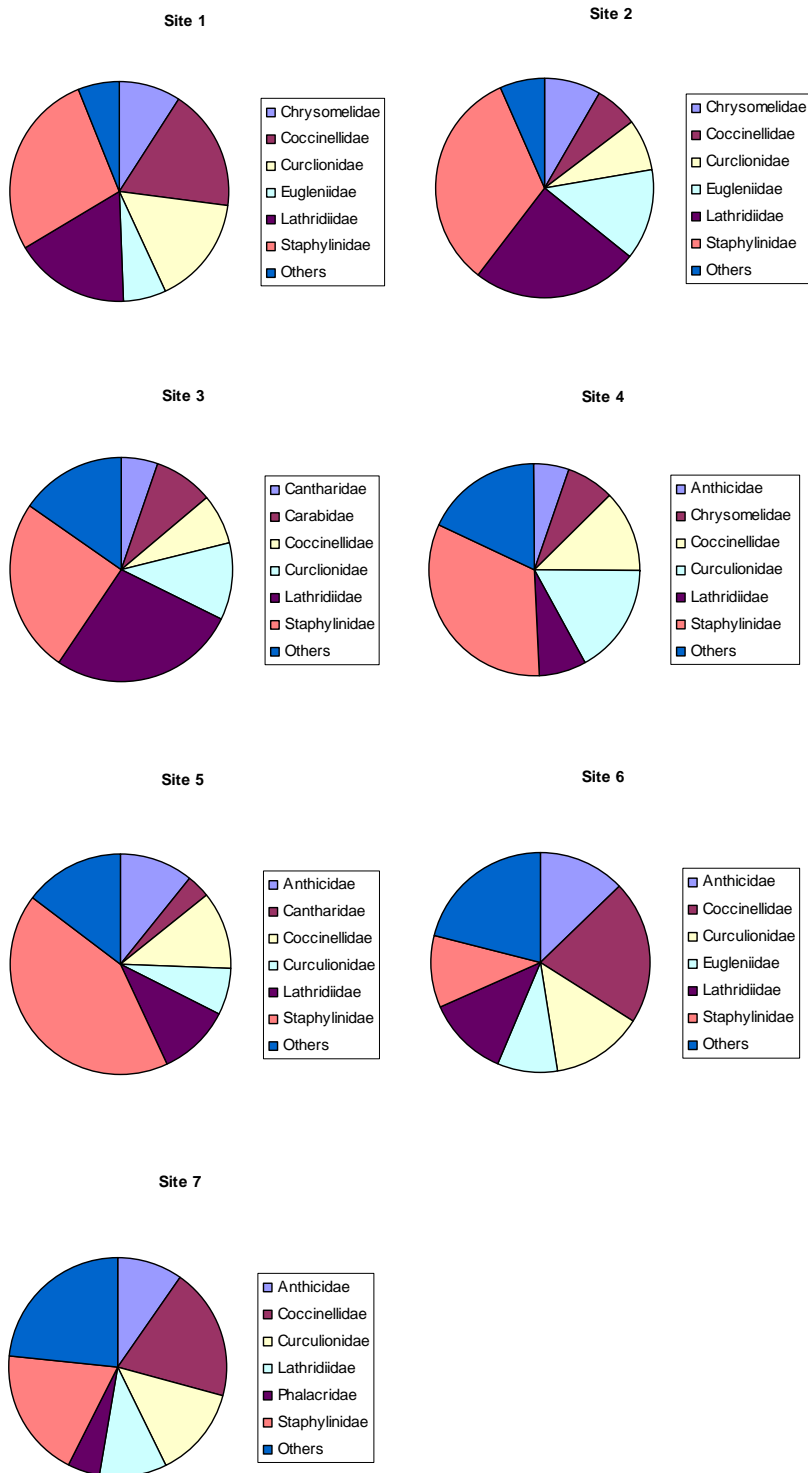


Figure 47: Pie graphs showing numbers of the major families collected at each of the sampling sites. 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford).

Assemblage Composition

Consideration of trends at the richness and abundance level can mask trends in individual species. Thus, the effect of tree species on the beetle assemblage as a whole was also examined using ordinations. Figure 48 shows the similarity of the beetle assemblages at the first period of sampling in October 2005, while Figure 49 shows the similarity of the beetle assemblages at the second period of sampling in May 2006. Similar trends are apparent between the assemblages resident on the same tree species at both times of sampling. Assemblages on the same type of vegetation, for example remnant species, as opposed to mallee species, also tended to have similar beetle assemblages.

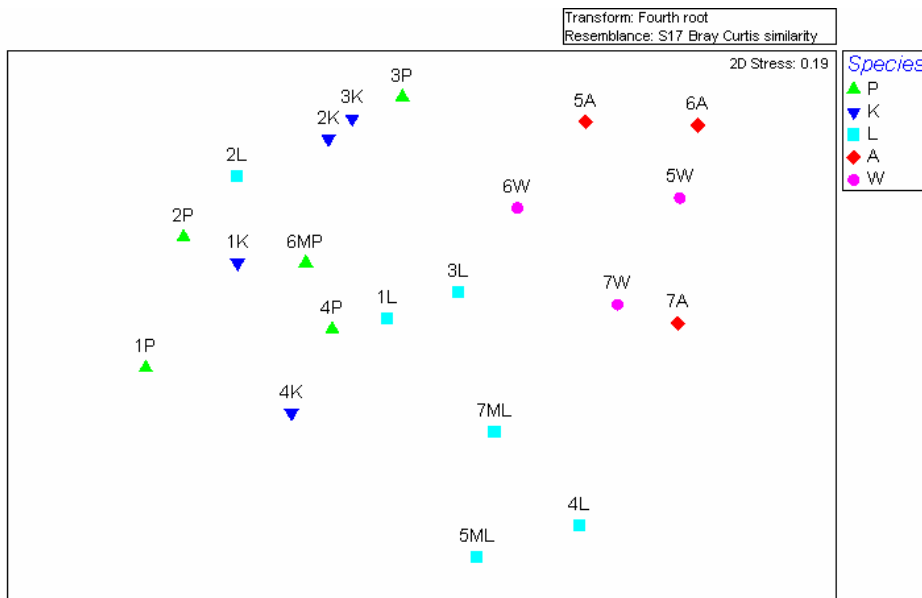


Figure 48: MDS ordination showing the similarity of beetle assemblages between each tree species and site for the October 2005 sampling period. Numbers represent site: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters and colours indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data.

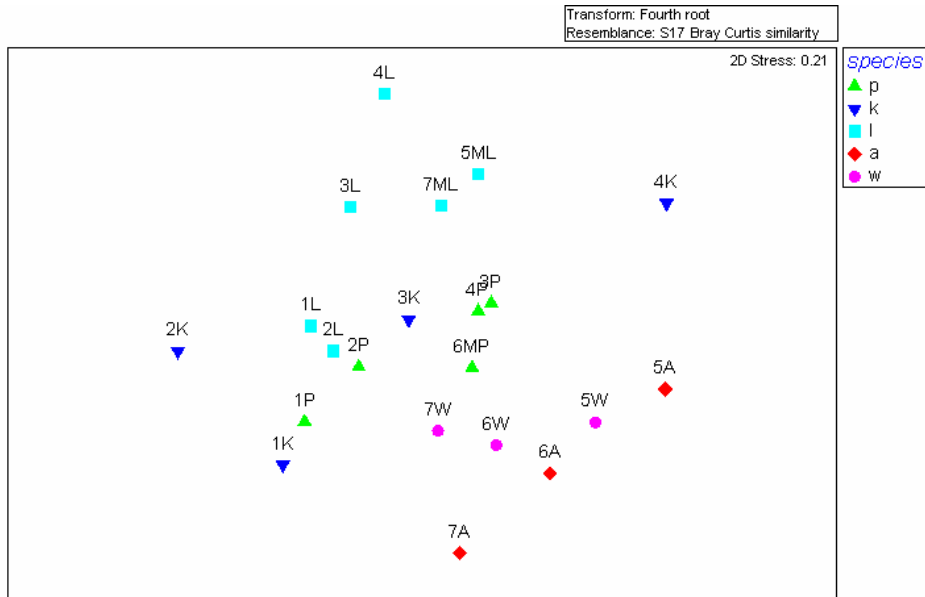


Figure 49: MDS ordination showing the similarity of beetle assemblages between each tree species and site for the May 2006 sampling period. Numbers represent site: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters and colours indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data.

The ordinations (Figures 48 and 49) illustrate a number of interesting trends. The remnant assemblages (coloured red and pink) are clearly distinct from those of the mallee assemblages. There is no clear distinction however, between the two remnant species assemblages. Mallees located close to remnant vegetation (indicated by the first letter M on the ordinations), are no more similar in their assemblages to the remnant vegetation than those of the mallees located distant from the remnant. The fauna of *E. kochii* subsp. *borealis* and *E. polybractea* (represented by blue and green triangles respectively), are difficult to separate from each other, while the other mallee species, *E. loxophleba* subsp. *lissophloia*, appears to have a somewhat more distinct fauna.

ANOSIMs (Table 30) showed that the distinctions observed in Figures 48 and 49 between the beetle assemblages of different tree species and vegetation types at both times of sampling were significant.

Table 30: ANOSIM global test results for difference in beetle assemblage between various combinations of tree species and habitat types at sampling period 1 (October 2005) and sampling period 2 (May 2006). A random sample of 999 permutations was used in each instance.

Factor	Sampling period	Sample statistic (Global R)	Significance level	Permuted statistics > global R
Species	1	0.509	0.001	0
Species	2	0.327	0.002	1
Mallee/Remnant	1	0.526	0.001	0
Mallee/Remnant	2	0.353	0.004	3

Pairwise tests (Table 31) at the first period of sampling (October 2005) revealed significant differences between several of the tree species sampled. The only pairs which were not significantly different from each other in their beetle assemblages were the mallee species, *E. polybractea* and *E. kochii* subsp. *borealis*, and the two remnant species, *E. wandoo* subsp. *wandoo* and *E. astringens*.

Table 31: ANOSIM pairwise tests of factor levels for tree species at the first time of sampling (October 2005). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*. Bold text indicates a significant difference at the P<0.05 significance level. All possible permutations were used.

Pairwise test	R statistic	Significance level	Permuted statistics > R statistic
P vs. K	0.056	0.341	43
P vs. L	0.363	0.026	12
P vs. A	0.867	0.018	1
P vs. W	0.651	0.018	1
K vs. L	0.433	0.019	4
K vs. A	0.907	0.029	1
K vs. W	0.833	0.029	1
L vs. A	0.660	0.012	1
L vs. W	0.463	0.024	2
A vs. W	0.296	0.400	4

Pairwise tests (Table 32) at the second time of sampling (May 2006) also revealed significant differences between some of the tree species sampled. The pairs which had significantly different beetle assemblages were *E. polybractea* and *E. astringens*, *E.*

polybractea and *E. wandoo* subsp. *wandoo*, *E. loxophleba* subsp. *lissophloia* and *E. astringens*, plus *E. loxophleba* subsp. *lissophloia* and *E. wandoo* subsp. *wandoo*. Generally speaking then, the mallee species, apart from *E. kochii* subsp. *borealis*, significantly differed in their beetle assemblages from the remnant species. The overall trends of the second set of pairwise tests were the same as at the first, except that some of the significance was lost, possibly due to there being less species and variance in the data from the second sampling period.

Table 32: ANOSIM pairwise tests of factor levels for tree species at the second time of sampling (May 2006). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*. Bold text indicates a significant difference at the P<0.05 significance level. All possible permutations were used.

Pairwise test	R statistic	Significance level	Permuted statistics > R statistic
P vs. K	0.088	0.246	31
P vs. L	0.205	0.074	34
P vs. A	0.846	0.018	1
P vs. W	0.682	0.018	1
K vs. L	0.218	0.105	22
K vs. A	0.370	0.114	4
K vs. W	0.352	0.086	3
L vs. A	0.722	0.012	1
L vs. W	0.500	0.012	1
A vs. W	-0.037	0.700	7

Soil Nutrients

The relationship between each of the soil nutrients and beetle assemblage similarity is demonstrated by bubble plots (Figure 50). It is clear from Figure 50 that nutrient levels varied widely between sites, although this was not always reflected in the groupings of the beetle assemblages. The strongest relationship between soil nutrients and beetle assemblage grouping appeared to be for soil phosphorus (Figure 50b), with remnant species (which were grouped together) tending to have much lower levels of soil phosphorus than the mallee species.

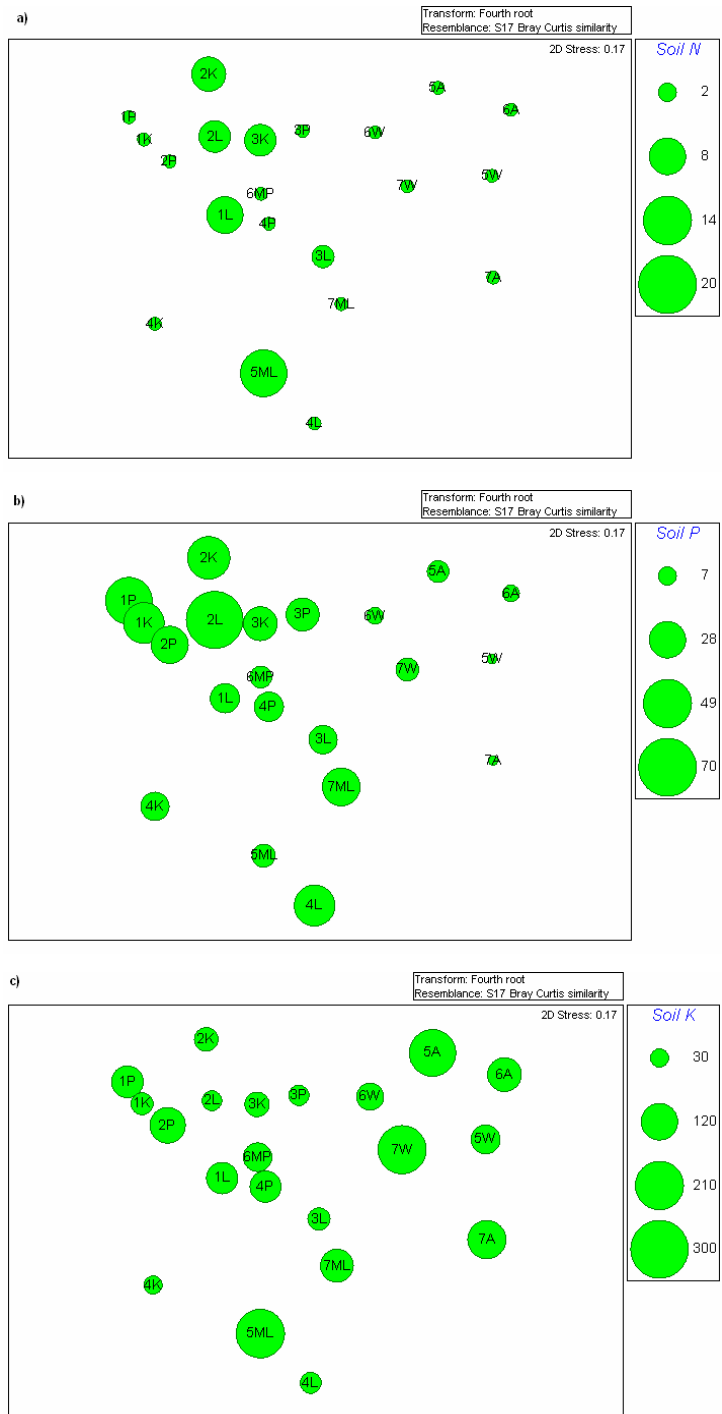


Figure 50: Bubble plot of soil nutrients. Soil nutrient levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of soil nutrients are represented by coloured bubbles a) soil N, b) soil P, c) soil K. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined.

The influence of soil nutrients on beetle abundance and richness was examined by coefficient of correlation (Table 33). There were no significant correlations between soil nutrients and beetle species richness or abundance at either the remnant, or the mallee sites, or when all sites were combined.

Table 33: Coefficient of correlation results for effect of soil nutrients on beetle (Coleoptera) species richness and abundance. There were no significant relationships at the P<0.05 significance level.

	Habitat type	Nitrogen	Phosphorus	Potassium
Richness	Remnant	-0.06	0.24	-0.31
Abundance	Remnant	-0.13	0.12	-0.19
Richness	Mallee	-0.02	-0.30	0.55
Abundance	Mallee	0.07	-0.07	0.01
Richness	Combined	-0.25	-0.14	0.07
Abundance	Combined	-0.01	0.07	-0.16

Leaf Nutrients

The relationship between each of the leaf nutrients and beetle assemblage similarity is illustrated by bubble plots in Figure 51. In most cases the levels of the various leaf nutrients did not appear to differ markedly between the different sites or species. The strongest relationship between leaf nutrients and beetle assemblage grouping appeared to be for leaf phosphorus (Figure 51b), with remnant species (which were grouped together) tending to have somewhat lower levels of leaf phosphorus than the mallee species.

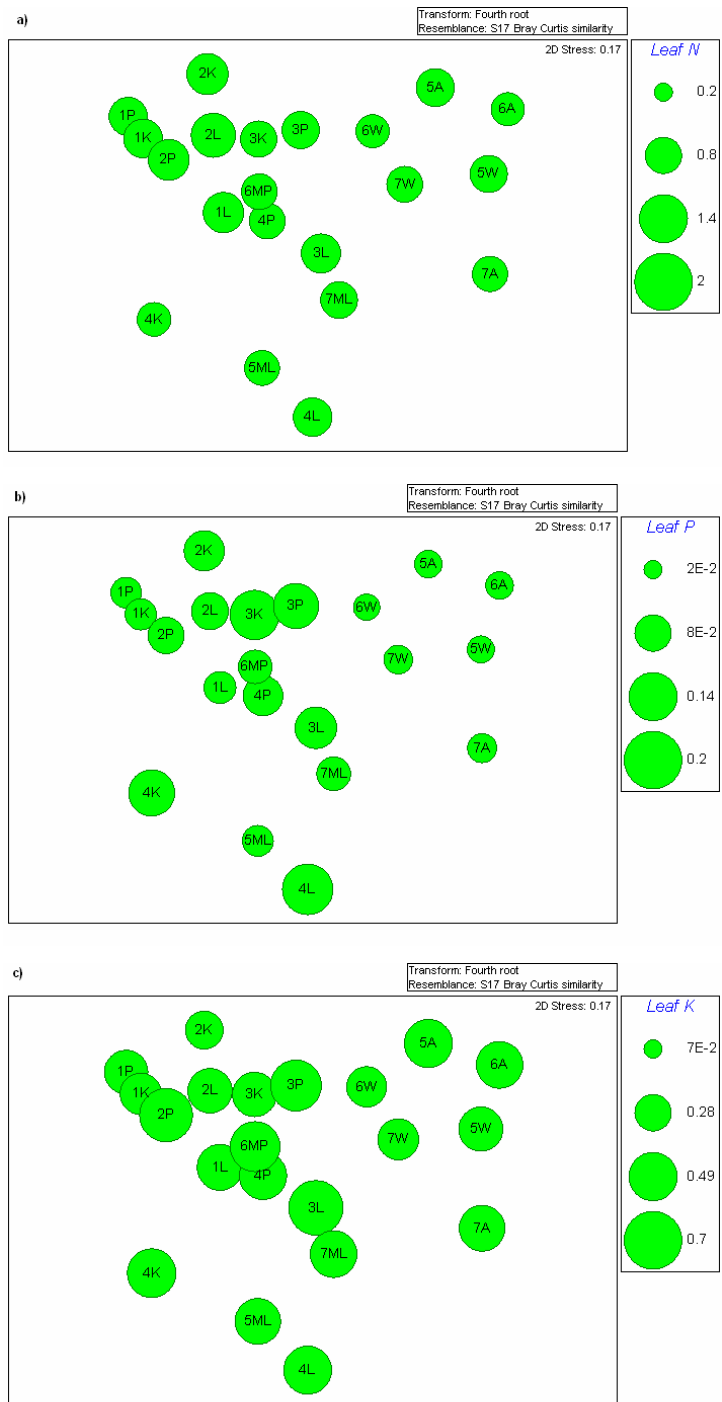


Figure 51: Bubble plot of leaf nutrients. Leaf nutrient levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of leaf nutrients are represented by coloured bubbles a) leaf N, b) leaf P, c) leaf K. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined.

The influence of leaf nutrients on beetle species richness and abundance was also examined by coefficient of correlation (Table 34). There were no significant correlations between leaf nutrients and beetle species richness or abundance at either the remnant, or the mallee sites, or when all sites were combined.

Table 34: Coefficient of correlation results for effect of leaf nutrients on beetle (Coleoptera) species richness and abundance. There were no significant relationships at the P<0.05 significance level.

	Habitat type	Nitrogen	Phosphorus	Potassium
Richness	Remnant	0.11	0.50	0.26
Abundance	Remnant	0.17	0.59	0.64
Richness	Mallee	-0.06	-0.34	0.17
Abundance	Mallee	0.00	-0.18	-0.14
Richness	Combined	-0.03	-0.17	0.19
Abundance	Combined	0.11	0.05	0.11

Essential Oils

Gas chromatographic (GC) conditions used during gas chromatography and results of the analysis are presented in Appendix L. Only the two major leaf essential oils are examined here: cineole and pinene.

The relationship between each of the leaf essential oils and beetle assemblage similarity is illustrated by bubble plots in Figure 52. Both cineole and pinene showed variations which appeared to relate at least partially to beetle assemblage groupings. Cineole levels tended to be lower in the remnant species (which were grouped together), while pinene levels were high in *E. loxophleba* subsp. *lissophloia* and the remnant species.

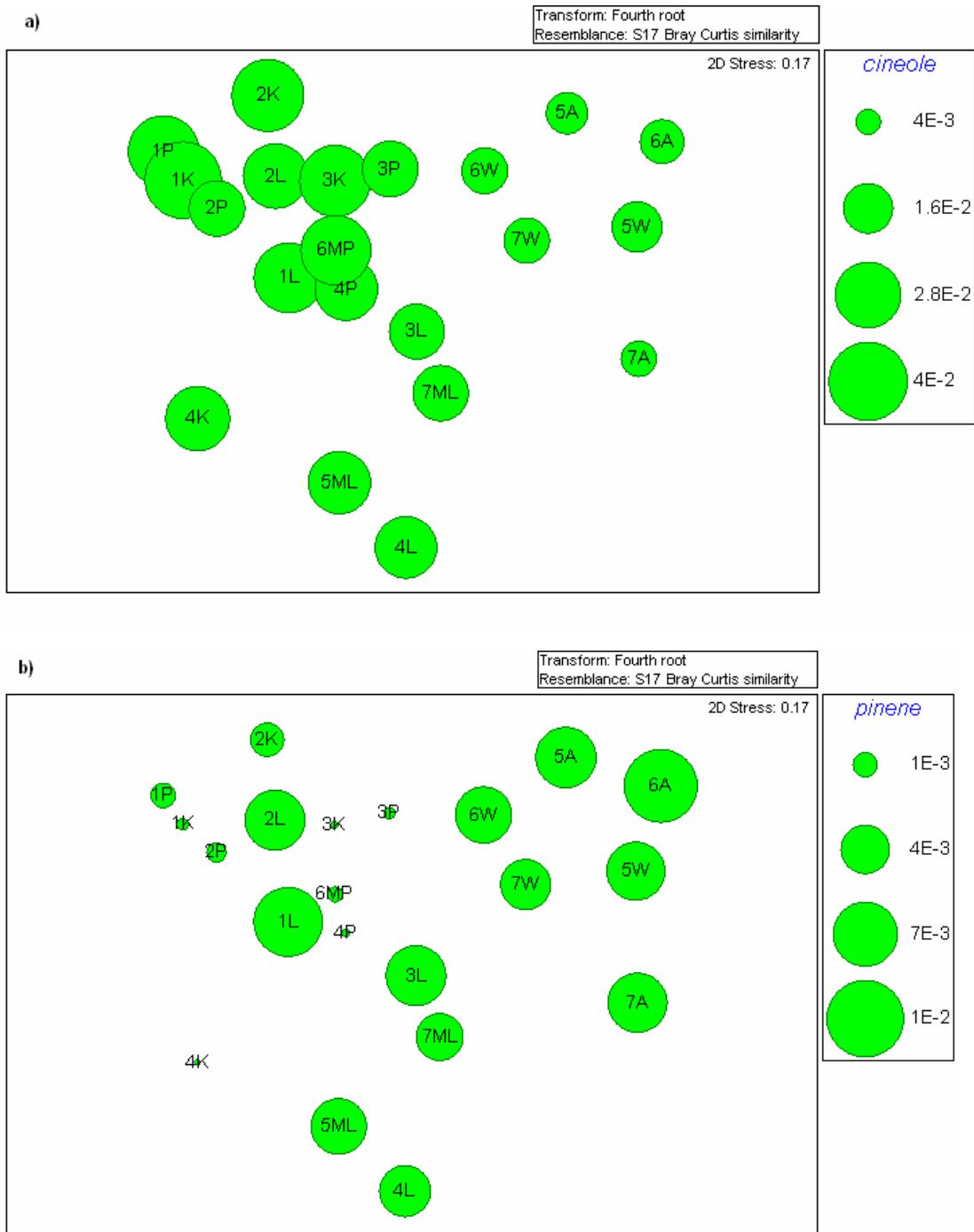


Figure 52: Bubble plot of leaf essential oils. Essential oil levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of leaf essential oils are represented by coloured bubbles a) cineole, b) pinene. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined.

The influence of cineole and pinene on beetle species richness and abundance was also examined by coefficient of correlation (Table 35). Cineole levels did not appear to strongly affect beetle species richness or abundance in either a positive or negative way. Pinene levels also did not show any significant relationship with beetle species richness. As already mentioned in Chapter 7, beetle abundance was significantly negatively correlated with leaf pinene levels.

Table 35: Coefficient of correlation results for effect of essential oils on beetle (Coleoptera) species richness and abundance. Figures in bold indicate significant relationships at the P<0.05 significance level.

	Pinene	Cineole
Richness	0.06	-0.08
Abundance	-0.22	0.10

Sideroxylonals

Only the most prevalent and important (in terms of arthropod ecology) of the formylated phloroglucinol compounds, the sideroxylonals, are examined here. Levels of sideroxylonals obtained are presented in Appendix M. There are three common types of sideroxylonals A, B and C. However, data are presented here as total sideroxylonals.

The relationship between sideroxylonals and beetle assemblage similarity is illustrated by a bubble plot in Figure 53. It is immediately apparent that sideroxylonals levels are strongly related to species, though not necessarily responsible for beetle assemblage groupings. The remnant species showed low levels of sideroxylonals, while the mallee species were split between *E. loxophleba* subsp. *lissophloia*, which exhibits extremely high levels, and the other two species which contain no sideroxylonals at all.

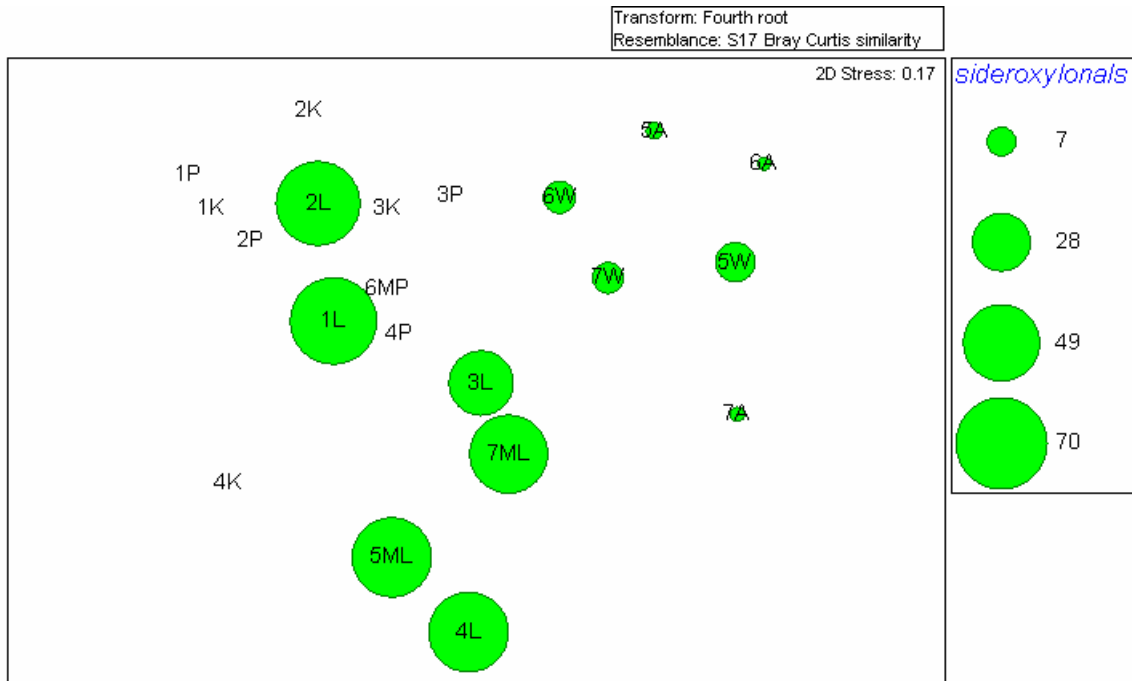


Figure 53: Bubble plot of sideroxylonals. Sideroxylonal levels are superimposed over an ordination showing the similarity of the beetle assemblages between each tree species and site for combined seasons. Levels of sideroxylonals are represented by coloured bubbles. Numbers represent site numbers: 1 = site 1 (Parnell), 2 = site 2 (McDougall), 3 = site 3 (Marshall), 4 = site 4 (Hassel), 5 = site 5 (Tutanning), 6 = site 6 (Sprigg), 7 = site 7 (Hesford). Letters indicate tree species: L = *E. loxophleba* subsp. *lissophloia*; K = *E. kochii* subsp. *borealis*; P = *E. polybractea*; W = *E. wandoo* subsp. *wandoo*; A = *E. astringens*; M = mallee species. Ordination constructed by non-metric multidimensional scaling based on Bray-Curtis similarities applied to fourth root transformed abundance data for both seasons combined.

The influence of sideroxylonals on beetle species richness and abundance was examined by coefficient of correlation (Table 36). Total sideroxylonals did not show any significant relationship with either beetle species richness or abundance.

Table 36: Coefficient of correlation results for effect of sideroxylonals on beetle (Coleoptera) species richness and abundance. There were no significant relationships at the P<0.05 significance level.

	Total Sideroxylonals
Richness (orders)	0.06
Abundance	-0.01

A large number of factors with potential to influence beetle assemblages have been examined in this chapter. A BEST analysis (Table 37) was conducted to determine

which of the environmental factors examined best explain the pattern of beetle assemblages observed.

Table 37: BEST selections calculated from Spearman rank correlations between the beetle (Bray-Curtis) matrix and the environmental (Euclidean distance) matrix of beetle assemblages. Factors included in the analysis: cineole, pinene, sideroxylonals, soil N, soil P, soil K, leaf N, leaf P and leaf K.

Number of variables	Correlation	Selections
1	0.436	cineole
2	0.429	cineole + pinene
5	0.284	cineole + pinene + soil N + soil P + leaf P
4	0.284	cineole + soil N + soil P + leaf P
4	0.283	cineole + pinene + soil N + soil P
3	0.283	soil N + soil P + leaf P
3	0.283	cineole + soil N + soil P
4	0.283	pinene + soil N + soil P + leaf P
5	0.282	cineole + soil N + soil P + leaf P + leaf K
4	0.282	soil N + soil P + leaf P + leaf K

This analysis indicated that a single environmental factor, cineole, best explained the pattern of beetle diversity observed. Adding more factors to the model only reduced its explanatory power. The strength of this single factor in predicting the pattern of beetle assemblages indicates that cineole is a powerful driver of beetle diversity. On closer examination of Figure 48a, it is apparent that *E. astringens* and *E. wandoo* both have lower cineole and a distinct beetle community. However, since both of these species are in the remnant habitat it seems that the result is somewhat confounded by site and species. So while cineole does appear to have a strong influence, it may not be solely responsible for the different beetle communities observed.

DISCUSSION

At the beginning of this chapter, I pointed out the importance of beetles as one of the most abundant and speciose orders on the planet. Beetles are ecologically extremely diverse, occurring in almost every terrestrial and freshwater environment (Grove and Stork 2000). They fill a huge variety of niches as predators, detritivores, folivores, wood borers, leaf miners, seed eaters and carrion feeders (CSIRO 1991).

In the current study, the most species-rich family collected at both times of sampling was the Curculionidae, weevils. Curculionidae are herbivores that contain representatives which feed on almost every part of the plant. The Curculionidae include wood borers, species that feed in galls, under bark, in seed pods, directly on foliage, or which are leaf miners. The most abundant of the beetle families collected was Staphylinidae in sampling period 1 (October 2005) and Coccinellidae in sampling period 2 (May 2006). The Staphylinidae, or rove beetles, are predators and carrion feeders, while Coccinellidae, or ladybird beetles, are predators.

It is interesting that the most species-rich family is herbivorous, while the most abundant beetle families are predators and carrion feeders. It is possible that the vegetational complexity of the tree species used in this study has provided a huge variety of possible feeding situations for herbivores, in turn leading to an abundance of feeding opportunities for predators. According to Woodcock *et al.* (2007), vegetational complexity is of enormous import to the abundance and richness of arthropods. This is supported by Lawton (1983), who described how more arthropod species were supported by hosts with greater plant architectural complexity and how, in general, larger sized trees were more complex than smaller ones.

The season of arthropod collection had some impact on the richness and abundance of beetles collected. Both beetle richness and abundance were found to be higher during sampling period 1. Previously, in Chapter 3, it was found that repeated sampling had no significant impact on ordinal richness and abundance. This was largely unexpected, as other workers (Stork 1991, Stork and Hammond 1997, Azarbayjani *et al.* 1999) have

found that recolonization can be a slow process, taking between three months to over a year to complete. Perhaps the influence of repeated sampling is more apparent at the species level. Alternatively, the greater beetle richness and abundance observed in sampling period 1 might be explained by seasonal factors. For instance, the first sampling took place during mid-spring, while the second sampling was conducted in late autumn. Weather conditions alone are extremely different between these two sampling periods, and there is also the increased presence of flowers and pollen at the spring sampling. These, or any number of other possible changes in environmental conditions, could help to explain the change in beetle abundance and richness observed between the two seasons.

During this study, seven sites were used. Beetle richness was not greatly influenced by site, although there was some effect of site on beetle abundance during the second period of sampling. This could possibly be caused by changes in the nutritional quality of foliage, brought about by fertilizer use at some, but not all, sites. Site 6 had the greatest abundance of all the sites and, though fertilizers were used at this site, it is unclear if this was the cause.

One of the most significant factors influencing beetle species (especially herbivores) is host tree species. Azarbayjani *et al.* (1999) questioned whether the species found on an individual host tree are a random collection of available species or if the unique attributes of each host tree make it suitable for particular arthropod species. This study tested five tree species, three being mallees, *Eucalyptus polybractea*, *E. kochii* subsp. *borealis*, and *E. loxophleba* subsp. *lissophloia*, and two being remnant species, *Eucalyptus wandoo* subsp. *wandoo* and *E. astringens*. *E. polybractea* consistently had the highest beetle species richness, while *E. kochii* had generally low beetle richness values. In terms of abundance, *E. polybractea* again had one of the highest levels. Differences in abundance levels between the species were, however, only significant at the first sampling. Interestingly *E. polybractea* appears to be the only species with some agreement between beetle abundance and richness figures, while all the other host tree species had either high abundance or richness, but not both. Humphrey *et al.* (1999)

suggested that differences in canopy structure and light interception might be important in determining arthropod species diversity, and this could potentially be the case in the current study. Other species-specific factors, such as leaf nutrition and chemistry, are also likely to influence beetle (and other arthropod) richness and abundance.

Beetle assemblages were also influenced by the host tree species. Strong similarities were apparent between beetle assemblages resident on the same tree species. This is likely because the same tree species provide the same kinds of foods and shelter, favouring a certain suite of beetle species. The same types of vegetation (mallee/remnant) also tended to support similar beetle assemblages. This is because the environments that the two vegetation types are located in are quite different. Thomas and Marshall (1999) found that higher floral diversity, like that experienced in the remnant, increased arthropod diversity. Clough *et al.* (2007) also suggest that the composition of the vegetation matrix could play an important role in colonization by arthropods, indicating that in the remnant stands where other species were present, the colonization of mallees would be better. This is supported by Kavanagh *et al.* (2007), who claim that efforts to reverse degradation using eucalypt planting would be more likely to succeed if trees were planted close to remnant vegetation.

This chapter, which focuses on a single order, the Coleoptera (beetles), found many interesting relationships between environmental and chemical factors and the richness, abundance and structure of beetle assemblages. Soil nutrients have been suggested as being important (Braithwaite *et al.* 1983, Landsberg *et al.* 1990, Blanche and Westoby 1995) due to their influence on plant nutrition. In this study, however, no significant impact of soil nutrients on either beetle richness or abundance was observed. There was, however, some influence of soil nutrients on beetle assemblages. The strongest relationship was for soil phosphorus, with the remnant species (which had similar assemblages) tending to have lower soil phosphorus than the mallees. This is interesting, as other studies (Woodcock *et al.* 2007) suggest that high levels of soil nutrient levels, especially those caused by the application of fertilizers, can cause reductions in beetle species richness and abundance. Studies by Dudt and Shure (1994), on the other hand,

found that while the use of fertilizers increases plant biomass, it has no apparent influence on levels of leaf chemicals or on arthropod herbivory.

Like soil nutrients, leaf nutrients had no significant impact on beetle richness and abundance. The beetle assemblage, however, was somewhat influenced by leaf nutrients, especially leaf phosphorus, which was somewhat lower in the remnant species than the mallees. This is most likely linked to soil nutrition, with the same pattern occurring with soil phosphorus. The levels of nitrogen in foliage are widely reported to be important factors contributing to arthropod nutrition, growth, and grazing patterns (Gordon 1972, Fox and Macauley 1977, Slansky and Feeny 1977, White 1978, Basset 1992). This was not evident in the current study and, apart from Ohmart's study of chrysomelids (1991), there is little evidence to suggest that nitrogen limits beetle numbers in eucalypts.

In some studies, it has been shown that high levels of 1,8-cineole can act as a feeding deterrent or anti-feedant against most insects (Stone and Bacon 1994). Studies conducted by Edwards (1993) found that trees with high levels of cineole in their leaves experienced very little defoliation by arthropod herbivores compared to those with lower cineole levels. In the current study, however, the only significant influence of essential oils on beetle species richness and abundance was a negative one on beetle abundance in response to high leaf pinene. The composition of beetle assemblages, however, was affected by both leaf essential oils. Cineole was lower in the remnant species (which tended to have similar assemblages), while pinene was high in *E. loxophleba* subsp. *lissophloia* and the remnant species. It seems that, in the case of the remnant species, high pinene and low cineole may well combine to create assemblages quite different from those in the mallee vegetation.

Sideroxylonal levels had no significant impact on beetle richness or abundance. While strong variations between the various species in relation to sideroxylonal levels are apparent, they do not adequately explain the pattern of beetle assemblage similarities. Although phenols such as sideroxylonal are commonly regarded as feeding deterrents for phytophagous insects, there have been instances where they have been used, rather

than avoided by arthropods (Chapman and Blaney 1979, Bernays and Woodhead 1982, Ohmart and Larsson 1989, Herms and Mattson 1992). This might well help to explain the lack of influence of this supposedly highly toxic secondary plant compound in this study.

All of the aforementioned variables can contribute together to influence canopy arthropod assemblages. According to the BEST analysis, however, cineole alone best explains the patterns of beetle assemblages observed and so it seems to be the most important factor affecting beetle assemblage similarities in this study. While a large number of factors can influence beetle assemblages, and arthropods in general, it is clear that in some cases the presence or absence of a single factor can dominate the pattern of assemblage similarities observed. It is important, however, to recognise the contribution that interaction between factors is likely to have made to the beetle assemblages. This is supported by Provenza *et al.* (2003), who suggest that interactions between nutrients and toxins may influence food and habitat selection, and thus tend to regulate plant-herbivore interactions. Therefore, this result should be treated with caution, as the presence of so many factors interacting is sure to cause some confounding of results.

CONCLUSION

In this chapter, the ways in which arthropod biodiversity may be influenced were examined in more detail using the example of the order Coleoptera. It was interesting to see that the most species rich family was phytophagous (Curculionidae), while the most abundant families were predators (Staphylinidae and Coccinellidae).

Season of sampling was found to have an influence, with both beetle richness and abundance being higher at the first than the second sampling. Site, on the other hand, had very little influence. Of the tree species, *E. polybractea* had both the highest beetle species richness, and high levels of beetle abundance. In terms of the beetle assemblage

strong similarities were apparent between beetle assemblages resident on the same tree species.

No influence of soil or leaf nutrients on either beetle richness or abundance was observed. However, beetle abundance was found to have a negative relationship with leaf pinene. Cineole levels were lower in the remnant species (which tended to have similar assemblages), while pinene was high in *E. loxophleba* subsp. *lissophloia* and the remnant species. The other major leaf chemical examined, sideroxylonal had no significant impact on beetle richness or abundance. Statistical analysis selected cineole as the single factor best explaining the pattern of beetle assemblages observed, though this result should be treated with caution due to possible confounding of the results as a consequence of interaction between the factors.

One thing that is clear from this chapter is that it is very difficult to untangle the vast array of variables influencing both the plant and the associated arthropods. It is also clear that host tree species, factors related to the growth and development of the tree, such as leaf chemistry and soil nutrition, and interactions between nutrients and toxins are vital in determining the diversity and abundance of beetle assemblages.

CHAPTER 9

GENERAL DISCUSSION

INTRODUCTION

The broad aim of this research was to determine the habitat value of oil mallee plantings in agricultural landscapes in terms of canopy arthropod biodiversity. In order to achieve this aim, a series of supporting objectives were formulated. These objectives are stated below as a set of questions which must be answered in order to accomplish the stated aim:

- Does the canopy-dwelling arthropod assemblage vary in abundance, diversity and composition between oil mallee species?
- Are canopy-dwelling arthropod assemblages that are present on planted oil mallees similar in terms of abundance, diversity and composition to those found on remnant eucalypt vegetation?
- Do soil and leaf nutrient levels influence canopy-dwelling arthropod assemblages?
- Do leaf essential oils and secondary plant compounds influence canopy-dwelling arthropod assemblages?

In this chapter I will address each of these points in turn, following with a short comparison of ground and canopy fauna, a discussion of some of the benefits of oil mallees, and finishing with some general conclusions and recommendations.

DOES THE CANOPY-DWELLING ARTHROPOD ASSEMBLAGE VARY IN ABUNDANCE, DIVERSITY AND COMPOSITION BETWEEN OIL MALLEE SPECIES?

The question has often been asked whether the species collected from an individual host tree are a random collection of the local species, or if the unique attributes of each host tree make it suitable for a particular suite of arthropod species (Azarbayjani *et al.* 1999). It appears that in the current study there are very few differences between host trees of the three mallee species examined when it comes to their arthropod biodiversity. Table 38 provides a comparative summary of the species used in this study, summarising the trends in a range of factors which showed significant differences between species. Each of the three mallee species used in this study appear quite different in a number of ways, and could feasibly have their own characteristic arthropod assemblages as a result. *E. polybractea* was the largest of the three mallee species, generally being taller and more architecturally complex than the other two species (Table 38). In contrast, *E. loxophleba* subsp. *lissophloia* was much less architecturally complex, with a spreading habit and low foliage density. *E. kochii* subsp. *borealis* was somewhat intermediate, being significantly shorter than *E. polybractea* but with much denser foliage than *E. loxophleba* and possessing slender leaves which were quite different from either of the other two species.

Studies suggest that the larger size and greater architectural complexity of trees such as *E. polybractea* should tend to increase its attractiveness to arthropods and other animals (Lawton 1983, Kuris *et al.* 1980). This is because of the wider range of habitats and resources expected to be provided by this species compared to the other mallees. Conversely, with a spreading habit, open canopy and generally the lowest density of foliage of the three mallee species, *E. loxophleba* would be expected to have a lower richness and abundance of arthropods than the other species. The reason for this expectation is that arthropods resident on this species would be relatively vulnerable to predators and adverse environmental conditions (Lawton 1983).

Table 38: Species comparison summary table. Factors highlighted red were significantly lower than in other species, factors highlighted green were significantly higher than in other species and factors coloured yellow were intermediate between the high and low species. Only factors with statistically significant differences are presented here, except for tree characteristics which are subjective rankings. Significant differences are at p=0.05. Beetle assemblages of tree species highlighted dark blue are distinct from those of tree species highlighted light blue.

	Mallee species			Mallee next to remnant	Remnant species	
	<i>E. loxophleba</i> subsp. <i>lissophloia</i>	<i>E. kochii</i> subsp. <i>borealis</i>	<i>E. polybractea</i>	Mallee sp.	<i>E. astringens</i>	<i>E. wandoo</i> subsp. <i>wandoo</i>
<u>Invertebrate Characteristics</u>						
Leaf folding	Low	High	Low	Low	Low	Low
Psyllid	High					
Beetle richness		Low				
Beetle abundance	Low		High			Low
Beetle assemblage	Distinct				Distinct	
Beetle assemblage	Distinct	Distinct				
<u>Tree Characteristics</u>						
Tree size	Low	Low			High	High
Foliage density	Low	High				
Architectural complexity	Low		High		High	High
<u>Chemical Characteristics</u>						
Leaf N		High				
Leaf P	High	High	High	High	Low	Low
Cineole		High			Low	Low
Pinene	High	Low	Low		High	
Sideroxylonals	High	Low	Low		Low	

This was not supported by most of the current work, however, as no significant differences were observed in the richness, abundance or number of individuals of particular orders collected from the three tree species. The influence of architectural complexity was evident, however, on beetles at the species level. *E. polybractea* consistently had the highest beetle species richness, while *E. kochii* had generally low beetle richness values. At the first sampling period, *E. polybractea* also had the highest levels of beetle abundance, while *E. loxophleba* had the lowest. This suggests that the influence of plant architectural complexity, while not evident at the ordinal level, may be operating on arthropods at the species level.

According to some sources (Lawton and Price 1979, Campbell and Norman 1989), canopy size and structure is also an important factor influencing the diversity of arthropods. While the three species differed in their height, they all had similar canopy volumes. This, combined with the distinct lack of variability in the arthropod assemblages of the three species, suggests that canopy volume might be a stronger indicator of the abundance and diversity of arthropod assemblages than tree height.

While the three mallee species did not differ in their total arthropod abundance or their ordinal richness, there were some small differences in the types of leaf blemishes and sedentary arthropods present on the different species. Psyllids were more common on *E. loxophleba* than on the other mallee species and leaf folding was almost entirely restricted to *E. kochii* (Table 38). Yen (2002) suggests that many psyllids are specific to a single *Eucalyptus* species. The general absence of psyllids on the other two mallees indicates that the species of psyllids specific to those species were not located at these sites. This makes sense, as of the three mallee species, *E. loxophleba* is closest to its natural range in the study area, while *E. polybractea* is native to the eastern states and *E. kochii* has a more northern distribution in Western Australia than that of the study area. Since *E. loxophleba* is in its native environment, it seems entirely possible that psyllid species which have adapted to specialize on this species should be present in the study area.

E. kochii was also observed to have large amounts of leaf folding, which was almost unseen in the other species (Table 38). Leaf folds were observed to be constructed by a single spider species, which I have referred to as ‘origami’ spiders. Lawton (1983) considered that some arthropod diversity might be explained by the ease with which arthropod species could exploit the different leaf shapes exhibited by host tree species. The small, slender leaves of *E. kochii* appear to have been the main attraction for the leaf folding spiders. The leaves of *E. kochii* appear to be amenable to the folding conducted by the spiders for their shelter building. The other mallees had broader leaves, which would be much harder for the small spiders to manipulate, and it is my contention that leaf size is a major limiting factor in the occurrence of the leaf folding (origami) spiders.

While certain arthropod orders were more prevalent on particular mallee species than on others, most of these differences were not statistically significant. The few significant differences which were observed appear to be attributable to host specificity and the suitability of one mallee species over another in terms of providing a particular resource for particular arthropods. Overall, however, there are very few differences between the three mallee species examined in terms of their canopy arthropod assemblages, or alternatively, existing differences were not detected during this study due to the generally coarse level of identification used.

ARE CANOPY-DWELLING ARTHROPOD ASSEMBLAGES THAT ARE PRESENT ON PLANTED OIL MALLEES SIMILAR IN TERMS OF ABUNDANCE, DIVERSITY AND COMPOSITION TO THOSE FOUND ON REMNANT EUCALYPT VEGETATION?

In the current study, I compared five eucalypt species; three planted oil mallees and two tree species selected from remnant vegetation. The various factors which exhibited significant differences between species are presented in Table 38. Ordinal richness, total abundance, and the numbers of individuals in particular orders were not influenced significantly by tree species and so are not presented in the table. Beetle assemblages

were, however, influenced by host species. Beetle richness was particularly low in *E. kochii* but not significantly different between the other species. Beetle abundance was high in *E. polybractea*, low in *E. loxophleba* and *E. wandoo* and intermediate in the other species. The beetle assemblages of the mallees were distinct from those of the remnant species. However, *E. loxophleba* also appeared to have a beetle assemblage which was distinct from that of the other two mallees (Table 38).

As previously discussed, many authors (Lawton 1983, Humphrey *et al.* 1999, Thomas and Marshall 1999) would suggest that the higher architectural complexity and floral diversity of the remnant vegetation should attract and support a greater number and diversity of arthropods than the mallee vegetation. This, however, was not supported by the current study, despite the low diversity and relative simplicity of the mallee vegetation the mallees were able to support similar levels of arthropod richness and abundance to those of the far more complex and floristically diverse remnant.

One reason for the lack of variation between the tree species examined in this study could be a low level of specificity of the arthropods encountered in this area. Alternatively, the assertion of Novotny *et al.* (2002) that most specialization is at the genus level, occurring most often in large genera (such as *Eucalyptus*) could prove correct in this instance. Since the current study used five *Eucalyptus* species, all from the same Symphyomyrtus subgenus, it seems unlikely that any specialisation present in the arthropod assemblage would be detected in a species level comparison of tree species. This, combined with the coarse level of arthropod identification used in this study, make it difficult to detect differences in arthropod richness between the host tree species. This does not however, explain the similarities observed in arthropod abundance between the mallee and remnant vegetation types.

Proximity of Mallee Species to Remnant Vegetation

One possible explanation for the similarities between the mallee and remnant arthropod assemblages could be the proximity of the mallees to the remnant. Clough *et al.* (2007) suggest that the composition of the vegetation matrix could play an important role in

colonization by arthropods, indicating that mallees located near the remnant stands could have experienced more rapid recolonization than mallees located away from remnants. In the species comparison (Table 38) Mallee sp., the mallees growing adjacent to remnant vegetation, tended to have characteristics intermediate between those of the remnant species and those of the mallees growing distant from remnants. Perhaps some of the greater diversity of arthropods supported by the remnant was able to be transferred to the mallee species, which were located close to the remnants in this study. Kavanagh *et al.* (2007) support this, claiming that efforts to reverse degradation using eucalypt plantings would be more likely to succeed if trees were planted close to remnant vegetation. This does not explain, however, why mallees not planted near to remnant vegetation also had similar arthropod assemblages.

So are canopy-dwelling arthropod assemblages that are present on planted oil mallees similar to those found on remnant eucalypt vegetation? The answer is ... simply 'Yes'. It appears that the similarities between the trees are related to individual tree species characteristics, and the relatedness of the species examined, rather than to the origin of the vegetation. While some might find it disconcerting that there were so few differences between planted mallees and remnant vegetation, it is encouraging to think that this could mean mallees are a good fit for revegetation programs aimed at supporting biodiversity in agricultural areas. This is supported by Kavanagh *et al.* (2007), who claim that any eucalypt plantings are useful in improving agricultural landscapes by adding to and supporting remnant vegetation and biodiversity as a whole. They believe that even the types of eucalypt plantings used in this work, which generally consist of only a single species, are still important in the conservation of birds and other fauna in rural areas.

DO SOIL AND LEAF NUTRIENT LEVELS INFLUENCE CANOPY-DWELLING ARTHROPOD ASSEMBLAGES?

Soil nutrient levels are considered to be important factors contributing to arthropod nutrition, growth, and grazing (Braithwaite *et al.* 1983, Landsberg *et al.* 1990, Blanche and Westoby 1995) due to their influence on plant nutrition. Table 39 provides a comparison of the seven sites used in this study, summarising the trends in the few factors which showed statistical differences between sites. Generally speaking, the mallee sites had higher levels of soil phosphorus, and higher beetle abundance, while the remnant sites had higher levels of soil potassium. Other factors which might have been influenced by site, such as soil nitrogen, ordinal richness, total abundance, beetle species richness and the number of individuals in any particular order showed no significant differences between sites.

Table 39: Site comparison summary table. Factors highlighted green are significantly higher than at other sites, factors highlighted red are significantly lower than at other sites, factors coloured yellow are intermediate between the high and low sites. Only factors with statistically significant differences are presented here. Significant differences are at $p=0.05$.

	Mallee sites				Remnant sites		
	Parnell	McDougall	Marshall	Hassel	Tutanning	Sprigg	Hesford
Soil P	Yellow	High	Yellow	Yellow	Low	Low	Yellow
Soil K	Yellow	Low	Low	Low	High	Yellow	Yellow
Beetle abundance	Yellow	Yellow	High	Low	Low	Yellow	Low

nb: Soil variables at the remnant sites represent an amalgamation of samples from both remnant vegetation and adjacent planted mallee.

Of the sites that showed statistical differences, site 2 (McDougall) had higher levels of soil phosphorus than all the other sites and had one of the lowest levels of soil potassium. Site 3 (Marshall) had the highest levels of beetle abundance of all the sites and also had one of the lowest values for soil potassium. Site 5 (Tutanning), the only site with very high soil potassium, had very low levels of both soil phosphorus and low beetle abundance compared to other sites. It is suggested that the average to high levels of soil phosphorus at the mallee sites are the result of the use of phosphorus fertilizers on crops and pastures sown between the mallee alleys. The low levels of soil potassium at the mallee sites are most likely the result of the removal of soil nutrients in harvested

crops (Nye and Greenland 1964, Stoorvogel *et al.* 1993), exacerbated by the limited application of potassium-containing fertilizers (Stoorvogel *et al.* 1993).

While there was no significant impact of soil nutrients on arthropod ordinal richness, both soil phosphorus and soil potassium appeared to have an impact on arthropod abundance. In general terms, higher levels of soil phosphorus were associated with higher arthropod abundance, while high levels of soil potassium tended to be associated with lower arthropod abundance. This is likely the result of the influence of fertilizers at the mallee sites increasing plant nutritive quality and in turn arthropod numbers (Fox and Morrow 1992).

The richness and abundance of beetles was not influenced by soil nutrients in the current study. However, there was some influence of soil nutrients on beetle assemblages. The strongest relationship was for soil phosphorus, with the trees in remnants which tended to have lower soil phosphorus, having assemblages which were distinct from those of the mallee species, which tended to have higher levels of soil phosphorus (Table 38 & Table 39). This suggests that the application of fertilizers might actually support the development of assemblages distinct from those occurring in natural vegetation.

Leaf nutrition was more variable between both sites and tree species than was soil nutrition. The levels of leaf nutrients are regarded as being important factors in the growth and development of herbivorous arthropods (Mattson 1980). Nitrogen, in particular, has been implicated as a limiting factor in many plant-herbivore relationships (Soo Hoo and Fraenkel 1966a, 1966b, Gordon 1972, Fox and Macauley 1977, Slansky and Feeny 1977, Onuf 1978, White 1978). One of the main findings of the current study in terms of the effects of nutrition was that, while soil borne nitrogen apparently had little or no effect, high levels of leaf nitrogen were associated with higher arthropod ordinal richness at the mallee sites and higher arthropod abundance at all sites. Of the mallee sites, site 2 (McDougall) had greater levels of leaf nitrogen than any of the other sites, perhaps related to the greater levels of soil nutrition, which probably resulted from fertilizers having been applied at this site. The higher arthropod richness and abundance

observed in relation to leaf nitrogen could be due to the higher nutritional quality of the leaves attracting, and being able to sustain, a greater variety of leaf-eating arthropods and, in turn, their predators (Moran and Hamilton 1980, Basset 1992).

Leaf phosphorus was greater at sites 3 (Marshall) and 4 (Hassel) than any of the other sites. These were mallee sites with slightly heavier soil. Overall, the mallee species, whether *E. loxophleba* subsp. *lissophloia*, *E. kochii* subsp. *borealis*, or *E. polybractea*, had higher levels of leaf phosphorus than the tree species in the remnant vegetation, regardless of whether they were at a mallee or remnant site (Table 38). This suggests a possible difference in the biology or physiology of these plants, although the difference could also be attributed to differences in fertilizer use. It has been suggested that different leaf nutrient levels between species located in the same area might be related to differences in their root physiologies or their systems of nutrient storage (Majer *et al.* 1992). In the current study, high levels of leaf phosphorus were associated with lower arthropod ordinal richness, implicating fertilizers in the reduction of arthropod richness in agricultural areas.

Like soil nutrients, leaf nutrients had no significant impact on beetle richness and abundance. The beetle assemblage, however, was somewhat influenced by leaf nutrients, especially leaf phosphorus, which tended to be lower in the remnant species than the mallees (Table 38). This is most likely linked to soil nutrition, which is heavily influenced by fertilizers, with the same pattern occurring with soil phosphorus.

Overall, leaf nutrition was greater in the mallee species than the remnant species. The higher leaf nutrition of the mallee species is likely to be related to fertilizer use. This suggestion is supported by work conducted by Fox and Morrow (1992), which found that levels of foliage nutrients were improved by the application of a balanced (NPK) fertilizer.

DO LEAF ESSENTIAL OILS AND SECONDARY PLANT COMPOUNDS INFLUENCE CANOPY-DWELLING ARTHROPOD ASSEMBLAGES?

Leaves are one of the major plant tissues consumed by herbivorous arthropods, so it is reasonable to expect that the chemistry and nutritional value of leaves should be significant determinants of arthropod richness and abundance. In the current study, the leaf chemicals cineole, pinene and sideroxylonal were investigated in relation to their influence on arthropod assemblages.

It is widely accepted that secondary plant compounds are one of the determining factors in the attractiveness of plants to herbivores (Atsatt and O'Dowd 1976). In the present study, one of the major secondary plant compounds tested for was the terpenoid 1,8-cineole. The remnant species, *E. astringens* and *E. wandoo*, had lower levels of this essential oil than the mallee species (Table 38). *E. kochii* had the highest levels of all the species tested, followed by *E. loxophleba*. The most obvious explanation for this is the simple genetic variation in the production of leaf essential oils between species, although it might be argued that the exposure of mallee species to fertilizers could also be responsible (O'Reilly-Wapstra *et al.* 2005). Some studies have suggested that cineole acts as a feeding deterrent to arthropods (Edwards 1993, Stone and Bacon 1994). This is not supported by the current work, however, as there was a significant positive relationship between leaf cineole content and arthropod abundance. A number of individual orders including, Hemiptera, Hymenoptera, and Psocoptera, were also in greater numbers in the presence of high levels of leaf cineole. *E. kochii*, which had the highest levels of leaf cineole, also had the highest total arthropod abundance, as well as the highest individual abundances of several orders. This suggests that cineole may have attractant properties to certain arthropod groups at high concentrations. This is supported by a number of studies which have found 1-8 cineole attractive to groups as diverse as orchid bees (Ricklefs *et al.* 1969), western flower thrips (Chermenskaya *et al.* 2001), and banana weevils (Ndiege *et al.* 1996).

The levels of the other terpenoid tested, pinene, did not follow the same pattern as that of cineole. The mallee species, *E. polybractea* and *E. kochii*, had very low levels of leaf pinene, while the remnant species, *E. astringens*, had the highest pinene levels, followed by another mallee, *E. loxophleba* (Table 38). The remnant sites had higher levels of leaf pinene compared to the mallee sites. This was due to the high levels exhibited by the remnant species, *E. astringens* and *E. loxophleba*, the latter being present as the mallee species comparison at two of the three remnant sites. Variability in leaf pinene contents between species could help to explain their variability in arthropod abundances, as there was a significant negative relationship between leaf pinene content and arthropod abundance. *E. astringens*, in particular, with its very high levels, had low total arthropod abundance and low or very low numbers of all the examined arthropod orders.

While leaf cineole and pinene levels had no impact on arthropod ordinal richness, when individual orders were examined, some significant relationships could be seen. Cineole had a generally positive influence, with high levels being correlated with high numbers of Hemiptera, Hymenoptera, and Psocoptera, while pinene had a generally negative effect, with high levels being correlated with low numbers of Acarina, Coleoptera, Hymenoptera and Psocoptera. There also appears to be a balance between the two essential oils, which enhances these effects. It appears that low levels of pinene, coupled with high cineole could possibly be stimulatory to arthropods. The opposite scenario, of high levels of pinene with low cineole, may inhibit arthropods, reducing their abundance and the prevalence of certain orders. The beetle assemblage was also influenced by interactions between leaf essential oils. The remnant species both had high pinene and low cineole, which appears to have resulted in a beetle assemblage quite different from that on those mallees which generally had low pinene and high cineole levels (Table 38). Leaf essential oils had impacts on arthropod abundance, individual orders, and on the beetle assemblage. However, there was a distinct lack of influence on arthropod ordinal richness. It is suggested that this might be related to dose, meaning that below a certain threshold of total oil concentration, no effects may be apparent (Morrow and Fox 1980).

The third leaf chemical examined in this study, sideroxylonal was present in very high levels in the mallee species, *E. loxophleba*, while the other mallee species had none. The two remnant species, *E. astringens* and *E. wandoo*, had lower levels (Table 38). Despite this, the current study revealed no significant effect of total leaf sideroxylonals on arthropod abundance. Arthropod ordinal richness, however, was significantly negatively correlated with total sideroxylonals. Although phenols like sideroxylonals are commonly regarded as feeding deterrents for phytophagous insects, in the current study high levels did not necessarily correlate to low arthropod abundances. For example, *E. loxophleba* had very high sideroxylonal levels and high arthropod abundance, while *E. wandoo* had moderate sideroxylonal levels but a low number of arthropods. The relationship between arthropods and sideroxylonal levels is not a clear-cut one. While some arthropods are adversely affected, others are able to use sideroxylonals as nutrients or sequester them in their bodies for use in defense against predators (Chapman and Blaney 1979, Ohmart and Larsson 1989). Undoubtedly, some level of adaptation or host specificity is at work here. However, the concentration of sideroxylonals present in the foliage is also likely to have an influence in determining the pattern of arthropod assemblages observed (Andrew *et al.* 2007). It is of interest to note that while essential oils had strong impacts (cineole attractant, pinene repellent) on arthropod abundance, and no effect on ordinal richness, sideroxylonals had a significant negative impact on ordinal richness and no impact on arthropod abundance. Put simply, it appears that sideroxylonals might have more influence on the types of species present, while essential oils have more impact on the number of individuals present.

The leaf chemicals examined here differed markedly in their relationships with arthropod ordinal richness, abundance and on the prevalence of particular arthropod orders. This supports much of the existing literature. However, Fox and Macauley (1977) and Ohmart and Larsson (1989) claim that herbivore-plant relations are not influenced by variations in the leaf concentrations of tannins, essential oils, phenols and other secondary plant compounds. In the current study the main purpose of leaf chemicals appears to be as feeding deterrents or attractants. However, a range of other functions of leaf chemicals have been suggested, including acting as antioxidants,

protecting leaves from photodamage, osmotic effects and other stresses, acting in the attraction of pollinators, and in providing allelopathic defense against other plants (Bennett and Wallsgrove 1994, Close and McArthur 2002). Whatever their purpose, it is clear that there is a range of plant secondary compounds present in the eucalypts studied, and some level of influence on arthropods should be generally accepted.

COMPARISON OF GROUND AND CANOPY FAUNA

In work by an honours student, Mei Chen Leng (2006), ground beetles were collected from one of the same sites used in the current study (Hesford). This provided an opportunity to compare ground and canopy beetle fauna. A comparison of the species collected showed that 36% of beetle species collected from the ground were also present in the canopy. Three of the species collected from the ground in the remnant and five of those collected from the paddock were also collected in the current study. Nine species were found on the ground in both the remnant and the paddock and were also present in the canopy. This suggests that species which were versatile enough to occur in both the remnant and the paddock were more likely to be found in the canopy as well as on the ground. It also indicates that while remnants and mallee plantings have some overlap in their fauna, both habitat types have some unique species which help to make up their own distinct fauna. This implies that the addition of mallees to agricultural systems, even in the presence of remnants, could improve the overall biodiversity of an area.

BENEFITS OF OIL MALLEE PLANTINGS

Aside from the financial benefits to farmers from the sale of oil and other products of oil mallees, there are a number of other less conspicuous benefits associated with oil mallee plantings in agricultural areas. This section will discuss two of them:

- Benefits to avifauna; and
- Harbours of beneficial arthropods.

Benefits to Avifauna

The benefits to insectivorous birds and other avifauna from oil mallees might appear obvious, but nevertheless are worth discussing. As we have seen from the current study, large numbers of insects and other arthropods can be found in the mallees. Insectivorous birds obtain a great deal of nutrition from these insects (Recher *et al.* 1985) and so would find the mallees an attractive feeding location. In a study of native birds (Recher *et al.* 1983), bird numbers were found to fluctuate with arthropod abundance. Arthropod numbers were at their highest in summer, and consequently the numbers and diversity of bird species also increased in summer.

From a bird's point of view, large areas of the wheatbelt must seem extremely barren, being devoid of any vegetation taller than a wheat plant. Many native birds are migratory, but will not fly huge distances across open farmland (Recher *et al.* 1983). In these areas especially, mallees would provide a haven in an otherwise desolate landscape. Unlike migratory birds in the northern hemisphere, Australian native birds rarely accumulate fat, opting instead to stop regularly to feed (Recher 2000). Hence, well designed mallee belts can provide connectivity, or a least rest points, between native vegetation remnants, helping birds to reach feeding and nesting sites, and stabilizing population numbers. As well as providing insect prey, mallees, and other eucalypts also supply alternate sources of food, such as nectar and seed, for omnivorous bird species when other food sources are limited (Recher 2000).

Oil mallees, especially mixed stands of local species, are preferable to plantations of introduced species like pines, for native bird conservation. Native tree species are always preferable in conservation, as native animals have evolved with them and are adapted to make the best use of them. Also, plantations are generally grown in monoculture with all undergrowth removed. This makes the habitat architecturally simple and unattractive to most native birds, while contributing to the spread of exotic bird species (Barrett 2000). Greater numbers and diversity of arthropods are supported

by a diverse habitat including a range of species and sizes of vegetation. Recher *et al.* (1985) noted that forest productivity was a good predictor of the abundance of arthropods and other bird foods. This suggests that mixed plantings of mallees, or even mallees grown in alley systems with crops or pastures, would support more types and numbers of arthropods than any plantation. This in turn provides a range of food sources and other opportunities, thus encouraging native bird species, in a region of Western Australia where 93% of the original native vegetation has been cleared.

Beneficial Arthropods

Apart from being a food source for insectivorous birds, some arthropods are beneficial and worth encouraging in their own right. Two of the major benefits to agriculture from arthropods are from pollination services (Recher 1981) and predation. A large number of arthropods pollinate our crops and pastures and are essential to production. Several studies have also found that arthropod diversity provides an important ecosystem function in the form of natural pest control (Ives *et al.* 2000, Wilby and Thomas 2002, and Gurr *et al.* 2003). Along with resistant varieties and cultural control measures, predatory and parasitic arthropods which attack crop and pasture pests are an integral part of integrated pest management systems (Sunderland *et al.* 1987). Without pest control by natural enemies, we become heavily reliant on chemical and cultural practices which are less effective, and generally less desirable than biological controls, to producers and consumers alike. This is supported by Ostman *et al.* (2003) who suggest that chemical control measures are expensive, have negative side effects on the environment and, over time, become ineffective as arthropods develop resistance to commonly used insecticides. In Europe, enhancing the potential of naturally occurring predators and parasitoids to control crop pests is a recognised pest control strategy referred to as Conservation Biological Control (CBC) (Eilenberg *et al.* 2001).

In many cases, however, beneficial arthropods are unable to travel the distances required to perform these functions or simply do not occur in an area due to a lack of resources. Seasonal movements between agricultural fields and field margins are common for

various beneficial arthropods, and have been described as an adaptation to the cyclic pattern of disturbance in arable landscapes (Wissinger 1997). One of the most important factors contributing to pest suppression by natural enemies in agricultural landscapes is the provision of overwintering structures. This is supported by a large number of studies (Oberger *et al.* 2008, Pywell *et al.* 2005, Wissinger 1997, Weisser and Siemann 2004). For example, Wratten and Thomas (1990) found that predatory beetles have a seasonal dependence on field margin habitats. Studies have also shown that there is considerable movement between field boundaries and the crop by beneficial arthropods seeking prey (Wratten and Thomas 1990). In European studies, beetles (Coleoptera) were found to use hedgerows and field margins as sources of food and overwintering sites in agricultural areas (Pollard 1968, Sotherton 1984, Dennis *et al.* 1994). However, beneficial arthropods will not remain in areas without the overwintering sites, shelter and adequate food supplies needed to survive (Dix and Leatherman 1988, Pasek 1988, Heisler and Dix 1991). Hence the provision of these resources is vital to the success of pest control by natural enemies.

One of the ways of maximizing the number and quality of overwintering sites is by increasing habitat heterogeneity and structural diversity. Landscape heterogeneity has been shown to contribute to greater invertebrate diversity and to the increased effectiveness of natural enemies (Colunga-Garcia *et al.* 1997, Bommarco 1998, Holland and Fahrig 2000, Bianchi *et al.* 2006, Marino *et al.* 2006). Work by Tsitsilas *et al.* (2006) suggested that shelterbelts with ground cover could harbour a diversity of beneficial organisms with the potential to suppress pest numbers in adjacent pastures. Ameixa and Kindlmann (2008), however suggest that the abundance of biocontrol agents, rather than their species diversity is the most important factor. Other studies have found that care should be taken when selecting plants for field boundaries as specific plants may encourage either pest species or natural enemies (Baggen *et al.* 1999, Wäckers 2001).

Structural diversity within agro-ecosystems, in the form of increased plant species diversity and increased plant architectural complexity (Lawton 1983), has been found to

lead to community stability and the encouragement of arthropod diversity, resulting in a reduction in damage to crops by pests. Reductions in structural diversity, on the other hand, had the opposite effect, resulting in insect communities dominated by pests (Ryskowski *et al.* 1993). A study by Ameixa and Kindlmann (2008) however found no clear evidence that agricultural intensification lead to a decrease in predator abundance. Landis *et al.* (2000) however, claim that the loss of invertebrate diversity due to agricultural practices can be countered to some extent by increasing vegetation diversity through provision of non-crop habitats such as those provided by shelterbelts, linear field boundaries, road edges and remnant vegetation. This is supported by Thomas, Wratten and Sotherton (1992), who found that increased structural diversity within the agro-ecosystem led to community stability, which resulted in some regulatory effect on pest populations by predators moving into the field from edge habitats. A review of conservation biological control by Bianchi *et al.* (2006) found that ‘landscape complexity enhanced natural enemy populations in 74% of cases’. This supports the general consensus of a large number of researchers, that the provision of diversified landscapes offering a range of crop and non-crop habitats are one of the best ways of conserving biodiversity and sustaining natural pest control functions (Bianchi *et al.* 2006).

It is clear from these studies that well designed oil mallee plantings, using native species, could provide the necessary resources to enhance beneficial arthropod populations in the Western Australian wheatbelt. In turn, this could lead to a range of environmental, as well as financial, benefits. However, despite the benefits, oil mallee plantings, and other forms of conservation biological control, are not as widely utilised as might be expected. One of the major obstacles to the widespread use of conservation based biological control is the perceived cost of its implementation (Griffiths *et al.* 2008). This is largely due to the lack of studies quantifying the costs and benefits in financial terms. Farming is a business, and any attempts to change farming systems for the benefit of nature conservation must recognize that the benefits to farming communities should be emphasized, in order to maximize adoption.

CONCLUSIONS AND RECOMENDATIONS

At the beginning of this chapter I outlined the objectives and aim of this thesis. I have dealt with the objectives individually and would now briefly like to address the overall aim of this thesis, to determine whether oil mallee plantings enhance arthropod biodiversity in agricultural landscapes.

This thesis has shown that oil mallees do have a good level of biodiversity. Overall, the mallees had a level of diversity not dissimilar to that of high quality remnant vegetation. When planted in alleys across agricultural fields, they represent a significant change in the vegetative and architectural diversity of the landscape. This has allowed for the development of new niches for native arthropods, which in turn will provide food for birds, lizards and other native animals, while reducing dryland salinity, improving the aesthetics of the wheatbelt, and possibly reducing the need for pesticides.

Aside from the environmental benefits, oil mallees also provide the potential for farmers to make an income from something designed to ameliorate environmental problems. In order for oil mallee planting to be adopted on a large scale, however, the right incentives need to be in place and the system must be well developed. This may require financial incentives to farmers for plantings, making sure that processing and transport logistics are worked out well in advance, and most importantly developing guidelines for selecting the right mallee species for each location and situation. If all of these measures are put into place, then the successes of the first farmers to take up the system will entice other farmers and encourage further investment. If oil mallee farming can be developed appropriately, it has the potential to be one of a rare breed of agricultural ventures, producing both financial and environmental benefits.

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APPENDICES

Appendix A

Weather Conditions

Weather conditions at times of sampling

Date	Weather conditions
<i>Sampling 1 - 2005</i>	
Mon 17 Oct	Rainy
Tues 18 Oct	Fine, still
Wed 19 Oct	Fine, still
Thu 20 Oct	Fine, windy
Fri 21 Oct	Overcast
Mon 31 Oct	Fine, light breeze
Tues 1 Nov	Fine, light breeze
Wed 2 Nov	Fine, breezy
Thu 3 Nov	Fine, wind gusts
Fri 4 Nov	Fine, gusty winds in the afternoon
<i>Sampling 2 - 2006</i>	
Mon 1 May	Fine, still
Tue 2 May	Overcast, slight breeze
Wed 3 May	Fine, clear, still (fog in the morning), afternoon drizzle
Thu 4 May	Fine, clear, still
Fri 5 May	Light drizzle, still
Mon 15 May	Showers, no wind
Tue 16 May	Overcast, light breeze
Wed 17 May	Cloudy, light breeze
Thu 18 May	Fog, then fine and clear, light breeze

Appendix B

Material Safety Data Sheet

Appendix C

Spray Study Means and Frequencies

Mean arthropod numbers collected from three species of *Eucalyptus* trees sprayed once H2 and twice H1 in the Wickepin and Pingelly shires during May 2006.

LOCATION		Site 7 H1			Site 7 H2		
TREE SPECIES		lox.	wandoo	astrin.	lox.	wandoo	astrin.
Arachnida	Araneae	15.8	35.0	13.2	9.6	22.0	19.8
	Acarina	2.3	3.2	3.0	1.0	6.2	3.8
	Pseudoscorpionida	0.0	3.5	1.5	0.0	18.0	1.0
Collembola		3.4	2.8	2.0	2.8	4.4	2.0
Insecta	Archaeognatha	0.0	0.0	0.0	0.0	0.0	0.0
	Thysanura	0.0	0.0	0.0	0.0	0.0	0.0
	Odonata	0.0	0.0	0.0	0.0	0.0	0.0
	Blattodea	1.0	3.5	1.0	1.0	2.0	1.0
	Isoptera	1.0	0.0	0.0	3.0	1.0	1.0
	Mantodea	0.0	0.0	0.0	0.0	0.0	0.0
	Dermaptera	0.0	1.3	1.0	0.0	1.0	0.0
	Orthoptera	0.0	1.0	0.0	0.0	0.0	2.0
	Psocoptera	4.0	11.3	5.5	4.2	49.6	2.0
	Hemiptera	17.4	17.8	11.8	14.8	24.2	20.3
	Thysanoptera	3.3	2.5	1.8	4.0	35.8	9.0
	Neuroptera	2.0	0.0	0.0	0.0	0.0	0.0
	Coleoptera	23.2	43.0	24.6	24.6	35.4	39.0
	Diptera	11.0	95.0	38.6	8.8	82.4	21.8
	Lepidoptera	1.0	6.0	1.8	1.0	2.8	0.0
	Hymenoptera	101.2	44.6	40.8	100.6	89.8	49.4
	larvae	2.3	6.0	17.5	2.6	19.2	7.0

Frequency of arthropod occurrence per five trees collected from three species of *Eucalyptus* sprayed once H2 and twice H1 in the Wickepin and Pingelly shires during May 2006.

LOCATION		Site 7 H1			Site 7 H2		
TREE SPECIES		lox.	wandoo	astrin.	lox.	wandoo	astrin.
Arachnida	Araneae	5	5	5	5	5	5
	Acarina	3	5	4	1	5	4
	Pseudoscorpionida	0	4	2	0	4	1
Collembola		5	4	3	5	5	5
Insecta	Archaeognatha	0	0	0	0	0	0
	Thysanura	0	0	0	0	0	0
	Odonata	0	0	0	0	0	0
	Blattodea	1	2	1	2	3	1
	Isoptera	2	0	0	2	1	1
	Mantodea	0	0	0	0	0	0
	Dermaptera	0	3	1	0	2	0
	Orthoptera	0	1	0	0	0	1
	Psocoptera	4	4	4	5	5	2
	Hemiptera	5	5	5	5	5	4
	Thysanoptera	3	4	5	4	5	3
	Neuroptera	1	0	0	0	0	0
	Coleoptera	5	5	5	5	5	5
	Diptera	5	5	5	5	5	5
	Lepidoptera	2	5	4	1	5	0
	Hymenoptera	5	5	5	5	5	5
	larvae	3	5	4	5	5	5

Appendix D

Mallee Site Tree Measurements

Tree measurements: Mallees, Sampling 1 (Spring)

Tree No.	Width N-S (m)	Width E-W (m)	Radius (m)	Canopy base ht (m)	Tree ht (m)	Canopy ht (m)	Canopy vol (m ³)
001	2.10	2.11	1.05	1.10	4.00	2.90	6.73
002	2.05	2.50	1.14	0.50	4.34	3.84	10.30
003	2.15	2.20	1.09	0.45	3.49	3.04	7.53
004	2.47	2.90	1.34	0.40	3.37	2.97	11.14
005	2.25	2.39	1.16	1.00	4.92	3.92	11.04
006	3.10	3.50	1.65	0.40	3.00	2.60	14.77
007	2.65	2.40	1.26	0.60	2.50	1.90	6.33
008	2.40	2.25	1.16	0.50	2.60	2.10	5.94
009	2.14	2.80	1.24	0.25	2.75	2.50	7.84
010	3.40	3.15	1.64	0.50	3.00	2.50	14.02
011	2.70	2.60	1.33	0.80	3.60	2.80	10.29
012	2.30	2.50	1.20	0.70	3.20	2.50	7.53
013	2.01	3.35	1.34	0.55	2.50	1.95	6.88
014	2.36	2.40	1.19	0.90	3.67	2.77	8.21
015	3.40	3.38	1.70	1.00	3.60	2.60	15.64
016	1.68	1.87	0.89	0.80	2.24	1.44	2.37
017	2.15	1.70	0.96	0.70	2.50	1.80	3.44
018	1.70	1.55	0.81	0.40	2.50	2.10	2.90
019	1.90	2.00	0.98	0.40	2.65	2.25	4.48
020	2.00	1.95	0.99	0.40	2.35	1.95	3.98
021	2.20	2.56	1.19	0.45	1.50	1.05	3.10
022	2.70	3.10	1.45	0.60	2.20	1.60	7.01
023	2.50	2.70	1.30	0.50	2.66	2.16	7.63
024	2.40	2.30	1.18	0.50	2.60	2.10	6.07
025	2.65	2.95	1.40	1.00	2.70	1.70	6.96
026	2.20	3.70	1.48	1.20	4.60	3.40	14.49
027	2.50	3.40	1.48	2.50	5.20	2.70	12.02
028	3.50	3.50	1.75	1.80	5.50	3.70	23.73
029	2.15	2.80	1.24	1.60	4.80	3.20	10.09
030	2.60	2.70	1.33	0.80	5.00	4.20	15.44
031	2.50	2.40	1.23	0.60	2.50	1.90	5.97
032	3.10	3.00	1.53	1.00	3.40	2.40	11.69
033	2.50	2.40	1.23	0.80	2.90	2.10	6.60
034	2.40	2.05	1.11	0.60	1.80	1.20	3.09
035	2.30	2.15	1.11	1.60	2.70	1.10	2.85
036	3.10	2.90	1.50	1.20	4.00	2.80	13.18
037	2.00	2.00	1.00	2.50	4.80	2.30	4.82
038	2.30	1.60	0.98	3.28	5.10	1.82	3.51
039	3.50	3.00	1.63	2.30	4.40	2.10	11.55
040	3.80	3.00	1.70	2.00	4.40	2.40	14.33
041	2.60	2.50	1.28	1.50	4.40	2.90	9.87
042	3.30	3.30	1.65	1.20	5.00	3.80	21.67
043	3.10	2.20	1.33	1.40	4.90	3.50	12.50
044	2.50	1.95	1.11	1.20	4.30	3.10	7.91
045	2.40	2.15	1.14	1.60	5.10	3.50	9.46
046	2.50	2.90	1.35	2.00	6.20	4.20	15.94
047	2.55	2.40	1.24	1.00	3.40	2.40	7.69
048	2.55	2.10	1.16	1.40	5.10	3.70	10.37
049	2.40	2.70	1.28	2.10	5.40	3.30	11.20
050	2.40	3.00	1.35	1.90	5.10	3.20	12.06
051	2.40	1.90	1.08	0.55	2.50	1.95	4.66
052	2.90	2.50	1.35	1.15	2.80	1.65	6.26
053	2.40	2.40	1.20	0.85	2.00	1.15	3.47
054	3.10	3.00	1.53	0.85	2.70	1.85	9.01
055	2.90	2.10	1.25	1.05	2.90	1.85	5.90
056	3.00	3.30	1.58	0.10	3.10	3.00	15.55
057	2.70	3.20	1.48	0.90	4.30	3.40	15.38
058	1.70	1.70	0.85	0.80	4.50	3.70	5.60
059	2.25	2.00	1.06	0.50	2.70	2.20	5.18
060	1.90	2.10	1.00	1.10	2.90	1.80	3.76

Tree measurements: Mallees, Sampling 2 (Autumn)

Tree No.	Width N-S (m)	Width E-W (m)	Radius (m)	Canopy base ht (m)	Tree ht (m)	Canopy ht (m)	Canopy vol (m ³)
001	2.50	2.90	1.35	1.20	5.10	3.90	14.80
002	2.20	2.65	1.21	0.40	4.80	4.40	13.43
003	2.00	2.60	1.15	0.50	3.80	3.30	8.98
004	2.30	3.10	1.35	0.60	4.50	3.90	14.56
005	2.80	3.10	1.48	0.80	4.80	4.00	18.18
006	3.10	3.40	1.63	0.20	4.30	4.10	22.63
007	3.00	2.55	1.39	0.50	2.70	2.20	8.81
008	2.70	2.50	1.30	0.50	2.90	2.40	8.48
009	2.50	2.70	1.30	0.10	2.80	2.70	9.54
010	3.90	3.90	1.95	0.30	3.60	3.30	26.28
011	3.20	3.40	1.65	0.60	3.80	3.20	18.23
012	3.10	3.50	1.65	0.40	4.00	3.60	20.45
013	2.90	3.10	1.50	0.40	2.50	2.10	9.89
014	3.10	3.30	1.60	0.80	3.70	2.90	15.53
015	3.50	5.30	2.20	1.20	4.40	3.20	31.08
016	1.75	2.40	1.04	0.40	2.50	2.10	4.62
017	2.70	0.40	0.78	0.20	2.90	2.70	1.53
018	2.10	2.10	1.05	0.40	3.20	2.80	6.47
019	1.80	2.30	1.03	0.40	3.00	2.60	5.64
020	2.60	2.70	1.33	0.10	3.00	2.90	10.66
021	2.30	2.65	1.24	0.20	2.10	1.90	6.06
022	2.50	3.30	1.45	0.50	2.60	2.10	9.07
023	2.50	2.80	1.33	0.20	2.70	2.50	9.16
024	2.90	2.30	1.30	0.50	2.60	2.10	7.33
025	2.80	3.50	1.58	1.20	2.70	1.50	7.70
026	2.50	2.90	1.35	1.40	4.70	3.30	12.53
027	2.40	3.80	1.55	2.00	5.20	3.20	15.28
028	3.70	3.90	1.90	1.80	5.50	3.70	27.96
029	1.90	3.00	1.23	1.70	4.80	3.10	9.25
030	2.60	3.10	1.43	0.50	4.90	4.40	18.57
031	2.90	2.75	1.41	0.50	2.90	2.40	10.02
032	3.80	3.70	1.88	0.70	3.70	3.00	22.09
033	2.85	2.70	1.39	0.65	3.40	2.75	11.08
034	2.20	1.90	1.03	0.75	1.80	1.05	2.30
035	2.45	2.30	1.19	1.60	2.70	1.10	3.25
036	3.70	2.95	1.66	1.40	4.30	2.90	16.57
037	2.40	1.95	1.09	2.60	4.80	2.20	5.39
038	2.60	2.05	1.16	3.40	5.20	1.80	5.02
039	3.65	3.40	1.76	1.90	4.70	2.80	18.19
040	3.80	3.05	1.71	0.85	4.80	3.95	23.97
041	2.65	2.25	1.23	1.10	4.50	3.40	10.61
042	3.80	3.30	1.78	1.00	5.00	4.00	26.26
043	2.90	2.45	1.34	1.30	5.20	3.90	14.51
044	2.50	2.00	1.13	1.20	4.40	3.20	8.38
045	2.40	2.10	1.13	1.40	5.20	3.80	10.03
046	2.50	3.30	1.45	2.80	6.30	3.50	15.12
047	2.65	2.40	1.26	1.00	3.70	2.70	8.99
048	2.90	2.30	1.30	1.70	4.90	3.20	11.18
049	2.70	3.00	1.43	1.70	5.60	3.90	16.54
050	2.70	2.80	1.38	2.00	5.10	3.10	12.27
051	2.55	1.95	1.13	0.80	2.70	1.90	4.95
052	3.20	2.40	1.40	1.50	3.40	1.90	7.64
053	2.90	2.75	1.41	0.90	2.60	1.70	7.10
054	3.30	3.00	1.58	0.80	2.90	2.10	10.89
055	3.20	2.50	1.43	1.00	3.70	2.70	11.31
056	3.70	4.00	1.93	0.20	3.60	3.40	26.35
057	2.80	3.40	1.55	1.00	4.80	3.80	18.94
058	2.60	2.50	1.28	1.20	4.70	3.50	11.91
059	2.70	2.50	1.30	0.60	3.60	3.00	10.60
060	2.40	2.40	1.20	1.00	4.10	3.10	9.35

Appendix E

Mallee Site Flowering and Site Information

Flowering, sampling and site information: Mallees, Sampling 1 (Spring)

Tree No.	Date sampled	Taxon	Site	Locality	Alley position	Alley formation	Flowering
001	18/10/2005	<i>E. polybractea</i>	Parnell	Harrismith	N - C	4 row	Just finished
002	18/10/2005	<i>E. polybractea</i>	Parnell	Harrismith	S - C	4 row	Just finished
003	18/10/2005	<i>E. polybractea</i>	Parnell	Harrismith	N - C	4 row	Just finished
004	18/10/2005	<i>E. polybractea</i>	Parnell	Harrismith	S	4 row	Just finished
005	18/10/2005	<i>E. polybractea</i>	Parnell	Harrismith	N	4 row	Just finished
006	18/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	Not started
007	18/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	S	4 row	Not started
008	18/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	Not started
009	18/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	Not started
010	18/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N	4 row	Not started
011	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S - C	4 row	Just started
012	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	N	4 row	Just started
013	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S	4 row	Just started
014	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	N - C	4 row	Not started
015	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S - C	4 row	Just started
016	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	Not started
017	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W	4 row	Not started
018	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	E	4 row	Not started
019	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	Not started
020	19/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	Not started
021	19/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E - C	3 x 2 row	Finnished
022	19/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	W - C	3 x 2 row	Not started
023	19/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E - C	3 x 2 row	Not started
024	19/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E	3 x 2 row	Not started
025	19/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	W - C	3 x 2 row	Not started
026	21/10/2005	<i>E. polybractea</i>	McDougal	Harrismith	W	2 x 2 row	Finnished
027	21/10/2005	<i>E. polybractea</i>	McDougal	Harrismith	W - C	2 x 2 row	Not started
028	21/10/2005	<i>E. polybractea</i>	McDougal	Harrismith	E - C	2 x 2 row	Finnished
029	21/10/2005	<i>E. polybractea</i>	McDougal	Harrismith	E - C	2 x 2 row	Not started
030	21/10/2005	<i>E. polybractea</i>	McDougal	Harrismith	E	2 x 2 row	Finnished
031	20/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S	2 x 3 row	Finnished
032	20/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	N - C	2 x 3 row	Finnished
033	20/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S - C	2 x 3 row	Finnished
034	20/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S - C	2 x 3 row	Finnished
035	20/10/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	N	2 x 3 row	Finnished
036	20/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	Some
037	20/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	Some
038	20/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N	2 x 2 row	Some
039	20/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	Some
040	20/10/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	S	2 x 2 row	Some
041	20/10/2005	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S	2 x 2 row	Not started
042	20/10/2005	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	Finnished
043	20/10/2005	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	N	2 x 2 row	Finnished
044	20/10/2005	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	Finnished
045	20/10/2005	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	Finnished
046	31/10/2005	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N	4 row	Not started
047	31/10/2005	<i>E. polybractea</i>	Hassell / Hall	Pingelly	S	2 x 2 row	Finnished
048	31/10/2005	<i>E. polybractea</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	Finnished
049	31/10/2005	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	Finnished
050	31/10/2005	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	Finnished
051	1/11/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	N	2 x 2 row	Finnished
052	1/11/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	Finnished
053	1/11/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S	2 x 2 row	Not started
054	1/11/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	Not started
055	1/11/2005	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	Finnished
056	1/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S	2 x 2 row	Some
057	1/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	N	2 x 2 row	Some
058	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	Some
059	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	Some
060	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	Some

Flowering, sampling and site information: Mallees, Sampling 2 (Autumn)

Tree No.	Date sampled	Taxon	Site	Locality	Alley position	Alley formation	Flowering
001	17/5/06	<i>E. polybractea</i>	Parnell	Harrismith	N - C	4 row	buds
002	17/5/06	<i>E. polybractea</i>	Parnell	Harrismith	S - C	4 row	some
003	17/5/06	<i>E. polybractea</i>	Parnell	Harrismith	N - C	4 row	some
004	17/5/06	<i>E. polybractea</i>	Parnell	Harrismith	S	4 row	some
005	17/5/06	<i>E. polybractea</i>	Parnell	Harrismith	N	4 row	some
006	17/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	nil
007	17/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	S	4 row	nil
008	17/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	nil
009	17/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N - C	4 row	nil
010	17/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Parnell	Harrismith	N	4 row	nil
011	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S - C	4 row	just started
012	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	N	4 row	buds
013	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S	4 row	buds
014	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	N - C	4 row	few
015	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Parnell	Harrismith	S - C	4 row	nil
016	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	nil
017	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W	4 row	nil
018	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	E	4 row	nil
019	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	nil
020	16/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	McDougal	Harrismith	W - C	4 row	nil
021	16/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E - C	3 x 2 row	buds
022	16/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	W - C	3 x 2 row	buds
023	16/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E - C	3 x 2 row	nil
024	16/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	E	3 x 2 row	buds
025	16/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	McDougal	Harrismith	W - C	3 x 2 row	buds
026	16/5/06	<i>E. polybractea</i>	McDougal	Harrismith	W	2 x 2 row	buds
027	16/5/06	<i>E. polybractea</i>	McDougal	Harrismith	W - C	2 x 2 row	buds
028	16/5/06	<i>E. polybractea</i>	McDougal	Harrismith	E - C	2 x 2 row	buds
029	16/5/06	<i>E. polybractea</i>	McDougal	Harrismith	E - C	2 x 2 row	nil
030	16/5/06	<i>E. polybractea</i>	McDougal	Harrismith	E	2 x 2 row	buds
031	3/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S	2 x 3 row	nil
032	3/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	N - C	2 x 3 row	nil
033	3/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S - C	2 x 3 row	nil
034	3/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	S - C	2 x 3 row	nil
035	3/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Marshall /Lyon	Pingelly	N	2 x 3 row	nil
036	4/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	nil
037	4/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	nil
038	4/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N	2 x 2 row	nil
039	4/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	nil
040	4/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Marshall /Lyon	Pingelly	S	2 x 2 row	nil
041	4/5/06	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S	2 x 2 row	nil
042	4/5/06	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	some
043	4/5/06	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	N	2 x 2 row	60%
044	4/5/06	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	N - C	2 x 2 row	nil
045	4/5/06	<i>E. polybractea</i>	Marshall /Lyon	Pingelly	S - C	2 x 2 row	just started
046	17/5/06	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N	4 row	some
047	17/5/06	<i>E. polybractea</i>	Hassell / Hall	Pingelly	S	2 x 2 row	nil
048	17/5/06	<i>E. polybractea</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	nil
049	17/5/06	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	nil
050	17/5/06	<i>E. polybractea</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	finished
051	18/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	N	2 x 2 row	nil
052	18/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	nil
053	18/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S	2 x 2 row	nil
054	18/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	nil
055	18/5/06	<i>E. kochii</i> subspecies <i>borealis</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	nil
056	18/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S	2 x 2 row	nil
057	18/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	N	2 x 2 row	nil
058	18/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	nil
059	18/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	S - C	2 x 2 row	nil
060	18/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hassell / Hall	Pingelly	N - C	2 x 2 row	just started

Appendix F

Arthropod Mean and Frequency Data

Mean arthropod numbers collected from five species of *Eucalyptus* in the Wickepin and Pingelly shires during October 2005.

LOCATION		Site 1 P			Site 2 McD			Site 3 ML			Site 4 HH			Site 5 T			Site 6 S			Site 7 H		
TREE SPECIES		poly.	kochii	lox.	lox.	kochii	poly.	kochii	lox.	poly.	poly.	kochii	lox.	astrin.	lox.	wandoo	poly.	astrin.	wandoo	lox.	wandoo	astrin.
Arachnida	Araneae	35.4	34.0	40.4	9.6	21.4	9.4	18.2	10.4	30.8	35.0	27.8	11.2	7.4	5.8	15.8	22.6	5.4	16.4	10.4	11.6	7.6
	Acarina	30.0	10.0	8.8	1.0	5.8	4.0	34.0	14.4	54.2	4.0	4.3	17.0	3.8	1.5	13.8	35.2	4.4	21.0	10.8	12.8	6.5
	Pseudoscorpionida	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.0	0.0	2.0	0.0	2.3	1.0
Chilopoda		1.0	1.0	2.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Collembola		12.4	29.2	10608.2	4.6	6.0	6.0	16.8	1.5	2.0	2.7	2.7	1.0	35.8	23.6	4.4	3.2	1.0	5.0	2.3	7.0	2.8
Insecta	Thysanura	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	4.5	0.0	2.0	0.0	0.0	1.0	0.0
	Odonata	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Blattodea	4.6	18.0	3.5	0.0	2.5	2.5	2.0	1.0	1.0	1.3	2.0	0.0	0.0	0.0	1.5	2.0	2.7	9.8	0.0	1.7	1.0
	Isoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.5	0.0	0.0	0.0	0.0	0.0	0.0	1.0	17.0	84.0	1.0	3.7	0.0
	Mantodea	0.0	0.0	0.0	0.0	1.5	0.0	0.0	0.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Dermaptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	1.0	0.0
	Orthoptera	2.5	1.4	2.3	0.0	3.4	5.8	2.0	0.0	0.0	1.3	1.0	0.0	0.0	1.7	1.0	4.5	2.0	3.5	0.0	1.5	2.0
	Psocoptera	92.0	50.8	26.2	16.8	17.0	20.2	104.0	23.0	144.2	90.2	25.2	42.2	4.8	23.4	15.6	73.0	7.0	19.0	5.2	4.2	4.5
	Hemiptera	80.2	74.8	440.0	149.2	43.2	30.8	262.0	136.2	158.6	80.2	14.2	39.6	11.4	63.2	70.4	42.4	18.0	88.0	41.6	21.0	23.0
	Thysanoptera	9.2	23.0	29.4	12.2	42.8	4.3	53.6	47.6	46.4	18.4	13.4	52.2	36.2	62.6	30.6	25.8	4.4	11.8	39.5	6.6	4.2
	Neuroptera	2.3	3.0	3.6	4.0	0.0	0.0	2.3	3.0	5.8	3.2	3.5	2.7	1.0	12.0	0.0	8.3	0.0	0.0	1.3	3.0	3.0
	Coleoptera	44.0	147.4	44.2	33.8	96.6	30.4	69.0	63.0	247.6	124.4	21.8	29.8	104.0	59.4	26.2	100.4	30.4	41.6	78.0	28.2	75.0
	Mecoptera	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Diptera	52.4	154.4	167.6	19.4	54.6	18.2	49.0	29.8	169.0	88.4	30.0	31.4	20.4	71.4	57.6	105.8	34.4	39.2	161.8	48.0	40.4
	Lepidoptera	1.0	1.0	1.8	1.0	0.0	1.0	0.0	0.0	1.0	1.0	1.0	0.0	1.7	1.0	1.0	1.0	1.0	1.0	1.0	1.0	2.5
	Hymenoptera	131.8	215.2	117.0	68.2	184.4	47.6	101.6	40.2	72.4	98.4	92.8	83.8	39.2	37.0	82.8	117.8	27.2	114.4	79.0	52.0	44.4
	larvae	9.2	12.8	2.6	1.8	5.8	6.2	6.6	4.8	5.2	26.4	22.4	10.0	4.6	2.0	6.8	8.0	3.2	7.2	7.2	4.6	7.4

Frequency of arthropod occurrence per five trees collected from five species of *Eucalyptus* in the Wickepin and Pingelly shires during October 2005.

LOCATION		Site 1 P			Site 2 McD			Site 3 ML			Site 4 HH			Site 5 T			Site 6 S			Site 7 H		
TREE SPECIES		poly.	kochii	lox.	lox.	kochii	poly.	kochii	lox.	poly.	poly.	kochii	lox.	astrin.	lox.	wandoo	poly.	astrin.	wandoo	lox.	wandoo	astrin.
Arachnida	Araneae	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Acarina	4	4	4	2	4	2	5	5	5	5	4	1	4	2	5	5	5	5	4	5	4
	Pseudoscorpionida	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	2	0	3	2
Chilopoda		2	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Collembola		5	5	5	5	5	2	5	2	2	3	3	1	5	5	5	5	1	5	4	1	5
Insecta	Thysanura	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	1	0	0	2	0
	Odonata	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Blattodea	5	5	4	0	2	4	1	1	2	3	1	0	0	0	4	3	3	4	0	3	1
	Isoptera	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	1	1	3	2	3	0
	Mantodea	0	0	0	0	2	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
	Dermaptera	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0
	Orthoptera	2	5	3	0	5	4	1	0	0	3	1	0	0	3	3	2	2	2	0	2	2
	Psocoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	4	5	5	2
	Hemiptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Thysanoptera	5	5	5	5	5	4	5	5	5	5	5	5	5	5	5	4	5	5	4	5	5
	Neuroptera	3	2	5	1	0	0	3	2	5	5	4	3	1	2	0	4	0	0	3	3	2
	Coleoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Mecoptera	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Diptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Lepidoptera	3	2	4	1	0	1	0	0	3	3	2	0	3	1	2	1	1	1	1	1	2
	Hymenoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	larvae	5	5	5	5	5	5	5	5	5	5	5	5	5	3	4	5	5	5	5	5	5

Mean arthropod numbers collected from five species of *Eucalyptus* in the Wickepin and Pingelly shires during May 2006.

LOCATION		Site 1 P			Site 2 McD			Site 3 ML			Site 4 HH			Site 5 T		Site 6 S			Site 7 H			
TREE SPECIES		poly.	kochii	lox.	lox.	kochii	poly.	kochii	lox.	poly.	poly.	kochii	lox.	astrin.	lox.	wandoo	poly.	astrin.	wandoo	lox.	wandoo	astrin.
Arachnida	Araneae	38.2	52.8	93.4	20.4	30.2	25.0	71.6	34.6	30.8	45.0	23.4	10.0	13.6	19.0	25.0	47.4	11.4	32.2	15.8	35.0	13.2
	Acarina	6.5	1.8	8.8	5.8	9.2	5.4	1.0	0.0	1.3	1.7	3.8	2.0	2.3	2.0	5.0	3.5	2.0	5.5	2.3	3.2	3.0
	Pseudoscorpionida	0.0	0.0	0.0	0.0	0.0	2.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0	8.0	1.0	4.7	6.3	0.0	3.5	1.5
Collembola		2.5	6.6	27.8	331.0	526.6	243.0	4.3	114.0	3.0	33.0	217.3	148.6	2.0	1.8	3.8	0.0	2.0	9.0	3.4	2.8	2.0
Insecta	Archaeognatha	0.0	0.0	0.0	0.0	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Thysanura	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	1.0	6.0	0.0	0.0	2.0	0.0	0.0	0.0
	Odonata	0.0	1.0	0.0	2.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Blattodea	8.0	11.5	3.8	1.0	5.8	1.4	2.0	2.7	1.0	1.3	2.8	1.0	2.3	0.0	12.0	3.8	5.0	9.2	1.0	3.5	1.0
	Isoptera	0.0	0.0	2.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0	10.5	1.0	0.0	0.0
	Mantodea	0.0	3.5	0.0	0.0	1.3	0.0	0.0	0.0	0.0	0.0	2.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Dermaptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.0	1.0	0.0	1.0	3.3	0.0	1.3	1.0
	Orthoptera	0.0	1.0	2.4	0.0	3.0	1.8	0.0	1.3	1.0	8.5	0.0	0.0	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0	0.0
	Psocoptera	9.8	10.8	16.0	7.4	3.6	4.6	32.0	3.4	13.8	7.6	6.2	3.0	4.0	1.0	10.6	8.8	6.0	4.0	4.0	11.3	5.5
	Hemiptera	79.4	47.6	312.8	178.6	46.8	31.4	160.8	27.2	34.6	13.0	13.6	17.8	18.2	19.2	20.8	37.4	7.8	16.4	17.4	17.8	11.8
	Thysanoptera	107.6	4.3	42.2	24.3	10.4	14.2	5.0	1.8	1.3	13.3	1.0	2.0	3.0	2.0	2.0	73.4	5.5	45.3	3.3	2.5	1.8
	Neuroptera	0.0	0.0	2.0	0.0	1.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.0	2.0	1.0	2.0	0.0	0.0
	Coleoptera	56.6	43.6	60.8	85.0	15.2	36.0	54.8	46.0	29.2	74.0	11.4	16.4	15.4	15.8	25.4	135.2	25.8	66.4	23.2	43.0	24.6
	Diptera	27.0	25.0	96.6	14.2	86.0	58.8	29.0	19.0	19.8	37.8	118.0	8.2	9.4	8.6	9.2	98.4	21.0	31.8	11.0	95.0	38.6
	Lepidoptera	2.5	1.2	1.5	1.5	1.0	1.7	1.7	1.4	1.0	1.0	2.0	0.0	2.0	2.0	2.8	1.3	1.8	1.7	1.0	6.0	1.8
	Hymenoptera	69.4	37.0	338.0	171.0	37.0	48.4	413.6	56.8	31.6	165.0	85.6	20.2	84.0	42.2	59.2	97.6	45.0	56.2	101.2	44.6	40.8
	larvae	12.2	9.0	23.0	8.8	3.6	12.6	5.3	3.7	6.0	11.2	7.3	2.4	2.8	4.0	4.2	17.0	2.0	5.8	2.3	6.0	17.5

Frequency of arthropod occurrence per five trees collected from five species of *Eucalyptus* in the Wickepin and Pingelly shires during May 2006.

LOCATION		Site 1 P			Site 2 McD			Site 3 ML			Site 4 HH			Site 5 T		Site 6 S			Site 7 H			
TREE SPECIES		poly.	kochii	lox.	lox.	kochii	poly.	kochii	lox.	poly.	poly.	kochii	lox.	astrin.	lox.	wandoo	poly.	astrin.	wandoo	lox.	wandoo	astrin.
Arachnida	Araneae	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Acarina	4	4	5	4	5	5	1	0	3	3	4	1	4	4	2	4	3	4	3	5	4
	Pseudoscorpionida	0	0	0	0	0	2	0	0	1	0	0	0	0	0	1	2	3	3	0	4	2
Collembola		4	5	5	5	5	4	3	3	4	5	4	5	2	4	4	0	2	3	5	4	3
Insecta	Archaeognatha	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Thysanura	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	0	1	0	0	0
	Odonata	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Blattodea	3	4	4	1	4	5	4	3	2	3	4	1	3	0	3	4	2	5	1	2	1
	Isoptera	0	0	4	0	0	0	0	0	0	0	0	1	0	0	0	0	0	2	2	0	0
	Mantodea	0	4	0	0	3	0	0	0	0	0	1	1	0	0	0	0	0	0	0	0	0
	Dermaptera	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1	0	2	3	0	3	1
	Orthoptera	0	3	5	0	3	4	0	3	1	4	0	0	0	1	0	3	0	1	0	1	0
	Psocoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	2	5	5	3	3	4	4	4
	Hemiptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Thysanoptera	5	4	5	4	5	5	1	4	3	3	1	2	4	2	3	5	2	3	3	4	5
	Neuroptera	0	0	1	0	1	1	0	0	0	0	0	0	0	0	2	0	1	1	1	0	0
	Coleoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Diptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	Lepidoptera	4	5	2	2	4	3	3	5	2	2	1	0	4	2	5	3	5	3	2	5	4
	Hymenoptera	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5	5
	larvae	5	5	5	4	5	5	3	3	4	5	4	5	5	4	5	5	4	5	3	5	4

Appendix G

Sedentary Arthropod Data

Frequency of sedentary arthropod types collected by leaf clipping.

Tree	Galls	Leaf folding	Scale	Psyllid	Leaf miner	Webbing	Blistering
1	0	0	1	0	0	0	1
2	1	0	1	0	0	0	0
3	1	0	1	0	1	0	1
4	1	0	0	0	1	0	1
5	0	0	0	0	1	0	0
6	1	0	1	0	0	0	0
7	1	1	0	1	0	0	1
8	1	0	1	0	0	0	0
9	1	0	0	1	0	0	1
10	1	1	1	1	0	1	0
11	0	0	0	1	0	1	0
12	1	0	0	1	0	0	0
13	1	0	0	1	0	0	0
14	1	0	0	1	0	1	0
15	1	0	1	1	0	0	0
16	1	0	1	1	1	1	0
17	1	0	1	1	1	0	1
18	1	0	0	1	1	1	0
19	1	0	0	1	1	0	0
20	1	0	0	1	1	1	0
21	0	0	1	0	1	1	0
22	0	0	1	0	1	0	0
23	0	0	0	0	1	1	0
24	0	1	0	0	0	0	0
25	0	0	0	0	0	1	0
26	0	0	0	0	1	0	0
27	1	0	1	0	1	1	0
28	0	0	1	0	1	1	1
29	0	0	0	0	1	1	0
30	0	0	1	1	1	1	0
31	0	0	1	1	1	0	0
32	1	0	0	0	0	0	0
33	0	0	0	0	0	0	0
34	0	1	0	1	0	1	0
35	0	0	0	0	0	0	0
36	1	0	0	1	1	0	0
37	0	0	0	1	0	1	0
38	1	0	0	0	0	0	0
39	0	0	1	1	0	1	0
40	1	0	0	1	0	1	1
41	0	0	0	0	0	0	1
42	0	0	0	0	0	0	0
43	0	0	0	1	0	0	0
44	0	1	0	1	0	1	1
45	0	0	1	1	0	1	0
46	0	0	0	1	0	1	0
47	1	0	1	1	0	1	0
48	0	0	1	0	0	1	0
49	0	0	1	0	1	0	0
50	1	0	1	1	0	0	1
51	1	1	1	0	0	0	0
52	1	0	0	0	0	0	1
53	1	0	1	0	0	1	0
54	1	0	1	0	0	1	0
55	1	1	1	0	0	0	0
56	1	0	0	1	0	0	0
57	1	0	0	1	0	1	0
58	1	0	0	1	0	1	0
59	1	0	1	1	0	1	0
60	1	0	0	1	0	0	0
61	1	0	0	1	0	1	0
62	1	0	1	1	1	1	0
63	1	0	0	1	1	1	0
64	1	0	0	1	0	0	0
65	1	0	0	1	0	0	0
66	1	0	0	0	0	1	0
67	1	0	0	1	0	1	0
68	1	0	0	1	0	0	0
69	1	0	0	1	0	0	0
70	1	0	0	1	1	1	1
71	1	0	1	0	0	0	0
72	1	0	0	0	0	0	0
73	1	1	0	1	1	1	0
74	1	0	0	1	1	1	1
75	1	0	0	1	1	1	0
76	0	0	1	0	1	1	0
77	1	0	1	1	1	0	0
78	0	0	1	1	1	0	0
79	1	0	1	0	0	0	0
80	1	1	1	0	0	0	1
81	1	0	0	1	1	1	0
82	1	0	1	1	0	1	0
83	1	0	0	1	0	0	0
84	1	1	0	1	0	1	0
85	0	1	0	1	0	0	0
86	1	0	0	1	0	1	0
87	1	0	1	1	0	1	0
88	1	0	0	1	0	1	0
89	1	0	0	1	0	1	0
90	1	0	0	1	0	1	0
91	1	0	0	1	1	0	0
92	1	0	0	1	0	0	0
93	1	0	0	1	0	0	0
94	1	0	0	0	1	0	0
95	1	0	0	1	0	0	0
96	1	0	0	1	0	1	0
97	1	0	0	0	0	1	0
98	1	0	0	1	0	1	0
99	1	0	0	1	0	1	0
100	1	0	0	1	0	1	0
101	1	0	0	1	0	1	1
102	1	0	0	0	0	0	0
103	1	0	0	1	0	0	0
104	0	0	0	1	0	0	0
105	1	0	0	1	0	1	0

Appendix H

Remnant Site Tree Measurements

Tree measurements: Remnant, Sampling 1 (Spring)

Tree No.	Width N-S (m)	Width E-W (m)	Radius (m)	Canopy base ht (m)	Tree ht (m)	Canopy ht (m)	Canopy vol (m ³)
061	2.50	2.55	1.26	1.25	4.50	3.25	10.85
062	2.50	2.30	1.20	2.00	6.70	4.70	14.15
063	2.60	2.80	1.35	1.20	6.20	5.00	19.06
064	2.10	2.50	1.15	3.00	4.70	1.70	4.67
065	3.50	3.20	1.68	2.85	5.50	2.65	15.54
066	1.30	1.45	0.69	0.45	2.50	2.05	2.02
067	2.40	2.30	1.18	0.25	3.70	3.45	9.97
068	2.50	2.00	1.13	0.55	2.70	2.15	5.63
069	1.70	1.55	0.81	0.35	3.80	3.45	4.76
070	1.95	2.00	0.99	0.10	3.75	3.65	7.45
071	2.50	2.00	1.13	1.90	3.70	1.80	4.71
072	1.70	2.60	1.08	2.50	5.70	3.20	7.41
073	1.40	1.80	0.80	0.85	2.70	1.85	2.44
074	3.20	3.70	1.73	0.01	5.70	5.69	35.27
075	1.70	2.90	1.15	1.05	5.10	4.05	10.45
076	2.25	2.10	1.09	0.45	4.50	4.05	10.02
077	1.85	2.00	0.96	1.25	4.10	2.85	5.52
078	3.10	3.00	1.53	1.05	5.30	4.25	20.70
079	3.90	3.70	1.90	0.65	5.10	4.45	33.62
080	2.10	2.20	1.08	0.65	2.70	2.05	4.96
081	3.10	2.50	1.40	1.50	4.30	2.80	11.36
082	1.80	2.40	1.05	2.30	4.50	2.20	4.98
083	2.20	2.00	1.05	2.80	5.50	2.70	6.22
084	1.80	2.00	0.95	2.70	6.70	4.00	7.54
085	2.30	3.10	1.35	2.10	5.50	3.40	12.69
086	1.80	1.60	0.85	1.50	3.00	1.50	2.26
087	2.60	2.35	1.24	2.50	4.90	2.40	7.68
088	3.60	2.80	1.60	1.50	5.90	4.40	23.22
089	2.30	2.80	1.28	2.30	5.50	3.20	10.79
090	3.90	3.50	1.85	1.45	5.70	4.25	30.38
091	1.40	1.50	0.73	0.45	2.00	1.55	1.70
092	2.65	2.25	1.23	0.25	2.70	2.45	7.65
093	2.70	2.30	1.25	0.45	2.90	2.45	7.97
094	2.50	2.35	1.21	0.25	2.60	2.35	7.23
095	2.10	2.20	1.08	0.55	3.10	2.55	6.17
096	1.40	1.90	0.83	1.30	3.70	2.40	3.34
097	2.80	3.40	1.55	2.50	5.20	2.70	13.46
098	3.80	4.45	2.06	2.70	5.70	3.00	26.56
099	1.40	1.70	0.78	2.30	4.20	1.90	2.37
100	3.20	4.30	1.88	0.00	5.20	5.20	37.46
101	3.90	4.45	2.09	2.30	5.70	3.40	30.90
102	1.80	1.40	0.80	1.50	4.10	2.60	3.43
103	1.90	1.90	0.95	2.30	5.50	3.20	6.05
104	1.40	1.90	0.83	2.70	4.90	2.20	3.06
105	1.90	2.50	1.10	1.90	5.40	3.50	8.70

Tree measurements: Remnant, Sampling 2 (Autumn)

Tree No.	Width N-S (m)	Width E-W (m)	Radius (m)	Canopy base ht (m)	Tree ht (m)	Canopy ht (m)	Canopy vol (m ³)
061	2.30	2.90	1.30	1.50	4.80	3.30	11.52
062	2.65	2.00	1.16	1.90	6.00	4.10	11.38
063	2.05	2.10	1.04	1.00	6.20	5.20	11.72
064	2.65	2.90	1.39	2.20	5.90	3.70	14.89
065	2.10	2.10	1.05	3.00	5.00	2.00	4.62
066	1.20	1.35	0.64	1.00	3.20	2.20	1.87
067	2.70	2.45	1.29	0.25	4.50	4.25	14.72
068	2.20	2.25	1.11	1.00	3.70	2.70	7.00
069	1.70	1.70	0.85	1.00	4.50	3.50	5.30
070	1.90	2.30	1.05	0.40	4.00	3.60	8.24
071	2.33	1.75	1.02	1.75	3.70	1.95	4.16
072	2.00	2.60	1.15	2.30	4.80	2.50	6.81
073	1.40	1.60	0.75	1.00	3.00	2.00	2.35
074	3.20	3.25	1.61	0.00	5.30	5.30	28.86
075	1.70	2.10	0.95	0.90	4.90	4.00	7.48
076	2.30	2.30	1.15	0.30	4.70	4.40	12.19
077	1.90	2.10	1.00	1.40	4.50	3.10	6.48
078	4.30	3.20	1.88	0.70	5.60	4.90	35.30
079	3.80	2.80	1.65	0.50	5.50	5.00	27.86
080	1.70	3.30	1.25	0.50	3.20	2.70	7.93
081	3.15	2.10	1.31	1.20	3.90	2.70	9.35
082	1.40	2.10	0.88	2.40	4.60	2.20	3.39
083	1.70	2.40	1.03	3.50	6.00	2.50	5.34
084	1.70	1.90	0.90	2.20	6.20	4.00	6.76
085	2.70	3.20	1.48	1.50	6.20	4.70	21.26
086	2.10	1.70	0.95	0.90	3.20	2.30	4.30
087	2.40	2.10	1.13	1.10	4.90	3.80	10.03
088	2.60	2.30	1.23	1.40	5.90	4.50	14.09
089	3.10	3.20	1.58	1.50	5.10	3.60	18.70
090	4.00	3.30	1.83	0.50	5.20	4.70	32.48
091	1.40	1.50	0.73	0.60	2.60	2.00	2.20
092	2.20	2.40	1.15	0.30	3.30	3.00	8.29
093	2.20	3.00	1.30	0.50	3.70	3.20	11.06
094	1.90	2.00	0.98	0.20	3.40	3.20	6.37
095	2.00	2.10	1.03	0.20	3.60	3.40	7.48
096	1.65	1.90	0.89	0.50	3.80	3.30	5.42
097	2.65	2.70	1.34	2.30	5.20	2.90	10.86
098	2.80	3.09	1.47	2.90	4.90	2.00	9.06
099	1.40	1.45	0.71	2.20	3.90	1.70	1.81
100	3.90	4.00	1.98	0.00	4.90	4.90	40.02
101	3.30	3.60	1.73	1.30	5.20	3.90	24.26
102	2.00	1.65	0.91	1.10	4.50	3.40	5.87
103	1.65	2.05	0.93	2.10	5.80	3.70	6.55
104	1.20	1.45	0.66	2.50	4.75	2.25	2.05
105	1.75	2.50	1.06	1.70	5.90	4.20	9.62

Appendix I

Remnant Site Flowering and Site Information

Flowering, sampling and site information: Remnant, Sampling 1 (Spring)

Tree No.	Date sampled	Taxon	Site	Locality	Alley position	Alley formation	Flowering
061	2/11/2005	<i>E. astringens</i>	Tutanning	Pingelly	N - W	remnant	Not started
062	2/11/2005	<i>E. astringens</i>	Tutanning	Pingelly	N - E	remnant	Not started
063	1/11/2005	<i>E. astringens</i>	Tutanning	Pingelly	N - E	remnant	Not started
064	1/11/2005	<i>E. astringens</i>	Tutanning	Pingelly	S - W	remnant	Not started
065	1/11/2005	<i>E. astringens</i>	Tutanning	Pingelly	S - W	remnant	Just started
066	1/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	N - C	2 x 2 row	Not started
067	1/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	S - C	2 x 2 row	Some
068	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	S - C	2 x 2 row	Not started
069	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	N - C	2 x 2 row	Not started
070	2/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	NN - C	2 x 2 row	Some
071	2/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S	remnant	Not started
072	2/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S - W	remnant	Not started
073	2/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S - E	remnant	Not started
074	2/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	N	remnant	Not started
075	2/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	N	remnant	Not started
076	3/11/2005	<i>E. polybractea</i>	Spriggs	Narrogin	N	2 x 2 row	Not started
077	3/11/2005	<i>E. polybractea</i>	Spriggs	Narrogin	S - C	2 x 2 row	Not started
078	3/11/2005	<i>E. polybractea</i>	Spriggs	Narrogin	S	2 x 2 row	Not started
079	3/11/2005	<i>E. polybractea</i>	Spriggs	Narrogin	S - C	2 x 2 row	Finnished
080	3/11/2005	<i>E. polybractea</i>	Spriggs	Narrogin	N - C	2 x 2 row	Finnished
081	3/11/2005	<i>E. astringens</i>	Spriggs	Narrogin	N - E	remnant	Not started
082	3/11/2005	<i>E. astringens</i>	Spriggs	Narrogin	N	remnant	Not started
083	3/11/2005	<i>E. astringens</i>	Spriggs	Narrogin	N - W	remnant	Not started
084	3/11/2005	<i>E. astringens</i>	Spriggs	Narrogin	W	remnant	Not started
085	3/11/2005	<i>E. astringens</i>	Spriggs	Narrogin	S - E	remnant	Just started
086	3/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N - E	remnant	Not started
087	3/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N	remnant	Not started
088	3/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N - W	remnant	Not started
089	3/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	S - W	remnant	Not started
090	3/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	S - E	remnant	Not started
091	4/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	S	4 row	Not started
092	4/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	S - C	4 row	Some
093	4/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N - C	4 row	Some
094	4/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N - C	4 row	Full
095	4/11/2005	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N	4 row	Some
096	4/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	Not started
097	4/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	Not started
098	4/11/2005	<i>E. astringens</i>	Hesford	Wickepin	N - W	remnant	Not started
099	4/11/2005	<i>E. astringens</i>	Hesford	Wickepin	N - W	remnant	Full
100	4/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	Not started
101	4/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	Not started
102	4/11/2005	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	Not started
103	4/11/2005	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	Just started
104	4/11/2005	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	Some
105	4/11/2005	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	Some

Flowering, sampling and site information: Remnant, Sampling 2 (Autumn)

Tree No.	Date sampled	Taxon	Site	Locality	Alley position	Alley formation	Flowering
061	3/5/06	<i>E. astringens</i>	Hassell / Hall	Pingelly	N - W	remnant	nil
062	3/5/06	<i>E. astringens</i>	Tutanning	Pingelly	N - E	remnant	nil
063	3/5/06	<i>E. astringens</i>	Tutanning	Pingelly	N - E	remnant	nil
064	3/5/06	<i>E. astringens</i>	Tutanning	Pingelly	S - W	remnant	nil
065	3/5/06	<i>E. astringens</i>	Tutanning	Pingelly	S - W	remnant	nil
066	3/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	N - C	2 x 2 row	nil
067	3/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	S - C	2 x 2 row	nil
068	3/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	S - C	2 x 2 row	nil
069	3/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	N - C	2 x 2 row	nil
070	3/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Tutanning	Pingelly	NN - C	2 x 2 row	just started
071	3/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S	remnant	nil
072	3/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S - W	remnant	nil
073	3/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	S - E	remnant	nil
074	3/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	N	remnant	nil
075	3/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Tutanning	Pingelly	N	remnant	nil
076	4/5/06	<i>E. polybractea</i>	Tutanning	Narrogin	N	2 x 2 row	some
077	4/5/06	<i>E. polybractea</i>	Spriggs	Narrogin	S - C	2 x 2 row	nil
078	4/5/06	<i>E. polybractea</i>	Spriggs	Narrogin	S	2 x 2 row	some
079	4/5/06	<i>E. polybractea</i>	Spriggs	Narrogin	S - C	2 x 2 row	80%
080	4/5/06	<i>E. polybractea</i>	Spriggs	Narrogin	N - C	2 x 2 row	75%
081	5/5/06	<i>E. astringens</i>	Spriggs	Narrogin	N - E	remnant	nil
082	5/5/06	<i>E. astringens</i>	Spriggs	Narrogin	N	remnant	nil
083	5/5/06	<i>E. astringens</i>	Spriggs	Narrogin	N - W	remnant	nil
084	5/5/06	<i>E. astringens</i>	Spriggs	Narrogin	W	remnant	nil
085	5/5/06	<i>E. astringens</i>	Spriggs	Narrogin	S - E	remnant	nil
086	5/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N - E	remnant	nil
087	5/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N	remnant	nil
088	5/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	N - W	remnant	nil
089	5/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	S - W	remnant	nil
090	5/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Spriggs	Narrogin	S - E	remnant	nil
091	1/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Spriggs	Wickepin	S	4 row	nil
092	1/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	S - C	4 row	nil
093	1/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N - C	4 row	nil
094	1/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N - C	4 row	nil
095	1/5/06	<i>E. loxophleba</i> subspecies <i>lissophloia</i>	Hesford	Wickepin	N	4 row	finished
096	2/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	nil
097	2/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	nil
098	2/5/06	<i>E. astringens</i>	Hesford	Wickepin	N - W	remnant	nil
099	2/5/06	<i>E. astringens</i>	Hesford	Wickepin	N - W	remnant	nil
100	2/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	nil
101	2/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	nil
102	2/5/06	<i>E. wandoo</i> subspecies <i>wandoo</i>	Hesford	Wickepin	N - W	remnant	nil
103	2/5/06	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	nil
104	2/5/06	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	nil
105	2/5/06	<i>E. astringens</i>	Hesford	Wickepin	N - E	remnant	nil

Appendix J

Soil Nutrient Analysis Results

Soil nutrition test results

CSBP ANALYSIS REPORT
 CSBP LIMITED ABN: 81 008 668 371

UNITS		%			mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	%	mg/kg	dS/m	pH	pH
SAMPLE_ID	SERIAL_NO	Texture	Gravel	Colour	NO ₃ ⁻	NH ₄ ⁺	P	K	S	Organic C	Fe	Conductivity	pH_CaCl ₂	pH_H ₂ O
WAN 5	TUTANNING	1.5		GR	1	1	2	76	3.2	1.68	357	0.034	5.2	6.3
LOX 5	TUTANNING	1.5		BR	13	16	11	212	11.3	3.35	989	0.11	5.1	6
AST 5	TUTANNING	2		DKBR	1	37	10	196	186	4.51	1689	2.792	4.6	5
POLY 1	PARNELL	2		GR	1	3	46	91	7	1.09	791	0.085	4.6	5.7
LOX 1	PARNELL	2	5	GR	8	2	18	89	4.8	2.57	751	0.158	5.2	6.2
LOX 4	PARNELL	1.5		LTGR	1	8	35	40	12.9	1.1	503	0.216	4.7	5.4
LOX 7	PARNELL	2		BR	1	3	30	98	11.6	2.84	1725	0.143	5	5.8
WAN 7	HESFORD	2		BR	1	9	11	209	32.9	4.36	2653	0.474	4.4	4.9
AST 7	HESFORD	2.5	75-80	LTBR	1	1	2	132	19.7	0.89	588	0.146	4.9	5.8
HOR 4	HESFORD	1.5		GR	1	3	17	30	8.1	1.41	295	0.077	4.3	5.3
POLY 4	HASSEL	1.5		GR	1	4	18	88	8.3	1.56	568	0.068	4.8	5.8
HOR 1	HASSEL	2		GR	1	1	35	44	8	1.08	694	0.045	4.5	5.7
POLY 3	MARSHALL	1.5		GR	1	1	23	37	10	1.37	584	0.1	4.3	5.1
LOX 3	MARSHALL	1.5	5-10	GRBR	3	2	17	45	7.8	1.36	500	0.067	4.5	5.4
HOR 3	MARSHALL	1.5		GRBR	6	2	24	54	4.7	1.33	597	0.039	4.5	5.5
WAN 6	MARSHALL	1.5		GR	1	2	6	66	8.4	3.76	1212	0.144	5	6.1
AST 6	MARSHALL	1.5	15-20	GRBR	1	2	6	103	85.1	3.35	1456	0.967	5.1	5.4
POLY 6	MARSHALL	1.5		GR	1	12	10	74	19.3	1.92	1503	0.222	4.8	5.9
POLY 2	MCDUGAL	1.5		GR	1	2	29	114	15.2	2.31	582	0.155	4.6	5.8
LOX 2	MCDUGAL	2		GR	6	2	68	35	100	2.02	810	0.194	4.8	5.7
HOR 2	MCDUGAL	2		GR	7	1	38	51	21.5	2.64	634	0.164	5.1	6

Appendix K

Leaf Nutrient Analysis Results

Leaf nutrition test results

CSBP ANALYSIS REPORT
 CSBP LIMITED ABN: 81 008 668 371

UNITS		%	%	%	%	%	%	%	%	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	ug/kg	ug/kg	ug/kg
SAMPLE_ID	SERIAL_NUMBER	N	P	K	S	Na	Ca	Mg	Cl ⁻	Cu	Zn	Mn	Fe	NO ₃ ⁻	B	Mo	Co	Se
LOX 1	PARNELL	0.988	0.062	0.441	0.124	0.562	0.414	0.134	0.151	1.89	14.65	11.1	120.9	54	45.8	0	0	0
HOR 1	PARNELL	0.908	0.059	0.36	0.11	0.477	0.49	0.122	0.35	2.35	8.22	89.9	111.9	73	47.2	0	0	0
POLY 1	PARNELL	0.882	0.057	0.399	0.111	0.469	0.802	0.134	0.322	3.43	7.83	246.1	104.6	50	175	0	0	0
POLY 2	MCDUGALL	1.01	0.079	0.593	0.127	0.252	0.805	0.208	0.369	0.87	7.41	67	102.7	75	191	0	0	0
HOR 2	MCDUGALL	1.03	0.098	0.3	0.132	0.416	0.555	0.213	0.298	0.74	10.38	19.4	77.7	97	66.2	0	0	0
LOX 2	MCDUGALL	1.18	0.083	0.416	0.126	0.561	0.688	0.122	0.206	1.54	12.34	38.8	80.5	65	73.7	0	0	0
LOX 3	MARSHALL	0.926	0.105	0.622	0.109	0.583	0.838	0.175	0.385	4.3	22.21	283.7	103	71	23.4	0	0	0
HOR 3	MARSHALL	0.795	0.146	0.412	0.111	0.343	0.942	0.199	0.263	3.07	23.14	364.3	147.8	58	50.3	0	0	0
POLY 3	MARSHALL	0.843	0.123	0.544	0.108	0.216	1.371	0.337	0.666	1.81	13.34	693.8	118.3	71	85.2	0	0	0
POLY 4	HASELL	0.782	0.096	0.473	0.106	0.426	2.032	0.135	0.338	2	16.14	806.9	99.3	56	136	0	0	0
HOR 4	HASELL	0.684	0.128	0.5	0.092	0.394	0.717	0.152	0.341	1.95	11.27	310	70.9	58	47.4	0	0	0
LOX 4	HASELL	0.902	0.154	0.483	0.114	0.49	1.155	0.161	0.228	3.26	21.49	418.8	110.9	57	61.9	0	0	0
WAN 5	TUTANNING	0.84	0.044	0.409	0.109	0.499	0.463	0.2	0.722	2.43	7.12	35.5	115.5	55	113	0	0	0
LOX 5	TUTANNING	0.742	0.058	0.438	0.113	0.529	0.393	0.186	0.308	-0.5	9.67	15	83.4	53	48.9	0	0	0
AST 5	TUTANNING	0.881	0.046	0.486	0.113	0.639	0.293	0.125	0.686	-0.5	8.65	61.4	140.1	59	58.8	0	0	0
POLY 6	SPRIGG	0.776	0.068	0.524	0.107	0.382	0.967	0.151	0.424	3.17	7.14	219	79.3	60	139	0	0	0
WAN 6	SPRIGG	0.671	0.042	0.339	0.101	0.47	0.5	0.211	0.588	3.42	8.99	121.4	70.8	56	118	0	0	0
AST 6	SPRIGG	0.667	0.046	0.457	0.094	0.547	0.457	0.196	0.631	2.87	12.2	284.3	78.5	65	37.9	0	0	0
LOX 7	HESFORD	0.823	0.069	0.457	0.121	0.49	0.755	0.163	0.318	11.55	13.33	322.9	187.7	60	66	0	0	0
WAN 7	HESFORD	0.781	0.05	0.352	0.117	0.532	0.352	0.194	0.583	5.69	7.58	134.2	266.2	60	166	0	0	0
AST 7	HESFORD	0.753	0.051	0.441	0.1	0.581	0.343	0.18	0.712	4.68	11.99	160.9	165.5	55	65.6	0	0	0

Appendix L

Gas Chromatographic Conditions and Results

Gas Chromatographic (GC) Conditions

Column Phase: Alltech AT-35 (35% Phenyl, 65% Methylpolysiloxane)

Column dimensions: 60m x 0.25mm x 0.25 um

Temperature: 105°C for 5.5 minutes, 28°/min to 250° for 5 minutes

Carrier gas: Hydrogen @ 45.5 cm/sec

Detector: FID

Contact: Dr Peter Grayling
Natural Resources Branch
Department of Environment and Conservation
Locked Bag 104, Bentley Delivery Centre
WA, 6983, Australia
Phone: 61 8 9334 0139
Fax: 61 8 9334 0367

Results of Gas Chromatogram analysis for essential oils

Bottle No.	Leaf α-pinene	Leaf 1,8-cineole	Cineole (%)	Analysis Date	GC	Date Collected	Days Extracted	Taxon	Site
1	0.26%	3.51%	3.5	6-Dec-05	A	18-Oct-05	49	<i>E. polybractea</i>	Parnells
2	0.03%	2.92%	2.9	6-Dec-05	A	18-Oct-05	49	<i>E. polybractea</i>	Parnells
3	0.04%	3.70%	3.7	6-Dec-05	A	18-Oct-05	49	<i>E. polybractea</i>	Parnells
4	0.16%	2.78%	2.8	6-Dec-05	A	18-Oct-05	49	<i>E. polybractea</i>	Parnells
5	0.03%	3.34%	3.3	6-Dec-05	A	18-Oct-05	49	<i>E. polybractea</i>	Parnells
6	0.01%	3.30%	3.3	6-Dec-05	A	18-Oct-05	49	<i>E. kochii</i>	Parnells
7	0.03%	5.24%	5.2	6-Dec-05	A	18-Oct-05	49	<i>E. kochii</i>	Parnells
8	0.03%	3.74%	3.7	6-Dec-05	A	18-Oct-05	49	<i>E. kochii</i>	Parnells
9	0.01%	3.01%	3.0	6-Dec-05	A	18-Oct-05	49	<i>E. kochii</i>	Parnells
10	0.04%	3.83%	3.8	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	Parnells
11	0.68%	2.63%	2.6	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	Parnells
12	0.99%	3.15%	3.1	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	Parnells
13	0.72%	3.46%	3.5	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	Parnells
14	0.63%	3.44%	3.4	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	Parnells
15	0.99%	2.99%	3.0	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	Parnells
16	0.63%	2.56%	2.6	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	McDougalls
17	0.57%	2.48%	2.5	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	McDougalls
18	0.70%	2.72%	2.7	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	McDougalls
19	0.50%	2.57%	2.6	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	McDougalls
20	0.63%	3.27%	3.3	6-Dec-05	A	19-Oct-05	48	<i>E. loxophleba</i>	McDougalls
21	0.20%	3.25%	3.3	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	McDougalls
22	0.11%	3.15%	3.1	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	McDougalls
23	0.15%	3.11%	3.1	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	McDougalls
24	0.21%	3.58%	3.6	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	McDougalls
25	0.26%	3.74%	3.7	6-Dec-05	A	19-Oct-05	48	<i>E. kochii</i>	McDougalls
26	0.04%	3.00%	3.0	6-Dec-05	A	21-Oct-05	46	<i>E. polybractea</i>	McDougalls
27	0.07%	0.91%	0.9	6-Dec-05	A	21-Oct-05	46	<i>E. polybractea</i>	McDougalls
28	0.09%	1.15%	1.2	6-Dec-05	A	21-Oct-05	46	<i>E. polybractea</i>	McDougalls
29	0.08%	2.03%	2.0	6-Dec-05	A	21-Oct-05	46	<i>E. polybractea</i>	McDougalls
30	0.03%	2.97%	3.0	6-Dec-05	A	21-Oct-05	46	<i>E. polybractea</i>	McDougalls
31	0.00%	4.74%	4.7	6-Dec-05	A	20-Oct-05	47	<i>E. kochii</i>	Marshalls
32	0.00%	2.91%	2.9	6-Dec-05	A	20-Oct-05	47	<i>E. kochii</i>	Marshalls
33	0.02%	3.48%	3.5	6-Dec-05	A	20-Oct-05	47	<i>E. kochii</i>	Marshalls
34	0.01%	2.30%	2.3	6-Dec-05	A	20-Oct-05	47	<i>E. kochii</i>	Marshalls
35	0.02%	2.92%	2.9	6-Dec-05	A	20-Oct-05	47	<i>E. kochii</i>	Marshalls
36	0.77%	2.36%	2.4	6-Dec-05	A	20-Oct-05	47	<i>E. loxophleba</i>	Marshalls
37	0.64%	2.65%	2.7	6-Dec-05	A	20-Oct-05	47	<i>E. loxophleba</i>	Marshalls
38	0.60%	0.05%	0.0	6-Dec-05	A	20-Oct-05	47	<i>E. loxophleba</i>	Marshalls
39	0.45%	2.21%	2.2	6-Dec-05	A	20-Oct-05	47	<i>E. loxophleba</i>	Marshalls
40	0.59%	2.55%	2.6	6-Dec-05	A	20-Oct-05	47	<i>E. loxophleba</i>	Marshalls
41	0.11%	1.65%	1.6	6-Dec-05	A	20-Oct-05	47	<i>E. polybractea</i>	Marshalls
42	0.00%	2.55%	2.5	6-Dec-05	A	20-Oct-05	47	<i>E. polybractea</i>	Marshalls
43	0.00%	3.56%	3.6	6-Dec-05	A	20-Oct-05	47	<i>E. polybractea</i>	Marshalls
44	0.00%	1.45%	1.4	6-Dec-05	A	20-Oct-05	47	<i>E. polybractea</i>	Marshalls
45	0.00%	0.95%	0.9	6-Dec-05	A	20-Oct-05	47	<i>E. polybractea</i>	Marshalls
46	0.00%	1.49%	1.5	6-Dec-05	A	31-Oct-05	36	<i>E. polybractea</i>	Halls
47	0.00%	2.37%	2.4	6-Dec-05	A	31-Oct-05	36	<i>E. polybractea</i>	Halls
48	0.00%	2.93%	2.9	6-Dec-05	A	31-Oct-05	36	<i>E. polybractea</i>	Halls
49	0.00%	4.30%	4.3	6-Dec-05	A	31-Oct-05	36	<i>E. polybractea</i>	Halls
50	0.04%	1.66%	1.7	6-Dec-05	A	31-Oct-05	36	<i>E. polybractea</i>	Halls
51	0.00%	3.17%	3.2	6-Dec-05	A	01-Nov-05	35	<i>E. kochii</i>	Halls
52	-0.01%	2.73%	2.7	6-Dec-05	A	01-Nov-05	35	<i>E. kochii</i>	Halls
53	0.03%	2.20%	2.2	6-Dec-05	A	01-Nov-05	35	<i>E. kochii</i>	Halls
54	0.00%	3.07%	3.1	6-Dec-05	A	01-Nov-05	35	<i>E. kochii</i>	Halls
55	-0.01%	2.24%	2.2	6-Dec-05	A	01-Nov-05	35	<i>E. kochii</i>	Halls
56	0.53%	2.90%	2.9	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Halls
57	0.40%	1.59%	1.6	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Halls
58	0.23%	2.13%	2.1	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Halls
59	0.47%	3.04%	3.1	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Halls
60	0.55%	2.66%	2.7	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Halls
61	0.63%	0.89%	0.9	6-Dec-05	A	02-Nov-05	34	<i>E. astringens</i>	Tutanning
62	0.96%	1.17%	1.2	6-Dec-05	A	02-Nov-05	34	<i>E. astringens</i>	Tutanning
63	0.89%	1.00%	1.0	6-Dec-05	A	02-Nov-05	34	<i>E. astringens</i>	Tutanning
64	0.22%	1.52%	1.5	6-Dec-05	A	02-Nov-05	34	<i>E. astringens</i>	Tutanning
65	0.38%	0.99%	1.0	6-Dec-05	A	02-Nov-05	34	<i>E. astringens</i>	Tutanning
66	0.53%	2.42%	2.4	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Tutanning
67	0.40%	2.33%	2.3	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Tutanning
68	0.62%	3.28%	3.3	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Tutanning
69	0.32%	1.49%	1.5	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Tutanning
70	0.75%	3.04%	3.0	6-Dec-05	A	01-Nov-05	35	<i>E. loxophleba</i>	Tutanning
71	0.62%	1.99%	2.0	6-Dec-05	A	02-Nov-05	34	<i>E. wandoo</i>	Tutanning
72	0.62%	1.28%	1.3	6-Dec-05	A	02-Nov-05	34	<i>E. wandoo</i>	Tutanning
73	0.49%	1.53%	1.5	6-Dec-05	A	02-Nov-05	34	<i>E. wandoo</i>	Tutanning
74	0.56%	1.76%	1.8	8-Dec-05	A	2-Nov-05	36	<i>E. wandoo</i>	Tutanning
75	0.58%	1.61%	1.6	8-Dec-05	A	2-Nov-05	36	<i>E. wandoo</i>	Tutanning
76	0.01%	3.89%	3.9	8-Dec-05	A	3-Nov-05	35	<i>E. polybractea</i>	Spriggs
77	0.03%	4.15%	4.1	8-Dec-05	A	3-Nov-05	35	<i>E. polybractea</i>	Spriggs
78	0.08%	0.94%	0.9	8-Dec-05	A	3-Nov-05	35	<i>E. polybractea</i>	Spriggs
79	0.05%	4.03%	4.0	8-Dec-05	A	3-Nov-05	35	<i>E. polybractea</i>	Spriggs
80	0.03%	2.85%	2.9	8-Dec-05	A	3-Nov-05	35	<i>E. polybractea</i>	Spriggs
81	0.86%	1.01%	1.0	8-Dec-05	A	3-Nov-05	35	<i>E. astringens</i>	Spriggs
82	1.03%	1.37%	1.4	8-Dec-05	A	3-Nov-05	35	<i>E. astringens</i>	Spriggs
83	0.63%	1.08%	1.1	8-Dec-05	A	3-Nov-05	35	<i>E. astringens</i>	Spriggs
84	0.91%	1.42%	1.4	8-Dec-05	A	3-Nov-05	35	<i>E. astringens</i>	Spriggs
85	1.07%	1.35%	1.3	8-Dec-05	A	3-Nov-05	35	<i>E. astringens</i>	Spriggs
86	0.45%	1.41%	1.4	8-Dec-05	A	3-Nov-05	35	<i>E. wandoo</i>	Spriggs
87	0.60%	1.63%	1.6	8-Dec-05	A	3-Nov-05	35	<i>E. wandoo</i>	Spriggs
88	0.34%	1.03%	1.0	8-Dec-05	A	3-Nov-05	35	<i>E. wandoo</i>	Spriggs
89	0.76%	1.58%	1.6	8-Dec-05	A	3-Nov-05	35	<i>E. wandoo</i>	Spriggs
90	0.53%	1.21%	1.2	8-Dec-05	A	3-Nov-05	35	<i>E. wandoo</i>	Spriggs
91	0.58%	2.13%	2.1	8-Dec-05	A	4-Nov-05	34	<i>E. loxophleba</i>	Hesfords
92	0.21%	1.16%	1.2	8-Dec-05	A	4-Nov-05	34	<i>E. loxophleba</i>	Hesfords
93	0.19%	1.73%	1.7	8-Dec-05	A	4-Nov-05	34	<i>E. loxophleba</i>	Hesfords
94	0.53%	2.89%	2.9	8-Dec-05	A	4-Nov-05	34	<i>E. loxophleba</i>	Hesfords
95	0.35%	2.09%	2.1	8-Dec-05	A	4-Nov-05	34	<i>E. loxophleba</i>	Hesfords
96	0.65%	1.64%	1.6	8-Dec-05	A	4-Nov-05	34	<i>E. wandoo</i>	Hesfords
97	0.42%	1.34%	1.3	8-Dec-05	A	4-Nov-05	34	<i>E. wandoo</i>	Hesfords
98	0.95%	1.23%	1.2	8-Dec-05	A	4-Nov-05	34	<i>E. astringens</i>	Hesfords
99	0.49%	0.64%	0.6	8-Dec-05	A	4-Nov-05	34	<i>E. astringens</i>	Hesfords
100	0.43%	1.21%	1.2	8-Dec-05	A	4-Nov-05	34	<i>E. wandoo</i>	Hesfords
101	0.18%	0.73%	0.7	8-Dec-05	A	4-Nov-05	34	<i>E. wandoo</i>	Hesfords
102	0.47%	1.78%	1.8	8-Dec-05	A	4-Nov-05	34	<i>E. wandoo</i>	Hesfords
103	0.32%	0.49%	0.5	8-Dec-05	A	4-Nov-05	34	<i>E. astringens</i>	Hesfords
104	0.65%	0.96%	1.0	8-Dec-05	A	4-Nov-05	34	<i>E. astringens</i>	Hesfords
105	0.58%	0.78%	0.8	8-Dec-05	A	4-Nov-05	34	<i>E. astringens</i>	Hesfords

Individual Gas Chromatograms (CD)

Appendix M

Sideroxylonal Content Results

Results of phenol (sideroxyflonal) content analysis

Sample	Total Sideroxyflonals (A, B & C) mg/g dry matter
E. polybractea 1	0.00
E. polybractea 2	0.00
E. polybractea 3	0.00
E. polybractea 4	0.00
E. polybractea 5	0.00
E. kochii 6	0.00
E. kochii 7	0.00
E. kochii 8	0.00
E. kochii 9	0.00
E. kochii 10	0.00
E. loxopleba 11	54.03
E. loxopleba 12	74.54
E. loxopleba 13	70.45
E. loxopleba 14	63.47
E. loxopleba 15	50.65
E. loxopleba 16	49.21
E. loxopleba 17	73.73
E. loxopleba 18	56.47
E. loxopleba 19	39.24
E. loxopleba 20	76.65
E. kochii 21	0.00
E. kochii 22	0.00
E. kochii 23	0.00
E. kochii 24	0.00
E. kochii 25	0.00
E. polybractea 26	0.00
E. polybractea 27	0.00
E. polybractea 28	0.00
E. polybractea 29	0.00
E. polybractea 30	0.00
E. kochii 31	0.00
E. kochii 32	0.00
E. kochii 33	0.00
E. kochii 34	0.00
E. kochii 35	0.00
E. loxopleba 36	43.13
E. loxopleba 37	46.95
E. loxopleba 38	0.70
E. loxopleba 39	25.05
E. loxopleba 40	59.05
E. polybractea 41	0.00
E. polybractea 42	0.00
E. polybractea 43	0.00
E. polybractea 44	0.00
E. polybractea 45	0.00
E. polybractea 46	0.00
E. polybractea 47	0.00
E. polybractea 48	0.00
E. polybractea 49	0.00
E. polybractea 50	0.00
E. kochii 51	0.00
E. kochii 52	0.00
E. kochii 53	0.00
E. kochii 54	0.00
E. kochii 55	0.00
E. loxopleba 56	83.24
E. loxopleba 57	37.65
E. loxopleba 58	42.44
E. loxopleba 59	52.93
E. loxopleba 60	51.13
E.astringens 61	0.97
E.astringens 62	2.24
E.astringens 63	1.25
E.astringens 64	6.03
E.astringens 65	1.48
E. loxopleba 66	42.04
E. loxopleba 67	67.63
E. loxopleba 68	60.33
E. loxopleba 69	25.92
E. loxopleba 70	68.89
E. wandoo 71	11.34
E. wandoo 72	8.55
E. wandoo 73	9.05
E. wandoo 74	13.53
E. wandoo 75	23.48
E. polybractea 76	0.00
E. polybractea 77	0.00
E. polybractea 78	0.00
E. polybractea 79	0.00
E. polybractea 80	0.00
E.astringens 81	0.00
E.astringens 82	2.56
E.astringens 83	0.94
E.astringens 84	2.55
E.astringens 85	1.44
E. wandoo 86	7.74
E. wandoo 87	12.62
E. wandoo 88	7.44
E. wandoo 89	9.64
E. wandoo 90	5.79
E. loxopleba 91	62.95
E. loxopleba 92	33.08
E. loxopleba 93	50.25
E. loxopleba 94	62.56
E. loxopleba 95	46.98
E. wandoo 96	7.67
E. wandoo 97	5.33
E.astringens 98	3.28
E.astringens 99	0.93
E. wandoo 100	13.43
E. wandoo 101	7.75
E. wandoo 102	6.01
E.astringens 103	1.25
E.astringens 104	1.69
E.astringens 105	1.69

Appendix N

Coleopteran (Beetle) Raw Data

