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Stimulation of Western Australian Sandalwood (Santalum spicatum) oil production using multiple treatments [thesis]

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Stimulation of Western Australian Sandalwood (*Santalum spicatum*) oil production using multiple treatments

This thesis is presented in fulfilment of the degree of

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Abstract

Sandalwood is an important international commodity, recognised for its aromatic oil which is a key ingredient in many fragrances and cosmetics. Western Australian (WA) sandalwood (*Santalum spicatum*) is known to be a cheaper alternative for the superior Indian sandalwood (*Santalum album*) as it has a lower oil content and lower quality oil. The natural stocks of *S. album* have declined due to illegal poaching, mismanagement, and disease. WA sandalwood's natural stands have also reduced due to historical mismanagement. As a result, WA sandalwood (*S. spicatum*) has been established in plantations in the southern half of WA to attempt to meet the demands of the sandalwood industry. Plantation WA sandalwood is promoted to farmers as agroforestry, with the promise of economic and environmental benefits. While these benefits are attractive, sandalwood has an estimated 25 year rotation.

This research aimed to determine the effect of physical and chemical treatments on oil production and heartwood formation in WA sandalwood, with the aim being to increase oil production, thus allowing the time between establishment and harvesting to be reduced. This study was conducted over three plantations in the Wheatbelt region of Western Australia; 'Sandawindy', 'Kylie Reserve', and 'Brookton'. At each site, four treatments were applied: a dowel soaked with the plant hormone Methyl Salicylate (MeSA) and inserted into the tree (Treated Dowel treatment), a dowel with no MeSA inserted into the tree (Blank Dowel treatment), a drill hole left empty (Empty Drill treatment), and a section of bark removed from the tree (Bark Removed treatment), as well as a group of trees left as a control for comparison. The Blank Dowel and Empty Drill treatments were established to determine if any significant increases of sandalwood oil in the Treated Dowel treatment were a result of the MeSA, the foreign dowel, or drilling into the tree. The Bark Removed treatment was used to mimic dry-sidedness; a condition that occurs naturally in the Rangelands of WA as a result of sun scald.

The sandalwood trees were measured and treated in November of 2016. Plantations were divided into 30 evenly sized blocks per site, with 6 replicate blocks allocated to each treatment and control group. Two replicate blocks for every treatment and control group at each plantation were harvested in November of 2017, and all trees were remeasured. Of the approximate 300 trees harvested, 150 were cored using a 12 mm auger drill. These core samples were analysed for oil yield and composition at Wescorp's laboratory. The total oil was measured an analysed, as well as the oil constituents α-santalol, β-santalol, farnesol, nuciferol, and β-bisabalol oil compositions (percentage) and yield (%w/w). All trees that were harvested

were cut into 8 discs measuring 25 mm each, and the percentage of heartwood area at each height was measured and recorded. All data was statistically analysed using a univariate general linear model.

There was no treatment that consistently increased total oil or oil component yields, qualities, or heartwood area percentages. The Empty Drill treatment resulted in more oil production than the control group on the most occasions, however it did not consistently increase oil production. This showed that the presence of MeSA did not have a significant effect on oil production, and the physical wounding of the tree had the overall greatest effect.

The Kylie Reserve plantation showed low oil yield and low heartwood area percentages compared to the Sandawindy and Brookton plantations, although also showed the highest oil yields. This research, while not showing significant increases in oil production for the different treatments used, has giving a promising indication that a longer time between treatment and harvesting could influence the oil production. Further research extending this study should be conducted to give more information on the effect of the treatments on oil production.

Declaration

I certify that this thesis does not, to the best of my knowledge and belief:

i) incorporate without acknowledgement any material previously submitted for a degree or diploma in any institution of higher education;

ii) contain any material previously published or written by another person except where due reference is made in the text of the thesis; or

iii) contain any defamatory material.

Peta-Anne Smith

28th May 2019

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1. Introduction

1.1 Global Perspective on Sandalwood

Sandalwood (*Santalum* spp.) is a genus of hemi-parasitic trees whose fragrant heartwood, which is largely used in perfumes, pharmaceuticals, incense, and ornamental carvings, has made it the target for international exploitation (Kumar, Joshi, & Ram, 2012; Moniodis *et al.*, 2017). Sandalwood has important value for some religions, including Hinduism and Buddhism, where low grade sandalwood logs are powdered and made into incense (or joss) sticks that are used in religious ceremonies (Loneragan, 1990; Tonts & Selwood, 2003). Higher grade sandalwood heartwood is used for carvings, including statues, boxes, and beads. Sandalwood's most valuable asset, however, is its aromatic oil (Kumar *et al.*, 2012). The oil is obtained from the heartwood of the tree and occurs in different sections of the tree (Brand, Sawyer, & Evans, 2014), however it is most concentrated in the root and butt (150 mm above ground level to the below-ground root crown) of the sandalwood tree (Brand & Pronk, 2011). Sandalwood oil is used in perfumes, soaps, and cosmetics.

The most valuable sandalwood species is *Santalum album* (known commonly as Indian Sandalwood or East Indian Sandalwood). This species occurs naturally in India and some islands of Indonesia, and has been planted throughout South East Asia, China, and the Pacific (Clark, 2006; Doran & Turnbull, 1997; Kumar *et al.*, 2012). It is also grown in plantations in the tropical north of Australia (Radomiljac, 1998). The abundance of *S. album* has declined significantly from within its natural stands. This is due to unsustainable harvesting practices, illegal poaching, disease (including spike disease), which have increased in line with the high growth in in human populations throughout Asia (Kumar *et al.*, 2012). With the decline of *S. album*, and the continued demand for sandalwood products, other sandalwood species, such as WA sandalwood (*Santalum spicatum*), are being used as a substitute.

1.2 Sandalwood species in Australia

Apart from *S. spicatum*, there are five other *Santalum* species in Australia. These are *S. acuminatum* (Quandong or Candle Nut), *S. lanceolatum* (Plumbush or Northern Sandalwood), *S. murrayanum* (Bitter Quandong), *S. obtusifolium*, and *S. album* (Harbaugh, 2007; Loneragan, 1990). The first four mentioned are native to WA as well as other places of Australia, whereas *S. obtusifolium* is only found on the east coast of Australia (Atlas of Living Australia, n.d.). *S. acuminatum* and *S. murrayanum* do not produce aromatic fragrance (Loneragan, 1990). Only

S. lanceolatum and *S. album* produce fragrant oil, and are the only native sandalwood trees other than *S. spicatum* that is harvested for its fragrant timber (Harbaugh, 2007; McKinnell & Levinson, 2008). *S. acuminatum* is harvested for its edible fruit (quandong), which is used in the indigenous foods market (Harbaugh, 2007; Loneragan, 1990; Loveys, Tyerman, & Loveys, 2002). *S. album* occurs along the coast and on adjacent islands between Melville Island and Elcho Island in the Northern Territory (Linnaeus, 2019). is grown in the northern tropical regions commercially (Hettiarachchi *et al.*, 2012; Radomiljac, 1998).

1.3 *Santalum spicatum*

Western Australian Sandalwood, *Santalum spicatum* (hereafter referred to as sandalwood), is a root hemi-parasitic tree, native to Western Australia (Hewson & George, 1984). Sandalwood is naturally distributed across the southern half of Western Australia, across the inland regions of the state and as far north as Exmouth (Hewson & George, 1984; Kealley, 1991; Spooner, 1999). When the value of sandalwood was realised and first exported in 1845, sandalwood harvesters (known as pullers), were initially restricted in the areas that they could practically harvest from, reflecting the need to cart the timber over long distances to the ports at Bunbury, Fremantle, Geraldton, and Albany (Loneragan, 1990). As the railways expanded inland towards Southern Cross and Kalgoorlie, pullers were able to access and therefore harvest greater areas of sandalwood (Loneragan, 1990; Talbot, 1983). Historical overharvesting, land clearing for agriculture, a historical mismanagement of sandalwood, and the decline of the seed-dispersing woylie (*Bettongia penicillata*) has led to the decline in sandalwood's natural range and abundance (Forest Products Commission, 2015; Murphy *et al.*, 2015). Today, sandalwood can be found from Latitude 24 (Shark Bay area), through the inland regions of WA, and as far south as Latitude 35 (Loneragan, 1990). Establishing sandalwood in plantations, as well as through agroforestry on farmland has allowed for it to be widespread across WA, extending past its original range.

Exports of WA sandalwood timber first began in 1845 with a shipment of four tonnes from Fremantle to Ceylon (present-day Sri Lanka) (Loneragan, 1990). Today, the sandalwood industry is growing and evolving, with an increase in demand for timber, as well as a new demand for WA sandalwood oil (McKinnell & Levinson, 2008). WA sandalwood is in demand internationally for its fragrant oil and for the manufacture of incense and joss sticks in religious ceremonies. Major export countries are Hong Kong and China, but there are new emerging markets in Malaysia, Singapore, India and Thailand (Forest Products Commission, 2016; Hettiarachchi *et al.*, 2012).

WA sandalwood is sorted into five groups of differing values when harvested, depending on the oil composition in the wood. Sandalwood roots and butts will usually contain the highest oil composition. This is the followed by the stem, which is separated into 1st, 2nd, and 3rd grade wood, depending on the given diameter (Brand & Pronk, 2011).

Active regeneration, i.e. seed sowing, is required due to the significant reduction in natural regeneration as a result of increased grazing and drought, as well as the decline of the seed dispersing woylie (Forest Products Commission, 2015; Murphy *et al.*, 2015). Sandalwood cannot regenerate via coppice (Kealley, 1991). Historically, the native population has been threatened through clearing activities, fire, poor seed dispersal, grazing animals, and drought (Brand, Sawyer, & Evans, 2014; Kealley, 1989; Loneragan, 1990).

S. spicatum is a small and slow growing tree normally reaching heights of 3-10 m tall (Kealley, 1991). Sandalwood trees can be single- or multi-stemmed, and flower in years when there is sufficient summer and autumn rainfall, with the flowers forming between January and April (Kealley, 1991; Loneragan, 1990). From this flower, a red-brown fruit with leathery skin is developed (Kealley, 1989). Germination occurs after sufficient autumn and winter rains (Brand *et al.*, 2014) but the survival rate is very low; typically only 1-5% of seed germinate and establish successfully in wild plots, and less than 20% in plantations (Kealley, 1991).

As mentioned, sandalwood is a root hemi-parasite (Hewson & George, 1984) which means *S. spicatum* is capable of photosynthesis, but it needs a host species to survive. Fine roots are established on the lateral roots of the sandalwood tree. These then produce a lateral haustorium, a modified root that attaches the sandalwood plant to the root of the host species, allowing it to form a parasitic connection and obtain nutrients (Helms, 1998; Loneragan, 1990). Known host species include plants from the *Acacia*, *Cassia*, *Casuarina*, and *Eremophila* genera (Loneragan, 1990). Studies have been conducted to determine the superior host species for *S. spicatum*. The host species, to be successful, would need to promote maximum growth of the sandalwood tree, live as long as the sandalwood tree, and. *Acacia saligna* was identified as a promising host species, as in a trial it provided the best performing sandalwood trees. *A. saligna* was ultimately deemed unsuitable, however, as it would die on average 4 years after establishment, therefore not being able to provide nutrients for the sandalwood tree until harvest. ‘Jam’ species *Acacia acuminata* has been identified as a preferred host species, for its longevity and ability to provide adequate growing conditions for the sandalwood tree (Brand, 2009; Loneragan, 1990). It has even been suggested to plant both *A. saligna* and *A. acuminata*

with the sandalwood tree, to provide the best initial growth and continuous growth (Brand, Robinson & Archibald, 2003).

1.4 Sandalwood Industry in Australia

S. spicatum was first exported from Western Australia (WA) in 1845, two years after its value was first realised. Sandalwood was, and continues to be, a desirable commodity for export to Asian countries for use in incense manufacture. It was desirable not only for its high value, but also for its ease to harvest – it was cheaper and easier to exploit than other valuable export products such as heavy timber or whale products. In 1848 Sandalwood overtook wool as the colony's highest export commodity, providing 45% of its export income (Talbot, 1983). Traditionally, WA sandalwood is harvested from wild stands in the south west and central parts of Western Australia (Loneragan, 1990). While wild stands continue to be harvested, tree numbers are declining, which has forced the WA sandalwood industry to establish plantations for establishment and harvest, as well as active regeneration via seed sowing from native stands for future supplies (Moniodis *et al.*, 2017).

Agroforestry in Australia

Agroforestry is the practice of adding trees and shrubs onto farmland for benefits such as shade and shelter, soil and water conservation, wildlife habitat, aesthetics, and economic values (Helms, 1998; Reid *et al.*, 2015). Economic pressures and environmental degradation have caused farmers, both in Australia and internationally, to move away from the mass production of standardised commodities towards farming systems that encourage economic and environmental diversity. In some parts of Australia, the emergence of industries that encourage niche products, such as high quality timber, essential oils and exotic food and fibres, have benefitted otherwise strained rural economies (Tonts & Selwood, 2003).

At present, *S. spicatum* is promoted to farmers as a way of diversifying their income through agroforestry (Tonts & Selwood, 2003). Economically, sandalwood can benefit participating farmers when harvested for its highly valuable timber and oil. It can also benefit the health of the farmland by reducing waterlogging, wind erosion and salinity, improving soil structure and fertility, and acting as a windbreak (Woodall & Robinson, 2003). In addition to this, some host species can provide food and shelter for insects and small birds.

1.5 Oil Production

Sandalwood oil is desired for its aromatic quality and is used in perfumes and religious ceremonies (Clark, 2006). The global consumption of essential oils increases approximately 8-

10% annually, triggered by the increasing demand for essential oils by perfume and cosmetic companies, and the preference of natural materials over synthetic compounds by consumers (Kusuma & Mahfud, 2017). Sandalwood oil is produced in the heartwood of the tree. Currently, the heritability of oil production in the heartwood of *Santalum* species is not fully understood, and requires further investigation (Jones, Plummer, & Barbour, 2007). The trees are slow growing and typically require at least 20 years to acquire a substantial quantity of oil bearing wood (Brand & Pronk, 2011; Hettiarachchi *et al.*, 2012).

Due to the large and complex combination of sesquiterpenes found in sandalwood oil, and because the oil is localised (specifically in the heartwood), oil production could presumably serve as protection for the tree against wood-rotting fungal pathogens (Moniodis *et al.*, 2017). The quality of sandalwood oil is determined by the percentages of different sesquiterpenes within the oil, especially of α- and β-santalol, the most desirable fragrance compounds (Moniodis *et al.*, 2017). At present, the international standard for *S. album* oil is 41-55% α-santalol, and 16-24% β-santalol (ISO 3518:2002). The international standard for *S. spicatum* oil is 15-25% α-santalol, and 5-20% β-santalol (ISO 22769:2009). The oil of *S. spicatum* is considered to be chemically more complex than that of *S. album*, and as a result may have greater end uses with further research and development (McKinnell & Levinson, 2008). At present, there is a gap in knowledge regarding the components that comprise sandalwood oil, nor the biosynthetic pathways between these components.

1.6 Heartwood

Heartwood is a normally occurring part of the xylem in some trees. The heartwood of trees has properties that can significantly influence its usefulness as a product, such as a natural resistance to deterioration by insects and microorganisms. There is debate and some contradicting theories regarding heartwood formation. Many theorise that heartwood is formed as a response to a negative event during the tree's life. Ziegler (1967) deduced that heartwood was formed as a result of hormone imbalance associated with the increasing distance of the ray parenchyma cells from the cambium. Morrell (2002) suggests that heartwood is excess sapwood that has been converted in order to reduce energy demands. Rudman (1966), Stewart (1966) and Zeigler (1967) all propose that heartwood is formed due to a toxic build-up of polyphenols in the inner sapwood, resulting in the death of parenchyma cells which subsequently form heartwood. Chattaway (1949) indicates that heartwood is formed due to the presence of fungi in the inner sapwood of the given tree. Huber (1956) considers that decreased moisture content in the inner sapwood is the causation of heartwood. Bamber (1976) proposes

a different theory; Bamber theorises that heartwood formation is a developmental process of the tree, similar to processes of plant development such as fruit ripening and dormancy.

Sandalwood heartwood has not yet been studied in significant detail. There are gaps in the literature regarding what influences heartwood production. It has been theorised that heartwood may be formed in sandalwood as a reaction to stress (Page *et al.*, 2010; Rai, 1990), however no concluding evidence has yet been discovered.

1.7 Oil Induction Trials

Previous studies have been conducted into techniques to induce oil production in tree species. For example, a study to enhance oil production was performed in China, and looked at inducing agarwood production in *Aquilaria sinensis* trees. Like sandalwood oil, the resin from agarwood (otherwise known as agilawood) is valued as incense and perfume, as well as being a traditional medicine (Chen *et al.*, 2017; Gao *et al.*, 2012). Agarwood is found in the heartwood tissue of certain *Aquilaria* species (Cui *et al.*, 2013). Agarwood is formed as a result of external influences, such as animal grazing, insect attack, and microbial invasion, and cannot be formed in a normal healthy tree (Chen *et al.*, 2017; Gibson, 1977; Pojanagaroon & Kaewrak, 2005). Early studies into agarwood induction included physically wounding the tree, using axes among other tools (Chen *et al.*, 2017; Persoon & van Beek, 2008). When it was noticed that fungal infection would accompany the wound, the method of promoting agarwood production through fungal inoculation was developed (Chen *et al.*, 2017; Gibson, 1977). In a 2013 study, a random selection of healthy *A. sinensis* trees were injected with a fungus, YNAS04 strain of *Paraconiothyrium variabile*, which resulted in an improved agarwood production, allowing for a greater yield when harvesting agarwood resin (Cui *et al.*, 2013).

1.8 Methyl Salicylate

Methyl salicylate (MeSA) is a naturally occurring herbivory-induced volatile liquid produced by many plants (James & Price, 2004; Shulaev, Silverman, & Raskin, 1997). MeSA is produced in some plants as a response to stress, characterised for its ability to defend plants from attack, by attracting the predators of the attackers (James & Price, 2004). A study on soybeans in America observed the effect of MeSA when sprayed in high quantities on the soybeans (Zhu & Park, 2005). This study infested the crop with a predator, soybean aphid (*Aphis glycines*). It was observed that where the soybean plant produced a large amount of MeSA, seven-spot ladybirds (*Coccinella septempunctata*) and the Asian lady beetle (*Harmonia axyridis*) were found in the largest amounts. These insects are predators of *A. glycines* (Zhu & Park, 2005).

Similar studies in other plant species have identified MeSA as being produced as a response to predation by caterpillars, mites, aphids, and beetles (De Boer & Dicke, 2004; James, 2003; Van Den Boom *et al.*, 2004; Walling, 2000).

In tobacco plants, a study was conducted on plants inoculated with tobacco mosaic virus (TMV) (Shulaev *et al.*, 1997). This study observed that inoculated plants produced higher levels of MeSA. These higher levels of MeSA functioned as an airborne signal to the infected plants as well as the neighbouring plants. The airborne signal activates disease resistance and the expression of defence-related genes in both neighbouring plants and in the healthy tissues of the infected plant.

A study conducted in 2004 observed the attractiveness of MeSA to predatory insects (James & Price, 2004). Sticky cards on blocks were baited with MeSA, and insects caught on these sticky cards were collated and counted. The study found that cards that were baited with MeSA had collected the greatest amounts of five species of predatory insects. In this study, it was theorised that the controlled release of MeSA could have the ability to increase the amount of insects beneficial to the respective crop's survival and health.

In non-crop scenarios, MeSA has been known to be emitted when the plant is under herbivorous predation (Kessler & Baldwin, 2001; Van Den Boom *et al.*, 2004). Wild tobacco (*Nicotiana attenuata*) have displayed elevated levels of MeSA emission when attacked by tobacco hornworm larvae (*Manduca sexta*) (James & Price, 2004). Jimsonweed (*Datura stramonium*) and black locust (*Robinia pseudo-acacia*) produced high quantities of MeSA when attacked by the spider mite (*Tetranychus urticae*) (Van Den Boom *et al.*, 2004).

In laboratory-based studies, a predatory mite (*Phytoseiulus persimilis*) and predatory bug (*Anthocoris nemoralis*) were attracted to MeSA (Dicke *et al.*, 1990; Ozawa *et al.*, 2000; Sabelis & Dicke, 1987), whereas aphid pests are repelled by MeSA (Hardie *et al.*, 1994; James & Price, 2004; Pettersson *et al.*, 1994).

MeSA was chosen as a potentially effective treatment to increase oil yield in sandalwood trees for two reasons. Firstly, in a previous unpublished study located in WA, an increase of oil production in *S. album* was theorised with the addition of MeSA (Tungngoes *et al.*, 2015). Secondly, as MeSA is known to be a growth hormone and a tool for disease resistance, it was theorised that the introduction of this unknown chemical could stress the sandalwood tree and increase the oil yield.

1.9 The purpose of this study

The aim of this research was to determine the effect of chemical and physical treatments on oil production in Western Australian sandalwood. This was conducted over three plantations in the Wheatbelt region of Western Australia. Four treatments were applied to the sandalwood trees: a dowel soaked in methyl salicylate (MeSA), a blank dowel, a drill hole left empty, and a section of bark removed, as well as a group of trees left as a control for comparison.

The main objective was to explore the effects of MeSA as a stimulant of oil production in *S. spicatum* trees. As the method of induction involves physical disturbance of the tree, there were other treatments used to distinguish the effects of the chemical and the physical treatments on oil production.

This thesis contains five chapters: Chapter 2 describes the methods used in the study; Chapter 3 describes the results of the study; Chapter 4 discusses the results and explores the reasoning for the results, and Chapter 5 explores limitations of the study and potential further research needed.

2. Methods

2.1 Treatment techniques

2.1.1 Site Layout

At each of the three sandalwood plantations used, an area of land 120 m by 15 rows was designated for use in the experiment (see Figure 2.1). This area equated to approximately 0.9 ha at each plantation. At each site, the ≈0.9 ha plot was divided into 30 blocks. These blocks (known as replicate blocks) were 20 m long, and 3 rows wide. In each block, up to 12 sandalwood trees were measured and treated. Within each block, the first 12 trees were selected to be included in the study; trees were only excluded if they were too small (having a diameter at 300mm above ground of less than 20mm), if the tree was dead, or if there were more than 12 trees in the block. Each treatment type, including the control group of trees, were assigned to six replicates at each plantation. A linear plot layout was used for the longevity of this study. As this study is intended to last for a long time beyond that of the Masters project, there is a need for the trees to be easily accessible and identifiable in the future. The plots were also located within the plantations, reducing any potential edge effect.

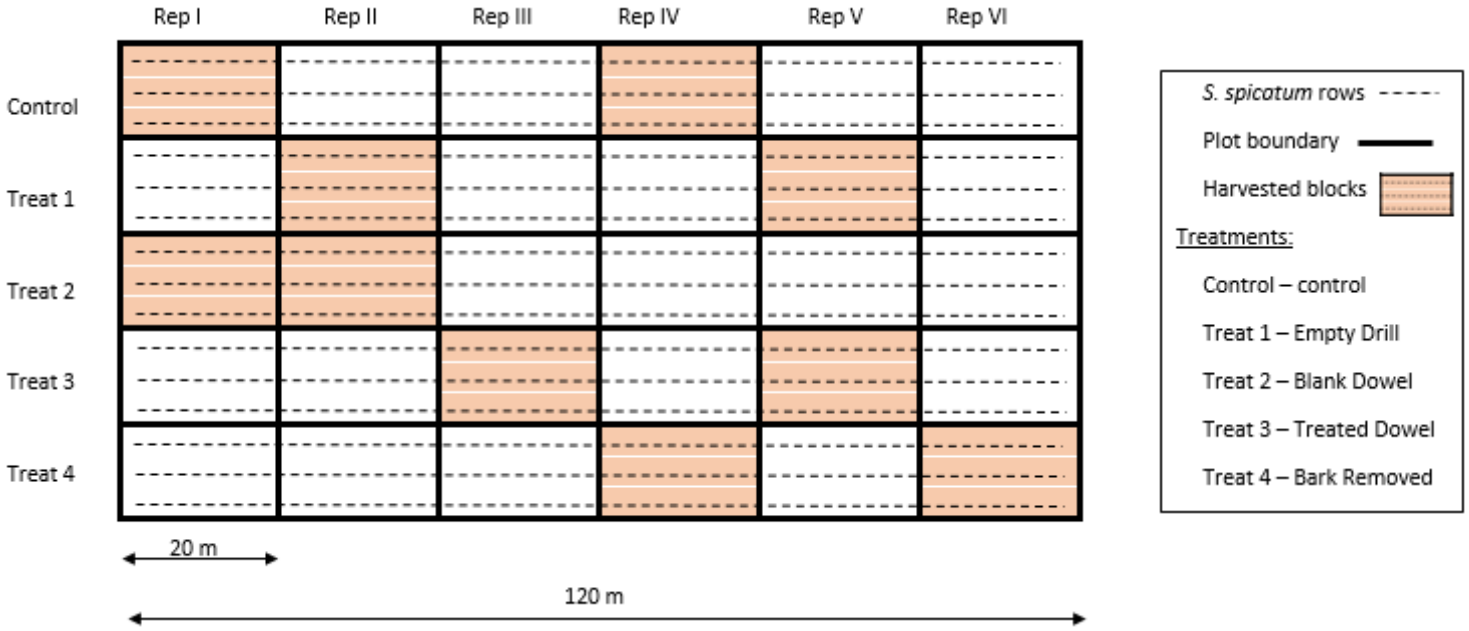

Figure 2.1: Plot design at each plantation.

2.1.2 Study Plantations

Three plantations were used for this study. Each were located in the Wheatbelt region of Western Australia. Each plantation had similar climatic characteristics, but differed in soil types. The sandalwood seed for all three plantations was derived from an FPC sharefarm

plantation located in Wandering, Western Australia (approximately 130 km south-east of Perth), which was seeded between 1998 and 1999.

Sandawindy

Sandawindy plantation is located near Moodiarrup, approximately 240 km south-east of Perth. This plantation has an area of 2.7 ha on a larger Forest Products Commission owned site. GPS coordinates for this site are: 33°36'29.8"S, 116°50'28.0"E. Sandalwood was planted in 2004, two years after the host species were planted. The host species within this site were *A. acuminata* (jam typical variant), *Acacia acuminata* (jam narrow-phyllode variant), and *Allocasuarina huegeliana* (rock sheoak). Host species were randomly distributed among the sandalwood. Rows were spaced 5 m apart, and were on red gravelly loam. This site was grazed by sheep, removing understorey plants.

Average annual rainfall for this area is 536.3 mm. Average minimum temperature is 9.4°C, and average maximum temperature is 21.5°C ("Qualeup (QA001)," n.d.). Rainfall and temperature data for Moodiarrup was unavailable, and so data from Qualeup was used. A total of 284 trees were used from this site, allowing approximately 60 trees for each of the four treatments applied to the sandalwood trees, as well as approximately 60 trees for the control group.

Kylie Reserve

Kylie Reserve is located in Bokal, approximately 235 km south-east of Perth. GPS coordinates are 33°25'42.6"S, 116°57'57.3"E. Sandalwood trees were planted in 2006, in rows spaced 5 m apart on sandy loam. The site contained 9-year-old *A. acuminata* (typical variant) hosts, which were planted one year before the sandalwood. The host species were stocked at 850 stems/ha, whilst sandalwood were 450 stems/ha.

Average rainfall is 482.1 mm/year, average minimum temperature is 9.8°C, and average maximum temperature is 23.0°C ("Mean Maximum Temperature - 010647 - Bureau of Meteorology," n.d.; "Mean Minimum Temperature - 010647 - Bureau of Meteorology," n.d.; "Monthly Rainfall - 010641 - Bureau of Meteorology," n.d.). Climatic and rainfall data were not available for Bokal; for rainfall, data from Maybrook station (7.2km away) was used instead. For temperature, data from Wagin (43 km east) was used. A total of 304 trees were used from this site; approximately 60 trees were designated for each treatment, as well as approximately 60 trees for the control group.

Brookton

The Brookton plantation is located near Brookton, in the western central Wheatbelt region of Western Australia. The property is approximately 135 km east of Perth. GPS coordinates for this plantation are 32°16'58.60"S, 117°3'11.81"E. Sandalwood trees were planted in 2006, two years after the host species, *A. acuminata* (typical variant) and *A. acuminata* (narrow-phyllode variant), were planted. The host trees were randomly distributed amongst the sandalwood trees. The sandalwood was growing on yellow sand in 'scalped' 1 m wide lines, spaced 4 m apart. Scalping is generally used on sandy soils to promote water retention and water availability near the developing planted seedlings. This is achieved by removing the top 10-20 cm of soil, which can be 'non-wetting' and also by removing the weeds in this top layer than will compete with the seedlings.

The average rainfall for Brookton is 450.2 mm/year ("Monthly Rainfall - 010524 - Bureau of Meteorology," n.d.). The average minimum temperature is 9.8°C, and the average maximum temperature is 24.3°C ("Mean Maximum Temperature - 010524 - Bureau of Meteorology," n.d.; "Mean Minimum Temperature - 010524 - Bureau of Meteorology," n.d.). At this site, 299 trees were used for this research; with approximately 60 trees used for each treatment and control group.

2.1.3 Measurement

Trees that were included in the study were measured for their overall height and their stem diameter at two heights above ground level. The height of sandalwood trees was measured using a PVC pipe that was marked in 0.1 m increments. One person would hold the pipe parallel to the tree, while another would stand facing the tree, so that the whole tree was in view, and read the height off the measuring pole. Heights of host trees were measured using the same method. This method of tree height measurement was chosen due to it being an uncomplicated and inexpensive method, as other methods such as the isosceles triangle method using a clinometer can be time consuming or require specialty equipment (Philip, 1998; Reid & Stephen, 2001). Stem diameter over bark was measured at 150 mm and 300 mm above ground, using a diameter tape. This method coincides with methods recommended in *The Farmers Forest* and *Measuring Trees and Forests* books (Philip, 1998; Reid & Stephen, 2001). If the tree had multiple stems, only the largest stem would be treated, and this stem along with the second-largest stem would be measured for diameter. Trees that were measured and included in the study were marked using uniquely labelled flagging tape. Flagging tape was attached to

a main branch of the selected sandalwood tree. The number of host trees in each replicate were counted and measured for height.

2.1.4 Treatments

Control group

The control group of trees did not have any treatment applied to them. This group was used as a comparison for the treated plants when looking at the oil composition and yield.

Empty Drill

Trees of the Empty Drill treatment had a hole, 8 mm in diameter, drilled through the stem of the sandalwood tree. The hole was drilled at 300 mm above ground, in a north-south direction completely through the tree stem. The hole was drilled using a cordless drill with an 8 mm auger piece, and was not sealed. This treatment was used to assist in separating the potential effects of the treated dowel treatment, by being able to potentially eliminate if the act of drilling the tree influenced the oil yield and composition.

Blank Dowel

The Blank Dowel treatment group repeated the Empty Drill treatment, with the addition of a piece of dowel. A piece of dowel, 8 mm in diameter, was inserted into the hole. The length of dowel inserted into each tree was 20 mm less than the diameter of the tree. The hole was then sealed at both ends using a gap sealant. The gap sealant was brown in colour, and of the brand Selleys, purchased from Bunnings Warehouse (model number 9300697116376). This treatment was able to identify if the foreign object inserted into the tree was an influencing factor on the oil yield and composition.

Treated Dowel

The Treated Dowel treatment group repeated the methods of the Blank Dowel treatment, except the dowel had been soaked in a plant hormone. The dowel had been dried at 103 °C for 24 hours in a drying oven, and then soaked in Methyl Salicylate (MeSA). The MeSA used was sourced from Australian Botanical Products Pty Ltd, and was of at least 98% concentration (determined via gas chromatography analysis) . The hole was then sealed using a gap sealant. This treatment was used to see if the chemical compound would have a significant impact on the production of oil within the sandalwood trees.

Bark Removed

In the Bark Removed treatment, a section of bark, which was 80 mm in height and went halfway around the tree, was removed using a chisel and mallet. The 80 mm section was

marked out as 40 mm above and below the 300 mm mark on the tree (*i.e.* between 260 mm and 340 mm), and had the centre point facing north. This treatment aimed to mimic occurrences in the rangelands, where dry-sidedness occurs on the north and western sides of sandalwood stems due to sun scald (Brand 1999).

2.2 Monitoring

The plantations were visited in May 2017, 6 months after they were treated. They were not measured at this time, but their health was observed. Only the Sandawindy and Brookton sites were visited. At these sites, all trees appeared to be healthy. The exposed sapwood on the bark removed trees had dried, hardened, and darkened in colour. At the Brookton site, where previously there had been no signs of fruit formation and only very few fallen nuts, buds were now developing on the sandalwood trees.

2.3 Harvesting and Preparation

2.3.1 Harvesting

The sandalwood trees were harvested 12 months after treatment. Before they were harvested, the sandalwood and host trees were measured, and the number of host trees were counted, according to the methods stated in the 'Measurement' section. For the Control group of trees, 300 mm above ground was marked using spray paint, so that this height could be identified during the coring stage. At each site, two entire replicate blocks from each treatment were harvested (see Figure 2.1). Only two blocks (of the 6 available) were harvested due to budget constraints and to allow the FPC to continue the study beyond this thesis. There were between 20 and 24 trees harvested per treatment type at each plantation, equalling approximately one third of trees at each site harvested. These blocks were chosen randomly. The trees were cut down using a chainsaw, operated by a current chainsaw ticket holder. The trees were first cut slightly below crown level, and then again at approximately 100 mm above ground. After they were felled, they were labelled using a permanent marker on the top cross section of the tree using unique codes that identified the site, treatment, replicate, and tree number. The trees were transported to FPC's Harvey Mill for storage until the coring and slicing stage.

2.3.2 Coring

A total of 150 trees were analysed for their oil quality and content. Ten trees from each treatment at each site were analysed for oil quality and volume. Of the 20-24 trees harvested per treatment type at each plantation, 10 were randomly selected for oil analysis. The logs were cored using a cordless drill with a 12 mm auger piece, 5 mm below the treatment site (*i.e.* 5mm

below 300mm above ground, or 295mm above ground). First, bark was removed from the log at the coring site using a rasp file. The trees were then drilled in a north-south direction. The shavings from the drill were collected, and stored in uniquely labelled paper bags. The bags were stored at the Harvey Mill until all the appropriate samples were cored. The samples were then delivered to Wescorp Sandalwood Pty Ltd for analysis of their oil yield and composition. This method of collecting shavings for analysis is supported in Braun, Meier & Hammerschmidt (2004) and Daramwar *et al*. (2012).

2.4 Oil Analysis

Tree cores were prepared to wood chip by direct attrition. The woodchip was then milled to a fine powder using a horizontal cutter mill and sieved to obtain particles below 2 mm. The oil was extracted by solvent extraction using the standard operating procedure of Wescorp Sandalwood Ltd (see Appendix 7.1). 1g of milled wood (accurately weighed) was transferred to a 20 mL Scintillation vial, 10 mL of hexane (with 1 % camphor) was added and the vial was sealed with a screw cap lid. The vial was placed in an ultrasonic bath and sonicated for 30 minutes at room temperature. The vial was then left to stand, and approximately 1 mL of the supernatant was transferred to a GC vial for analysis.

Calibration standards to measure % oil yield (w/w) were prepared, solutions of pure sandalwood oil at concentrations of 0.1-1 %(w/v) in hexane (containing 1 % camphor as the internal standard).

The extractions, calibration standards and blank (no oil in hexane) were analysed by a gas chromatograph (GC2010, Shimadzu Scientific, Japan) equipped with a flame ionisation detector. Full details are provided in Appendix 7.1.Separation was achieved on a 95% phenyl siloxane coated capillary column (Rt-5, Restek, USA). The column temperature gradient was 100°C to 140°C at 5°C/ min and then held at 140 °C for 25 min, and a gradient temperature resumed at 140-180°C at 5°C/min and held for 10 min. The detector was maintained at 220°C. 1 µL of sample was injected into the injector port which was held at 220°C at 110 kPa and with 50:1 split ratio.

A plot of mg oil/mg IS camphor) vs total peak area/peak of camphor was plotted and used to determine the % yield (w/w) of the milled wood.

The identity of the key analytes in the oil were determined by comparison of the retention times and retention index (Kovats Index). The concentration of the key sandalwood oil constituents

was determined by recording the peak area of the analyte of interest over the total peak area recorded for the sample (% composition).

2.4.1 Heartwood measurement

The spread of heartwood (the darkly stained oil-containing wood) through the sandalwood tree was recorded in all harvested sandalwood trees. The sandalwood log was cut into eight 25mm 'discs'; four above and four below the treatment point (300mm above ground). In this study, a disc refers to a cross-section of wood cut along the sandalwood log. Sandalwood logs were cut at FPC's Harvey Mill, using a radial arm saw. The size of the diameter of heartwood, transition wood, and clean wood was measured on each disc. Heartwood, if present, was in the centremost region of the disc, and was generally light to dark brown in colour. Transition wood, if present, would surround the heartwood, and was pink in colour. Sometimes there was transition wood but no heartwood present, with the transition wood in the centre of the disc. Sapwood surrounded any heartwood or transition wood, and was generally light yellow to cream in colour.

Measurements were taken from the top side of each disc, except for the bottom-most disc, which was measured on the top and bottom sides. This allowed for the spread of the heartwood from the treatment point to be mapped along the tree. Measurements were taken using a ruler, measuring in mm. Two diameters were recorded for each wood type. The diameters were taken at 90° angles to each other, and these measurements were used to get an area of each wood type on the disc, using the equation for an oval (Area = $\pi \times r_1 \times r_2$). These areas were then used to get a percentage of each type of wood for each sandalwood disc. Discs were numbered according to Table 2.1.

Table 2.1: Disc numbers and corresponding heights.

Disc Number	Height above ground (mm)
1	400
2	375
3	350
4	325
5	300
6	275
7	250
8 Top	225
8 Bottom	200

2.4.2 Statistical Analyses

Data Cleansing

For the oil yield (concentration), results that equalled a non-detect were recorded as half the detection level. The values did not necessarily mean zero, they just were below the detection limit. This method is supported by Clarke (1998) and Crogan & Egeghy (2003). For oil composition, data was excluded if it a non-detect was recorded or if it equalled 100%. Values of 100% were from samples where the oil composition was low, and only one oil component was detected, thus not truly equalling 100%.

Normality Tests

Statistical analyses were conducted using the software package SPSS. Data was tested for normality using a Shapiro-Wilk test, with a 95% significance level. If this test determined that data was not normally distributed, the range of the standardised residuals (following fitting linear models) were examined. If they were within the range of -4 to +4, they were accepted (as normal) and the raw data was used to analysis. If they were not within the correct residual range, the data was transformed, and then tested again for normality and residual range. Any required transformations for oil composition and yield was transformed using a log transformation. If a log transformation did not work, then a square root transformation was attempted. For heartwood area, square root transformations were used, as a log transformation is not appropriate for results equalling zero.

Normality test results and Transformations

For the oil concentrations, raw data was used for farnesol and nuciferol, while the data for total oil, α-santalol, and β-santalol was log transformed. Data for β-bisabalol was square root transformed. Raw data was used for the oil concentrations within the Sandawindy and Brookton plantations. Within the Kylie Reserve plantation, data for farnesol was log transformed, and all other oil components used raw data. For oil percentages, β-bisabalol used log transformed data, while the rest of the oil components used raw data. For the heartwood area, Disc 8 Bottom was log transformed, while the other discs used raw data. Results of normality tests are presented in Appendix 7.3.

Univariate General Linear Model

The data was analysed for statistically significant differences between the three different plantations (the three different sites) and treatments within plantations. This was done using a univariate general linear model (GLM), with a 95% confidence level, and with treatments nested within plantations. For the oil yield and % composition, as well as the disc comparisons, the univariate GLM examined if there were any significant differences between plantations, and between treatments within plantations, with the diameter measurement at 300 mm above ground used as a covariate. The stem diameter measurements were added as a covariate due to there being a modest correlation between stem diameter and oil yield, as determined when the data was being initially explored (note: GLMs were also done without stem diameter as covariates, for comparison). Correlations between stem diameter at 300 mm above ground and oil yield are as followed: Sandawindy $p = 0.153$, Kylie Reserve $p = 0.057$, Brookton $p = 0.108$. For the disc comparisons, there was also an examination of the differences between disc heights, to identify if there was a change in heartwood area within the height of the tree. Additional analysis occurred at each plantation separately, examining if there were significant differences between treatments within each of the plantations in terms of oil yields.

Simple Pair-wise Contrasts

If the univariate GLM resulted in a significance value of less than 0.05, simple pair-wise contrast with a significance level of 95% was conducted to determine between which pairs of treatments and/or plantations did such differences occur. Standard post-hoc tests could not be used due to the use of a covariate. A simple contrast was chosen as it allows all groups to be compared without penalisation. If the significance value was close to the 0.05 level, then a simple contrast was constructed to determine if there were any significant differences.

3. Results

In the first section (3.1 Oil Yields), the oil yields (in %w/w) are analysed and any significant differences between the plantations and between the treatments determined. Section '3.2 Oil Yields within the Plantations' looks at the effect of the treatments at each plantation individually. This allows removal of any potential plantation/site effects from potential differences between the treatment types. Section '3.3 Oil Quality' examines the composition of each oil component (i.e. as a percentage of the total amount of oil), a measure of oil quality. This is analysed at each plantation and for each treatment type. Lastly, section '3.4 Heartwood Area' describes the trends in the heartwood area (expressed as a percentage of the total stem cross-sectional area). Subsection 3.4.1 looks for differences in heartwood area between the disc heights, and any overall differences between the plantations or treatments when disc area percentages are combined. Subsection 3.4.2 looks at differences in heartwood area percentage between plantations and treatments, at each disc height individually.

3.1 Oil Yields

The sandalwood oil yield, measured as the percentage weight of oil per weight of the wood sample (% w/w) was compared between plantations (Figures 3.1 & 3.2), as well as between each treatment type across all the three plantations (Figures 3.3 & 3.4).

Mean values of total oil yield of trees in the three plantations ranged from 0.3 %w/w to 0.84 %w/w. In the treatments, total oil yield means ranged from 0.53 %w/w to 0.68 %w/w. For α-santalol, mean yields ranged from 0.05 %w/w to 0.035 %w/w in the plantations, and 0.05 %w/w and 0.1 %w/w in the treatments. For β-santalol, mean yields ranged from 0.01 %w/w in the plantations, to 0.035 %w/w in the treatments. For farnesol, mean yields ranged from 0.045 %w/w to 0.225 %w/w in the plantations, and from 0.1 %w/w to 0.205 %w/w in the treatments. For nuciferol mean yields, the plantations ranged from 0.025 %w/w to 0.125 %w/w, and from 0.05 %w/w to 0.105 %w/w for the treatments. For β-bisabalol, mean yields ranged from 0.03 %w/w to 0.11 %w/w in the plantations, and 0.055 %w/w to 0.075 %w/w in the treatments.

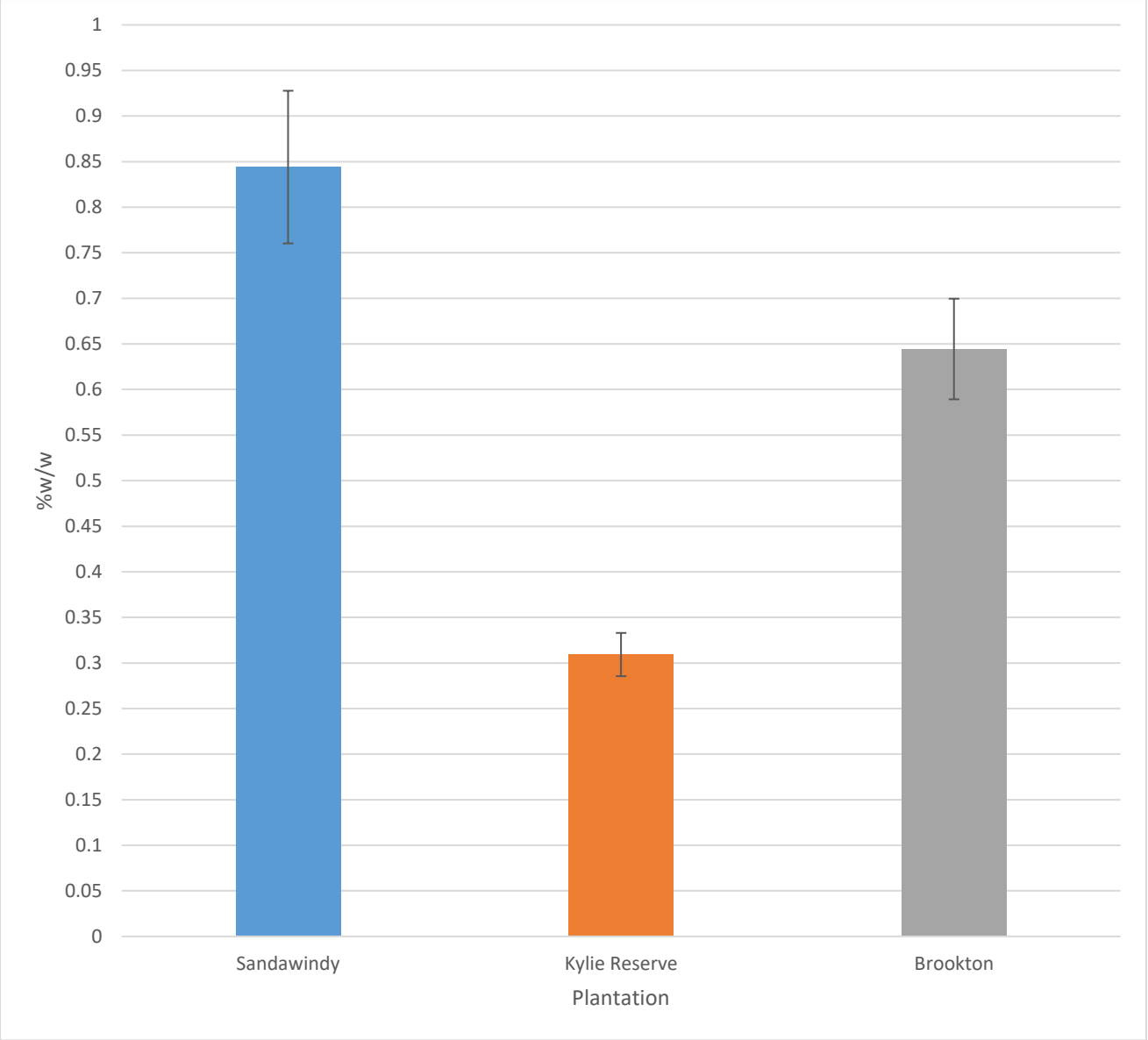

Figure 3.1: Mean yield of total sandalwood oil for each plantation, with standard error bars.

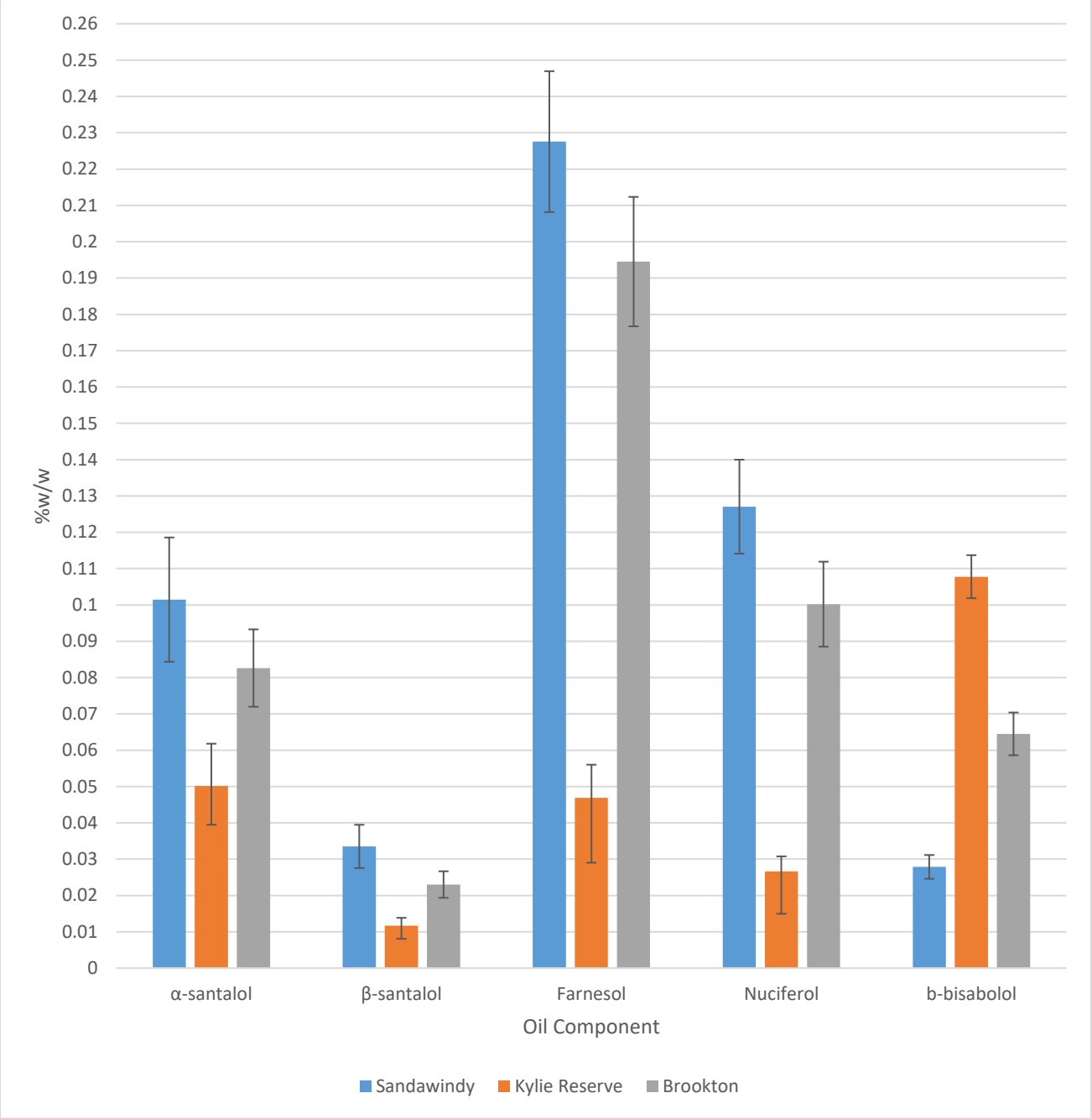

Figure 3.2: Mean yield of sandalwood oil components in each plantation, with standard error bars.

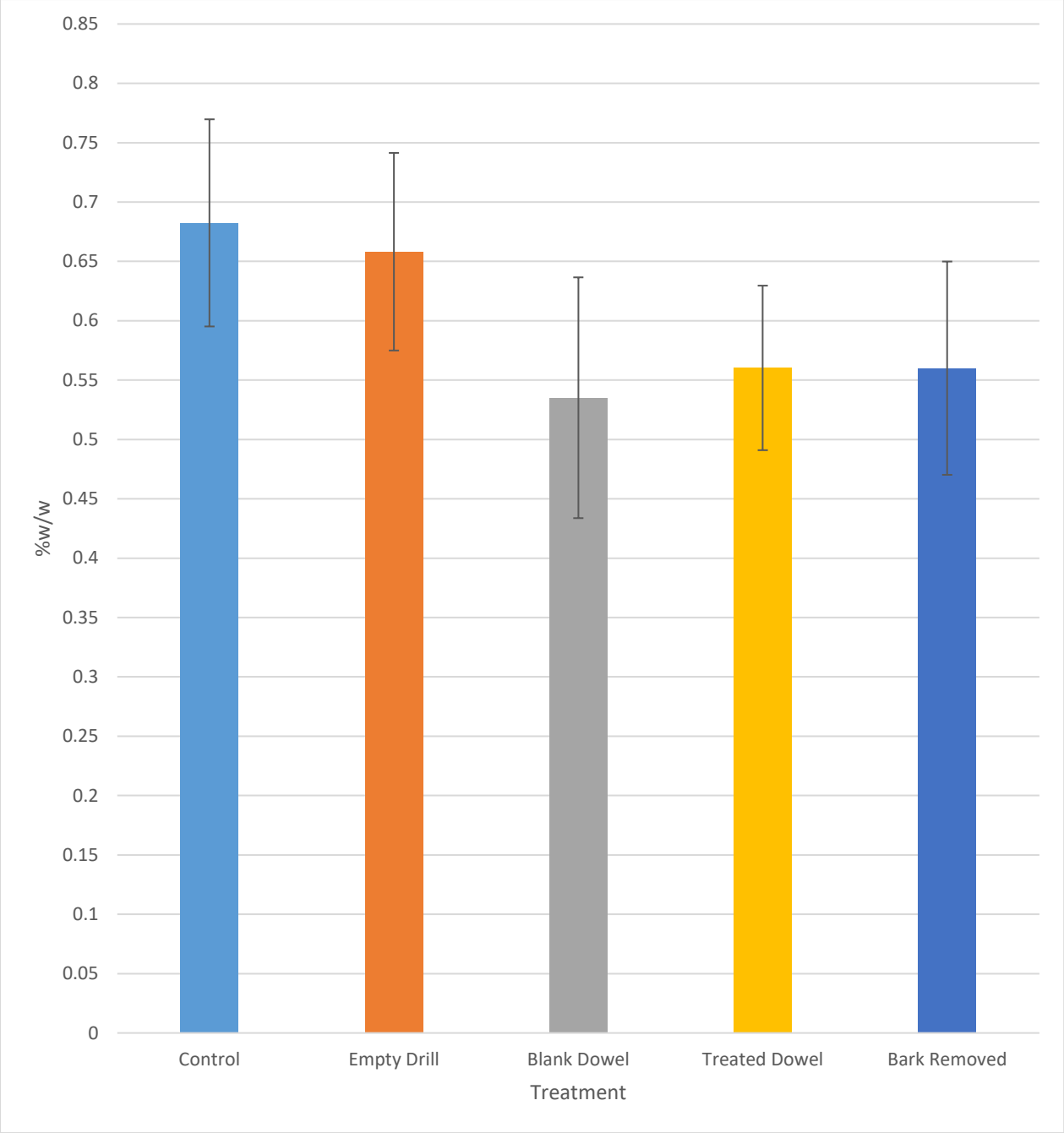

Figure 3.3: Mean total sandalwood oil yield for each treatment type, across all plantations, with standard error bars.

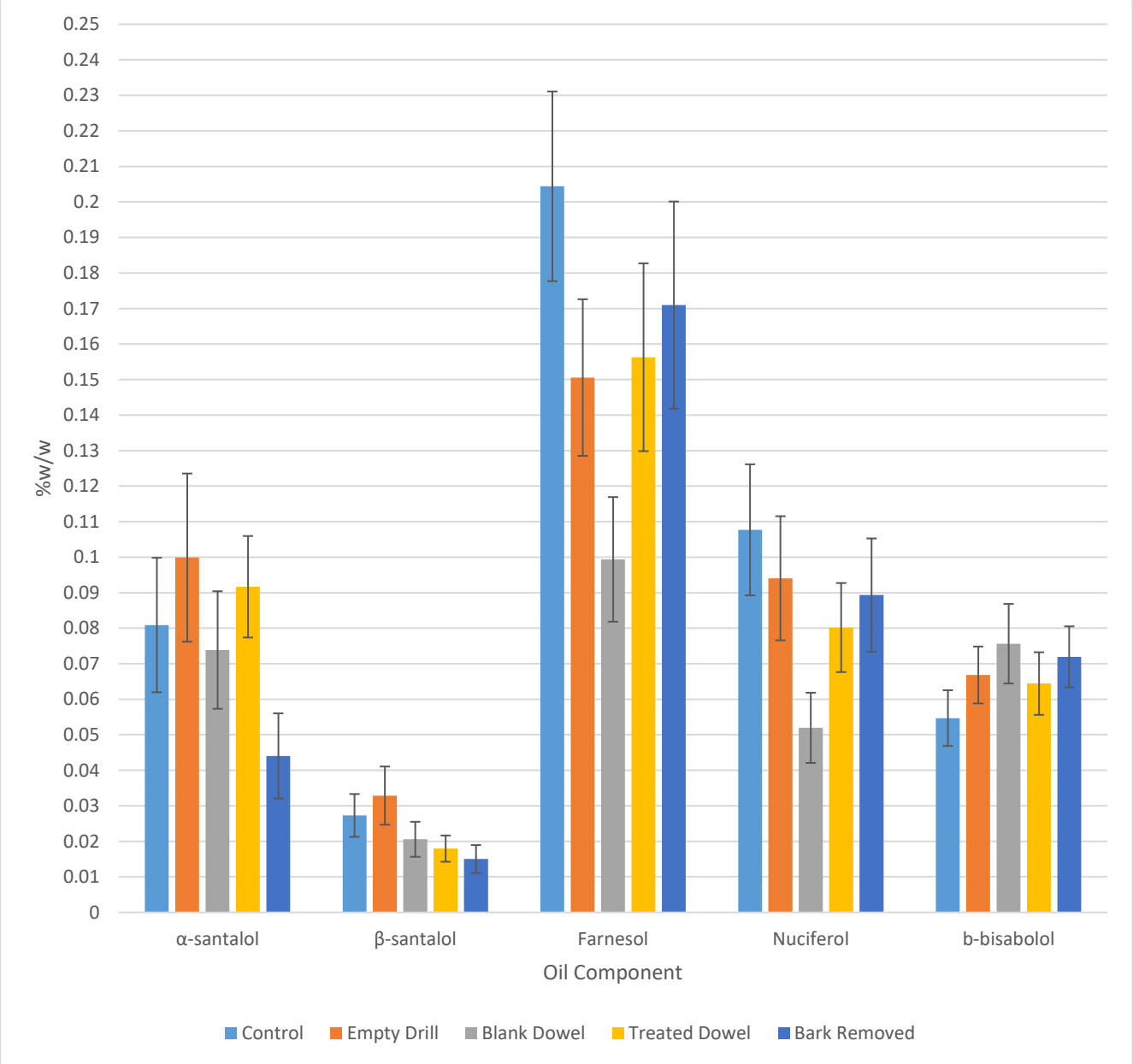

Figure 3.4: Mean yield of total sandalwood oil for each treatment type, with standard error bars.

3.1.1 Total oil yield

A univariate general linear model (GLM) determined that there were significant differences in total oil yield between the plantations ($p < 0.001$; Table 3.1). The treatments however did not show any significant differences in total oil yield ($p = 0.446$; Table 3.1). Kylie Reserve was significantly lower than the Sandawindy plantation ($p < 0.001$) and the Brookton plantation ($p < 0.001$; Table 3.2, Figure 3.1). Sandawindy and Brookton were not significantly different in total oil yield, however they did have a p value close to the significance level ($p = 0.061$; Table 3.2). Sandawindy was over 0.5 %w/w greater than Kylie Reserve on average, whilst Brookton was over 0.3 %w/w greater than Kylie Reserve. The Brookton and Sandawindy plantations

were close to being significantly different and were only 0.2 %w/w different from each other (Table 3.2, Figure 3.2). Diameter measurements were found to have a significant impact on the total oil yield (p = 0.004; Table 3.1).

Table 3.1: Univariate GLM for total oil %w/w yield.

Total Oil	***Type III SS***	***Df***	***MS***	***F***	***P***
Plantation	**9.596**	**2**	**4.798**	**16.166**	**< 0.001**
Diameter	**2.597**	**1**	**2.597**	**8.751**	**0.004**
Treatment(Plantation)	3.588	12	0.299	1.007	0.446

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.2: Simple contrast between plantations for total oil yield (%w/w).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**0.128**	**< 0.001**	**0.469**	**0.975**
	Brookton	0.109	0.061	-0.010	0.421
Kylie Reserve	**Sandawindy**	**0.128**	**< 0.001**	**-0.975**	**-0.469**
	Brookton	**0.127**	**< 0.001**	**-0.767**	**-0.266**
Brookton	Sandawindy	0.109	0.061	-0.421	0.010
	Kylie Reserve	**0.127**	**< 0.001**	**0.266**	**0.767**

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.1.2 α-santalol

Although no significant differences were found between the plantations (p = 0.098), there were significant differences in α-santalol yield between the different treatments (p = 0.013; Table 3.3). The Control group was significantly lower than the Treated Dowel treatment (p = 0.009; Table 3.4, Figure 3.4). The Bark Removed treatment was significantly lower than the Empty Drill treatment (p = 0.032), significantly lower than the Blank Dowel treatment (p = 0.018), and significantly lower the Treated Dowel treatment (p < 0.001; Table 3.4, Figure 3.4). Diameter measurements were found to have a significant effect on α-santalol yield (p < 0.001).

Table 3.3: Univariate GLM for α-santalol yield (%w/w).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	7.240	2	3.620	2.360	0.098
Diameter	**22.415**	**1**	**22.415**	**14.612**	**< 0.001**
Treatment(Plantation)	**41.173**	**12**	**3.431**	**2.237**	**0.013**

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.4: Simple contrast test between treatments for α-santalol yield (%w/w).

				95% CI	
		SE	***P***	***LL***	***UL***
Control	Empty Drill	0.321	0.337	-0.943	0.326
	Blank Dowel	0.323	0.787	-0.726	0.551
	Treated Dowel	**0.325**	**0.009**	**-1.499**	**-0.214**
	Bark Removed	0.325	0.233	-0.253	1.033
Empty Drill	Control	0.321	0.337	-0.326	0.943
	Blank Dowel	0.320	0.491	-0.412	0.855
	Treated Dowel	0.321	0.091	-1.183	0.088
	Bark Removed	**0.322**	**0.032**	**0.062**	**1.334**
Blank Dowel	Control	0.323	0.787	-0.551	0.726
	Empty Drill	0.320	0.491	-0.855	0.412
	Treated Dowel	**0.320**	**0.018**	**-1.402**	**-0.136**
	Bark Removed	0.320	0.139	-0.156	1.110
Treated Dowel	**Control**	**0.325**	**0.009**	**0.214**	**1.499**
	Empty Drill	0.321	0.091	-0.088	1.183
	Blank Dowel	**0.320**	**0.018**	**0.136**	**1.402**
	Bark Removed	**0.320**	**< 0.001**	**0.614**	**1.879**
Bark Removed	Control	0.325	0.233	-1.033	0.253
	Empty Drill	**0.322**	**0.032**	**-1.334**	**-0.062**
	Blank Dowel	0.320	0.139	-1.110	0.156
	Treated Dowel	**0.320**	**< 0.001**	**-1.879**	**-0.614**

Note: SE = Standard Error, P = Significance, CI = Confidence Interval, LL = Lower Limit, UL = Upper Limit. Bold denotes significant differences.

3.1.3 β-santalol

Significant differences were not found between plantations (p = 0.243) or treatments within plantations (p = 0.338) for β-santalol yield. The diameter measurements had a significant impact on the β-santalol yields (p = 0.005; Table 3.5).

Table 3.5: Univariate GLM for β-santalol yield (%w/w).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	2.860	2	1.430	1.430	0.243
Diameter	**8.001**	**1**	**8.001**	**8.003**	**0.005**
Treatment(Plantation)	13.609	12	1.134	1.134	0.338

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

3.1.4 Farnesol

A univariate GLM determined that significant differences in farnesol oil yield were found between the plantations (p < 0.001; Table 3.6). Simple contrasts were conducted and determined that significant differences were found between the Sandawindy plantation and the Kylie Reserve plantation (p < 0.001) and between the Brookton plantation and the Kylie Reserve plantation (p < 0.001; Table 3.7, Figure 3.2). Sandawindy had a farnesol level 0.18 % w/w greater than Kylie Reserve, and Brookton was 0.15 %w/w greater, on average (Figure 3.2). Significant differences were not found between the treatments (p = 0.067), however this result was close to the significance level and so simple contrasts were performed. These determined that the Blank Dowel treatment was significantly lower than the Control group (p = 0.001), Treated Dowel treatment (p = 0.039), and the Bark Removed treatment (p = 0.010; Figure 3.4). Diameter measurements were close to being significant towards farnesol yields (p = 0.053).

Table 3.6: Univariate GLM for farnesol yield (%w/w).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**0.453**	**2**	**0.227**	**19.057**	**< 0.001**
Diameter	0.045	1	0.045	3.820	0.053
Treatment(Plantation)	0.247	12	0.021	1.728	0.067

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.7: Simple contrast between plantations and between treatments for farnesol yield (%w/w).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**0.026**	**< 0.001**	**0.104**	**0.205**
	Brookton	0.022	0.143	-0.011	0.075
Kylie Reserve	**Sandawindy**	**0.026**	**< 0.001**	**-0.205**	**-0.104**
	Brookton	**0.025**	**< 0.001**	**-0.173**	**-0.072**
Brookton	Sandawindy	0.022	0.143	-0.075	0.011
	Kylie Reserve	**0.025**	**< 0.001**	**0.072**	**0.173**
Control	Empty Drill	0.028	0.081	-0.11	0.006
	Blank Dowel	**0.028**	**0.001**	**-0.15**	**-0.041**
	Treated Dowel	0.029	0.180	-0.095	0.018
	Bark Removed	0.029	0.420	-0.080	0.033
Empty Drill	Control	0.028	0.081	-0.006	0.11
	Blank Dowel	0.028	0.094	-0.10	0.008
	Treated Dowel	0.028	0.690	-0.045	0.067
	Bark Removed	0.028	0.360	-0.030	0.082
Blank Dowel	**Control**	**0.028**	**0.001**	**0.041**	**0.15**
	Empty Drill	0.028	0.094	-0.008	0.10
	Treated Dowel	**0.028**	**0.039**	**0.003**	**0.12**
	Bark Removed	**0.028**	**0.010**	**0.018**	**0.13**
Treated Dowel	Control	0.029	0.180	-0.018	0.095
	Empty Drill	0.028	0.690	-0.067	0.045
	Blank Dowel	**0.028**	**0.039**	**-0.12**	**-0.003**
	Bark Removed	0.028	0.600	-0.041	0.071
Bark Removed	Control	0.029	0.420	-0.033	0.080
	Empty Drill	0.028	0.360	-0.082	0.030
	Blank Dowel	**0.028**	**0.010**	**-0.13**	**-0.018**
	Treated Dowel	0.028	0.600	-0.071	0.041

Note: SE = Standard Error, P = Significance, CI = Confidence Interval, LL = Lower Limit, UL = Upper Limit. Bold denotes significant differences.

3.1.5 Nuciferol

Significant differences occurred between the plantations ($p < 0.001$), and between treatments ($p = 0.031$) for nuciferol oil yields (Table 3.8). Simple contrasts determined that, between the plantations, the Sandawindy plantation had a significantly greater yield (by a mean value of 0.1%) than the Kylie Reserve plantation ($p < 0.001$), and the Brookton plantation had a mean yield 0.7 %w/w greater than the Kylie Reserve plantation ($p < 0.001$; Table 3.9, Figure 3.2). Within the treatments, it was found that the Control group had twice the yield of nuciferol compared to the Blank Dowel treatment ($p = 0.004$), the Empty Drill treatment had approximately 0.03 %w/w less of nuciferol compared to the Blank Dowel treatment ($p = 0.025$), and the Bark Removed treatment had a mean nuciferol level of 0.055 %w/w greater than the Blank Dowel treatment ($p = 0.038$; Figure 3.4). Diameter measurements did not have a significant impact on the nuciferol yields ($p = 0.363$).

Table 3.8: Univariate GLM for nuciferol yield (%w/w).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**0.157**	**2**	**0.079**	**16.025**	**< 0.001**
Diameter	0.004	1	0.004	0.835	0.363
Treatment(Plantation)	**0.116**	**12**	**0.010**	**1.972**	**0.031**

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.9: Simple contrast between plantations and treatments for nuciferol yield (%w/w).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**0.016**	**< 0.001**	**0.060**	**0.125**
	Brookton	0.014	0.060	-0.001	0.054
Kylie Reserve	**Sandawindy**	**0.016**	**< 0.001**	**-0.125**	**-0.060**
	Brookton	**0.016**	**< 0.001**	**-0.098**	**-0.034**
Brookton	Sandawindy	0.014	0.060	-0.054	0.001
	Kylie Reserve	**0.016**	**< 0.001**	**0.034**	**0.098**
Control	Empty Drill	0.018	0.497	-0.024	0.048
	Blank Dowel	**0.018**	**0.004**	**0.017**	**0.090**
	Treated Dowel	0.018	0.183	-0.012	0.061
	Bark Removed	0.018	0.405	-0.021	0.052
Empty Drill	Control	0.018	0.497	-0.048	0.024
	Blank Dowel	**0.018**	**0.025**	**0.005**	**0.077**
	Treated Dowel	0.018	0.502	-0.024	0.048
	Bark Removed	0.018	0.869	-0.033	0.039
Blank Dowel	**Control**	**0.018**	**0.004**	**-0.090**	**-0.017**
	Empty Drill	**0.018**	**0.025**	**-0.077**	**-0.005**
	Treated Dowel	0.018	0.144	-0.065	0.007
	Bark Removed	**0.018**	**0.038**	**-0.074**	**-0.002**
Treated Dowel	Control	0.018	0.183	-0.061	0.012
	Empty Drill	0.018	0.502	-0.048	0.024
	Blank Dowel	0.018	0.114	-0.007	0.065
	Bark Removed	0.018	0.611	-0.045	0.027
Bark Removed	Control	0.018	0.405	-0.052	0.021
	Empty Drill	0.018	0.869	-0.039	0.033
	Blank Dowel	**0.018**	**0.038**	**0.002**	**0.074**
	Treated Dowel	0.018	0.611	-0.027	0.045

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.1.6 β-bisabalol

There were significant differences between the plantations ($p < 0.001$), whilst the treatments did not show significant differences ($p = 0.451$; Table 3.10) in β-bisabalol yields. A simple contrast test determined that Sandawindy was significantly lower than Kylie Reserve ($p < 0.001$), Brookton had a β-bisabalol level double that of Sandawindy ($p < 0.001$), and Brookton was significantly lower than Kylie Reserve ($p < 0.001$; Table 3.11, Figure 3.2). Diameter measurements did not have a significant impact on the β-bisabalol yields ($p = 0.704$).

Table 3.10: Univariate GLM for β-bisabalol yield (%w/w).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**0.522**	**2**	**0.261**	**45.225**	**< 0.001**
Diameter	0.001	1	0.001	0.145	0.704
Treatment(Plantation)	0.069	12	0.006	1.002	0.451

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.11: Simple contrast between plantations for β-bisabalol yield.

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**0.018**	**< 0.001**	**-0.201**	**-0.131**
	Brookton	**0.015**	**< 0.001**	**-0.118**	**-0.058**
Kylie Reserve	**Sandawindy**	**0.018**	**< 0.001**	**0.131**	**0.201**
	Brookton	**0.018**	**< 0.001**	**0.043**	**0.113**
Brookton	**Sandawindy**	**0.015**	**< 0.001**	**0.058**	**0.118**
	Kylie Reserve	**0.018**	**< 0.001**	**-0.113**	**-0.043**

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.2 Oil Yields within Plantations

The oil yields were analysed for significant differences between the different treatment types, at each plantation separately. This method allowed for any potential plantation effects to be removed from the analysis.

3.2.1 Sandawindy

In the Sandawindy plantation, mean oil yields among the treatments ranged from 0.04 %w/w to 0.15 %w/w for α-santalol. For β-santalol, there was a range of 0.03 %w/w. Farnesol mean yields ranged from 0.16 %w/w to 0.27 %w/w. Nuciferol means ranged from 0.09 %w/w to 0.16 %w/w. β-bisabalol yields had the smallest range, ranging from 0.023 %w/w to 0.032 %w/w.

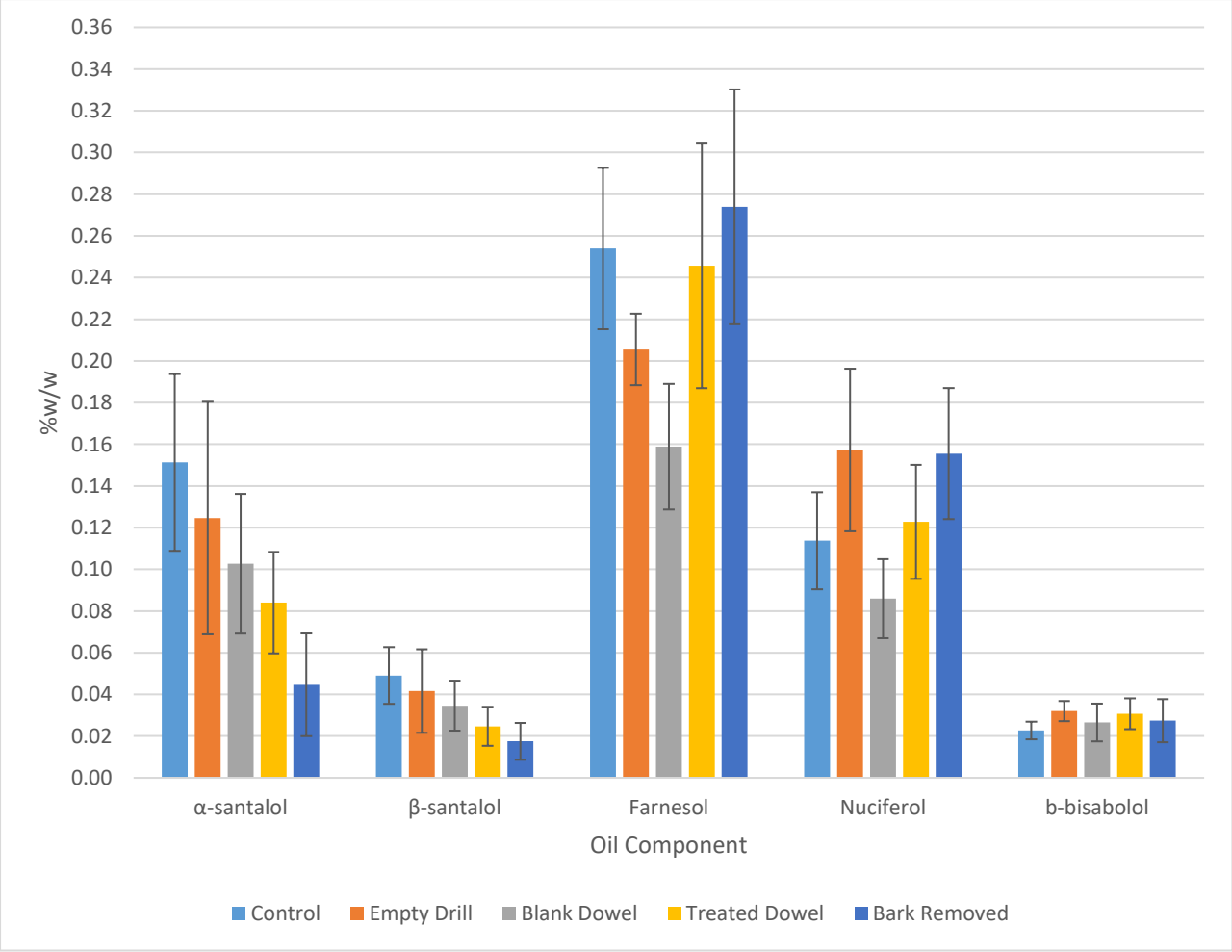

Figure 3.5: Mean yield of each oil component for each treatment type, with standard error bars, at the Sandawindy plantation.

A univariate GLM determined that there were no significant differences between any of the treatments within the Sandawindy plantation for any oil component (Table 3.12). Diameter

measurements were found to have a significant influence only for the α-santalol yield for Sandawindy (p = 0.030). The diameter measurements for β-santalol had a significance close to the significance limit (p = 0.059). When analysed for differences without the presence of diameter as a covariate, there still were no significant differences between treatments within the Sandawindy plantation for any oil component (Appendix 7.4, Table 7.16).

Table 3.12: Univariate GLM for α-santalol yield at the Sandawindy plantation.

		Type III SS	***MS***	***F***	***P***
α-santalol	*Treatment*[a]	0.029	0.007	0.555	0.697
	Diameter[b]	**0.067**	**0.067**	**5.007**	**0.030**
β-santalol	*Treatment*[a]	0.002	0.001	0.353	0.841
	Diameter[b]	0.006	0.006	3.765	0.059
farnesol	*Treatment*[a]	0.089	0.022	1.213	0.319
	Diameter[b]	0.033	0.033	1.795	0.187
nuciferol	*Treatment*[a]	0.035	0.009	1.040	0.398
	Diameter[b]	0.005	0.005	0.594	0.445
β-bisabalol	*Treatment*[a]	0.001	0.000	0.267	0.898
	Diameter[b]	$9.816e^{-5}$	$9.816e^{-5}$	0.169	0.683

Note: SS = Sum of squares, MS = Mean square, F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. a: df = 4, b: df = 1. Bold denotes below significance level (0.05).

3.2.2 Kylie Reserve

In the Kylie Reserve plantation, oil yields ranged from 0.02 %w/w to 0.10 %w/w for α-santalol. For β-santalol, yields had the smallest range, ranging from 0.01 %w/w to 0.017 %w/w. Farnesol mean yields ranged from 0.018 %w/w to 0.07 %w/w. Nuciferol means ranged from 0.014 %w/w to 0.035 %w/w. β-bisabalol ranged from 0.09 %w/w to 0.13 %w/w.

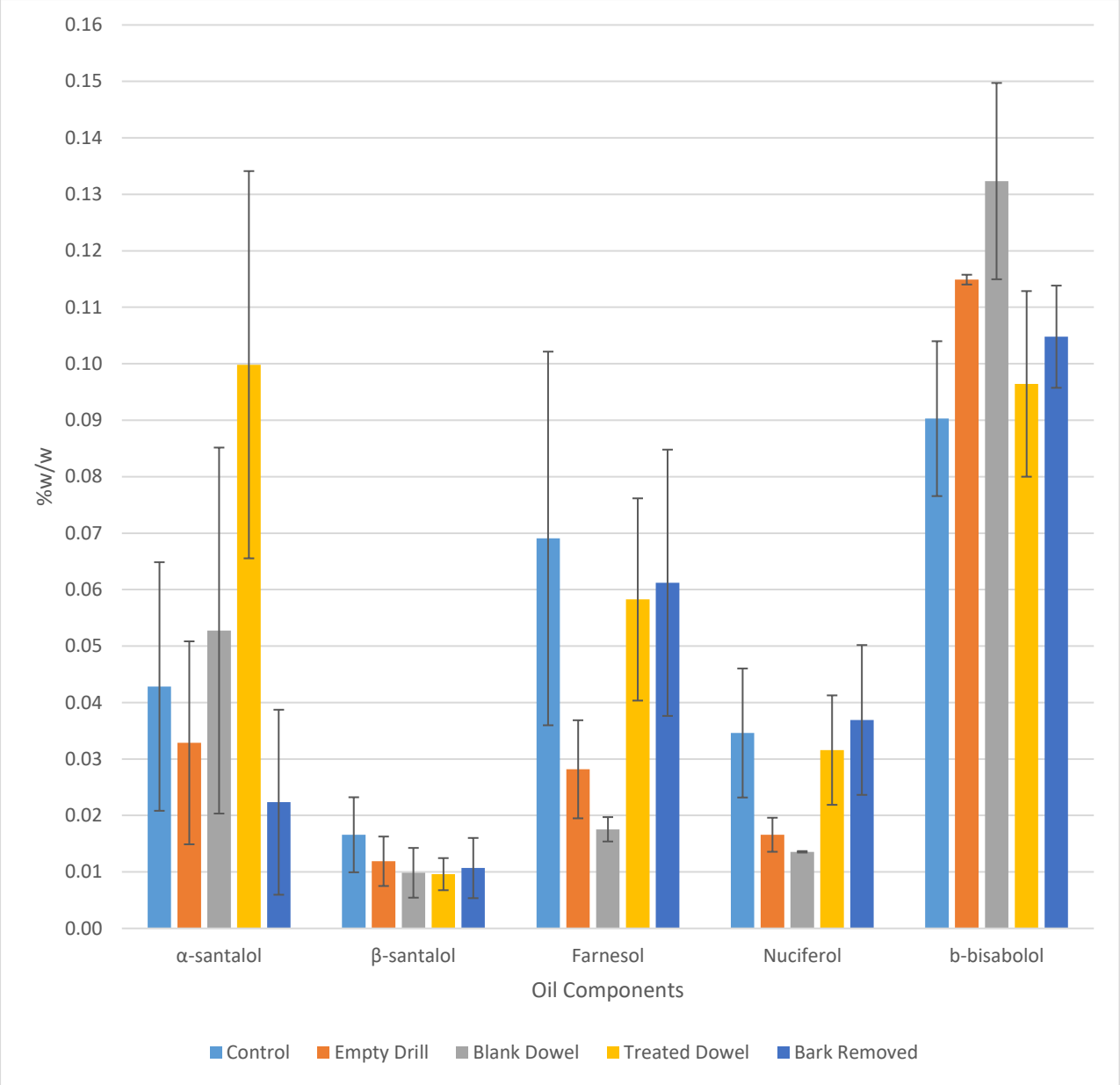

Figure 3.6: Mean yields of each oil component for each treatment type, with standard error bars, at the Kylie Reserve plantation.

A univariate GLM determined that there were no significant differences between the treatments for any oil component within the Kylie Reserve plantation (Table 3.13). The diameter measurements were found to have a significant influence on the α-santalol yield ($p = 0.004$), β-santalol yield ($p < 0.001$), and farnesol yield ($p = 0.006$). The diameter measurement had a close to significant positive relationship with nuciferol yields ($p = 0.056$).When the diameter measurements at 300 mm above ground were removed as a covariate, there were still no differences between the treatments for any oil component (Appendix 7.4, Table 7.17).

Table 3.13: Univariate GLM for α-santalol yield at the Kylie Reserve plantation (%w/w).

		Type III SS	***MS***	***F***	***P***
α-santalol	*Treatment*[a]	0.016	0.004	0.731	0.575
	Diameter[b]	**0.053**	**0.053**	**9.478**	**0.004**
β-santalol	*Treatment*[a]	0.001	< 0.001	1.637	0.182
	Diameter[b]	**0.003**	**0.003**	**18.243**	**< 0.001**
farnesol	*Treatment*[a]	5.483	1.371	1.833	0.140
	Diameter[b]	**6.205**	**6.205**	**8.296**	**0.006**
nuciferol	*Treatment*[a]	0.006	0.002	1.948	0.119
	Diameter[b]	0.003	0.003	3.851	0.056
β-bisabalol	*Treatment*[a]	0.012	0.003	1.870	0.133
	Diameter[b]	0.003	0.003	1.765	0.191

Note: SS = Sum of squares, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. a: df = 4, b: df = 1. Bold denotes below significance level (0.05).

3.3.3 Brookton

In the Brookton plantation, α-santalol yields ranged from 0.05 %w/w to 0.14 %w/w among the treatments. For β-santalol, yields had the smallest range, ranging from 0.02 %w/w to 0.04 %w/w. Farnesol mean yields ranged from 0.12 %w/w to 0.29 %w/w. Nuciferol means ranged from 0.06 %w/w to 0.17 %w/w. β-bisabalol yields ranged from 0.05 %w/w to 0.08 %w/w.

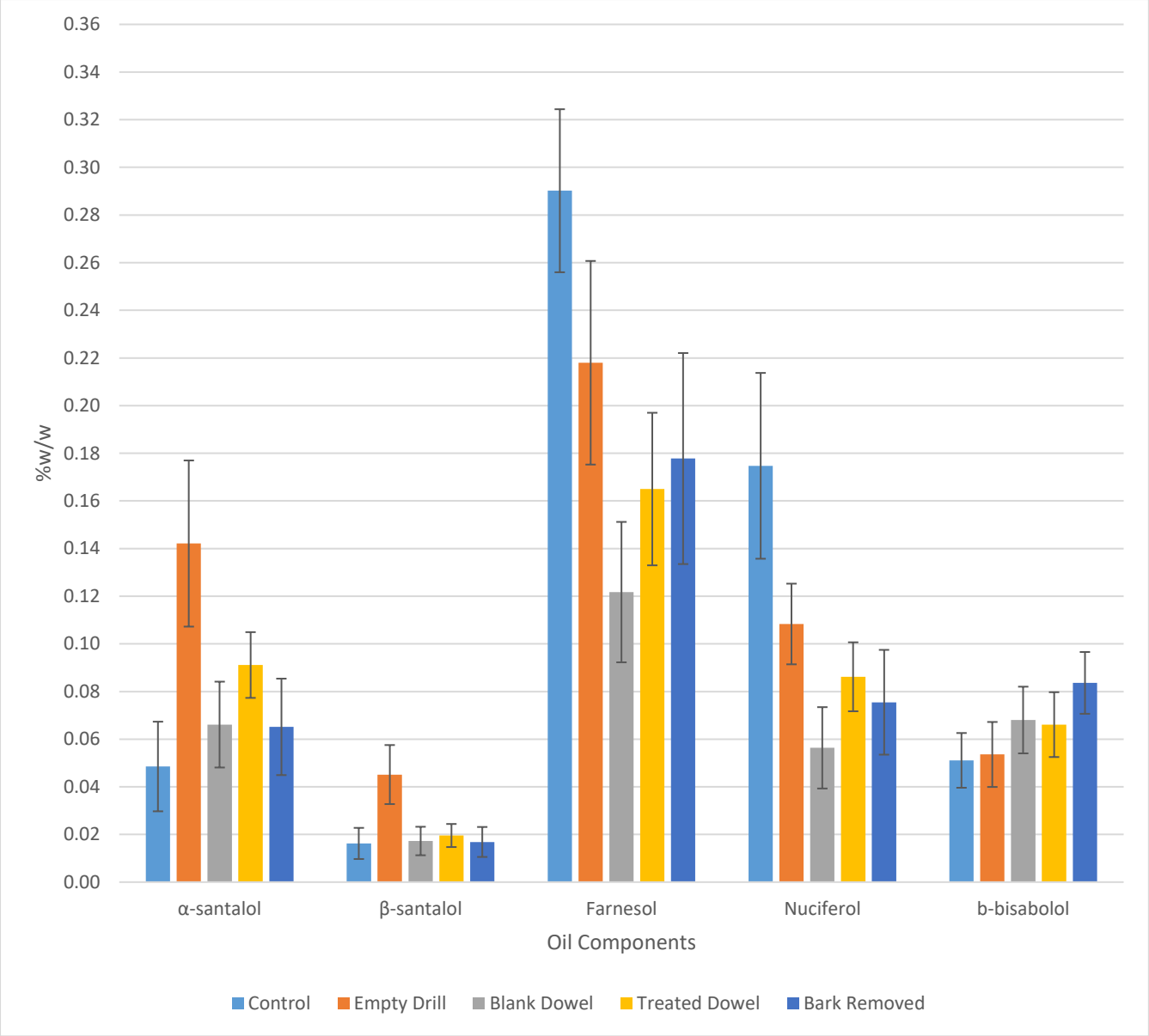

Figure 3.7: Mean yield of each oil component for each treatment type, with standard error bars, at the Brookton plantation.

There were significant differences between the treatments for α-santalol ($p = 0.035$), β-santalol ($p = 0.046$), and nuciferol ($p = 0.015$) within the Brookton plantation (Table 3.14). Farnesol had a p value close to the significance level ($p = 0.052$), and so a simple contrast was conducted. Diameter measurements did not have an effect on oil yield within the Brookton plantation for any oil component. When analysed without the diameter measurement as a covariate, significant differences still occurred for α-santalol ($p = 0.043$), β-santalol ($p = 0.047$), farnesol ($p = 0.030$), and nuciferol ($p = 0.010$; Appendix 7.4, Table 7.18).

Table 3.14: Univariate GLM for α-santalol yield at the Brookton plantation (%w/w).

		Type III SS	***MS***	***F***	***P***
α-santalol	***Treatment[a]***	**0.057**	**0.014**	**2.838**	**0.035**
	Diameter[b]	0.005	0.005	0.967	0.331
β-santalol	***Treatment[a]***	**0.006**	**0.002**	**2.652**	**0.046**
	Diameter[b]	0.000	0.000	0.489	0.488
farnesol	*Treatment[a]*	0.142	0.035	2.560	0.052
	Diameter[b]	0.008	0.008	0.582	0.450
nuciferol	***Treatment[a]***	**0.079**	**0.020**	**3.454**	**0.015**
	Diameter[b]	$4.91e^{-5}$	$4.91e^{-5}$	0.009	0.927
β-bisabalol	*Treatment[a]*	0.006	0.002	0.886	0.480
	Diameter[b]	0.000	0.000	0.203	0.654

Note: SS = Sum of squares, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. a: df = 4, b: df = 1. Bold denotes below significance level (0.05).

The α-santalol yield was highest for the Empty Drill treatment, and was significantly greater than the Control group (p = 0.005) by 0.09 % w/w on average, significantly greater than the Blank Dowel treatment (p = 0.020) by 0.07 %w/w, and significantly greater than the Bark Removed treatment (p = 0.019) by a mean value of 0.07 % w/w (Table 3.15, Figure 3.7). Without the diameter as a covariate, significant differences occurred between the same treatments, and significance levels remained the same (Appendix 7.4, Tables 7.18 & 7.19).

Table 3.15: Simple contrast between treatments for α-santalol yield in Brookton plantation.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	**Empty Drill**	**0.032**	**0.005**	**-0.16**	**-0.030**
	Blank Dowel	0.032	0.580	-0.081	0.046
	Treated Dowel	0.032	0.180	-0.11	0.021
	Bark Removed	0.032	0.600	-0.080	0.047
Empty Drill	**Control**	**0.032**	**0.005**	**0.030**	**0.16**
	Blank Dowel	**0.032**	**0.020**	**0.012**	**0.14**
	Treated Dowel	0.032	0.110	-0.013	0.11
	Bark Removed	**0.032**	**0.019**	**0.013**	**0.14**
Blank Dowel	Control	0.032	0.580	-0.046	0.081
	Empty Drill	**0.032**	**0.020**	**-0.14**	**-0.012**
	Treated Dowel	0.032	0.430	-0.089	0.039
	Bark Removed	0.032	0.980	-0.063	0.065
Treated Dowel	Control	0.032	0.180	-0.021	0.11
	Empty Drill	0.032	0.110	-0.11	0.013
	Blank Dowel	0.032	0.430	-0.039	0.089
	Bark Removed	0.032	0.420	-0.038	0.090
Bark Removed	Control	0.032	0.600	-0.047	0.080
	Empty Drill	**0.032**	**0.019**	**-0.14**	**-0.013**
	Blank Dowel	0.032	0.980	-0.065	0.063
	Treated Dowel	0.032	0.420	-0.090	0.038

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

For β-santalol, the Empty Drill treatment was significantly greater than all other treatments and the Control group. It was significantly greater than the Control group (p = 0.011) by a mean of 0.03 % w/w, significantly greater than the Blank Dowel treatment (p = 0.014) by a mean of 0.03 %w/w. significantly greater than the Treated Dowel treatment (p = 0.023) by a mean of 0.025 %w/w and significantly greater than the Bark Removed treatment (p = 0.012) by a mean

of 0.03 %w/w (Table 3.16, Figure 3.7). If the covariate was removed, the same treatments had significant differences, with the same significance levels (Appendix 7.4, Tables 7.18 & 7.20).

Table 3.16: Simple contrast between treatments for β-santalol yield in Brookton plantation.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	**Empty Drill**	**0.011**	**0.011**	**-0.051**	**-0.0070**
	Blank Dowel	0.011	0.930	-0.023	0.021
	Treated Dowel	0.011	0.760	-0.025	0.019
	Bark Removed	0.011	0.950	-0.023	0.021
Empty Drill	**Control**	**0.011**	**0.011**	**0.0070**	**0.051**
	Blank Dowel	**0.011**	**0.014**	**0.0060**	**0.050**
	Treated Dowel	**0.011**	**0.023**	**0.0037**	**0.048**
	Bark Removed	**0.011**	**0.012**	**0.0064**	**0.050**
Blank Dowel	Control	0.011	0.925	-0.021	0.023
	Empty Drill	**0.011**	**0.014**	**-0.050**	**-0.0060**
	Treated Dowel	0.011	0.830	-0.024	0.020
	Bark Removed	0.011	0.970	-0.022	0.022
Treated Dowel	Control	0.011	0.760	-0.019	0.025
	Empty Drill	**0.011**	**0.023**	**-0.048**	**-0.0037**
	Blank Dowel	0.011	0.833	-0.020	0.024
	Bark Removed	0.011	0.810	-0.019	0.025
Bark Removed	Control	0.011	0.950	-0.021	0.023
	Empty Drill	**0.011**	**0.012**	**-0.050**	**-0.0064**
	Blank Dowel	0.011	0.970	-0.022	0.022
	Treated Dowel	0.011	0.810	-0.025	0.019

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

For farnesol, significant differences occurred between the Control group and the Blank Dowel treatment (p = 0.004), and between the Control group and the Treated Dowel treatment (p = 0.035). The Control group and the Bark Removed group had a significance close to the limit

(p = 0.057; Table 3.17, Figure 3.7). With the covariate removed, significant differences occurred between the Control group and the Blank Dowel treatment (p = 0.002), between the Control group and the Treated Dowel treatment (p = 0.021), and between the Control group and the Bark Removed treatment (p = 0.037; Appendix 7.4, Tables 7.18 & 7.21).

Table 3.17: Simple contrast between treatments for farnesol yield in Brookton plantation (%w/w).

				95% CI	
		SE	**P**	**LL**	**UL**
Control	Empty Drill	0.053	0.230	-0.173	0.043
	Blank Dowel	**0.053**	**0.004**	**-0.269**	**-0.053**
	Treated Dowel	**0.054**	**0.035**	**-0.225**	**-0.008**
	Bark Removed	0.054	0.057	-0.213	0.003
Empty Drill	Control	0.053	0.230	-0.043	0.173
	Blank Dowel	0.053	0.074	-0.202	0.010
	Treated Dowel	0.053	0.331	-0.158	0.054
	Bark Removed	0.053	0.453	-0.146	0.066
Blank Dowel	**Control**	**0.053**	**0.004**	**0.053**	**0.269**
	Empty Drill	0.053	0.074	-0.010	0.202
	Treated Dowel	0.053	0.403	-0.062	0.150
	Bark Removed	0.053	0.290	-0.050	0.162
Treated Dowel	**Control**	**0.054**	**0.035**	**0.008**	**0.225**
	Empty Drill	0.053	0.331	-0.054	0.158
	Blank Dowel	0.053	0.403	-0.150	0.062
	Bark Removed	0.053	0.821	-0.094	0.118
Bark Removed	Control	0.054	0.057	-0.003	0.213
	Empty Drill	0.053	0.453	-0.066	0.146
	Blank Dowel	0.053	0.290	-0.162	0.050
	Treated Dowel	0.053	0.821	-0.118	0.094

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

For nuciferol, significant differences occurred between the Control group and the Blank Dowel treatment ($p = 0.001$), between the Control group and the Treated Dowel treatment ($p = 0.015$), and between the Control group and the Bark Removed treatment ($p = 0.006$). The Control group and the Empty Drill treatment had a significance close to the limit ($p = 0.062$; Table 3.18, Figure 3.7). With the covariate excluded, significant differences occurred between the Control group and the Empty Drill treatment ($p = 0.001$), between the Control group and the Treated Dowel treatment ($p = 0.011$), and between the Control group and the Bark Removed treatment ($p = 0.005$). The Control group and the Empty Drill treatment had a significance level of $p = 0.053$ (Appendix 7.4, Tables 7.18 & 7.22).

Table 3.18: Simple contrast between treatments for nuciferol yield in Brookton plantation.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	Empty Drill	0.034	0.062	-0.135	0.003
	Blank Dowel	**0.034**	**0.001**	**-0.187**	**-0.049**
	Treated Dowel	**0.035**	**0.015**	**-0.157**	**-0.018**
	Bark Removed	**0.034**	**0.006**	**-0.168**	**-0.029**
Empty Drill	Control	0.034	0.062	-0.003	0.135
	Blank Dowel	0.034	0.131	-0.120	0.016
	Treated Dowel	0.034	0.517	-0.090	0.046
	Bark Removed	0.034	0.337	-0.101	0.035
Blank Dowel	**Control**	**0.034**	**0.001**	**0.049**	**0.187**
	Empty Drill	0.034	0.131	-0.016	0.120
	Treated Dowel	0.034	0.381	-0.038	0.098
	Bark Removed	0.034	0.574	-0.049	0.087
Treated Dowel	**Control**	**0.035**	**0.015**	**0.018**	**0.157**
	Empty Drill	0.034	0.517	-0.046	0.090
	Blank Dowel	0.034	0.381	-0.098	0.038
	Bark Removed	0.034	0.752	-0.079	0.057
Bark Removed	**Control**	**0.034**	**0.006**	**0.029**	**0.168**
	Empty Drill	0.034	0.337	-0.035	0.101
	Blank Dowel	0.034	0.574	-0.087	0.049
	Treated Dowel	0.034	0.752	-0.057	0.079

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.3 Oil Quality

In comparison to the international standard for *S. spicatum*, no oil sample met the required international standard for all oil components (ISO, 2009). When the oil components were observed independently of each other, 16% of samples met the requirement for α-santalol, 23% met the requirement for β-santalol, 6% met the requirement for farnesol, 33% met the requirement for nuciferol, and 37% met the requirement for β-bisabalol. If farnesol was

excluded, 5% of samples met the requirement for α-santalol, β-santalol, nuciferol, and β-bisabalol.

α-santalol mean percentage composition ranged from 13% to 25% in plantations, and from 13% to 19% in treatments. For β-santalol, the plantation means ranged from 6% to 8%. The β-santalol mean percentage composition for the treatments ranged from 5.5% to 7.5%. Mean percentage composition for farnesol ranged from 27.5% to 34% for the plantations, and from 26.5% to 39% for the treatments. Nuciferol mean percentage composition ranged from 17% to 17.5% for the plantations, and from 13% to 22% for the treatments. β-bisabalol mean percentages at the plantations ranged from 4.5% to 12.5%, and for the treatments ranged from 3.5% to 10%.

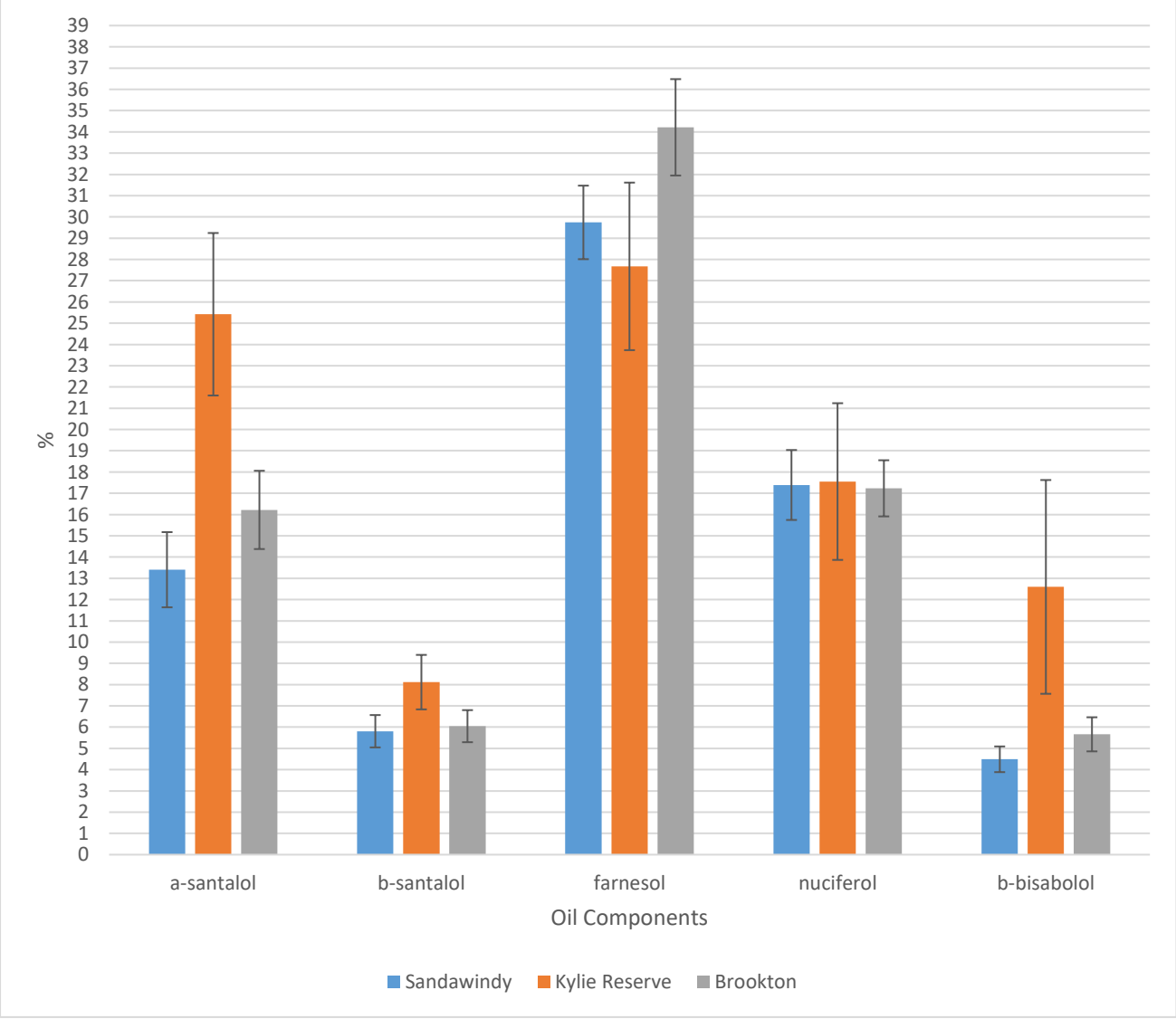

Figure 3.8: Mean percentage composition for each sandalwood oil component in each plantation, with standard error bars.

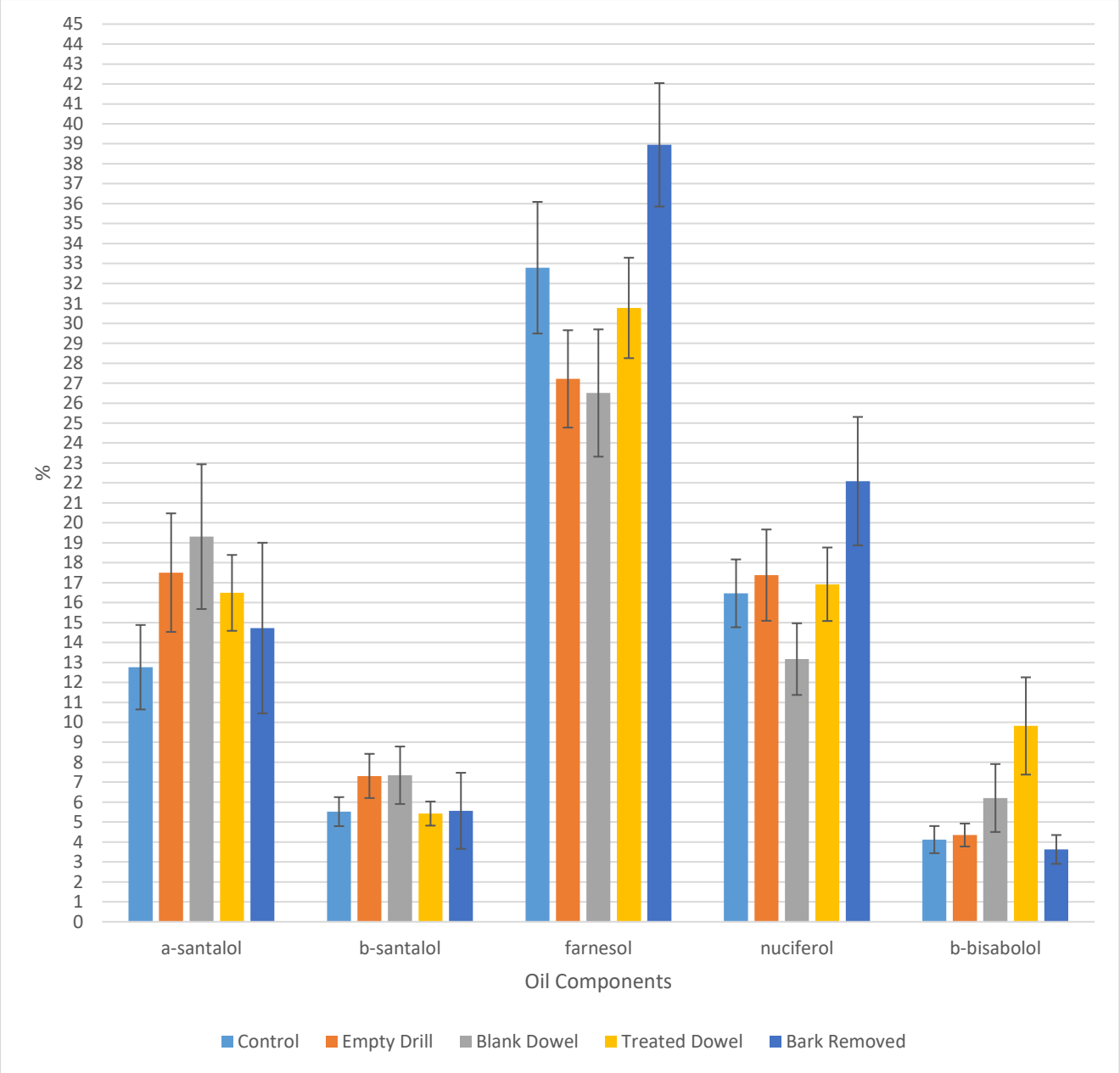

Figure 3.9: Mean percentage composition for each sandalwood oil component for each treatment type, with standard error bars.

3.3.1 α-santalol

There were significant differences between the treatments ($p = 0.017$) and between the plantations ($p < 0.001$) in the percentage α-santalol composition of the oil (Table 3.19). A simple contrast determined that significant differences occurred between the Sandawindy plantation and the Kylie Reserve plantation ($p < 0.001$), where Kylie Reserve was almost double that of Sandawindy, and between the Brookton plantation and the Kylie Reserve plantation ($p < 0.001$), where Kylie Reserve was an extra 9% higher than Brookton (Figure 3.8). For the treatments, the Control group was 5% lower than the Empty Drill treatment on

average (p = 0.017), and 6.5% lower than the Blank Dowel treatment (p = 0.002), and the Blank Dowel treatment was approximately 5% higher than the Treated Dowel treatment (p = 0.019). The Blank Dowel treatment was approximately 4% higher than the Bark Removed treatment, however did not meet the significance level (p = 0.071; Table 3.20, Figure 3.9). The diameter measurements did not have a significant impact on the α-santalol composition (p = 0.410).

Table 3.19: Univariate GLM for α-santalol composition (%).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**2332.652**	**2**	**1166.326**	**9.869**	**< 0.001**
Diameter	81.125	1	81.125	0.686	0.410
Treatment(Plantation)	**3185.727**	**12**	**265.477**	**2.246**	**0.017**

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.20: Simple contrast between plantations and treatments for α-santalol composition (%).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**4.304**	**< 0.001**	**-27.70**	**-10.55**
	Brookton	2.468	0.172	-8.32	1.51
Kylie Reserve	**Sandawindy**	**4.304**	**< 0.001**	**10.55**	**27.70**
	Brookton	**4.247**	**< 0.001**	**7.26**	**24.18**
Brookton	Sandawindy	2.468	0.172	-1.51	8.32
	Kylie Reserve	**4.247**	**< 0.001**	**-24.18**	**-7.26**
Control	**Empty Drill**	**4.123**	**0.017**	**-18.29**	**-1.87**
	Blank Dowel	**4.935**	**0.002**	**-25.30**	**-5.64**
	Treated Dowel	3.699	0.27	-11.48	3.26
	Bark Removed	5.042	0.349	-14.79	5.29
Empty Drill	**Control**	**4.123**	**0.017**	**1.87**	**18.29**
	Blank Dowel	5.103	0.294	-15.55	4.77
	Treated Dowel	3.921	0.132	-1.84	13.78
	Bark Removed	5.206	0.309	-5.038	15.70
Blank Dowel	**Control**	**4.935**	**0.002**	**5.64**	**25.30**
	Empty Drill	5.103	0.294	-4.77	15.55
	Treated Dowel	**4.729**	**0.019**	**1.94**	**20.78**
	Bark Removed	5.857	0.071	-0.94	22.38
Treated Dowel	Control	3.699	0.270	-3.26	11.48
	Empty Drill	3.921	0.132	-13.78	1.84
	Blank Dowel	**4.729**	**0.019**	**-20.78**	**-1.94**
	Bark Removed	4.819	0.895	-10.23	8.96
Bark Removed	Control	5.042	0.349	-5.29	14.79
	Empty Drill	5.206	0.309	-15.70	5.038
	Blank Dowel	5.857	0.071	-22.38	0.94

	Treated Dowel	4.819	0.895	-8.96	10.23

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.3.2 β-santalol

Significant differences occurred between the plantations (p = 0.050) for β-santalol percentage composition (Table 3.21). No significant differences were found between the treatment types (p = 0.507). Significant differences were found between Sandawindy plantation and Kylie Reserve (p = 0.017), where Kylie Reserve was 2% higher than Sandawindy, and between Kylie Reserve and Brookton (p = 0.030), where Kylie Reserve was 2% higher than Brookton (Table 3.22, Figure 3.8). The diameter measurements did not have a significant influence on the β-santalol composition (p = 0.457).

Table 3.21: Univariate GLM for β-santalol composition (%).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**89.701**	**2**	**44.851**	**3.225**	**0.050**
Diameter	7.853	1	7.853	0.565	0.457
Treatment(Plantation)	158.957	12	13.246	0.952	0.507

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.22: Simple contrast between plantations β-santalol composition (%).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**1.671**	**0.017**	**-7.521**	**-0.771**
	Brookton	1.139	0.663	-2.800	1.800
Kylie Reserve	**Sandawindy**	**1.671**	**0.017**	**0.771**	**7.521**
	Brookton	**1.627**	**0.030**	**0.360**	**6.932**
Brookton	Sandawindy	1.139	0.663	-1.800	2.800
	Kylie Reserve	**1.627**	**0.030**	**-6.932**	**-0.360**

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.3.3 Farnesol

A univariate GLM was conducted and determined that there were significant differences between the plantations' farnesol composition (p = 0.046; Table 3.23). There were no significant differences between the treatments (p = 0.127). Kylie Reserve was significantly lower than Brookton (p = 0.021) by 7% (Table 3.24, Figure 3.8). The diameter measurements did not have a significant impact on the farnesol composition percentage (p = 0.070).

Table 3.23: Univariate GLM for farnesol composition (%).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**1082.965**	**2**	**541.482**	**3.185**	**0.046**
Diameter	570.377	1	570.377	3.355	0.070
Treatment(Plantation)	3131.661	12	260.972	1.535	0.127

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.24: Simple contrast between plantations for farnesol composition (%).

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	Kylie Reserve	4.575	0.160	-2.609	15.575
	Brookton	2.764	0.117	-9.870	1.115
Kylie Reserve	Sandawindy	4.575	0.160	-15.575	2.609
	Brookton	**4.603**	**0.021**	**-20.007**	**-1.713**
Brookton	Sandawindy	2.764	0.117	-1.115	9.870
	Kylie Reserve	**4.603**	**0.021**	**1.713**	**20.007**

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.3.4 Nuciferol

A univariate GLM was conducted and determined that no significant differences were found between the plantations (p = 0.866) or between the treatments (p = 0.403). The diameter measurements did not have a significant influence on the nuciferol compositions (p = 0.215; Table 3.25).

Table 3.25: Univariate GLM for nuciferol composition (%).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	29.429	2	14.714	0.144	0.866
Diameter	159.745	1	159.745	1.561	0.215
Treatment(Plantation)	1194.044	11	108.549	1.061	0.403

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground.

3.3.5 β-bisabalol

No significant differences were found between the plantations (p = 0.150) or between the treatments (p = 0.400) for β-bisabalol percentage composition. The diameter measurements did not have a significant influence on the β-bisabalol percentage composition (p = 0.232; Table 3.26).

Table 3.26: Univariate GLM for β-bisabalol composition (%).

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	2.657	2	1.329	1.952	0.150
Diameter	0.989	1	0.989	1.454	0.232
Treatment(Plantation)	7.251	10	0.725	1.066	0.400

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground.

3.4 Heartwood Area

The heartwood area percentage (HW area %) was measured and analysed for differences between plantations, treatments and disc heights (i.e. different stem heights above ground level). Disc heights were compared to each other for differences, and all discs were combined and analysed for differences between plantations and treatments (subsection 3.4.1). All disc heights were then analysed separately, identifying any differences between plantations or treatments (subsection 3.4.2). Between the disc heights, the HW area % at Sandawindy ranged from 13.75% to 17%. At Kylie Reserve, the HW area % ranged from 2% to 3.5%, whilst at Brookton the HW area % ranged from 20.5% to 22% (Figure 3.10). In the treatments, HW area % ranged from 12.5% to 14.5% for the Control group. In the Empty Drill treatment, HW area % ranged from 15% to 16.75%. For the Blank Dowel treatment, HW area % ranged from 11% to 12.5%. In the Treated Dowel treatment, HW area % ranged from 11.75% to 13.75%. In the Bark Removed treatment, HW area % ranged from 11.25% to 13% (Figure 3.11).

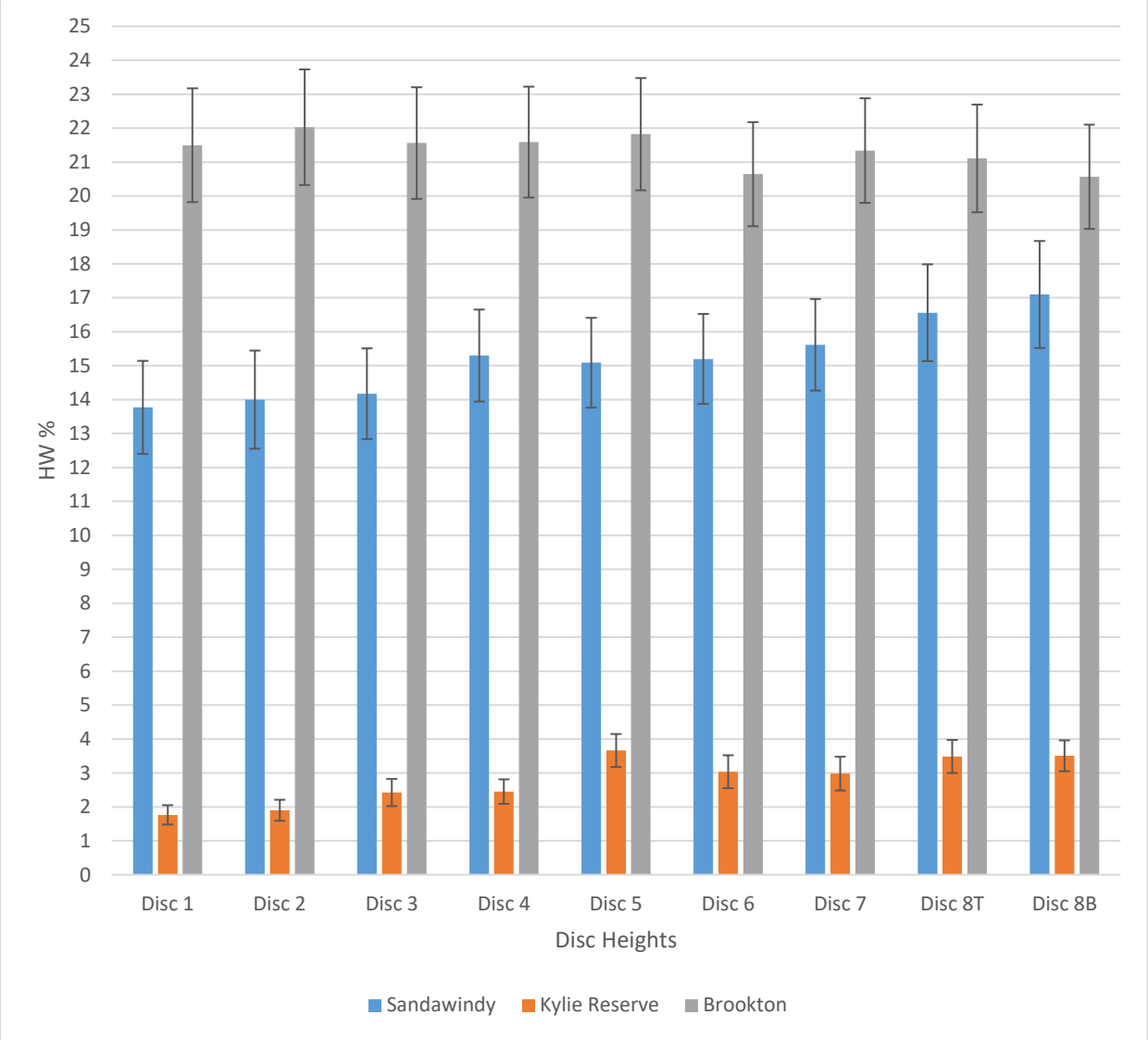

Figure 3.10: Mean percentage of heartwood area at each disc height at each plantation, with standard error bars.

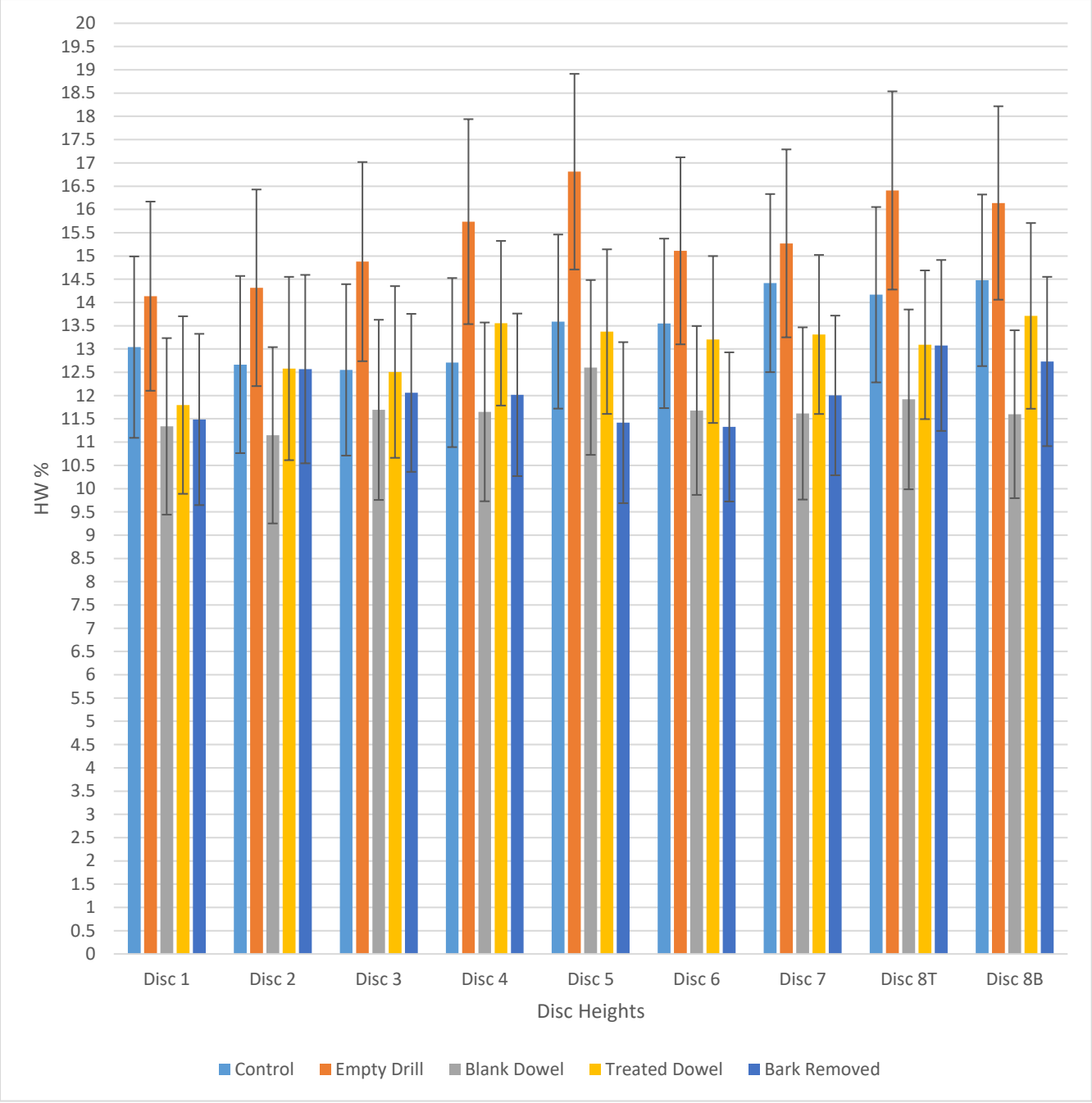

Figure 3.11: Mean percentage of heartwood area at each disc height for each treatment type, with standard error bars.

3.4.1 Comparisons Between Discs

Comparison was conducted between all discs, to determine if there were changes in HW area % between disc heights, between the plantations, and between the treatment types. The HW area % for the disc height comparison ranged from 12.5% to 13.5% (Figure 3.12). For the plantations, there was a range from 3% to 21% (Figure 3.13). For the treatments, HW area % ranged from 11.5% to 15.5% (Figure 3.14).

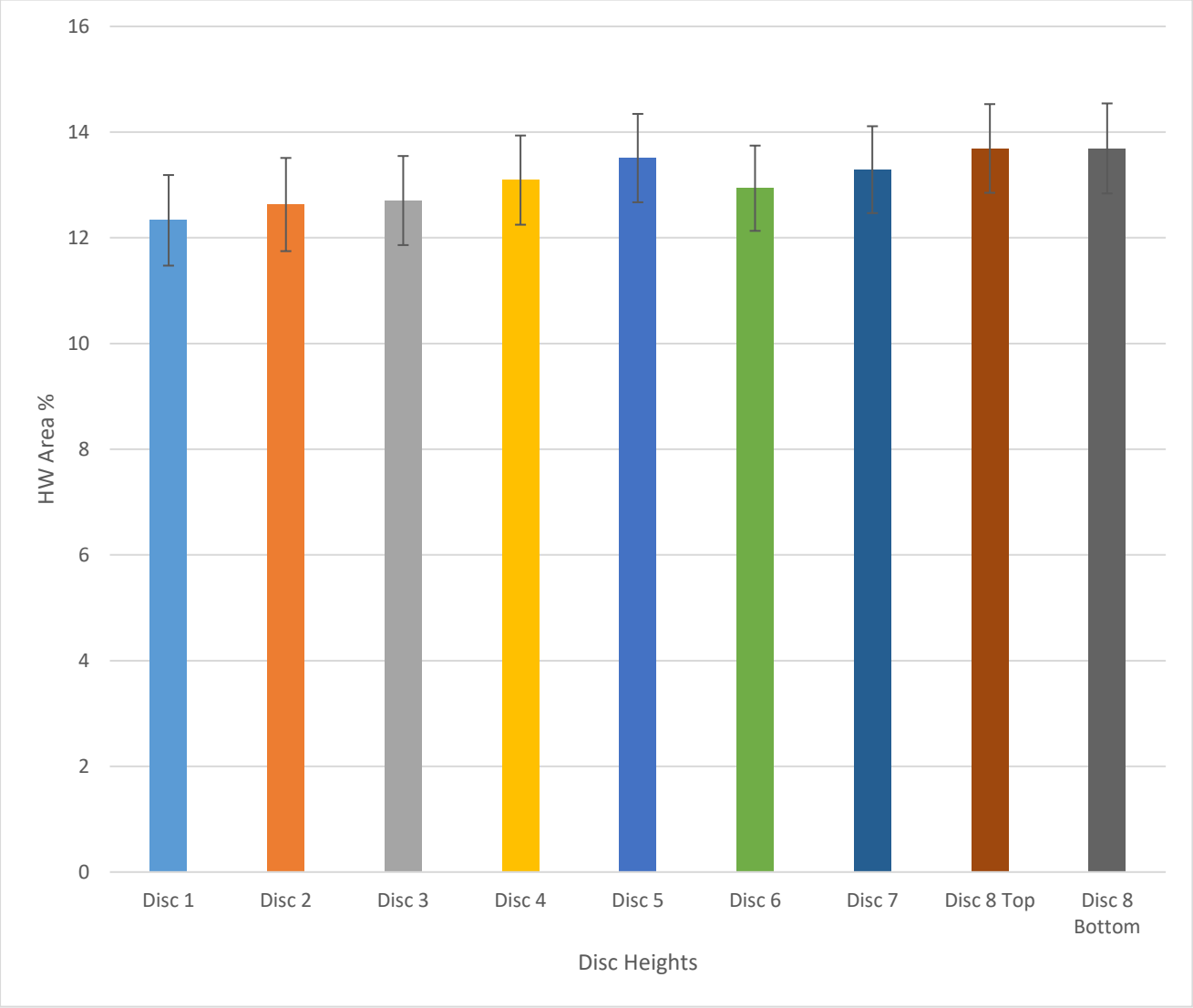

Figure 3.12: Mean HW area %, between all disc heights, with standard error bars.

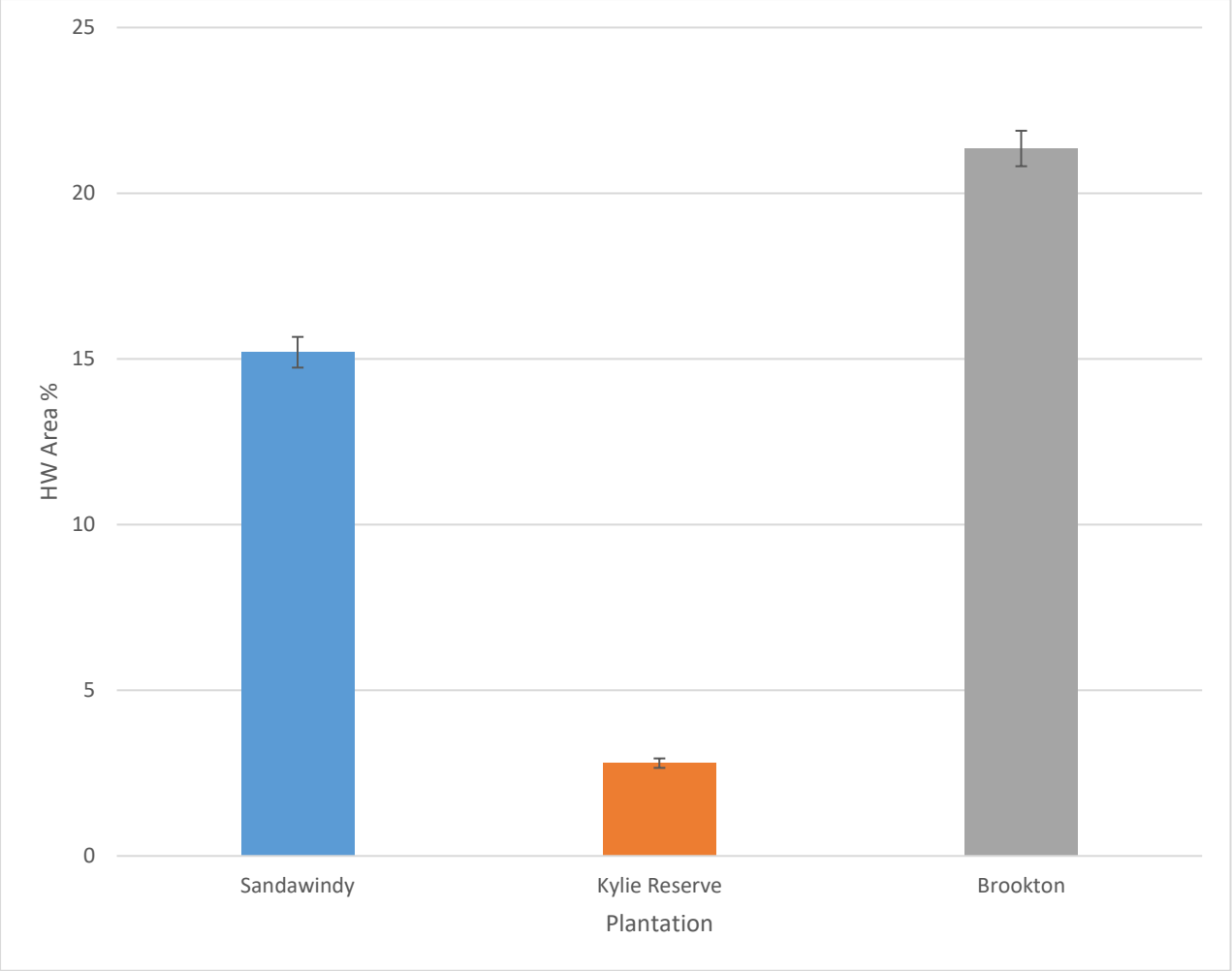

Figure 3.13: Mean HW area %, between all plantations, with standard error bars.

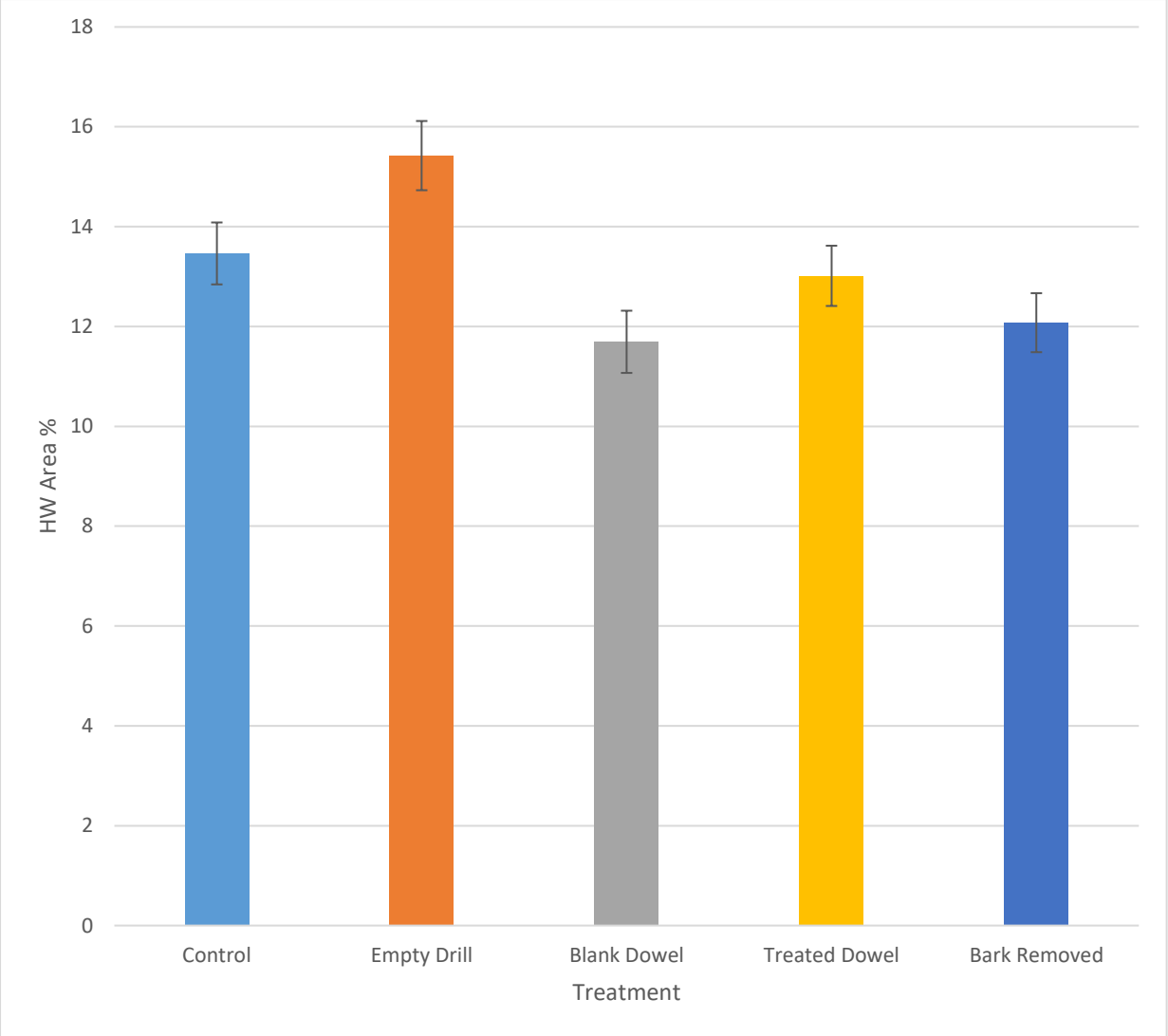

Figure 3.14: Mean HW area %, between all treatment types, with standard error bars.

No significant differences in % heartwood area occurred between the disc heights ($p = 0.118$). Significant differences were however present between the plantations ($p < 0.001$) and between the treatments ($p < 0.001$). In the plantations, Brookton was significantly greater in heartwood area % than Kylie Reserve ($p < 0.001$) and Sandawindy ($p < 0.001$). Sandawindy was significantly greater in heartwood area % than Kylie Reserve ($p < 0.001$; Figure 3.13). In the treatment types, significant differences were found between the Control group and the Blank Dowel treatment ($p = 0.002$), between the Empty Drill treatment and the Blank Dowel treatment ($p = 0.002$), and between the Blank Dowel treatment and the Treated Dowel treatment ($p = 0.027$; Figure 3.14). The difference between the Control group and the Bark Removed treatment ($p = 0.085$) and the Bark Removed treatment and Empty Drill treatment ($p = 0.092$) had significance levels close to the significance level (Table 3.27). Diameter measurements had a significant influence on % HW area ($p < 0.001$; Table 3.28).

Table 3.27: Univariate GLM for all discs % HW area.

	Type III SS	***Df***	***MS***	***F***	***P***
Plantation	**1866.487**	**2**	**933.244**	**321.996**	**< 0.001**
Disc	37.208	8	4.651	1.605	0.118
Diameter	**740.279**	**1**	**740.279**	**255.418**	**< 0.001**
Treatment(Plantation)	**470.384**	**12**	**39.199**	**13.525**	**< 0.001**

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

Table 3.28: Simple contrast between plantations and treatments for disc % HW area.

				95% CI	
		SE	***P***	***LL***	***UL***
Sandawindy	**Kylie Reserve**	**0.088**	**< 0.001**	**1.410**	**1.753**
	Brookton	**0.078**	**< 0.001**	**-0.809**	**-0.501**
Kylie Reserve	**Sandawindy**	**0.088**	**< 0.001**	**-1.753**	**-1.410**
	Brookton	**0.089**	**< 0.001**	**-2.411**	**-2.062**
Brookton	**Sandawindy**	**0.078**	**< 0.001**	**0.501**	**0.809**
	Kylie Reserve	**0.089**	**< 0.001**	**2.062**	**2.411**
Control	Empty Drill	0.103	0.999	-0.201	0.201
	Blank Dowel	**0.100**	**0.002**	**0.118**	**0.511**
	Treated Dowel	0.100	0.351	-0.103	0.291
	Bark Removed	0.100	0.085	-0.024	0.370
Empty Drill	Control	0.103	0.999	-0.201	0.201
	Blank Dowel	**0.102**	**0.002**	**0.114**	**0.516**
	Treated Dowel	0.102	0.362	-0.107	0.294
	Bark Removed	0.103	0.092	-0.028	0.375
Blank Dowel	**Control**	**0.100**	**0.002**	**-0.511**	**-0.118**
	Empty Drill	**0.102**	**0.002**	**-0.516**	**-0.114**
	Treated Dowel	**0.100**	**0.027**	**-0.418**	**-0.025**
	Bark Removed	0.100	0.156	-0.337	0.054
Treated Dowel	Control	0.100	0.351	-0.291	0.103
	Empty Drill	0.102	0.362	-0.294	0.107
	Blank Dowel	**0.100**	**0.027**	**0.025**	**0.418**
	Bark Removed	0.100	0.427	-0.117	0.276
Bark Removed	Control	0.100	0.085	-0.370	0.024
	Empty Drill	0.103	0.092	-0.375	0.028
	Blank Dowel	0.000	0.156	-0.054	0.337
	Treated Dowel	0.100	0.427	-0.276	0.117

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

3.4.2 Comparisons Within Discs

All discs had significant differences between the plantations (Table 3.29). No significant differences were found between the treatments at any disc level for heartwood area percentage. At each disc level, the diameter measurement had a significant impact on the heartwood area percentage.

Table 3.29: Univariate GLM for all disc heights for HW area %.

		Type III SS	Df	MS	F	P
Disc 1	**Plantation**	**9564.242**	**2**	**4782.121**	**31.763**	**< 0.001**
	Diameter	**3701.000**	**1**	**3701.000**	**24.582**	**< 0.001**
	Treatment(Plantation)	2960.202	12	246.683	1.638	0.080
Disc 2	**Plantation**	**9847.954**	**2**	**4923.977**	**29.825**	**< 0.001**
	Diameter	**3941.394**	**1**	**3941.394**	**23.874**	**< 0.001**
	Treatment(Plantation)	2084.856	12	173.738	1.052	0.401
Disc 3	**Plantation**	**9030.149**	**2**	**4515.075**	**30.117**	**< 0.001**
	Diameter	**3349.400**	**1**	**3349.400**	**22.342**	**< 0.001**
	Treatment(Plantation)	2496.286	12	208.024	1.388	0.170
Disc 4	**Plantation**	**9176.446**	**2**	**4588.223**	**30.341**	**< 0.001**
	Diameter	**2992.635**	**1**	**2992.635**	**19.790**	**< 0.001**
	Treatment(Plantation)	2100.119	12	175.010	1.157	0.314
Disc 5	**Plantation**	**8300.954**	**2**	**4150.477**	**27.096**	**< 0.001**
	Diameter	**2633.013**	**1**	**2633.013**	**17.189**	**< 0.001**
	Treatment(Plantation)	2696.389	12	224.699	1.467	0.136
Disc 6	**Plantation**	**7448.904**	**2**	**3724.452**	**26.876**	**< 0.001**
	Diameter	**3091.215**	**1**	**3091.215**	**22.306**	**< 0.001**
	Treatment(Plantation)	2441.151	12	203.429	1.468	0.135
Disc 7	**Plantation**	**7840.329**	**2**	**3920.165**	**27.873**	**< 0.001**
	Diameter	**3696.076**	**1**	**3696.076**	**26.280**	**< 0.001**
	Treatment(Plantation)	2125.725	12	177.144	1.260	0.242
Disc 8 Top	**Plantation**	**7379.007**	**2**	**3689.503**	**24.277**	**< 0.001**
	Diameter	**3261.866**	**1**	**3261.866**	**21.463**	**< 0.001**
	Treatment(Plantation)	3155.726	12	262.977	1.730	0.060
Disc 8 Bottom	**Plantation**	**160.186**	**2**	**80.093**	**25.896**	**< 0.001**
	Diameter	**91.930**	**1**	**91.930**	**29.723**	**< 0.001**
	Treatment(Plantation)	55.054	12	4.588	1.483	0.129

Note: SS = Sum of squares, df = degrees of freedom, MS = Mean square F = F statistic, P = Significance. Diameter refers to diameter measurement at 300mm above ground. Bold denotes below significance level (0.05).

For Discs 1 – 8 Top, significant differences occurred between all plantations (Table 3.30). The Brookton plantation is significantly greater than the both Sandawindy and Kylie Reserve, and Sandawindy is significantly greater than Kylie Reserve. For Disc 8 Bottom, Brookton and Sandawindy are both significantly greater than Kylie Reserve (Figure 3.10).

At Disc 1 and Disc 8 Top, the significance levels for the treatments were close to the 0.05 (Table 3.29), and so a simple contrast was conducted. This test determined that there were no significant differences between the treatment types (Appendix 7.5, Tables 7.23 & 7.24).

Table 3.30: Simple contrast between plantations for all disc heights HW area %.

					95% CI	
			SE	***P***	***LL***	***UL***
Disc 1	*Sandawindy*	**Kylie Reserve**	**1.893**	**< 0.001**	**4.152**	**11.603**
		Brookton	**1.693**	**< 0.001**	**-10.714**	**-4.049**
	Kylie Reserve	**Sandawindy**	**1.893**	**< 0.001**	**-11.603**	**-4.152**
		Brookton	**1.925**	**< 0.001**	**-19.047**	**-11.472**
	Brookton	**Sandawindy**	**1.693**	**< 0.001**	**4.049**	**10.714**
		Kylie Reserve	**1.925**	**< 0.001**	**11.472**	**19.047**
Disc 2	*Sandawindy*	**Kylie Reserve**	**1.983**	**< 0.001**	**3.900**	**11.703**
		Brookton	**1.773**	**< 0.001**	**-11.147**	**-4.168**
	Kylie Reserve	**Sandawindy**	**1.983**	**< 0.001**	**-11.703**	**3.900**
		Brookton	**2.015**	**< 0.001**	**-19.425**	**-11.493**
	Brookton	**Sandawindy**	**1.773**	**< 0.001**	**4.168**	**11.147**
		Kylie Reserve	**2.015**	**< 0.001**	**11.493**	**19.425**
Disc 3	*Sandawindy*	**Kylie Reserve**	**1.889**	**< 0.001**	**4.096**	**11.531**
		Brookton	**1.690**	**< 0.001**	**-10.357**	**-3.706**
	Kylie Reserve	**Sandawindy**	**1.889**	**< 0.001**	**-11.531**	**-4.096**
		Brookton	**1.921**	**< 0.001**	**-18.625**	**-11.066**
	Brookton	**Sandawindy**	**1.690**	**< 0.001**	**3.706**	**10.357**
		Kylie Reserve	**1.921**	**< 0.001**	**11.066**	**18.625**
Disc 4	*Sandawindy*	**Kylie Reserve**	**1.897**	**< 0.001**	**5.355**	**12.822**
		Brookton	**1.697**	**0.001**	**-9.276**	**-2.597**
	Kylie Reserve	**Sandawindy**	**1.897**	**< 0.001**	**-12.822**	**-5.355**
		Brookton	**1.929**	**< 0.001**	**-18.821**	**-11.229**
	Brookton	**Sandawindy**	**1.697**	**0.001**	**2.597**	**9.276**
		Kylie Reserve	**1.929**	**< 0.001**	**11.229**	**18.821**
Disc 5	*Sandawindy*	**Kylie Reserve**	**1.910**	**< 0.001**	**4.092**	**11.608**
		Brookton	**1.708**	**< 0.001**	**-9.778**	**-3.055**

	Kylie Reserve	**Sandawindy**	**1.910**	**< 0.001**	**-11.608**	**-4.092**
		Brookton	**1.941**	**< 0.001**	**-18.086**	**-10.446**
	Brookton	**Sandawindy**	**1.708**	**< 0.001**	**3.055**	**9.778**
		Kylie Reserve	**1.941**	**< 0.001**	**10.446**	**18.086**
Disc 6	*Sandawindy*	**Kylie Reserve**	**1.816**	**< 0.001**	**4.828**	**11.977**
		Brookton	**1.625**	**0.002**	**-8.326**	**-1.932**
	Kylie Reserve	**Sandawindy**	**1.816**	**< 0.001**	**-11.977**	**-4.828**
		Brookton	**1.847**	**< 0.001**	**-17.165**	**-9.898**
	Brookton	**Sandawindy**	**1.625**	**0.002**	**1.932**	**8.326**
		Kylie Reserve	**1.847**	**< 0.001**	**9.898**	**17.165**
Disc 7	*Sandawindy*	**Kylie Reserve**	**1.830**	**< 0.001**	**4.908**	**12.110**
		Brookton	**1.637**	**0.001**	**-8.598**	**-2.156**
	Kylie Reserve	**Sandawindy**	**1.830**	**< 0.001**	**-12.110**	**-4.908**
		Brookton	**1.860**	**< 0.001**	**-17.547**	**-10.225**
	Brookton	**Sandawindy**	**1.637**	**0.001**	**2.156**	**8.598**
		Kylie Reserve	**1.860**	**< 0.001**	**10.225**	**17.547**
Disc 8 Top	*Sandawindy*	**Kylie Reserve**	**1.902**	**< 0.001**	**5.445**	**12.931**
		Brookton	**1.701**	**0.014**	**-7.547**	**-0.851**
	Kylie Reserve	**Sandawindy**	**1.902**	**< 0.001**	**-12.931**	**-5.445**
		Brookton	**1.934**	**< 0.001**	**-17.192**	**-9.581**
	Brookton	**Sandawindy**	**1.701**	**0.014**	**0.851**	**7.547**
		Kylie Reserve	**1.934**	**< 0.001**	**9.581**	**17.192**
Disc 8 Bottom	*Sandawindy*	**Kylie Reserve**	**0.271**	**< 0.001**	**1.000**	**2.067**
		Brookton	0.243	0.110	-0.867	0.088
	Kylie Reserve	**Sandawindy**	**0.271**	**< 0.001**	**-2.067**	**-1.000**
		Brookton	**0.276**	**< 0.001**	**-2.466**	**-1.380**
	Brookton	Sandawindy	0.243	0.110	-0.088	0.867
		Kylie Reserve	**0.276**	**< 0.001**	**1.380**	**2.466**

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

4. Discussion

4.1 Effect of treatments

Of all the treatments used for this study, none consistently impacted the oil yield, oil composition, or heartwood area percentage. When compared to the Control group, the 'Bark Removed' treatment was not significantly greater or lower for each measured variable, so it appears this treatment had no effect. The 'Empty Drill' treatment was significantly greater than the Control group for α-santalol (Table 3.15) and β-santalol yield within the Brookton plantation (Table 3.16), and significantly greater than the Control group for α-santalol composition (Table 3.20). The Blank Dowel treatment was significantly lower than the Control group for farnesol and nuciferol yield (Table 3.7 & Table 3.9), and significantly higher than the Control group for farnesol yield within the Brookton plantation (Table 3.17). The Treated Dowel treatment was significantly higher than the Control group for α-santalol yield (Table 3.4) and farnesol yield within the Brookton plantation (Table 3.17).

Due to lack of consistency among the results, it can be said that there was no one treatment that overall affected sandalwood oil or heartwood production during the experimental period (12 months). It is more apparent that the different plantations, and their differing environmental and genetic elements, and perhaps age, had a larger impact on the oil production and quality.

4.2 Treated Dowel treatment

The Treated Dowel treatment did not significantly increase the total amount of oil or heartwood. This treatment was thought to have an effect on heartwood formation due to the nature of MeSA. Although not found in sandalwood, MeSA is produced in some plants when they experience stress (De Boer & Dicke, 2004; James, 2003; James & Price, 2004; Van Den Boom *et al.*, 2004; Walling, 2000). As it has been observed that heartwood is formed in sandalwood as a reaction to stress (Page *et al.*, 2010; Rai, 1990), it was theorised that by artificially introducing MeSA to the sandalwood tree, in response the tree would produce greater quantities of heartwood.

The results of the oil analyses did not support the theory of greater heartwood or oil production with the presence of MeSA. Although the Treated Dowel treatment was significantly higher than the Control group for α-santalol yield (Table 3.4) and for farnesol yield within the Brookton plantation (Table 3.17), it was not different to the Empty Drill treatment and so the effect cannot be attributed to MeSA alone. However, the Treated Dowel treatment performed better than the Blank Dowel treatment for α-santalol yield (Table 3.4), as well as heartwood

area between discs (Table 3.28). This treatment however also performed worse for α-santalol yield compared to the Blank Dowel treatment (Table 3.20), and for β-santalol yield within the Brookton plantation compared with the Empty Drill treatment (Table 3.16).

Although the Treated Dowel treatment performed better on some occasions, it was not consistently superior to the Control group or other treatments to improve the quantity of oil produced, including the oil components, nor the percentage of heartwood. As it did perform better on some occasions, it would be worthwhile to monitor longer term effects of the MeSA on sandalwood trees. Given the trees in this study were only monitored for one year post treatment, monitoring for at least another three years is recommended. As only 1/3 of trees were harvested at each plantation during this study, it is possible to extend the monitoring of sandalwood trees with the treatments using the remaining 2/3 of trees remaining.

4.3 Empty Drill treatment

Although there was no treatment that was consistently greater than the Control group, the Empty Drill treatment showed the highest oil contents on the most occasions. The Empty Drill treatment was significantly higher than the Blank Dowel treatment for Heartwood Area between discs (Table 3.28), nuciferol yield (Table 3.9), and α-santalol and β-santalol yield within the Brookton plantation (Tables 3.15 & 3.16). It was also significantly greater than the Treated Dowel treatment for β-santalol yield within the Brookton plantation (Table 3.16), and significantly greater than the Bark Removed treatment for α-santalol yield (Table 3.4), and α-santalol and β-santalol oil yield within the Brookton plantation (Tables 3.15 & 3.16).

These results support the rationale for separating the MeSA treatment into its individual components. This study was designed to separate the effects of the Treated Dowel treatment, to distinguish if a significant result was due to the wounding of the tree, the foreign object insertion, or the plant hormone. In a previous unpublished study examining the effect of MeSA on Indian Sandalwood (*Santalum album*), it was concluded that MeSA was responsible for an increase in oil yield (Tungngoes *et al.*, 2015). This study, however, failed to separate the physical and chemical components of the treament, and so their results cannot confidently state that the MeSA was the cause of the increase in oil (Tungngoes *et al*., 2015).

The Empty Drill treatment results support the idea that the wounding of the tree was the factor that triggered the oil production, rather than the dowel insertion or the MeSA. Other factors of the treatments such as the drying out of the drilled hole and the lack of gap sealant in the Empty Drill treatment could have influenced the results. However, longer term monitoring and more

study is required to confirm the positive effect of drilling into the stem. If shown to be effective, this treatment would be beneficial as it is an easier treatment to conduct compared to the treated dowel, given there is no need to source the MeSA or the dowel. This result is supported by studies in other tree species that use wounding as a way of increasing volatile oil. In *Aquilaria* trees, agarwood production can be stimulated by wounding the trees using methods such as cutting, holing, and hammering nails into the trunk (Chen *et al.*, 2017; Chhipa, Chowdhary, & Kaushik, 2016).

4.4 Oil spread throughout the stem

When testing for changes in heartwood area throughout the sandalwood tree, it was determined that there was no significant differences throughout the stem. There was no visible 'pooling' of heartwood at the location of the treated area for any treatment type, as is typically seen with agarwood in *Aquilaria* trees (Chen *et al.*, 2017; Li Zhang *et al.*, 2012; Liu *et al.*, 2013).

When sandalwood is harvested, the wood is sorted into up to five categories according to the oil content. The roots and butt typically have the best oil compositions, followed by the stem, which is sorted into 1st grade wood, 2nd grade wood, and 3rd grade wood depending on the diameter (Brand & Pronk, 2011). The wood harvested as part of this study ranged from 1st to 2nd grade wood. As it is known that the highest composition of sandalwood oil is found in the roots and the butt (Brand & Pronk, 2011), it is likely that, if the treatments made a significant impact, that the roots and butt would contain the most heartwood. As harvesting and oil analyses are expensive, extra analyses of the butt and roots was not feasible.

A study looking at the growth of Indian sandalwood clones (*Santalum album*) with different host trees in different locations of Western Australia examined the heartwood at multiple heights in the stem (McComb, 2009). This study measured percentage heartwood at the base of the trunk, and at 1.5 m above ground level. The results indicated that there were differences between the two heights, with the heartwood percentage decreasing at the 1.5 m height. However, McComb (2009) did not state whether there were significant heartwood percentage differences between the two heights. In this present study, it needs to be stated that not enough height was examined, which may have prevented significant differences in heartwood percentage being observed.

When looking at the discs individually, the treatments did not have a significant influence on the heartwood areas at any disc height (Table 3.29). The plantations did have significant influences; this occurred at every disc height except for Disc 8 Bottom. There were significant

differences between all plantations, with Brookton having the highest heartwood percentage (Table 3.30). At Disc 8 Bottom, there were significant differences between Brookton and Kylie Reserve, and Sandawindy and Kylie Reserve (Table 3.30). This indicates that while there were not any significant heartwood percentage changes within the stems for each treatment, there were differences between sites. Differences between the plantations could be due to genetic or environmental influence, or a combination of both. Moderate levels of genetic diversity occur across sandalwood's geographical range (Brand & Norris, 2017; Byrne, MacDonald, Broadhurst, & Brand, 2003) and could be a factor that has influenced the differences in heartwood area percentage.

4.5 Kylie Reserve had quality over quantity

The trees from the Kylie Reserve plantation had the lowest oil yield for total oil, farnesol, and nuciferol (Tables 3.2, 3.7 & 3.9). Studies have determined that sandalwood oil concentrations increase concurrently with age (Brand & Norris, 2017; Brand & Pronk, 2011), however Kylie Reserve was planted only two years after Sandawindy, and in the same year as Brookton (2006). Kylie Reserve produced, on average, the smallest diameter trees, which would have likely contributed to the lower yields of oil. The diameter at 300 mm above ground at Kylie Reserve averaged 59 mm, while Sandawindy had an average of 85 mm and Brookton had an average of 87 mm. Environmental factors, such as climate and soil, and/or genetic differences were more likely to have influenced the oil yield. While the seed from all three plantations was collected from a single source plantation, the source plantation was established with seed from multiple sites. This could cause potential genetic differences between the plantations. Environmental conditions have been identified as effecting essential oil production in plants (Figueiredo, Barroso, Pedro, & Scheffer, 2008).

Oil composition results concluded that the highest quality sandalwood oil was at the Kylie Reserve plantation, achieving the highest % composition of α-santalol and β-santalol (Tables 3.20 & 3.22), as well as the lowest composition of farnesol (Table 3.24). α-santalol and β-santalol are known to give sandalwood oil its distinct scent (Adams, Bhatnagar, & Cookson, 1975; Brand, Norris, & Dumbrell, 2012), and so are required in high amounts. Farnesol has been identified to be an allergen (Moniodis, 2014; Moniodis *et al.*, 2017), and is preferred to be in lower quantities.

Kylie Reserve also had the lowest heartwood area percentages at each disc, and between disc levels (Table 3.30). The study has shown that the presence of a large amount of heartwood

doesn’t necessarily denote higher quality sandalwood oil. Further research will need to be performed analysing the heartwood structure, to determine if oil is evenly spread within the heartwood, or if it centres in the primary heartwood (most inner point of the heartwood) and either lessens or is not present in the secondary heartwood (outer section of the heartwood).

The Kylie Reserve plantation has displayed an inverse relationship between oil yield and quality. This indicates that Kylie Reserve has the most favourable conditions of the three plantations utilised for this study for production of the highest quality oil. Further studies will need to explore this inverse relationship, and the relationship between sandalwood tree stress and oil quality, in addition to the known relationship between stress and oil yield (Page *et al*., 2010; Rai, 1990).

5. Synthesis and Conclusions

The objective in this study was to examine the short-term effects of a chemical treatment, and multiple physical treatments, on the production of sandalwood oil and heartwood. This was done by treating the trees, and evaluating the response in heartwood development, oil production and oil composition after one year. Of the treatments applied, there was none that consistently increased oil production, quality, or heartwood production, when compared to the control group. The Treated Dowel treatment, when compared to the Blank Dowel and Empty Drill treatments, did not significantly increase oil production, quality, or heartwood production. This signifies that the plant hormone MeSA did not increase the oil production, quality, or heartwood production in the treated sandalwood as there was no clear chemical effect above that of the physical effects of drilling and dowel insertion. The physical effect of drilling into the tree generally had the greatest effect on increasing oil production, with the Empty Drill treatment having significantly greater yield and quality compared to the Control group, on the most occasions.

The plantations were shown to have a greater impact on oil production compared to the treatments. The Brookton and Sandawindy plantations produced more oil and more heartwood compared to Kylie Reserve. Kylie Reserve, while producing the least amount of oil, produced the highest quality oil. This evidence indicates that soil, age, genetics, or tree size could have a larger impact on sandalwood oil compared to the tried treatments.

While the treatments have not shown to have a significant or consistent impact on oil or heartwood production in a one year timeframe, the results have shown that there is a potential for the treatments to do this over a longer period of time. This study has provided important information for future studies in oil inducing techniques for WA sandalwood. Future studies could potentially have more positive results and could be used to reduce the time between establishment and harvesting of sandalwood in a plantation setting.

5.1 Study's Limitations

5.1.1 Length of the Study

This project was limited by the ability to only complete one harvest. The long-term effects of the treatments on the oil and heartwood formation in the sandalwood trees was not able to be explored. Sandalwood is a slow growing tree, and oil production normally begins at 10 years of age. Therefore, with the trees between the ages of 10-12 at the time of treatment, a longer period between treatment and harvesting (of several years) may be required to detect full effects

of treatments. The experiments were set up to allow continued monitoring as only one third of the treated trees were harvested.

5.1.2 Budget

Budgetary restraints restricted the number of oil analyses and harvests conducted in this study. The oil analyses were priced at approximately AU$110 per sample. Therefore, only 150 samples were able to be analysed. The budget also restricted the number of harvests conducted, as the cost of resources and personnel for the harvest and subsequent coring and cutting of the sandalwood logs was too great to conduct more harvests.

5.1.3 Number of Plantations

The number of plantations that were used were limited by the amount that were available. Plantations had to meet certain requirements to be utilised for the study; sandalwood had to be between 10-12 years old at the time of treatment, with certain host species and enough trees for the study. The trees also needed not to be involved in any other studies or agreements that would prevent them from being treated or harvested for this study.

5.2 Recommendations

5.2.1 Increase Study Time

For further research, it is recommended that the trees be left for longer after treatment before their harvest. As sandalwood is a slow growing tree, it is important to explore the longer-term effects of the treatments on the sandalwood trees. A longer wait time between treatment and harvesting could have a significant effect of the amount of sandalwood oil and heartwood in the trees. The study should be extended for a further 10 years, with monitoring and sampling occurring every 3-5 years. This will allow the potential long term effects of the treatments used to be examined.

5.2.2 Treat at Different Ages

The age in which the sandalwood trees are treated could be explored. Further studies could examine treating the trees at different ages, to determine a preferred age for treatment. In this study, the trees were treated at an age where they typically start to produce oil (age 10-12). Treating the trees before they begin to produce oil, or later in their production stage, could have impacts on the amount of oil and heartwood produced.

5.2.3 Apply Multiple Treatments

In this study, the sandalwood trees were treated only once before they were harvested. Treating the sandalwood trees multiple times during their life before they are harvested could influence the amount of sandalwood oil or heartwood that is produced. It may be beneficial to also observe the effect of other stressing factors on the oil and heartwood production of sandalwood. This may include artificial droughts or floods, or pathogens or infestations.

5.2.4 Use Other Chemicals

The use of other chemicals apart from MeSA could be further examined. MeSA is a plant hormone but does not occur in sandalwood. Hormones that do naturally occur in sandalwood could be used, to see if they influenced the oil production. As MeSA is not naturally occurring in sandalwood, the hormone may not have imitated a typically stressful situation. Having a hormone that is familiar to the sandalwood tree could influence the amount of heartwood and sandalwood oil found in the tree.

5.2.5 Harvesting Method

As heartwood is found throughout the whole tree, comparing more parts of the tree to each other would give greater insights to the effects of the treatments on oil and heartwood production. Examination of the roots and butt, which is where the greatest amount of heartwood is, could lead to more information about the effect of the treatments. Only a sample of the tree was harvested in this study which may not give an indication to the effect that the treatment has to the sandalwood tree as a whole.

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7. Appendix

7.1 Sandalwood Extraction and Gas Chromatographic Analysis Method

Protocol for sandalwood Extraction

1. Weigh 1000 mg ± 10 mg of the sample in to a 20ml scintillation glass vial with screw cap and record the weight.
2. Transfer 10ml±10µL of internal standard solution precisely (1% camphor in n-Hexane).
3. Close the vial tightly and set on the ultrasonic extraction, 40kHz for 30minutes.
4. Remove the vials and stand to settle and transfer 1ml±10 µL solution from the top of the solution into a GC vial.

Preparation and Calibration of Internal Standard (IS) Solution for Sandalwood Analysis.

1. Dissolve 10g of (-)-camphor (Sigma-Aldrich, USA) in 1000 mL of n-hexane (Ajax UNICHROM, Australia) to make the internal standard (IS) solution.
2. Stock solution of a standard sandalwood oil (AS2112:2003) of 100mg oil in 10 mL (1% w/v) using the above IS solution.
3. Make dilutions by additions using the IS solution as follows; 7.5mL to 10 mL(0.75% w/v), 5 mL to 10mL (0.5%w/v), 2.5mL to 10 mL (0.25%w/v) and 1mL to 10mL (0.1% w/v).
4. Analyse the samples composition in Rtx5_Sandalwood method.
5. Enter the following data to a MS Office Excel© spreadsheet; area of IS, total volatile area and concentration.
6. Plot a X Y scatter (linear graph) for total volatile are/ IS area on Y axis against concentration on x axis.
7. Check the regression of the line of best fit (> 0.98)
8. Calculate the slope of the line and intercept.
9. Use the above equation to calculate the concentration (x) of samples by measuring the Area of volatile and Area of IS.

Gas Chromatography Method

1. Gas Chromatogram (GC2010, Shimadzu Scientific, Japan) equipped with a flame ionisation detector. 95% phenyl siloxane coated capillary column (Rt-5, Restek, USA) was used.
2. 1 µL of this solution was injected into the injector was at 220°C at 110 kPa with 50:1 split ratio.
3. Oven was programmed for 100°C to 140°C at 5°C/ min gradient and hold for 25 min, 140-180°C at 5°C/min and hold for 10 min.

4. Detector was kept at 220°C
5. Data were processed by Labsolutions© software (Shimadzu Scientific, Japan).
6. Compounds were identified and quantified using Kovat's retention indices against alkane series (Subasinghe *et al.* 2013; Subasinghe et al 2016).
7. Compounds were verified using the above column in Thermo Scientific™ ISQ™ series quadrupole GC-MS System. Oven program and injector parameters remained the same, whereas the injector volume was reduced to 0.1 µL and the mass spectra interface was kept at 220^{o}C and signals were measured at m/z in 40 ms intervals using scan mode. Compounds were identified by comparing the mass fragmentation patterns with published data and online database library (NIST-17 Library, NIST, USA).
8. Method limit of detection was 2.4 ng/mL and the limit of quantification was calculated to be 48 ng/mL per each reported compound.

7.2 Descriptive Statistics Tables

Table 7.1: Mean total oil composition and mean oil composition for a-santalol, b-santalol farnesol, nuciferol and b-bisabolol in the oil with standard deviations and 95% confidence intervals in composition at each plantation.

			95% CI	
Oil Component	**Plantation**	**M ± SD**	**LL**	**UL**
Total Oil	Sandawindy	0.84 ± 0.59	0.66	1.012
	Kylie Reserve	0.31 ± 0.17	0.26	0.36
	Brookton	0.64 ± 0.39	0.53	0.76
α-santalol	Sandawindy	0.1 ± 0.12	0.067	0.14
	Kylie Reserve	0.05 ± 0.082	0.027	0.074
	Brookton	0.083 ± 0.075	0.061	0.10
β-santalol	Sandawindy	0.034 ± 0.042	0.022	0.045
	Kylie Reserve	0.012 ± 0.015	0.0075	0.016
	Brookton	0.023 ± 0.026	0.016	0.031
Farnesol	Sandawindy	0.23 ± 0.14	0.19	0.27
	Kylie Reserve	0.047 ± 0.065	0.028	0.065
	Brookton	0.19 ± 0.13	0.16	0.23
Nuciferol	Sandawindy	0.13 ± 0.092	0.10	0.15
	Kylie Reserve	0.027 ± 0.029	0.018	0.035
	Brookton	0.1 ± 0.083	0.077	0.12
β-bisabalol	Sandawindy	0.028 ± 0.023	0.021	0.034
	Kylie Reserve	0.11 ± 0.042	0.096	0.12
	Brookton	0.064 ± 0.042	0.053	0.076

Note: n = 50 for each plantation. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.2: Mean total oil composition (%w/w) and mean composition for all oil components, with standard deviations and 95% confidence intervals for each treatment type.

			95% CI	
		M ± SD	***LL***	***UL***
Total oil	Control	0.68 ± 0.48	0.50	0.86
	Empty Drill	0.66 ± 0.46	0.49	0.83
	Blank Dowel	0.54 ± 0.56	0.33	0.74
	Treated Dowel	0.56 ± 0.38	0.42	0.70
	Bark Removed	0.56 ± 0.49	0.38	0.74
α-santalol	Control	0.08 ± 0.10	0.042	0.12
	Empty Drill	0.10 ± 0.13	0.051	0.15
	Blank Dowel	0.074 ± 0.091	0.040	0.11
	Treated Dowel	0.092 ± 0.078	0.062	0.12
	Bark Removed	0.044 ± 0.066	0.020	0.069
β-santalol	Control	0.027 ± 0.033	0.015	0.040
	Empty Drill	0.033 ± 0.045	0.016	0.050
	Blank Dowel	0.021 ± 0.027	0.010	0.031
	Treated Dowel	0.092 ± 0.078	0.010	0.026
	Bark Removed	0.015 ± 0.022	0.0070	0.023
Farnesol	Control	0.20 ± 0.15	0.15	0.26
	Empty Drill	0.15 ± 0.12	0.11	0.20
	Blank Dowel	0.099 ± 0.096	0.064	0.14
	Treated Dowel	0.16 ± 0.14	0.10	0.21
	Bark Removed	0.17 ± 0.16	0.11	0.23
Nuciferol	Control	0.11 ± 0.10	0.070	0.15
	Empty Drill	0.094 ± 0.096	0.058	0.13
	Blank Dowel	0.052 ± 0.054	0.032	0.072
	Treated Dowel	0.080 ± 0.069	0.055	0.11
	Bark Removed	0.089 ± 0.087	0.057	0.12
β-bisabalol	Control	0.055 ± 0.043	0.039	0.071
	Empty Drill	0.067 ± 0.044	0.050	0.083
	Blank Dowel	0.076 ± 0.061	0.053	0.099
	Treated Dowel	0.064 ± 0.048	0.046	0.082

	Bark Removed	0.072 ± 0.047	0.054	0.089

Note: n = 30 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.3: Mean total oil composition and oil component composition with standard deviations and 95% confidence intervals at the Sandawindy plantation.

			95% CI	
		M ± SD	***LL***	***UL***
α-santalol	Control	0.15 ± 0.13	0.055	0.25
	Empty Drill	0.12 ± 0.18	-0.0016	0.25
	Blank Dowel	0.1 ± 0.11	0.027	0.18
	Treated Dowel	0.084 ± 0.077	0.029	0.14
	Bark Removed	0.045 ± 0.078	-0.011	0.10
β-santalol	Control	0.049 ± 0.043	0.018	0.080
	Empty Drill	0.042 ± 0.063	-0.0037	0.087
	Blank Dowel	0.035 ± 0.038	0.0075	0.062
	Treated Dowel	0.025 ± 0.03	0.0035	0.046
	Bark Removed	0.017 ± 0.028	-0.0025	0.037
Farnesol	Control	0.25 ± 0.12	0.17	0.34
	Empty Drill	0.21 ± 0.054	0.17	0.24
	Blank Dowel	0.16 ± 0.095	0.091	0.23
	Treated Dowel	0.25 ± 0.19	0.11	0.38
	Bark Removed	0.27 ± 0.18	0.15	0.40
Nuciferol	Control	0.11 ± 0.074	0.061	0.17
	Empty Drill	0.16 ± 0.12	0.069	0.25
	Blank Dowel	0.086 ± 0.06	0.043	0.13
	Treated Dowel	0.12 ± 0.086	0.061	0.19
	Bark Removed	0.16 ± 0.099	0.084	0.23
β-bisabalol	Control	0.023 ± 0.013	0.013	0.032

Empty Drill	0.032 ± 0.015	0.021	0.043
Blank Dowel	0.027 ± 0.029	0.0060	0.047
Treated Dowel	0.031 ± 0.023	0.014	0.047
Bark Removed	0.027 ± 0.033	0.0040	0.051

Note: n = 10 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.4: Mean total oil composition and oil component composition with standard deviations and 95% confidence intervals in composition at the Kylie Reserve plantation.

			95% CI	
		M ± SD	***LL***	***UL***
α-santalol	Control	0.043 ± 0.07	-0.0070	0.093
	Empty Drill	0.033 ± 0.057	-0.0078	0.074
	Blank Dowel	0.053 ± 0.1	-0.021	0.13
	Treated Dowel	0.1 ± 0.11	0.022	0.18
	Bark Removed	0.022 ± 0.052	-0.015	0.059
β-santalol	Control	0.017 ± 0.021	0.0016	0.032
	Empty Drill	0.012 ± 0.014	0.0020	0.022
	Blank Dowel	0.0098 ± 0.014	-0.00012	0.020
	Treated Dowel	0.0096 ± 0.009	0.0032	0.016
	Bark Removed	0.011 ± 0.017	-0.0014	0.023
Farnesol	Control	0.069 ± 0.1	-0.0058	0.14
	Empty Drill	0.028 ± 0.027	0.0085	0.048
	Blank Dowel	0.018 ± 0.0068	0.013	0.022
	Treated Dowel	0.058 ± 0.057	0.018	0.099
	Bark Removed	0.061 ± 0.075	0.0079	0.11
Nuciferol	Control	0.035 ± 0.036	0.0088	0.060
	Empty Drill	0.017 ± 0.0095	0.0098	0.023
	Blank Dowel	0.014 ± 0.00041	0.013	0.014
	Treated Dowel	0.032 ± 0.031	0.0096	0.054
	Bark Removed	0.037 ± 0.042	0.0069	0.067
β-bisabalol	Control	0.09 ± 0.043	0.059	0.12
	Empty Drill	0.11 ± 0.0027	0.11	0.12
	Blank Dowel	0.13 ± 0.055	0.093	0.17
	Treated Dowel	0.096 ± 0.052	0.059	0.13
	Bark Removed	0.1 ± 0.029	0.084	0.13

Note: n = 10 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.5: Mean total oil composition and oil component compositions with standard deviations and 95% confidence intervals in composition at the Brookton plantation.

			95% CI	
		M ± SD	***LL***	***UL***
α-santalol	Control	0.049 ± 0.059	0.0060	0.091
	Empty Drill	0.14 ± 0.11	0.063	0.22
	Blank Dowel	0.066 ± 0.057	0.025	0.11
	Treated Dowel	0.091 ± 0.044	0.060	0.12
	Bark Removed	0.065 ± 0.064	0.019	0.11
β-santalol	Control	0.016 ± 0.021	0.0014	0.031
	Empty Drill	0.045 ± 0.039	0.017	0.073
	Blank Dowel	0.017 ± 0.019	0.0037	0.031
	Treated Dowel	0.02 ± 0.015	0.0086	0.031
	Bark Removed	0.017 ± 0.02	0.0026	0.031
Farnesol	Control	0.29 ± 0.11	0.21	0.37
	Empty Drill	0.22 ± 0.14	0.12	0.31
	Blank Dowel	0.12 ± 0.093	0.055	0.19
	Treated Dowel	0.16 ± 0.1	0.093	0.24
	Bark Removed	0.18 ± 0.14	0.078	0.28
Nuciferol	Control	0.17 ± 0.12	0.087	0.26
	Empty Drill	0.11 ± 0.054	0.070	0.15
	Blank Dowel	0.056 ± 0.054	0.018	0.095
	Treated Dowel	0.086 ± 0.046	0.053	0.12
	Bark Removed	0.075 ± 0.07	0.026	0.13
β-bisabalol	Control	0.051 ± 0.036	0.025	0.077
	Empty Drill	0.054 ± 0.043	0.023	0.084
	Blank Dowel	0.068 ± 0.044	0.036	0.10
	Treated Dowel	0.066 ± 0.043	0.035	0.097
	Bark Removed	0.084 ± 0.041	0.054	0.11

Note: n = 10 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.6: Mean composition percentage of oil component with standard deviations and 95% confidence intervals at each plantation.

			95% CI	
Plantation	***Oil Component***	**M ± SD**	***LL***	***UL***
Sandawindy	α-santalol	13.40 ± 11.18	9.83	16.98
	β-santalol	5.81 ± 3.81	4.24	7.38
	Farnesol	29.74 ± 11.73	26.26	33.22
	Nuciferol	17.39 ± 11.16	14.076	20.70
	β-bisabalol	4.49 ± 3.96	3.27	5.71
Kylie Reserve	α-santalol	25.42 ± 13.24	17.0087	33.83
	β-santalol	8.12 ± 3.84	5.16	11.072
	Farnesol	27.67 ± 14.75	19.16	36.19
	Nuciferol	17.55 ± 11.66	9.21	25.89
	β-bisabalol	12.60 ± 15.084	1.0007	24.19
Brookton	α-santalol	16.22 ± 11.79	12.50	19.94
	β-santalol	6.045 ± 3.62	4.48	7.61
	Farnesol	34.21 ± 15.034	29.64	38.79
	Nuciferol	17.23 ± 8.56	14.57	19.90
	β-bisabalol	5.66 ± 4.54	4.025	7.30

Note: n = 50 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.7: Mean percentage composition of total oil and oil component with standard deviations and 95% confidence intervals for each treatment type.

			95% CI	
Treatment	***Oil Component***	***M ± SD***	***LL***	***UL***
Control	α-santalol	12.76 ± 9.23	8.31	17.21
	β-santalol	5.52 ± 2.62	3.94	7.11
	Farnesol	32.79 ± 15.48	25.93	39.65
	Nuciferol	16.46 ± 7.98	12.93	20.0023
	β-bisabalol	4.12 ± 3.045	2.70	5.55
Empty Drill	α-santalol	17.50 ± 13.62	11.31	23.70
	β-santalol	7.31 ± 4.15	4.92	9.71
	Farnesol	27.21 ± 11.19	22.12	32.31
	Nuciferol	17.38 ± 10.23	12.59	22.17
	β-bisabalol	4.35 ± 2.44	3.14	5.56
Blank Dowel	α-santalol	19.31 ± 14.52	11.57	27.043
	β-santalol	7.35 ± 4.55	4.088	10.60
	Farnesol	26.51 ± 13.15	19.74	33.27
	Nuciferol	13.17 ± 6.96	9.31	17.022
	β-bisabalol	6.21 ± 6.37	2.53	9.88
Treated Dowel	α-santalol	16.49 ± 9.32	12.55	20.42
	β-santalol	5.43 ± 2.093	4.097	6.76
	Farnesol	30.77 ± 12.34	25.56	35.98
	Nuciferol	16.92 ± 8.64	13.090	20.75
	β-bisabalol	9.82 ± 10.90	4.71	14.92
Bark Removed	α-santalol	14.72 ± 15.42	5.40	24.045
	β-santalol	5.57 ± 5.39	1.058	10.073
	Farnesol	38.95 ± 13.83	32.47	45.42
	Nuciferol	22.089 ± 14.036	15.32	28.85
	β-bisabalol	3.63 ± 2.50	2.038	5.22

Note: n = 30 for each cell. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.8: Mean percentage of heartwood area at each disc height with standard deviations and 95% confidence intervals at each plantation.

			95% CI	
Disc	**Plantation**	**M ± SD**	**LL**	**UL**
1	Sandawindy[a]	13.77 ± 13.97	11.055	16.49
	Kylie Reserve[b]	1.76 ± 2.95	1.20	2.33
	Brookton[b]	21.50 ± 17.33	18.17	24.82
2	Sandawindy[a]	14.00 ± 14.74	11.13	16.87
	Kylie Reserve[b]	1.90 ± 3.20	1.29	2.51
	Brookton[b]	22.028 ± 17.62	18.65	25.40
3	Sandawindy[a]	14.17 ± 13.63	11.52	16.83
	Kylie Reserve[b]	2.42 ± 4.15	1.63	3.22
	Brookton[b]	21.56 ± 17.019	18.30	24.82
4	Sandawindy[a]	15.30 ± 13.82	12.61	17.99
	Kylie Reserve[b]	2.45 ± 3.74	1.73	3.17
	Brookton[b]	21.59 ± 16.91	18.35	24.83
5	Sandawindy[a]	15.087 ± 13.50	12.46	17.71
	Kylie Reserve[b]	3.66 ± 5.025	2.70	4.62
	Brookton[b]	21.82 ± 17.10	18.54	25.10
6	Sandawindy[a]	15.20 ± 13.53	12.57	17.83
	Kylie Reserve[b]	3.036 ± 5.020	2.074	4.00
	Brookton[b]	20.64 ± 15.88	17.60	23.69
7	Sandawindy[a]	15.62 ± 13.75	12.94	18.29
	Kylie Reserve[b]	2.98 ± 5.15	1.99	3.97
	Brookton[b]	21.34 ± 15.94	18.28	24.39
8 Top	Sandawindy[a]	16.56 ± 14.55	13.73	19.39
	Kylie Reserve[b]	3.48 ± 5.040	2.52	4.45
	Brookton[b]	21.11 ± 16.42	17.96	24.25
8 Bottom	Sandawindy[a]	17.097 ± 16.080	13.97	20.22
	Kylie Reserve[b]	3.51 ± 4.72	2.60	4.41
	Brookton[b]	20.57 ± 15.89	17.52	23.61

Note: a: n = 104, b: n = 107. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.9: Mean percentage of heartwood area at each disc height with standard deviations and 95% confidence intervals for each treatment type.

			95% CI	
Disc	***Treatment***	***M ± SD***	***LL***	***UL***
1	Control[a]	13.040 ± 15.60	9.14	16.94
	Empty Drill[b]	14.14 ± 15.74	10.069	18.20
	Blank Dowel[c]	11.34 ± 15.30	7.55	15.13
	Treated Dowel[a]	11.80 ± 15.26	7.98	15.61
	Bark Removed[c]	11.49 ± 14.83	7.81	15.16
2	Control[a]	12.67 ± 15.23	8.86	16.47
	Empty Drill[b]	14.32 ± 16.36	10.088	18.54
	Blank Dowel[c]	11.15 ± 15.27	7.36	14.93
	Treated Dowel[a]	12.58 ± 15.76	8.64	16.52
	Bark Removed[c]	12.57 ± 16.32	8.52	16.61
3	Control[a]	12.55 ± 14.73	8.87	16.23
	Empty Drill[b]	14.88 ± 16.58	10.59	19.16
	Blank Dowel[c]	11.69 ± 15.61	7.83	15.56
	Treated Dowel[a]	12.51 ± 14.76	8.82	16.19
	Bark Removed[c]	12.058 ± 13.69	8.67	15.45
4	Control[a]	12.71 ± 14.53	9.078	16.34
	Empty Drill[b]	15.74 ± 17.061	11.33	20.14
	Blank Dowel[c]	11.65 ± 15.49	7.81	15.49
	Treated Dowel[a]	13.55 ± 14.16	10.019	17.090
	Bark Removed[c]	12.016 ± 14.082	8.53	15.50
5	Control[a]	13.59 ± 14.96	9.85	17.33
	Empty Drill[b]	16.81 ± 16.28	12.61	21.018
	Blank Dowel[c]	12.60 ± 15.14	8.85	16.36
	Treated Dowel[a]	13.37 ± 14.15	9.84	16.91
	Bark Removed[c]	11.42 ± 13.95	7.96	14.87
6	Control[a]	13.55 ± 14.57	9.91	17.19
	Empty Drill[b]	15.11 ± 15.57	11.088	19.13
	Blank Dowel[c]	11.68 ± 14.63	8.055	15.30
	Treated Dowel[a]	13.20 ± 14.35	9.62	16.79

	Bark Removed[c]	11.33 ± 12.92	8.12	14.53
7	Control[a]	14.42 ± 15.31	10.59	18.24
	Empty Drill[b]	15.27 ± 15.64	11.23	19.31
	Blank Dowel[c]	11.62 ± 14.92	7.92	15.31
	Treated Dowel[a]	13.31 ± 13.67	9.90	16.73
	Bark Removed[c]	12.0026 ± 13.83	8.58	15.43
8 Top	Control[a]	14.17 ± 15.075	10.40	17.93
	Empty Drill[b]	16.41 ± 16.49	12.15	20.67
	Blank Dowel[c]	11.92 ± 15.58	8.058	15.78
	Treated Dowel[a]	13.091 ± 12.78	9.90	16.28
	Bark Removed[c]	13.077 ± 14.81	9.41	16.75
8 Bottom	Control[a]	14.48 ± 14.75	10.79	18.16
	Empty Drill[b]	16.14 ± 16.10	11.98	20.30
	Blank Dowel[c]	11.60 ± 14.55	7.99	15.20
	Treated Dowel[a]	13.71 ± 15.96	9.72	17.70
	Bark Removed[c]	12.73 ± 14.66	9.098	16.37

Note: a: n = 64, b: n = 60, c: n = 65. M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

Table 7.10: Mean percentage of heartwood area with standard deviations and 95% confidence intervals for each disc height, plantation, and treatment type.

				95% CI	
		n	***M ± SD***	***LL***	***UL***
Disc	1	318	12.33 ± 15.28	10.64	14.017
	2	318	12.63 ± 15.72	10.90	14.36
	3	318	12.71 ± 15.028	11.047	14.36
	4	318	13.092 ± 15.050	11.43	14.75
	5	318	13.51 ± 14.915	11.86	15.15
	6	318	12.94 ± 14.39	11.35	14.53
	7	318	13.29 ± 14.65	11.67	14.91
	8 Top	318	13.69 ± 14.96	12.040	15.34
	8 Bottom	318	13.69 ± 15.19	12.016	15.37
Plantation	Sandawindy	104	15.20 ± 14.18	14.29	16.11
	Kylie Reserve	107	2.80 ± 4.44	2.52	3.081
	Brookton	107	21.35 ± 16.63	20.30	22.40
Treatment	Control	64	13.46 ± 14.89	12.24	14.68
	Empty Drill	60	15.42 ± 16.11	14.060	16.79
	Blank Dowel	65	11.69 ± 15.070	10.47	12.92
	Treated Dowel	64	13.015 ± 14.48	11.83	14.20
	Bark Removed	65	12.076 ± 14.29	10.92	13.24

Note: n = number of samples, M = Mean, SD = Standard deviation, CI = Confidence interval, LL = Lower limit, UL = Upper limit.

7.3 Normality Tests

Table 7.11: Shapiro-Wilk tests of normality for all oil compositions, with standardised residuals.

						SR	
				Statistic	**P**	**LL**	**UL**
Total Oil	Raw Data	*Plantation*[a]	**Sandawindy**	**0.815**	**< 0.001**	-1.73	**5.79**
			Kylie Reserve	**0.738**	**< 0.001**		
			Brookton	**0.827**	**< 0.001**		
		Treatment[b]	**Control**	**0.926**	**0.038**		
			Empty Drill	**0.886**	**0.004**		
			Blank Dowel	**0.486**	**< 0.001**		
			Treated Dowel	**0.687**	**< 0.001**		
			Bark Removed	**0.675**	**< 0.001**		
	Log Transformed	*Plantation*[a]	Sandawindy	0.972	0.283	-3.06	3.05
			Kylie Reserve	**0.845**	**< 0.001**		
			Brookton	**0.940**	**0.014**		
		Treatment[b]	**Control**	**0.879**	**0.003**		
			Empty Drill	0.962	0.356		
			Blank Dowel	**0.871**	**0.002**		
			Treated Dowel	**0.863**	**0.001**		
			Bark Removed	**0.901**	**0.009**		
α-santalol	Raw data	*Plantation*[a]	**Sandawindy**	**0.778**	**< 0.001**	-1.59	**4.71**
			Kylie Reserve	**0.607**	**< 0.001**		
			Brookton	**0.874**	**< 0.001**		
		Treatment[b]	**Control**	**0.754**	**< 0.001**		

			Empty Drill	**0.754**	**< 0.001**		
			Blank Dowel	**0.775**	**< 0.001**		
			Treated Dowel	**0.829**	**< 0.001**		
			Bark Removed	**0.65**	**< 0.001**		
	Log Transformed	*Plantation*[a]	**Sandawindy**	**0.933**	**0.007**	-1.89	2.39
			Kylie reserve	**0.649**	**< 0.001**		
			Brookton	**0.912**	**0.001**		
		Treatment[b]	**Control**	**0.863**	**0.001**		
			Empty drill	**0.899**	**0.008**		
			Blank dowel	**0.822**	**< 0.001**		
			Treated dowel	**0.883**	**0.003**		
			Bark removed	**0.753**	**< 0.001**		
β-santalol	Raw data	*Plantation*[a]	**Sandawindy**	**0.725**	**< 0.001**	-1.72	**5.35**
			Kylie Reserve	**0.494**	**< 0.001**		
			Brookton	**0.734**	**< 0.001**		
		Treatment[b]	**Control**	**0.717**	**< 0.001**		
			Empty Drill	**0.684**	**< 0.001**		
			Blank Dowel	**0.646**	**< 0.001**		
			Treated Dowel	**0.669**	**< 0.001**		
			Bark Removed	**0.530**	**< 0.001**		
	Log Transformed	*Plantation*[a]	**Sandawindy**	**0.808**	**< 0.001**	-1.79	2.61
			Kylie Reserve	**0.530**	**< 0.001**		
			Brookton	**0.783**	**< 0.001**		
		Treatment[b]	**Control**	**0.755**	**< 0.001**		

			Empty Drill	**0.790**	**< 0.001**		
			Blank Dowel	**0.697**	**< 0.001**		
			Treated Dowel	**0.737**	**< 0.001**		
			Bark Removed	**0.618**	**< 0.001**		
Farnesol	Raw data	*Plantation*[a]	**Sandawindy**	**0.935**	**0.009**	-2.31	3.68
			Kylie Reserve	**0.569**	**< 0.001**		
			Brookton	0.955	0.056		
		Treatment[b]	Control	0.937	0.074		
			Empty Drill	**0.904**	**0.010**		
			Blank Dowel	**0.830**	**< 0.001**		
			Treated Dowel	**0.840**	**< 0.001**		
			Bark Removed	**0.876**	**0.002**		
Nuciferol	Raw data	*Plantation*[a]	**Sandawindy**	**0.918**	**0.002**	-2.00	3.95
			Kylie Reserve	**0.532**	**< 0.001**		
			Brookton	**0.845**	**< 0.001**		
		Treatment[b]	**Control**	**0.847**	**0.001**		
			Empty Drill	**0.801**	**< 0.001**		
			Blank Dowel	**0.742**	**< 0.001**		
			Treated Dowel	**0.862**	**0.001**		
			Bark Removed	**0.826**	**< 0.001**		
β-bisabalol	Raw data	*Plantation*[a]	**Sandawindy**	**0.819**	**< 0.001**	-2.16	**4.25**
			Kylie Reserve	**0.639**	**< 0.001**		
			Brookton	**0.797**	**< 0.001**		
		Treatment[b]	**Control**	**0.798**	**< 0.001**		

		Empty Drill	**0.781**	**< 0.001**		
		Blank Dowel	**0.801**	**< 0.001**		
		Treated Dowel	**0.871**	**0.002**		
		Bark Removed	**0.760**	**< 0.001**		
Square root transformed	*Plantation*[a]	**Sandawindy**	**0.928**	**0.005**	-2.20	2.62
		Kylie Reserve	**0.651**	**< 0.001**		
		Brookton	**0.836**	**< 0.001**		
	Treatment[b]	**Control**	**0.885**	**0.004**		
		Empty Drill	**0.820**	**< 0.001**		
		Blank Dowel	**0.893**	**0.006**		
		Treated Dowel	**0.927**	**0.041**		
		Bark Removed	**0.793**	**< 0.001**		

Note: a: n = 50, b: n = 30. P = significance. SR = Standardised residuals. LL = lower limit, UL = upper limit.

Table 7.12: Shaprio-Wilk test of normality for all oil compositions, with standardised residuals.

					SR	
			Statistic	**P**	**LL**	**UL**
α-santalol	Raw data	*Plantation*[a]	**Sandawindy**	**0.869**	**< 0.001**	-1.44	3.51
			Kylie Reserve	0.959	0.769		
			Brookton	**0.904**	**0.002**		
		Treatment[b]	Control	0.920	0.114		
			Empty Drill	**0.902**	**0.039**		
			Blank Dowel	0.898	0.075		
			Treated Dowel	0.942	0.184		
			Bark Removed	**0.776**	**0.004**		
β-santalol	Raw Data	*Plantation*[a]	**Sandawindy**	**0.915**	**0.039**	-1.75	2.49
			Kylie Reserve	0.976	0.942		
			Brookton	0.940	0.183		
		Treatment[b]	Control	0.977	0.963		
			Empty Drill	0.940	0.421		
			Blank Dowel	0.882	0.136		
			Treated Dowel	0.961	0.795		
			Bark Removed	**0.816**	**0.042**		
Farnesol	Raw data	*Plantation*[a]	Sandawindy	0.976	0.461	-1.90	2.51
			Kylie Reserve	0.956	0.655		
			Brookton	**0.930**	**0.010**		
		Treatment[b]	**Control**	**0.899**	**0.028**		
			Empty Drill	0.927	0.122		

			Blank Dowel	0.931	0.228		
			Treated Dowel	0.983	0.939		
			Bark Removed	0.971	0.772		
Nuciferol	Raw data	*Plantation*[a]	**Sandawindy**	**0.877**	**< 0.001**	-1.64	3.12
			Kylie Reserve	**0.807**	**0.018**		
			Brookton	**0.899**	**0.001**		
		Treatment[b]	Control	0.943	0.225		
			Empty Drill	0.916	0.084		
			Blank Dowel	**0.881**	**0.048**		
			Treated Dowel	**0.773**	**< 0.001**		
			Bark Removed	**0.848**	**0.006**		
β-bisabalol	Raw data	*Plantation*[a]	**Sandawindy**	**0.811**	**< 0.001**	-2.53	**4.17**
			Kylie Reserve	**0.698**	**0.001**		
			Brookton	**0.779**	**< 0.001**		
		Treatment[b]	**Control**	**0.860**	**0.008**		
			Empty Drill	0.948	0.388		
			Blank Dowel	**0.753**	**0.001**		
			Treated Dowel	**0.732**	**< 0.001**		
			Bark Removed	**0.836**	**0.024**		
	Log transformed	*Plantation*[a]	Sandawindy	0.971	0.337	-1.96	1.83
			Kylie Reserve	0.895	0.227		
			Brookton	0.976	0.666		
		Treatment[b]	Control	0.962	0.592		
			Empty Drill	**0.885**	**0.032**		

	Blank Dowel	0.985	0.993
	Treated Dowel	0.975	0.861
	Bark Removed	0.939	0.486

Note: a: n = 50, b: n = 30. P = significance. SR = Standardised residuals. LL = lower limit, UL = upper limit.

Table 7.13: Shapiro-Wilk test of normality for all heartwood area measurement, with standardised residuals.

							SR	
				Statistic	**Df**	**P**	**LL**	**UL**
Disc 1	Raw data	*Plantation*	**Sandawindy**	**0.864**	**104**	**< 0.001**	-2.60	3.24
			Kylie Reserve	**0.660**	**107**	**< 0.001**		
			Brookton	**0.931**	**107**	**< 0.001**		
		Treatment	**Control**	**0.809**	**64**	**< 0.001**		
			Empty Drill	**0.834**	**60**	**< 0.001**		
			Blank Dowel	**0.755**	**65**	**< 0.001**		
			Treated Dowel	**0.776**	**64**	**< 0.001**		
			Bark Removed	**0.781**	**65**	**< 0.001**		
Disc 2	Raw data	*Plantation*	**Sandawindy**	**0.847**	**104**	**< 0.001**	-2.45	3.29
			Kylie Reserve	**0.660**	**107**	**< 0.001**		
			Brookton	**0.919**	**107**	**< 0.001**		
		Treatment	**Control**	**0.806**	**64**	**< 0.001**		
			Empty Drill	**0.823**	**60**	**< 0.001**		
			Blank Dowel	**0.755**	**65**	**< 0.001**		
			Treated Dowel	**0.786**	**64**	**< 0.001**		
			Bark Removed	**0.765**	**65**	**< 0.001**		
Disc 3	Raw data	*Plantation*	**Sandawindy**	**0.880**	**104**	**< 0.001**	-2.60	3.28
			Kylie Reserve	**0.644**	**107**	**< 0.001**		
			Brookton	**0.928**	**107**	**< 0.001**		
		Treatment	**Control**	**0.810**	**64**	**< 0.001**		
			Empty Drill	**0.836**	**60**	**< 0.001**		

			Blank Dowel	**0.768**	**65**	**< 0.001**		
			Treated Dowel	**0.784**	**64**	**< 0.001**		
			Bark Removed	**0.826**	**65**	**< 0.001**		
Disc 4	Raw data	*Plantation*	**Sandawindy**	**0.900**	**104**	**< 0.001**	-2.53	3.47
			Kylie Reserve	**0.699**	**107**	**< 0.001**		
			Brookton	**0.935**	**107**	**< 0.001**		
		Treatment	**Control**	**0.823**	**64**	**< 0.001**		
			Empty Drill	**0.844**	**60**	**< 0.001**		
			Blank Dowel	**0.768**	**65**	**< 0.001**		
			Treated Dowel	**0.835**	**64**	**< 0.001**		
			Bark Removed	**0.819**	**65**	**< 0.001**		
Disc 5	Raw data	*Plantation*	**Sandawindy**	**0.906**	**104**	**< 0.001**	-2.41	3.53
			Kylie Reserve	**0.754**	**107**	**< 0.001**		
			Brookton	**0.939**	**107**	**< 0.001**		
		Treatment	**Control**	**0.840**	**64**	**< 0.001**		
			Empty Drill	**0.874**	**60**	**< 0.001**		
			Blank Dowel	**0.814**	**65**	**< 0.001**		
			Treated Dowel	**0.825**	**64**	**< 0.001**		
			Bark Removed	**0.805**	**65**	**< 0.001**		
Disc 6	Raw data	*Plantation*	**Sandawindy**	**0.905**	**104**	**< 0.001**	-2.50	3.25
			Kylie Reserve	**0.649**	**107**	**< 0.001**		
			Brookton	**0.941**	**107**	**< 0.001**		
		Treatment	**Control**	**0.845**	**64**	**< 0.001**		
			Empty Drill	**0.868**	**60**	**< 0.001**		

			Blank Dowel	**0.798**	**65**	**< 0.001**		
			Treated Dowel	**0.833**	**64**	**< 0.001**		
			Bark Removed	**0.827**	**65**	**< 0.001**		
Disc 7	Raw data	*Plantation*	**Sandawindy**	**0.908**	**104**	**< 0.001**	-2.53	3.20
			Kylie Reserve	**0.617**	**107**	**< 0.001**		
			Brookton	**0.942**	**107**	**< 0.001**		
		Treatment	**Control**	**0.846**	**64**	**< 0.001**		
			Empty Drill	**0.868**	**60**	**< 0.001**		
			Blank Dowel	**0.781**	**65**	**< 0.001**		
			Treated Dowel	**0.841**	**64**	**< 0.001**		
			Bark Removed	**0.811**	**65**	**< 0.001**		
Disc 8 Top	Raw data	*Plantation*	**Sandawindy**	**0.905**	**104**	**< 0.001**	-2.48	3.31
			Kylie Reserve	**0.722**	**107**	**< 0.001**		
			Brookton	**0.937**	**107**	**< 0.001**		
		Treatment	**Control**	**0.845**	**64**	**< 0.001**		
			Empty Drill	**0.876**	**60**	**< 0.001**		
			Blank Dowel	**0.775**	**65**	**< 0.001**		
			Treated Dowel	**0.865**	**64**	**< 0.001**		
			Bark Removed	**0.819**	**65**	**< 0.001**		
Disc 8 Bottom	Raw data	*Plantation*	**Sandawindy**	**0.884**	**104**	**< 0.001**	-2.28	**5.49**
			Kylie Reserve	**0.759**	**107**	**< 0.001**		
			Brookton	**0.934**	**107**	**< 0.001**		
		Treatment	**Control**	**0.867**	**64**	**< 0.001**		
			Empty Drill	**0.879**	**60**	**< 0.001**		

		Blank Dowel	**0.789**	**65**	**< 0.001**		
		Treated Dowel	**0.789**	**64**	**< 0.001**		
		Bark Removed	**0.817**	**65**	**< 0.001**		
Square Root Transformed	*Plantation*	**Sandawindy**	**0.970**	**104**	**0.019**	-2.58	3.45
		Kylie Reserve	**0.900**	**107**	**< 0.001**		
		Brookton	**0.942**	**107**	**< 0.001**		
	Treatment	**Control**	**0.950**	**64**	**0.012**		
		Empty Drill	**0.953**	**60**	**0.021**		
		Blank Dowel	**0.911**	**65**	**< 0.001**		
		Treated Dowel	**0.947**	**64**	**0.008**		
		Bark Removed	**0.915**	**65**	**< 0.001**		

Note: P = significance. SR = Standardised residuals. LL = lower limit, UL = upper limit.

Table 7.14: Shapiro-Wilk test of normality for heartwood area between discs, with standardised residuals.

						SR	
			Statistic	**Df**	**P**	**LL**	**UL**
Raw data	Disc	***1***	**0.798**	**318**	**< 0.001**	-2.57	**6.00**
		2	**0.791**	**318**	**< 0.001**		
		3	**0.815**	**318**	**< 0.001**		
		4	**0.826**	**318**	**< 0.001**		
		5	**0.843**	**318**	**< 0.001**		
		6	**0.840**	**318**	**< 0.001**		
		7	**0.840**	**318**	**< 0.001**		
		8T	**0.845**	**318**	**< 0.001**		
		8B	**0.839**	**318**	**< 0.001**		
	Plantation	***Sandawindy***	**0.894**	**936**	**< 0.001**		
		Kylie Reserve	**0.681**	**963**	**< 0.001**		
		Brookton	**0.939**	**963**	**< 0.001**		
	Treatment	***Control***	**0.838**	**576**	**< 0.001**		
		Empty Drill	**0.861**	**540**	**< 0.001**		
		Blank Dowel	**0.782**	**585**	**< 0.001**		
		Treated Dowel	**0.826**	**576**	**< 0.001**		
		Bark Removed	**0.813**	**585**	**< 0.001**		
Square root transformed	Disc	***1***	**0.913**	**318**	**< 0.001**	-2.91	3.66
		2	**0.910**	**318**	**< 0.001**		
		3	**0.923**	**318**	**< 0.001**		
		4	**0.932**	**318**	**< 0.001**		

	5	**0.946**	**318**	**< 0.001**
	6	**0.936**	**318**	**< 0.001**
	7	**0.934**	**318**	**< 0.001**
	8T	**0.941**	**318**	**< 0.001**
	8B	**0.944**	**318**	**< 0.001**
Plantation	***Sandawindy***	**0.970**	**936**	**< 0.001**
	Kylie Reserve	**0.869**	**963**	**< 0.001**
	Brookton	**0.946**	**963**	**< 0.001**
Treatment	***Control***	**0.936**	**576**	**< 0.001**
	Empty Drill	**0.952**	**540**	**< 0.001**
	Blank Dowel	**0.905**	**585**	**< 0.001**
	Treated Dowel	**0.933**	**576**	**< 0.001**
	Bark Removed	**0.915**	**585**	**< 0.001**

Note: P = significance. SR = Standardised residuals. LL = lower limit, UL = upper limit.

Table 7.15: Shapiro-Wilk test of normality for oil composition within plantations, with standardised residuals.

					SR	
			Statistic	***P***	***LL***	***UL***
Sandawindy	*α-santalol*	Control	0.916	0.328	-1.26	3.79
		Empty Drill	**0.68**	**0.001**		
		Blank Dowel	0.846	0.052		
		Treated Dowel	**0.804**	**0.016**		
		Bark Removed	**0.588**	**< 0.001**		
	β-santalol	Control	0.897	0.205	-1.22	3.86
		Empty Drill	**0.66**	**< 0.001**		
		Blank Dowel	**0.80**	**0.014**		
		Treated Dowel	**0.733**	**0.002**		
		Bark Removed	**0.523**	**< 0.001**		
	Farnesol	Control	0.947	0.635	-1.89	2.78
		Empty Drill	0.969	0.877		
		Blank Dowel	0.963	0.819		
		Treated Dowel	0.910	0.281		
		Bark Removed	0.956	0.741		
	Nuciferol	Control	0.951	0.680	-1.56	2.40
		Empty Drill	**0.800**	**0.015**		
		Blank Dowel	0.917	0.331		
		Treated Dowel	0.904	0.244		
		Bark Removed	0.940	0.554		
	β-bisabalol	Control	0.973	0.920	-1.29	3.68
		Empty Drill	0.940	0.553		
		Blank Dowel	**0.702**	**0.001**		
		Treated Dowel	**0.822**	**0.027**		
		Bark Removed	**0.677**	**< 0.001**		
Kylie Reserve	*α-santalol*	**Control**	**0.625**	**< 0.001**	-1.45	3.00
		Empty Drill	**0.531**	**< 0.001**		
		Blank Dowel	**0.54**	**< 0.001**		
		Treated Dowel	**0.774**	**0.007**		

		Bark Removed	**0.368**	**< 0.001**		
	β-santalol	**Control**	**0.624**	**< 0.001**	-1.71	2.86
		Empty Drill	**0.546**	**< 0.001**		
		Blank Dowel	**0.376**	**< 0.001**		
		Treated Dowel	**0.553**	**< 0.001**		
		Bark Removed	**0.373**	**< 0.001**		
	Farnesol Raw data	**Control**	**0.614**	**< 0.001**	-0.85	**4.07**
		Empty Drill	**0.550**	**< 0.001**		
		Blank Dowel	**0.426**	**< 0.001**		
		Treated Dowel	**0.763**	**0.005**		
		Bark Removed	**0.638**	**< 0.001**		
	Farnesol Log transformed	**Control**	**0.676**	**< 0.001**	-0.98	2.51
		Empty Drill	**0.559**	**< 0.001**		
		Blank Dowel	**0.462**	**< 0.001**		
		Treated Dowel	**0.787**	**0.010**		
		Bark Removed	**0.627**	**< 0.001**		
	Nuciferol	**Control**	**0.664**	**< 0.001**	-0.83	3.42
		Empty Drill	**0.397**	**< 0.001**		
		Blank Dowel	0.947	0.631		
		Treated Dowel	**0.671**	**< 0.001**		
		Bark Removed	**0.657**	**< 0.001**		
	β-bisabalol	**Control**	**0.694**	**0.001**	-1.97	3.80
		Empty Drill	0.913	0.306		
		Blank Dowel	**0.424**	**< 0.001**		
		Treated Dowel	0.872	0.105		
		Bark Removed	**0.458**	**< 0.001**		
Brookton	*α-santalol*	**Control**	**0.688**	**0.001**	-1.75	2.55
		Empty Drill	0.926	0.408		
		Blank Dowel	0.865	0.087		
		Treated Dowel	0.919	0.350		
		Bark Removed	**0.80**	**0.015**		
	β-santalol	**Control**	**0.595**	**< 0.001**	-1.60	2.43
		Empty Drill	0.861	0.078		

	Blank Dowel	**0.699**	**0.001**		
	Treated Dowel	**0.759**	**0.005**		
	Bark Removed	**0.636**	**< 0.001**		
Farnesol	Control	0.943	0.589	-1.73	2.42
	Empty Drill	0.937	0.522		
	Blank Dowel	0.926	0.407		
	Treated Dowel	0.868	0.094		
	Bark Removed	0.934	0.486		
Nuciferol	Control	0.895	0.192	-1.72	3.66
	Empty Drill	0.966	0.856		
	Blank Dowel	**0.815**	**0.022**		
	Treated Dowel	0.877	0.121		
	Bark Removed	**0.823**	**0.028**		
β-bisabalol	**Control**	**0.791**	**0.011**	-1.63	1.63
	Empty Drill	**0.753**	**0.004**		
	Blank Dowel	**0.826**	**0.030**		
	Treated Dowel	**0.803**	**0.016**		
	Bark Removed	**0.758**	**0.004**		

Note: Df = 10 for all. P = Significance. SR = Standardised residuals. LL = Lower Limit, UL = Upper Limit.

7.4 Within plantations without covariates

Table 7.16: Univariate GLM for oil component yield at the Sandawindy plantation, without diameter measurement as a covariate.

		Type III SS	***MS***	***F***	***P***
α-santalol	*Treatment*	0.066	0.016	1.13	0.353
β-santalol	*Treatment*	0.006	0.002	0.90	0.473
farnesol	*Treatment*	0.084	0.021	1.12	0.357
nuciferol	*Treatment*	0.036	0.009	1.085	0.375
β-bisabalol	*Treatment*	0.001	< 0.001	0.24	0.916

Note: SS = Sum of squares, MS = Mean square, F = F statistic, P = Significance. df = 4.

Table 7.17: Univariate GLM for oil component yield at the Kylie Reserve plantation, without diameter measurement as a covariate.

		Type III SS	***MS***	***F***	***P***
α-santalol	*Treatment*	0.036	0.009	1.36	0.262
β-santalol	*Treatment*	0.000	$8.174e^{-5}$	0.34	0.848
farnesol	*Treatment*	4.20	1.051	1.21	0.320
nuciferol	*Treatment*	0.036	0.009	1.085	0.375
β-bisabalol	*Treatment*	0.001	0.000	0.24	0.916

Note: SS = Sum of squares, MS = Mean square, F = F statistic, P = Significance. df = 4.

Table 7.18: Univariate GLM for oil component yield at the Brookton plantation, without diameter measurement as a covariate.

		Type III SS	***MS***	***F***	***P***
α-santalol	***Treatment***	**0.053**	**0.013**	**2.68**	**0.043**
β-santalol	***Treatment***	**0.006**	**0.002**	**2.62**	**0.047**
farnesol	***Treatment***	**0.16**	**0.040**	**2.95**	**0.030**
nuciferol	***Treatment***	**0.083**	**0.021**	**3.74**	**0.010**
β-bisabalol	*Treatment*	1.95	0.49	0.88	0.485

Note: SS = Sum of squares, MS = Mean square, F = F statistic, P = Significance. df = 4.

Bold denotes significant differences.

Table 7.19: Simple contrast between treatments for α-santalol yield in Brookton plantation, without diameter as a covariate.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	**Empty Drill**	**0.032**	**0.005**	**0.030**	**0.157**
	Blank Dowel	0.032	0.580	-0.046	0.081
	Treated Dowel	0.032	0.184	-0.021	0.106
	Bark Removed	**0.032**	**0.032**	**-0.047**	**0.080**
Empty Drill	**Control**	**0.032**	**0.005**	**-0.157**	**-0.030**
	Blank Dowel	**0.032**	**0.020**	**-0.140**	**-0.012**
	Treated Dowel	0.032	0.114	-0.115	0.013
	Bark Removed	**0.032**	**0.019**	**-0.141**	**-0.013**
Blank Dowel	Control	0.032	0.580	-0.081	0.046
	Empty Drill	**0.032**	**0.020**	**0.012**	**0.140**
	Treated Dowel	0.032	0.433	-0.039	0.089
	Bark Removed	0.032	0.976	-0.065	0.063
Treated Dowel	Control	0.032	0.184	-0.106	0.021
	Empty Drill	0.032	0.114	-0.013	0.115
	Blank Dowel	0.032	0.433	-0.089	0.039
	Bark Removed	0.032	0.415	-0.090	0.038
Bark Removed	Control	0.032	0.601	-0.080	0.047
	Empty Drill	**0.032**	**0.019**	**0.013**	**0.141**
	Blank Dowel	0.032	0.976	-0.063	0.065
	Treated Dowel	0.032	0.415	-0.038	0.090

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

Table 7.20: Simple contrast between treatments for β-santalol yield in Brookton plantation, without diameter as a covariate.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	**Empty Drill**	**0.011**	**0.011**	**0.007**	**0.051**
	Blank Dowel	0.011	0.925	-0.021	0.023
	Treated Dowel	0.011	0.761	-0.019	0.025
	Bark Removed	0.011	0.954	-0.021	0.023
Empty Drill	**Control**	**0.011**	**0.011**	**-0.051**	**-0.007**
	Blank Dowel	**0.011**	**0.014**	**-0.050**	**-0.006**
	Treated Dowel	**0.011**	**0.023**	**-0.047**	**-0.004**
	Bark Removed	**0.011**	**0.012**	**-0.050**	**-0.006**
Blank Dowel	Control	0.011	0.925	-0.023	0.021
	Empty Drill	**0.011**	**0.014**	**0.006**	**0.050**
	Treated Dowel	0.011	0.833	-0.020	0.024
	Bark Removed	0.011	0.971	-0.022	0.021
Treated Dowel	Control	0.011	0.761	-0.025	0.019
	Empty Drill	**0.011**	**0.023**	**0.004**	**0.047**
	Blank Dowel	0.011	0.833	-0.024	0.020
	Bark Removed	0.011	0.805	-0.025	0.019
Bark Removed	Control	0.011	0.954	-0.023	0.021
	Empty Drill	**0.011**	**0.012**	**0.006**	**0.050**
	Blank Dowel	0.011	0.971	-0.021	0.022
	Treated Dowel	0.011	0.805	-0.019	0.025

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

Table 7.21: Simple contrast between treatments for farnesol yield in Brookton plantation, without diameter as a covariate.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	Empty Drill	0.052	0.175	-0.178	0.033
	Blank Dowel	**0.052**	**0.002**	**-0.274**	**-0.063**
	Treated Dowel	**0.052**	**0.021**	**-0.231**	**-0.020**
	Bark Removed	**0.052**	**0.037**	**-0.218**	**-0.007**
Empty Drill	Control	0.052	0.175	-0.033	0.178
	Blank Dowel	0.052	0.073	-0.202	0.009
	Treated Dowel	0.052	0.317	-0.158	0.052
	Bark Removed	0.052	0.447	-0.146	0.065
Blank Dowel	**Control**	**0.052**	**0.002**	**0.063**	**0.274**
	Empty Drill	0.052	0.073	-0.009	0.202
	Treated Dowel	0.052	0.413	-0.062	0.149
	Bark Removed	0.052	0.290	-0.049	0.161
Treated Dowel	**Control**	**0.052**	**0.021**	**0.020**	**0.231**
	Empty Drill	0.052	0.317	-0.052	0.158
	Blank Dowel	0.052	0.413	-0.149	0.062
	Bark Removed	0.052	0.808	-0.093	0.118
Bark Removed	**Control**	**0.052**	**0.037**	**0.007**	**0.218**
	Empty Drill	0.052	0.447	-0.065	0.146
	Blank Dowel	0.052	0.290	-0.161	0.049
	Treated Dowel	0.052	0.808	-0.118	0.093

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

Table 7.22: Simple contrast between treatments for nuciferol yield in Brookton plantation, without diameter as a covariate.

				95% CI	
		SE	**P**	**LL**	**UL**
Control	Empty Drill	0.033	0.053	-0.134	0.001
	Blank Dowel	**0.033**	**0.001**	**-0.186**	**-0.051**
	Treated Dowel	**0.033**	**0.011**	**-0.156**	**-0.021**
	Bark Removed	**0.033**	**0.005**	**-0.167**	**-0.032**
Empty Drill	Control	0.033	0.053	-0.001	0.134
	Blank Dowel	0.033	0.127	-0.119	0.015
	Treated Dowel	0.033	0.511	-0.089	0.045
	Bark Removed	0.033	0.331	-0.100	0.034
Blank Dowel	**Control**	**0.033**	**0.001**	**0.051**	**0.186**
	Empty Drill	0.033	0.127	-0.015	0.119
	Treated Dowel	0.033	0.377	-0.037	0.098
	Bark Removed	0.033	0.570	-0.048	0.086
Treated Dowel	**Control**	**0.033**	**0.011**	**0.021**	**0.156**
	Empty Drill	0.033	0.511	-0.045	0.089
	Blank Dowel	0.033	0.377	-0.097	0.037
	Bark Removed	0.033	0.750	-0.078	0.057
Bark Removed	Control	0.033	0.005	0.032	0.167
	Empty Drill	0.033	0.331	-0.034	0.100
	Blank Dowel	0.033	0.570	-0.086	0.048
	Treated Dowel	0.033	0.750	-0.057	0.078

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

7.5 Comparisons Within Discs – Simple Contrasts Between Treatments

Table 7.23: Simple contrast between treatments for Disc 1 HW % area.

				95% CI	
		SE	***P***	***LL***	***UL***
Control	Empty Drill	2.216	0.706	-5.198	3.526
	Blank Dowel	2.168	0.522	-5.655	2.877
	Treated Dowel	2.171	0.437	-5.964	2.581
	Bark Removed	2.173	0.657	-5.240	3.311
Empty Drill	Control	2.216	0.706	-3.526	5.198
	Blank Dowel	2.215	0.803	-4.911	3.805
	Treated Dowel	2.216	0.700	-5.216	3.504
	Bark Removed	2.223	0.954	-4.503	4.245
Blank Dowel	Control	2.168	0.522	-2.877	5.655
	Empty Drill	2.215	0.803	-3.805	4.911
	Treated Dowel	2.165	0.889	-4.564	3.959
	Bark Removed	2.159	0.844	-3.824	4.672
Treated Dowel	Control	2.171	0.437	-2.581	5.964
	Empty Drill	2.216	0.700	-3.504	5.216
	Blank Dowel	2.165	0.889	-3.959	4.564
	Bark Removed	2.169	0.738	-3.542	4.996
Bark Removed	Control	2.173	0.657	-3.311	5.240
	Empty Drill	2.223	0.954	-4.245	4.503
	Blank Dowel	2.159	0.844	-4.672	3.824
	Treated Dowel	2.169	0.738	-4.996	3.542

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.

Table 7.24: Simple contrast between treatments Disc 8 Top HW % area.

				95% CI	
		SE	***P***	***LL***	***UL***
Control	Empty Drill	2.227	0.884	-4.057	4.708
	Blank Dowel	2.178	0.381	-6.196	2.376
	Treated Dowel	2.181	0.484	-5.821	2.765
	Bark Removed	2.183	0.820	-4.792	3.799
Empty Drill	Control	2.227	0.884	-4.708	4.057
	Blank Dowel	2.225	0.316	-6.614	2.143
	Treated Dowel	2.226	0.406	-6.234	2.527
	Bark Removed	2.233	0.713	-5.216	3.573
Blank Dowel	Control	2.178	0.381	-2.376	6.196
	Empty Drill	2.225	0.316	-2.143	6.614
	Treated Dowel	2.176	0.861	-3.900	4.663
	Bark Removed	2.169	0.515	-2.855	5.681
Treated Dowel	Control	2.181	0.484	-2.765	5.821
	Empty Drill	2.226	0.406	-2.527	6.234
	Blank Dowel	2.176	0.861	-4.663	3.900
	Bark Removed	2.180	0.636	-3.257	5.321
Bark Removed	Control	2.183	0.820	-3.799	4.792
	Empty Drill	2.233	0.713	-3.573	5.216
	Blank Dowel	2.169	0.515	-5.681	2.855
	Treated Dowel	2.180	0.636	-5.321	3.257

Note: SE = Standard error, P = Significance, CI = Confidence interval, LL = Lower limit, UL = Upper limit. Bold denotes significant differences.