Use of fungicides for the management of *Uromycladium acaciae* in *Acacia mearnsii* plantations, South Africa

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DECLARATION

I, Richard Guy Payn (211183024), hereby declare that the thesis for Master of Science is my own work and that it has not previously been submitted for assessment or completion of any postgraduate qualification to another University or for another qualification.

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PUBLICATIONS AND PRESENTATIONS

Information contained within this thesis has been published or submitted for publication and presented, the details of which are outlined below.

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Presentations

Information contained within this thesis has been presented at the following research meetings and field days by the author.

 Wattle Rust Working Committee meeting. 12th October 2015. Venue, Pietermaritzburg; Reported on completion and results of fungicide screening trial. Discussed events and treatments to be tested in adjuvant and fungicide application timing trials

Wattle Rust Working Committee 5th July 2016; Reported on completion and results of adjuvant and fungicide application timing trials. Reported progress

made on trying to determine link between wattle rust symptoms. Discussed approach to determine the economics of fungicide use

 Institute for Commercial Forestry Research Field Day. 25th October 2016. Venue, Paulpietersburg area. Presented results of fungicide screening trials and adjuvant and fungicide application timing trials, results of the attempt to link wattle rust Disease Expression to tree growth and results on the economics of fungicide use

ABSTRACT

South Africa has ca. 110 000 ha planted to Acacia mearnsii with 85% of the revenue from the species obtained from the timber, and 15% from the bark. Since its detection in 2013, wattle rust (recently identified as Uromycladium acaciae) has spread throughout the black wattle plantation area in KwaZulu-Natal, and from 2015 it was recorded in southern Mpumalanga. The pathogen affects trees of all age classes, causing a reduction in growth, as well as mortality with severe infection. Research has been initiated to determine a number of strategies for the management of the pathogen. These strategies include understanding wattle rust biology and epidemiology, planting tolerant or resistant black wattle, the testing and use of fungicide for management, and remote sensing and process based modelling to assess black wattle loss and high risk areas. These, with the outcomes from this research, will be combined into an overall Integrated Pest Management plan. Of the various strategies, the management of wattle rust with the use of fungicides is important, not only as it will have the potential to reduce the negative impacts of wattle rust, but it will also provide an interim solution until the other research areas provide alternative solutions. To address the current lack of fungicides available (and knowledge around their application) for the management of wattle rust, a series of trials were implemented to screen fungicides for their potential use, extend periods between the re-application of fungicide (if possible), the linking of symptoms to Disease Expression to aid with the timing of application, and the cost:benefits associated with fungicide use. Prior to the initiation of research into managing wattle rust, no fungicides were registered in South Africa for the control of wattle rust. In October/November 2014, three A. mearnsii trials were initiated in the KwaZulu-Natal Midlands and SE Mpumulanga where fungicides were tested at varying rates for the control of wattle rust. Wattle rust had a significant and negative impact on tree growth, irrespective of site and/or previous infection. All fungicides tested and at all the rates applied, proved effective for control. For the most effective control of wattle rust, fungicides should be applied as a preventative, rather than corrective measure.

In October 2015 a trial was initiated in southern KwaZulu-Natal to determine the effectiveness of varied application schedules and adjuvants of fungicides for the management of wattle rust. Two trials had initially been initiated but one had to be abandoned due to browsing damage. Wattle rust had a significant impact upon Groundline Diameter and Biomass Index but not Height. All of the adjuvants used and application schedules were effective in managing wattle rust. The most effective fungicide application used will therefore be based upon cost and in a manner that will reduce the likelihood of acquired resistance developing in wattle rust populations.

The timing of fungicide application is necessary for optimal use of these fungicides. Fungicide applications could potentially be linked to the emergence of different wattle rust symptoms to optimize fungicide use. Wattle rust symptoms were analysed from the untreated control plots of two trials, one in the KwaZulu-Natal midlands and one in southern KwaZulu-Natal, to determine whether wattle rust Disease Expression could be linked to black wattle tree growth. Regression trees were used for the analysis, as linear and multiple regression techniques would be unsuitable for the data. Regression trees were overfitted and attempts at testing the robustness of the model by cross-validation were unsuccessful. No individual symptom emerged as a significant predictor of tree growth, indicating that fungicide application should take place with the onset of any of the wattle rust symptoms tested.

The results from six trials testing the use of fungicides for managing wattle rust were compared to assess costs associated with fungicide use. Relative growth for Biomass Index was compared to untreated controls to obtain comparisons within and between sites. Costs versus benefit were compared using a two-way table to determine the most optimum treatment. The largest portion of treatment costs was attributed to the cost of fungicide. No single treatment was found to be optimal for the recommended rate of application. The use of adjuvants increased the cost of treatment, without additional benefit in growth. Control of wattle rust is beneficial, although costly if over-applied. Rotation-end data is required to determine whether fungicide use is economical for managing wattle rust over an extended period of time.

As a limited number of fungicides, from a limited number of fungicide groups were screened, the screening of additional fungicides from different fungicide groups will provide an additional selection of fungicides. If these are used in combination or alternation, the likelihood of acquired resistance developing among wattle rust populations will be reduced. Linking fungicide applications with wattle rust epidemiological and climatic data will aid in optimal use of fungicides, by timing applications to coincide with epidemiological and climatic cues. Rotation end research comparing final yield on fungicide treated versus untreated black wattle is needed to fully understand the economics of fungicide use. This will also aid in the understanding of the impact of wattle rust on tree age.

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CHAPTER 1: Introduction

Plantations in South Africa have been an important timber resource since their first establishment around the mid-1800s. Forest plantations are a necessary requirement to supply South Africa with wood as wood resources from indigenous reserves are inadequate to meet the demand. A number of different genera are grown in South Africa to provide the various needs for wood. Exotic timber plantations in South Africa, satisfying this need for wood, contribute significantly to South Africa's economy. Although South Africa imported approximately R 14.9 billion worth of forest products in 2013, R 19 billion worth of forest products was exported thereby contributing R 4.1 billion towards South Africa's population is dependent directly or indirectly on services provided by the forestry industry (Godsmark 2011).

Currently three genera of trees are grown in South Africa on a significant commercial scale. Various species of Eucalyptus, Pinus and Acacia mearnsii De Wild. (black wattle) are grown for the production of various forest products. Acacia *mearnsii* is a commercially significant source of timber in the forestry sector and is grown primarily for its tannin extracts and timber properties in South Africa as well as Brazil and south east Asia (Brown et al. 1997; Chan et al. 2015; Griffin et al. 2011). The primary use of extracts is for the tanning of leather in the leather industry (Dunlop and MacLennan 2002; Brown et al. 1997). Other uses for the tannin extracts include flocculants, adhesives in wood composites, and various other speciality pesticides for the food and beverage industries (Chan et al. 2015; Brown et al. 1997). South Africa currently has approximately 110 000 ha of A. mearnsii planted (Chan et al. 2015). The total value of tannin extracts during 2013 was ca. R 579 174 400 (Godsmark 2014). However, there has been an increased interest and use of A. mearnsii as a source of fibre for pulp production since the late 1990's (Dunlop and MacLennan 2002) to the extent that currently 85% of the revenue from this species is obtained by timber and the remaining 15% from bark (Chan et al. 2015). South Africa produces ca. 45 000 tonnes of bark extract annually, and in 2013, exported ca. 800 000 tonnes of black wattle wood chips (Chan et al. 2015). The only other significant secondary products produced by the species are firewood and charcoal (Chan et al. 2015). Acacia mearnsii plantations make up ca. 7% (88 882 ha)

(Forestry Economics Services 2014) of the total plantation area in South Africa and provide 36 000 people with both direct and indirect employment (Dunlop and MacLennan 2002).

Long-term sustainability of A. mearnsii is of importance to the established wattle industry. As for any land management scheme, challenges arise from a number of sources. Damage to plantations (in total, of all species) through biotic and abiotic factors have the potential to cause losses in growth and volume (Brown et al. 1997). In 2011-12 approximately 18 115 ha (Forestry Economics Services 2014) of plantation area was damaged by fire, insects, diseases and animals. Pests and diseases pose a challenge as they can induce mortality and growth reductions of infected areas (Brownet al. 1997). In 2011-12 approximately 2 637 ha were damaged by pests and diseases (Forestry Economics Services 2014). As the primary loss in the yield of a plantation is through a failure to achieve full stocking (Atkinson and MacLennan 1997), it is crucial to develop different management strategies for pests and diseases toachieve full stocking during re-establishment. In the past, management of pests and diseases was focused on forest hygiene and stress avoidance through silvicultural practices (Atkinson and MacLennan 1997). Current pest and disease management focuses on the use of corrective and preventative treatments in addition to hygiene and silvicultural treatments to reduce losses (Atkinson and MacLennan 1997). New pests and diseases are constantly emerging and pose challenges to plantation forestry worldwide (Wingfield et al. 2001). Therefore, ongoing research is required to provide information on how to manage new pests and diseases as they arise.

The most recent disease of *A. mearnsii* is *Uromycladium acacia* (Cooke) P. Syd. & Syd. 1914 (McTaggart et al. 2015) which has the potential to impact upon the sustainability of the species. The disease has spread throughout the wattle growing areas both in KwaZulu-Natal Midlands and Mpumalanga provinces since its detection in 2012-13. Unless managed, or controlled, the disease is a major concern for the wattle growing industry (Chan et al. 2015), as it has the potential to reduce productivity in *A. mearnsii* plantations. Strategies for managing wattle rust need to be developed and implemented.

1.1. Pests and diseases in plantations

South African timber plantations are even aged monocultural forest systems. Monocultures provide conditions conducive for both natural and introduced pests and diseases to flourish (Nyland 2002; Agrios 2005). The success of tree plantations throughout the Southern Hemisphere is partly attributed to the planting of exotic species in locations removed from their natural predators (Wingfield et al. 2001); (Wingfield et al. 2008; Wingfield and Swart 1994). South African plantations make use of exotic species which were initially absent from the source of their natural pests and diseases when introduced into South Africa. The forestry plantation species grown in South Africa are susceptible to a variety of pests and diseases, both local and introduced (Wingfield et al. 2011). When a natural pest or disease adapts its lifestyle to include exotic plantation species, it lacks its own natural predators which would keep it in check in its natural ecosystem (Atkinson and MacLennan 1997). Accidental introduction of natural pests and diseases over time has led to outbreaks that have resulted in timber yield losses through reductions in growth, poor tree form and tree mortalities (Liebhold et al. 1995). Plantation forests provide conditions suitable for pests and diseases to exist and spread without natural checks and balances, or until the introduction of some form of management practice.

Introduced pests and diseases arrive at an undesired location through different pathways. Contaminated or infected soil, untreated wood, seeds and plant debris transported around the world provide a means for pest and disease movement (Burgess and Wingfield 2002; Liebhold et al.1995; Wingfield et al. 2001). There is a constant increase in the number of emerging pests and diseases in South Africa and it is expected to continue in that manner due to globalisation and climate change (Ramsfield et al. 2016; Wingfield et al. 2001). There is a need to develop both local and global strategies (Wingfield et al. 2001) for the management of the spread of pests and diseases due to the costs associated with management and reduced productivity.

The emergence of a natural pest or disease of a plantation species is a risk to the yield productivity of a plantation (and therefor the economic viability) when the pest or disease reaches levels of economic significance (Radcliffe et al. 2009; Wingfield et al. 2001). Understanding pest biology, which includes taxonomy and

epidemiology, is the first step to effectively manage a pest or disease. In addition, understanding the interaction between host-pathogen-environment is crucial for implementing effective management strategies. These include silvicultural practices, resistance programmes and quarantine measures used (Wingfield et al. 2001).

1.1.1. Epidemiology and life cycle of pests and diseases

Understanding the epidemiology and life cycle of pests and diseases is required to determine an optimal management strategy. The combination of susceptible host plants, a pest or disease and favourable environmental conditions are factors associated with the epidemiology of pests and diseases (Agrios 2005). Once epidemiology is understood then management strategies can be implemented, optimally during vulnerable phases of the pest or disease life cycle (Agrios 2005). This ensures best use of resources and the most efficient strategy to minimise damage from the pest and/or disease with regard to its biology. Conversely, incorrectly timing the implementation of pest and disease movement strategies has the potential to be costly and wasteful.

1.1.2. Site and climate

Environmental conditions are linked to the occurrence and spread of pests and diseases. Fluctuations in climatic conditions influence the occurrence of pests and diseases (Coakley, et al. 1999). The "growth stage, succulence, and genetic susceptibility of host plants" are all impacted by environmental conditions (Agrios 2005). In addition, the "survival, vigour, rate of multiplication, sporulation, ease, dispersion, and distance of dispersal as well as the rate of spore germination and penetration" of the pathogen are also impacted by environmental conditions (Agrios 2005). The most significant climatic conditions that affect the epidemiology of a pest or disease are temperature and moisture (Agrios 2005).

Site specific factors are also linked to plant health, particularly soil factors. Soil pH and structure can be either conducive to, or hinder the development of soil pathogens (Agrios 2005; Norris et al. 2003; Robson 2012). Host plant nutrition, as obtained from soil, also has an impact upon plant health. Variations in available nitrogen, phosphorus, potassium, calcium, copper, manganese, magnesium and

silicone cause varying responses in soil plant pathogens (Agrios 2005; Norris et al. 2003). Understanding the impact of the interactions among site specific factors is necessary before managing plant health concerns that arise from nutrient deficiency or abundance, particularly for soil-borne pathogens.

1.1.3. Site occupation (stocking and species selection)

Species and planting density are matched to specific sites to optimise production from a given area. The stocking of a plantation compartment impacts tree health (increasing or reducing stress) therefore making it more or less susceptible to infection/infestation from pests and/or diseases. Increasing stocking can provide a buffer for losses that occur as a result of pest and disease damage, thereby offsetting the cost of losses (Norris et al. 2003). However, increasing stocking density with the purpose of offsetting losses could also increase the spread of pests and diseases (Norris et al. 2003) due to increasing stress on trees and increased abundance of food. Stocking levels have seemingly contradictory effects upon pest and disease losses. Therefore, a situational approach to stocking in the context of the specific pest or disease is needed, such as the avoidance of overstocked *Pinus* patula Schiede ex Schltdl. and Cham compartments for the management of Fusarium circinatum (Nirenberg and O'Donnell 1998; Mitchell et al. 2011). Site occupation (site species matching) is linked to site and climatic conditions. When linked together, site characteristics and climatic conditions will determine the optimal species and plantation compartment planting density. Selecting correct species for the conditions present at the planting site, followed by appropriate silvicultural practices are necessary to obtain optimal yields and mitigate any losses which could occur from risks present at the site.

1.1.4. Silvicultural interactions

Risks associated with pest and disease damage can be mitigated by strategies used at various tree growth stages through silvicultural operations. Strategies used in nurseries, during site preparation and planting, and during the tending phase in silvicultural operations can either increase or decrease the incidence of pest and disease damage (Agrios 2005; Nyland 2002). One of the aims of silvicultural practices is to manage the health of a forest area (Nyland 2002).

Implementation of the most cost effective and efficient method of managing pests and diseases during all silvicultural operations is necessary. This requires an understanding of the pest's biology and its interaction with the host and environment and site-species matching, such as delaying planting *Pinus* species after burning areas where *Rhizina undulata* occurs, to avoid mortalities caused by the disease (Atkinson 1999). Linking these operations into a silvicultural management regime will provide the best strategy for managing pests and diseases.

1.1.5. Management strategies for pests and diseases

The financial viability of a plantation is reduced when no options exist to manage pests and diseases, because of growth losses through pest and disease damage. Effectively controlling the damage caused by pests and diseases requires input from scientists involving knowledge of the problem and the subsequent development of management strategies using this knowledge (Wingfield et al. 2001). Management options can be then determined based upon the epidemiology, biology and genetics of the pest or disease (Wingfield et al. 2001).

Management can be achieved through methods that exclude the pathogen from the host, reduce or eradicate the pathogen inoculum, immunise or improve the resistance of the host organism and directly protect plants from pathogens (Agrios 2005). These actions are implemented through several methods for the management of pests and diseases within forestry silvicultural practices. Pesticide, biological, cultural and mechanical management methods, transgenic plants and quarantine protocols can be used to manage pests and diseases (Maredia et al. 2003).

Most often pest and disease management strategies aim at the reduction of pest populations, rather than eradication (Radcliffe et al. 2009). This is due to the costs of removing problem organisms entirely, which would be unjustifiably high for eradication methods. Establishing sustainable and cost effective management systems is desired (Radcliffe et al. 2009). Therefore, the goal of integrated pest management (IPM) is to keep pest levels at an acceptable and manageable level rather than eradicating them entirely (Norris et al. 2003; Maredia et al. 2003).

1.1.5.1. Cultural management

Establishing and maintaining forest hygiene in a plantation is essential to prevent outbreaks of pests and diseases. Cultural methods such as sanitation cutting, rotating crops, adequate sanitation and creating conditions unfavourable to the pest or disease are cultural practices that could potentially be used to manage forest pests and diseases (Agrios 2005; Nyland 2002). Reducing stress upon trees is a key factor in maintaining forest hygiene (Nyland 2002). Stressed trees are predisposed to infection and infestation from pests and diseases. Correct density at planting will result in stands that are not over-stocked when established, and hence less stressed. Adequate timing of thinning operations, such as thinning pine sawtimber stands to reduce damage from *Sirex noctilio* Fabricius (Dodds et al. 2007), and reducing competition from weeds are some of the necessary management practices to maintaining a healthy plantation.

Selective tree breeding is another area which has shown success in managing pests and diseases. Breeding genetically resistant crops to pathogens is a major tool for managing plant pathogens in particular (Agrios, 2005). Hybridisation between species offers opportunities to reduce the damage impact from a pest or disease (Denison and Kietzka 1993). Cultural control options currently being researched for wattle rust are discussed in section 1.6.1.

1.1.5.2. Biological management

Biological management uses a natural predator or pathogen of a problem pest or disease. If available, the use of biological management is best suited to pests and diseases which have been introduced to South Africa (Atkinson and MacLennan 1997). Importing a biological control agent to use on a pest or disease that has emerged from outside South Africa follows a strict screening and legislative process. In the context of biological management, populations of biological control agents will require their host (the actual pest or disease) to survive (Maredia et al. 2003). Therefore, populations of pests and/or diseases in plantations (for which biological management options are available) are maintained at a manageable level, rather than eradicated, otherwise biological management populations disappear with the host. Research to determine a biological management agent is lengthy in comparison with the research to determine pesticide management options. Therefore, pesticide management options can be determined and implemented as a more immediate (albeit intermediate) method of pest control while research is conducted into biological control methods. Both can then be incorporated as part of an IPM.

1.1.5.3. Pesticide management

Pesticide management is an increasingly used option to manage biological problems in forestry (Atkinson and MacLennan 1997). Pesticide management is used extensively in agricultural crops to manage pest and disease problems which arise. However, it must be noted that forestry uses less pesticides than other commercially grown crops in South Africa (Atkinson and MacLennan 1997). Coupled with this is the fact that pesticide use in forestry is also less frequent than other crops as the use may only occur once in every five to 25 years (Atkinson and MacLennan 1997). Due to the limited size of the South African forestry industry, it is often financially unfeasible for pesticide companies to perform testing of pesticides for registration purposes (Atkinson and MacLennan 1997). Realising this, the forestry industry in South Africa often performs such research itself as a means to provide managers with additional tools for pest and disease management (Atkinson and MacLennan 1997).

The nature of research required to determine such management methods is time consuming and therefore pesticide management can serve as a "bridge" (due to a reduced time span needed for pesticide research) while biological and cultural options are developed. Furthermore, forestry companies in South Africa which subscribe to certification organisations, such as the Forestry Stewardship Council (FSC), are required to comply with policies regulating pesticide use as set out by the certification organisation (FSC 2015a). It is logical then that when pesticides are used, they should be used as effectively as possible. The use of pesticide (natural or synthetic chemicals used for controlling or managing an unwanted species) is one potential tool to manage a pest or disease. Pesticide use needs to be applied in a responsible manner which also reduces off target damage as well as reduces the chance of the target pest or disease developing resistance to the pesticide.

1.1.5.3.1. Efficient pesticide use

The efficient use of pesticides to manage pests and diseases relies on the correct application volumes and methods of application as well as the use of adjuvants to increase the efficacy of the active ingredients. In addition, correctly timing the application of pesticides also aids in improving their effectiveness (Blandino et al. 2012). The use of adjuvants can also aid in improving the coverage, absorption, persistence, translocation and efficacy of a pesticide used to control a pest or disease, thus reducing the active ingredient used (Gent et al. 2003). Benefits of adjuvants have been determined through testing, but with relatively few pesticides and crop species under field conditions (Gent et al. 2003). A study on commercial crop adjuvants by Gent et al. (2003) stated that "a properly selected adjuvant appears to have the potential to considerably improve fungicide and bactericide performance and perhaps allow lower active ingredient per hectare and/or longer spray intervals". Therefore, to effectively manage a pest or disease using pesticides, correct volumes of application need to be determined as well as whether the use of adjuvants and alteration of application timing will have any effect.

1.1.5.3.2. Pesticide resistance

Research needs to be conducted to determine whether the use of pesticides is a feasible option and if so, what pesticides are effective and how are they best applied to manage a pest or disease. Once a pesticide has been determined to be effective in managing a pest or disease it is necessary to use the pesticide in a manner that will not encourage the development of resistant genotypes. Constant, repetitive use of a single pesticide to control a pest or disease may lead to the development of resistance in the target pest or disease population (Norris et al. 2003; Agrios 2005). The development of resistance has the potential to render the use of a pesticide ineffective against the target pest or disease (Norris et al. 2003). The development of resistance to pesticides must be avoided forpesticides to remain a part of a pest or disease management strategy.

1.1.6. Integrated Pest Management (IPM)

The management methods discussed are used in combination with knowledge of pest and disease life cycles and environmental conditions to form an ecological approach to pest management, known as Integrated Pest Management (Sandler 2010). Norris et al. (2003) defines IPM as "a decision support system for the selection and use of pest control tactics singularly or harmoniously coordinated into a management strategy, based upon the cost-benefit analysis that consider the interests and impacts on producers, society, and the environment". Therefore pest management decision making also includes cost-benefit analysis of control methods. Sandler (2010) states "an integrated approach to pest management is based upon dynamic principles, rather than a definitive set of rules for control of a particular pest situation" indicating that a flexible and 'tailor-made' plan to manage each specific pest or disease is optimal.

According to Norris et al. (2003) IPM has a series of operational goals. They are to:

- maintain economic reliability in managing pests;
- reduce the risk of crop loss;
- minimise selection pressure on pests to maintain the viability of tactics in future (particularly pesticides); and
- maintain environmental quality and avoid tactics that are environmentally detrimental to ecosystems.

These goals provide criteria for an IPM plan to be viable and effective. Environmental, economic and social consequences may arise should these goals not be met when implementing an IPM plan.

1.2. Fungal diseases in plantations

Pine, eucalypt and wattle species grown in South African plantations have been subject to numerous pathogenic fungi. Significant fungal diseases of pine species include *Rhizina undulata* Fries (Wingfield et al. 2001), *Dothistroma septospora* Doroguine, *Fusarium circinatum* Nirenberg & O'Donnell (Wingfield et al. 2008), *Sphaeropsis sapinea* Fr. Dyko & Sutton and *Diplodia pinea* Desm. In eucalypt species *Mycosphaerella spp.*, *Botryosphaeria dothidea* Ces. & De Not. and *Cryphonectria cubensis* Hodges are of the greatest concern (Wingfield et al. 2008). The fungal diseases of *A. mearnsii* are discussed in detail in section 1.3.2. Control of fungal diseases will be most efficient when combined into an IPM plan.

1.3. Management of fungal diseases in plantations

The management of pathogens in an IPM plan relies predominantly on the use of resistant varieties of crops to the pathogen. Biological control is used but is relatively minor when compared to the management of insects. Cultural methods are also used, as is the use of pesticides (Norris et al. 2003).

Fungicides have been used for the management of both agricultural and forestry plant pathogenic fungi. *Fusarium circinatum*, *R. undulata* and *D. pinea* are fungal plant pathogenic fungi of pine species which have had various fungicides tested with varying degrees of success (Atkinson 1999; Iturritxa et al. 2011; Crous 2005; Allen et al. 2004). However, most research focuses upon the use of these pesticides at the nursery and during the establishment phase (Rolando 2006). *Fusarium circinatum* is one of the most important plant pathogenic fungi in the world (Wingfield et al. 2008a). Although there is no singularly effective method for controlling *F. circinatum*, the disease can be managed economically through an integrated management approach which relies on a combination of silvicultural management, planting genetically resistant stock and pesticide methods (Wingfield et al. 2008). *Rhizina undulata* has also been managed successfully through an integrated management approach (Wingfield and Swart 1994). It is likely that a combination of management strategies will be used to manage wattle rust.

1.4. Pests and diseases of Acacia mearnsii in South Africa

Pests and diseases of *A. mearnsii* exist at both the seedling and establishment phases of *A. mearnsii* plantations (Dunlop and MacLennan 2002). Both above and below ground parts of the tree can be affected by both pests and diseases. Although several pests and diseases occur on *A. mearnsii* of local and foreign origin, only a small portion of the many pests and diseases are of economic significance.

1.4.1. Pests of A. mearnsii

A multitude of pests exist during the establishment phase of a wattle plantation in South Africa. Whitegrubs, cutworms, termites, grasshoppers, millipedes and nematodes are significant establishment pests (Dunlop and MacLennan 2002). Whitegubs, cutworms and termites have registered pesticides for their management within a forestry situation (Dunlop and MacLennan 2002). Significant post establishment pests include wattle bagworm (*Kotochalia junodi* Heylaerts), the brown wattle mirid (*Lygidolon laevigatum* Reut.) and lappet moths of the *Lasiocampidae* family. Biological, cultural and pesticide management options exist for the successful management of these pests (Dunlop and MacLennan 2002).

1.4.2. Diseases of A. mearnsii

Diseases of *A. mearnsii* have the potential to cause significant damage to plantations, and if severe, re-establishment may be required (Dunlop and MacLennan 2002). Having measures in place to reduce damage to wattle plantations by diseases is necessary to reduce loss in stocking and yield. Although a high number of pathogenic fungi have been associated with the species, many disease symptoms of the species remain unknown (Roux and Wingfield 1997). *Phythopthora spp., Ceratocystis spp.* and *Botryosphaeria spp.* are the three most notable disease causing pathogens of *A. mearnsii* (Dunlop and MacLennan 2002; Roux et al. 2001). Three foliar diseases of *A. mearnsii*; *Camptomeris albiziae* Petch, *C. verruculosa* Syd. & P. Syd. and *Uromycladium alpinum* McAlpine have been reported (Roux and Wingfield, 1997).

Wattle rust was previously reported in South Africa as *U. alpinum* but was present only in the uredinial stage, with less economic damage or impact than is currently reported (Morris et al. 1988). Recent research has revealed a change in the lifecycle of the fungus, and increased disease severity which is yet to be fully understood (McTaggart et al. 2015). As such, the recent emergence of wattle rust currently poses a risk to wattle plantations as increased disease severity of fungal diseases may affect the physiological processes of the host plants (Berger et al. 2007), resulting in growth reduction and yield loses (Berger et al. 2007). Due to its recent increased impacts, no management strategy currently exists and hence

processes need to be developed to minimise growth and financial losses caused by wattle rust.

1.5. Wattle rust

Wattle rust has spread throughout the KwaZulu-Natal Midlands and Mpumalanga growing region. Research is currently being conducted by the Tree Protection Co-operative Programme (TPCP) to determine the pathogens biology, population diversity and epidemiology, which will aid in the understanding of the disease. In addition a Wattle Rust Working Group has been established to coordinate research efforts (some of which are described in more detail below) for the overall management of wattle rust.

1.5.1. Current research on wattle rust

Several studies have been initiated to develop an integrated pest management plan. These, conducted by different research institutes, aim to identify the pathogen symptoms, understand its biology and spread (epidemiology), assess the damage impact and to develop management strategies.

Exclusion studies are conducted at the Institute of Commercial Forestry (ICFR) as part of quantifying yield loses and contributing towards research carried out by the TPCP for understanding the pathogen's biology. Remote sensing to monitor and map high and low risk areas for disease outbreak as a potential tool for management of the disease is also being researched at the ICFR. As an alternative to remote sensing, process-based models (such as 3-PG) are being developed to predict yield loses and predict high risk areas based on quantifiable tree indicators. As part of the studies, weather variables are being recorded to understand any interactions associated with climate.

Screening of breeding populations for wattle rust resistance/tolerance, development of screening methods in controlled environments and breeding disease tolerant *A. mearnsii* is to be conducted by the ICFR and TPCP. Preliminary results from trials show that no *A. mearnsii* progeny is resistant to wattle rust, only tolerant. Breeding *A. mearnsii* trees for tolerance will provide genetic stock which can be planted in field as a long-term management option for wattle rust.

The initial screening of pesticide products for potential registration and the refinement of application methods for the cost effective management of wattle rust is part of current wattle rust management research. This research serves to determine whether fungicides are effective for managing wattle rust, how fungicide use may be improved upon through the use of adjuvants and fungicide application timing and whether the use of fungicides is cost effective, based upon the treatments tested.

1.5.2. Wattle rust management using fungicides

Limited research has been conducted in South Africa with regard to the use of fungicides for managing in-field diseases of plantation trees. In contrast, postestablishment use of fungicides on trees grown for other purposes has received notable attention in South Africa (Kotze 1981; Darvas and Kotze 1987; de Villiers et al. 1997). In the case of *Puccinia psidii* Winter, a significant rust of tree species within the *Myrtaceae* family, research has been conducted on various species, including *Eucalyptus* species. This research has been successful but focused on trees in nurseries, as well as coppiced trees in-field in South America (Coutinho et al. 1998).

Research exists on the use fungicides for managing nursery related pathogenic fungi as well as on other agricultural crops. Most testing of fungicides has been on canker and root rot type fungi. These tests focus upon the use of fungicides on the seed and planting stages of the forest production cycle. Testing fungicides for controlling wattle rust in-field can serve as a base for further research on the pesticide control of rust fungi in future.

Trials were implemented to determine potential fungicides as well as their optimum rates of application. These trials will form part of the data in the thesis. A second series of trials were aimed at determining the correct timing of pesticide application and how the effects of the applied pesticides can be prolonged (thus reducing return time and cost).

1.5.2.1. Initial screening of fungicides

The first phase of using fungicides for the management of wattle rust was to register several fungicides with the Registrar of Fertiliser, Farm Feeds and Agricultural Remedies. Screening and registration procedures are set out as required by the Fertilisers, Farm Feeds, Agricultural Remedies and Stock Remedies Act 36 of 1947 and the subsequent amendments to the Act. Registration was required to permit the use of the selected fungicides for the control of wattle rust.

The fungicides screened were Amistar Top^{®1} (azoxystrobin and difenoconazole), Amistar Extra^{®1} (azoxystrobin and dyproconazole) and Ortiva^{®1} (azoxystrobin) produced by Syngenta. Azoxystrobin is a quonine outside inhibitor, a pesticide which inhibits mitochondrial respiration in fungi, and operates systemically (Fungicide Resistance Action Committee, 2016). Cyproconazole and difenoconazole are both demethylation inhibitors, which prevent sterol biosynthesis in fungi (Fungicide Action Resistance Committee, 2016).

1.5.2.2. Further refinement of fungicides

While testing appropriate fungicides for use is important, of equal importance is determining the most cost effective method of fungicide use. To determine this further refinement of the application methods is required. Several techniques can be used to improve the efficacy of the fungicides. Adjuvants can be tested, methods of application can be varied, timing can be altered and the role of buffers determined. Focus will be on refinement of fungicides through altering timing and prolonging the effect of fungicides through the use of adjuvants as both are interlinked.

1.5.2.3. Linking wattle rust Disease Expression to black wattle growth

Fungicide use can potentially be improved upon by timing applications to coincide with stages of wattle rust growth. This can be potentially timed according to the emergence of a particular wattle rust symptom or to climatic conditions conducive to wattle rust growth. Failing to correctly time applications reduces the effectiveness of fungicide use and impacts upon the cost:benefit of treatment.

¹ Use of trade names does not indicate endorsement by Nelson Mandela Metropolitan University or the author and are included for the reader's benefit only

1.6. Cost of pesticide management

The purpose of fungicide use for the control of wattle rust is to prevent and mitigate losses through preventative spraying. The use of a fungicide is justified as effective if it reduces the impact of a fungus on tree growth to an acceptable level and if the fungicide helps facilitate unhindered growth (when compared to the absence of fungicide application). However, the cost of application/s need to be offset by the growth achieved in order for the use of the treatment to be cost effective. Fungicide cost effectiveness may potentially be improved through the correct timing of fungicide application and/or use of adjuvants. Linking growth reductions in black wattle to Disease Expression of wattle rust may provide an indicator of when to time fungicide applications. Wattle growers need an understanding of the cost-benefit of treatments to decide on the use of fungicides for the management of wattle rust. The economic decision on whether or not to use a pest management method can be based upon the economic injury level (EIL) and economic threshold (ET) concepts by Stern et al (1959). Economic decisions behind whether or not to implement pest and disease management strategies are critical to economic and environmental sustainability in IPM strategies (Radcliffe, Hutchison et al. 2009). If a sound IPM strategy is to be developed, the costs associated with treatments needs to be determined prior to determining the EIL and ET.

1.7. Research objectives

To address the lack of fungicides available for use (and knowledge associated with effective application) for the management of wattle rust, a series of trials were implemented with the following objectives:

- Screening fungicides to determine their effectiveness and which rates at which they are best applied (**Chapter 2**)
- Extend the period between reapplication through determining the role of adjuvants and timing of applications (**Chapter 3**)
- Understand the link between wattle rust Disease Expression and tree growth to potentially link fungicide applications to wattle rust Disease Expression (Chapter 4)
- Determining the cost effectiveness of fungicide applications tested (Chapter 5)

Screening fungicides and determining the rate at which they are best applied, together with the role of adjuvants and application timing, combined with linking wattle rust to tree growth can be used to determine the most cost effective method of fungicide use for the management of wattle rust. This knowledge of fungicide use can then be incorporated as part an Integrated Pest Management plan which makes use of resistant or tolerant black wattle, growth and yield models, knowledge of wattle rust biology and epidemiology and climatic conditions associated with wattle rust to manage the pathogen in the most effective manner.

NOTE: The research chapters (Chapters 2, 3, 4 and 5) contained within this thesis were prepared as separate research outputs (papers), with their original format retained. As the contents of these chapters dealt with subject matter around a common theme (the use of fungicides for the management of wattle rust), some duplication of contents is inevitable, particularly with respect to the introduction and literature review sections.

CHAPTER 2: Screening of fungicides for the management of wattle rust (*Uromycladium acaciae*) in *Acacia mearnsii* plantations, South Africa

Abstract

South Africa has ca. 110 000 ha planted to Acacia mearnsii with 85% of the revenue from the species obtained from the timber, and 15% from the bark. Since its detection in 2013, wattle rust (recently identified as Uromycladium acaciae) has spread throughout the black wattle plantation area in KwaZulu-Natal, and from 2015 it was recorded in southern Mpumalanga. The pathogen affects trees of all age classes, causing a reduction in growth, as well as mortality with severe infection. No fungicides are currently registered in South Africa for the control of wattle rust. In October/November 2014, three A. mearnsii trials were initiated in the KwaZulu-Natal Midlands and SE Mpumulanga where fungicides were tested at varying rates for the control of wattle rust. The same trial design was used for all three trials and consisted of a 3 x 3 factorial with one additional control, replicated three times and laid out as a RCBD. The factorial combination consisted of three fungicides (azoxystrobin + difenoconazole; azoxystrobin + cyproconazole; azoxystrobin) applied at three rates (x; 2x and $\frac{1}{2}x$). The additional control was not treated with fungicides. Wattle rust had a significant and negative impact on tree growth, irrespective of site and/or previous infection. All fungicides tested and at all the rates applied, proved effective for control. The optimum rate selected, timing and frequency of application will be based on a combination of prevention of resistance as well as optimization of growth. For the most effective control of wattle rust, fungicides should be applied as a preventative, rather than corrective measure. However, in cases of severe infestation, a corrective application could also be used to aid the management of wattle rust.

2.1. Introduction

South Africa has ca. 110 000 ha planted to *Acacia mearnsii* De Wild. (black wattle) (Chan et al. 2015). The species is grown primarily for its bark tannin extracts and wood (Griffin et al. 2011). Currently 85% of the revenue from the species is obtained from the timber, and 15% from the bark (Chan et al. 2015). South Africa produces ca. 45 000 tonnes of bark extract annually and in 2015, exported 800 000

bone dry metric tonnes (BDMT) of black wattle wood chips (Chan et al. 2015). Of the area planted to black wattle, 78% (86 000 ha) is owned by 600 private commercial growers who rely on the species as one of their primary timber crops. Different species (from three genera: *Pinus, Eucalyptus* and *Acacia*) are planted by these private growers so that they are less susceptible to negative market forces, with the dual income (bark and timber) generated from black wattle enabling them to achieve this objective. The preservation and continuation of the black wattle industry in South Africa is thus important, and when challenges arise that negatively impact on the sustainability of this industry, these risks need to be adequately managed.

Black wattle is impacted negatively by numerous pests and diseases of economic significance. The most common pests of wattle include wattle bagworm (Kotochalia junodi Heylaerts), the brown wattle mirid (Lygidolon laevigatum Reut.) and various lappet moths of the Lasiocampidae family (Dunlop and MacLennan 2002). Phythopthora spp., Ceratocystis spp. and Botryosphaeria spp. are the three most notable disease causing pathogens of black wattle (Roux and Wingfield 1997). The most recent disease of black wattle in South Africa is wattle rust (Wood Southern Africa and Timber Times 2014), recently identified as Uromycladium acacia (Cooke) P. Sydow & Sydow. Since its detection in 2013, wattle rust has spread throughout the black wattle plantation area in KwaZulu-Natal, and from 2015 it was recorded in southern Mpumalanga. The pathogen affects trees of all age classes, causing a reduction in growth, as well as mortality with severe infection. Symptoms for rust fungi may include the various life stages spermagonia, aecia, telia, pycnia, uredinia and basidia. Variation of the life cycle stages exists among different rust fungi, with telia and basidia most commonly expressed by U. acaciae in New Zealand (Dick 2009). Determining management strategies to control wattle rust is essential to minimize negative impacts on black wattle productivity.

Within South African plantations, the historical trend for pest and disease management has been to mitigate losses through adequate forest hygiene and stress avoidance strategies (Atkinson and MacLennan 1997). To achieve this, the use of genetic plant resistance and tolerance to disease, biological management strategies and silvicultural practices (in general) have received prominence. An understanding of the epidemiology of the wattle rust also needs to be developed, together with any links to tree productivity, site (climate and physiography) and/or management. Although more holistic and sustainable, the incorporation of these components from inception, into an integrated strategy can be a lengthy process. The testing and use of appropriate pesticides (fungicides) requires a shorter development period, and has the potential to manage the problem while these other strategies are being developed. Pesticide management of wattle rust can therefore serve as an interim measure, and may also be incorporated into an Integrated Pest Management (IPM) plan once knowledge of the other components are known. At the time of this research, no fungicides were registered for the management of wattle rust.

Fungicide-use has received attention in South African plantation forestry, mainly in pine and eucalypt species. However, the majority of research conducted on the use of fungicides for managing fungal pathogens has occurred either within nurseries, or at establishment (planting) (Rolando 2006). Little published literature could be found regarding research conducted on the use of fungicides during the post-establishment growth phase of plantation forestry species in South Africa. In contrast, the post-establishment use of fungicides on trees grown for other uses (such as fruit orchards) has received notable research attention in South Africa, particularly on avocado, apple and citrus bearing trees (for example: Darvas and Kotze 1987; Kotze 1981; De Villiers et al. 1997). No research has been conducted on the use of fungicides in controlling diseases of black wattle in South Africa.

To effectively manage the impact of wattle rust, management strategies need to be established timeously. Within South Africa, registration is required for the legal use of fungicides according to the protocols of the Fertilisers, Farm Feeds, Agricultural Remedies and Stock Remedies Act 36 of 1947 (Department of Agriculture and Forestry, 2015). In addition, any fungicide used for the management of wattle rust needs to conform to any environmental and/or ecological constraints.

The South African forest industry subscribes to the Forest Stewardship Council (FSC) principles associated with responsible environmental, social and economic management (FSC 2015b), with ca. 85% of the afforested area certified according to FSC criteria (FSA 2012). As part of this, the FSC has a process regarding pesticide use in forestry, with three main objectives: the identification and avoidance of 'highly hazardous' pesticides; the promotion of 'non-pesticide' methods

of pest management as an element of an integrated pest and vegetation management strategy; and the appropriate use of the pesticides that are used (FSC 2007). At the time of the initiation of this research azoxystrobin (methyl (α E)-2-[[6-(2-cyanophenoxy)-4-pyrimidinyl]oxy]- α -(methoxymethylene)benzeneacetate),

cyproconazole¹(α -(4-chlorophenyl)- α -(1-cyclopropylethyl)-1H-1,2,4-triazole-1-

ethanol) and difenoconazole (1-[[2-[2-chloro-4-(4-chlorophenoxy)phenyl]-4-methyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole) were selected as they were not included on the FSC hazardous pesticides list, and they are currently registered within the agriculture and fruit orchard sector for the management of rusts. All three fungicides are systemic in action. Azoxystrobin (strobilurin fungicide group) and cyproconazole (triazole fungicide group), prevent mitochondrial respiration in fungi. Difenoconazole (triazole fungicide group) prevents cell membrane ergosterol biosynthesis (Pscheidt 2015; Yang et al. 2011). To avoid resistance developing in the target pathogen, it is necessary to test and register a number of fungicides from different fungicide groups, such that different pesticides can be applied and/or used in combination (Peever and Milgroom 1995; Radcliffe et al. 2009). In October/November 2014, three *A. mearnsii* trials were initiated in the KwaZulu-Natal Midlands and SE Mpumulanga where azoxystrobin, cyproconazole and difenoconazole were tested at varying rates for the control of wattle rust.

2.2. Materials and Methods

Three sites, City Forests and Etterby in KwaZulu-Natal Midlands, and Commondale in SE Mpumalanga were located in the wattle growing regions of South Africa on existing stands of *A. mearnsii* (**Table 1**). The sites were selected such that they fell within areas where wattle rust was known to occur. Sites were also selected where the trees were between 0.5 and 1.0 m in height at the time of trial initiation. This would allow for the continued manual spraying of the fungicides onto the foliage of the trees through the growing season when using a knapsack sprayer. In addition, the trees would be in their exponential growth phase, the period during which any negative/positive treatment impacts (if any) would most likely be expressed. The same trial design was used for all three trials and consisted of a 3×3 factorial with one additional control, replicated three times and laid out as a RCBD. The factorial combination consisted of three fungicides (azoxystrobin + difenoconazole;

azoxystrobin + cyproconazole; azoxystrobin) applied at three rates (x; 2x and $\frac{1}{2}x$) (**Table 2**).

Table 1. Site characteristics for three *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga for testing of fungicides for the control of wattle rust.

Region	Magisterial district	Pietermaritzburg	Richmond	Paulpietersburg
	Plantation/trial name	City Forest	Etterby	Commondale
Latitude Longitude		29° 34' 52.69" S 30° 19' 52.28" E	29° 50' 30.17" S 30° 11' 24.33" E	27° 18' 26.78" S 30° 46' 13.75" E
Altitude (m a.s.l.)		1 075	1 094	1 100
MAP (mm)		904	968	908
MAT (°C)		15.8	16.8	18.1
Aspect		uniform, gentle ENE- facing slope	uniform, moderate NW- facing slope	uniform, gentle S-facing slope
Selected soil physical and pesticide properties	Soil form	Inanda 1100	Griffon 2100	Clovelly
	Soil depth (m)	0.50	0.80	1.00
	Soil texture	Clay	Clay/Loam	Clay
Spacing (m) Stems per hectare (sph)		3 x 1.8 1 852 sph	3 x 1.8 1 852 sph	3 x 1.8 1 852 sph
Seed lot/orchard		PSO-10	PSO-10	PSO-10
Date planted		05/03/2014	02/12/2013	05/02/2014
Drought risk (%)*	>850 mm	45.1	74.9	70.9
	<650 mm	13.7	5.0	3.6
Potential productivity*	Climate zone	CT8 (cool temperate)	WT3 (warm temperate)	WT8 (warm temperate)
	Growing conditions for species planted	Optimum	Optimum	Optimum
	Site index (age 5)	15	18	20

*Smith et al. 2005

The additional control was not treated with fungicides, and was repeated twice within each replicate. Each treatment plot consisted of a single line of 22 trees, with the inner 20 trees being measured (2 buffer trees at each end). In addition, there were single lines of non-treated trees on either side of the treated rows so as to act as a buffer between adjacent plots. The fungicides were sprayed onto the foliage to run-off using a 16 L knapsack sprayer, fitted with an air-induction, twin-flat-fan TeeJet Turbo TwinJet[®] AITTJ60 nozzle. This nozzle was selected due to its ability for good canopy penetration and cover. The nozzle is also recommended for the broadcast application of fungicides where good drift control and coarse droplets are required (TeeJet Technologies 2015). Pressure was regulated to 1.5 KPa, resulting in a spraying volume of 888.6 L ha⁻¹. The dates when the fungicides were applied, together with the climatic conditions on the day of spraying were recorded (**Table 3**).
Table 2. Treatments, including fungicide-related information, tested for the control of wattle rust in three *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga.

Treatments	Trade Name	Active ingredient (g L ⁻¹)	Rate of application of product ha ¹ assuming a spray volume of 1000 L ha ⁻¹	Treatment abbreviation		
1			500 ml (0.5%)	Ortiva_0.5		
2	Ortiva [®]	azoxystrobin (strobilurin) (250 g L ⁻¹)	1000 ml (1.0%)	Ortiva_1.0		
3			2000 ml (2.0%)	Ortiva_2.0		
4		$(200 \text{ m})^{-1}$	500 ml (0.5%)	Amistar Top_0.5		
5	Amistar Top [®]	diference paralle (triazele) (125 g L^{-1})	1000 ml (1.0%)	Amistar Top _1.0		
6			2000 ml (2.0%)	Amistar Top _2.0		
7	Amistar Vtra®	$(200 \text{ m})^{-1}$	500 ml (0.5%)	Amistar Xtra_0.5		
8	Amistal Alla	azoxystrobin (strobilurin) (200 g L) cyproconazole (triazole) (80 g L $^{-1}$)	1000 ml (1.0%)	Amistar Xtra _1.0		
9			2000 ml (2.0%)	Amistar Xtra _2.0		
Additional Treatments (Controls)						
10	Control	-	-	Control		
11	Control	-	-	Control		

2.2.1. Assessments

Tree Height (Ht in meters) and Groundline Diameter (Gld in mm) were taken on three occasions, at trail initiation and then at one month after the second and fourth application of the fungicides (**Table 3**). Biomass Index was calculated as Gld² x Ht and provides a good index of overall tree performance in young trees (Eccles et al. 1997). For use as possible co-variates when analysing individual tree data, all trees were scored for the presence of blanking (replanting of dead seedlings), double stems, multiple leaders, browsing, dead tops or any physical damage to the base of the stems from manual weeding operations. Tree Condition was quantified through the visual estimation of a combination of defoliation, discolouration and disease symptoms. A modified Braun Blanquet method (Kent and Coker 1996) for the estimation of the area of each tree affected was used, whereby increasing values are assigned, based on increasing cover (Table 4). This method of overall assessment of Tree Condition was selected as cover estimates are not biased by tree size. As the visual expression of any fungal pathogen varies according to the type, stage and severity of infection, a number of symptoms were scored so as to quantify Disease Expression.

Trial	Trial City Forest				Etterby			Commondale					
Assessment dates		24/10/2014	09/12	2/2014	11/03/2015	24/10/2014	10/12	2/2014	11/03/2015	22/11/2014	12/01	/2015	03/04/2015
Tree age when assessed		233	2	79	371	326	3	73	464	290	3	41	422
Fungicide application date		1 st	2 nd	3 rd	4 th	1 st	2 nd	3 rd	4 th	1 st	2 nd	3 rd	4 th
		23/10/2014	20/11/2014	13/01/2015	03/02/2015	23/10/2014	20/11/2014	13/01/2015	04/02/2015	22/11/2014	10/12/2014	05/02/2015	09/03/2015
Days betwee	en re- application	0	28	54	21	0	28	54	22	0	18	57	32
	Time of spraying (hrs)	15:43 - 17:45	10:10 - 13:00	11:30 - 14:06	15:49 - 18:23	11:00 - 14:00	06:45 - 08:40	06:30 - 08:51	06:15 - 08:20	06:45 - 08:30	07:00 - 09:42	06:15 - 08:20	06:00 - 08:40
Conditions at time of	Temperature shade (°C)	20.9	27.3	37.6	25.7	23.8	20.8	26.4	21.1	21.6	21.2	26.2	17.1
application	Relative humidity (%)	58.8	48.7	43.0	54.4	45.2	72.7	52.5	71.8	65.2	78.9	66.6	81.3
	Wind speed (m s ⁻¹)	1.6	2.1	1.7	1.1	1.4	0.0	0.9	0.4	0.1	0.0	0.0	0.0

Table 3. Sequence of events in terms of assessments dates and the application of fungicides in three Acacia meansii

 trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga.

These were adapted from symptoms as described by Dick (2009) and included the scoring of trees for teliospore masses (brown pustules) on the leaves, deformed pinnules or pinnae and stem lesions (teliospore masses on the main stem, or branches) according to the volume of tree affected, where 0, 1, 2 and 3 = 0, 1-25%, 26-50% and 3 = +50% affected respectively.

Table 4. Acacia mearnsii "Tree Condition" quantified through the visual estimation of a combination of defoliation, discolouration and disease symptoms. A modified Braun Blanquet method (Kent and Coker, 1996) for the estimation of the area of each tree affected was used, whereby increasing values were assigned, based on increasing cover affected.

Percentage area affected	Median value used for analyses
rare: 1-2 leaflets (less than 5 %)	1
few: 3 - 4 leaflets (less than 5 %)	2
many: 5 - 10 leaflets (less than 5 %)	3
abundant: > 10 leaflets (less than 5 %)	4
5 -12 %	8.75
12.5 - 25 %	18.75
25 - 50 %	37.5
50 - 75 %	62.5
75 -100 %	87.5
	Percentage area affected rare: 1-2 leaflets (less than 5 %) few: 3 - 4 leaflets (less than 5 %) many: 5 - 10 leaflets (less than 5 %) abundant: > 10 leaflets (less than 5 %) $5 - 12 \%$ 12.5 - 25 % 25 - 50 % 50 - 75 % 75 - 100 %

As the trees at City Forest and Etterby showed signs of the presence of wattle rust disease from the 2013 growing season, the second assessment of Tree Condition and Disease Expression was calculated relative to the first assessment (**Table 3** and **Figures 1d** and **2**). For Commondale, wattle rust symptoms only became apparent from the second assessment onwards and as such the third assessment of Tree Condition and Disease Expression was calculated relative to the second assessment. Plot means from the final assessment date were analysed as a 3 x 3 factorial with an additional control using GenStat for Windows (VSN International, 2013). Prior to the analysis, the data were checked to ensure that the assumptions for a valid ANOVA were not violated (**Table 5**). Before any comparison between treatments, an *F*-test was carried out to determine the overall significance of the differences between all treatment means within the experiment.

Table 5. Summary of analyses of variance showing means squares for selected tree variates at the final assessment date for three Acacia mearnsii trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga for testing of fungicides for the control of wattle rust.

		(371 d	City Forest (371 days after planting)			Etterby days after pl	anting)	(422	Commondale (422 days after planting)		
Source of variation	d.f. ¹	<i>Ht</i> (m)	Gld (mm)	ВІ	<i>Ht</i> (m)	<i>Gld</i> (cm)	BI	Ht (m)	<i>Gld</i> (cm)	ВІ	
Rep	2 (2)	0.020	0.041	25.71	0.056	0.155	80.90	0.087	0.021	7.4	
control.rest of treatments	1 (1)	0.913**	3.477**	1489.63**	0.255**	1.121**	273.37*	0.361 ^{\$}	1.894*	951.9*	
control.fungicide	2 (2)	0.041	0.026	30.90	0.019	0.070	53.85	0.0217	0.153	111.7	
control.rate	2 (2)	0.022	0.132	64.04	0.011	0.069	47.22	0.010	0.032	38.8	
control.fungicide.rate	4 (2)	0.021	0.042	22.90	0.020	0.100	163.77	0.130	0.366	164.7	
Residual	21 (19)	0.038	0.111	65.98	0.018	0.063	454.43	0.111	0.277	131.8	
Total	32 (30)										
Grand Mean		2.48	3.53	58.40	1.94	2.98	43.96	2.90	4.09	78.80	
Standard error of the difference (control.fungicide.rate)	•	0.14	0.24	5.74	0.10	0.18	3.46	0.24	0.37	8.12	
Coefficient of variation (units)		7.8	9.5	13.9	7.0	8.4	11.1	11.5	12.9	15.1	
Bartlett's test for homogeneity of variance, (control.fungicide.rate) Chi-square on 2 df.		2.03 ^{ns}	0.38 ^{ns}	0.75 ^{ns}	0.26 ^{ns}	1.93 ^{ns}	5.80 ^{\$}	1.46 ^{ns}	0.98 ^{ns}	0.25 ^{ns}	
Shapiro-Wilk test for Normality		0.98 ^{ns}	0.98 ^{ns}	0.98 ^{ns}	0.97 ^{ns}	0.99 ^{ns}	0.99 ^{ns}	0.35 ^{ns}	0.30 ^{ns}	0.75 ^{ns}	

Note: ^{\$}, * and ** indicates significance at *F*-prob <0.10, 0.05 and 0.01; and ^{ns}, non-significance. ¹ values in parenthesis indicate degrees of freedom for Etterby due to the exclusion of two plots from the analysis.



Figure 1. Height (a), Groundline Diameter (b), Biomass Index (square root transformed) (c) and relative Tree Condition (d) for three *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga for testing of fungicides for the control of wattle rust. Letters on bars (in figures a, b, and c) indicate significance at p< 0.05. Standard Error bars are included on Figure d.



Figure 2. The relative occurrence for three *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga for testing of fungicides for the control of wattle rust. Treatments means are shown as solid bars, the 95% confidence levels by the boxes, and the standard deviation by the bars.

2.2.2. Analysis

Only if the *F*-value was significant, were treatment differences further investigated using least significant differences (*Isd's*). Tree form was used in addition to the initial tree measurements as co-variates for subsequent measures. As these were neither significant as co-variates and/or did not alter the levels of significance or interpretation of the results, these assessments were excluded from the final analysis. The variate of Biomass Index received a square root transformation prior to analysis so as to stabilize the variance. At Etterby, two plots were excluded from the final analysis as they were inadvertently not weeded during routine commercial weed control operations, resulting in severely suppressed trees in those two plots. Changes in Tree Condition and Disease Expression were calculated relative to initial assessments (and expressed as a percentage) and are displayed using descriptive graphing techniques (**Figures 1d** and **2**).

2.3. Results and Discussion

The three contrasting sites were selected so as to determine the impact of wattle rust on *A. mearnsii*, and any interaction with the fungicides tested under different growing conditions. Although wattle rust was present at trial initiation at City Forest and Etterby (but not at Commondale), by the end of the 2015 growing season, tree growth at all three sites was significantly impacted by the fungus (**Table 5** and **Figure 1**). In terms of predicted growth criteria for *A. mearnsii* (**Table 1**), Etterby is considered to have the highest productive capacity, and City Forest the lowest. Actual growth for the period over which the trial was conducted (October/November 2014 - April/May 2015) showed Commondale as the highest with Etterby the lowest.

At all three sites, the application of fungicides resulted in a significant improvement in all the tree growth variates measured (**Table 5** and **Figure 1**). No significant differences were detected between the three fungicides tested, nor at the three different rates of application (**Table 5**). All fungicides were effective for controlling wattle rust as shown by improved tree growth and Tree Condition (Control versus Rest Combined), together with a reduction in all the Disease Expression symptoms assessed (**Figures 1** and **2**). Not only was the expression of wattle rust

greater in the Control treatment (no fungicides applied), but the variability for each of the variates assessed was also greater than where fungicides were applied (Rest Combined) (**Figure 2**). Any negative tree growth impacts of the fungicides (and rates of application) tested could not easily be detected. This may be due to the incidence of wattle rust which may, or may not have similar symptoms as those that were assessed. For example, deformed and curling leaflets/pinnae/pinnules, defoliation and phytoxticity (which was recorded as dicolouration in the trial) may all be symptoms of either wattle rust or phytotoxicity. However, the lack of the incidence of wattle rust during the initial period of tree growth at Commondale indicated the minor expression of phytoxicity in terms of curling leaflets. Despite this, the application of a fungicide had significantly greater benefits than that of any phytotoxicity.

The scoring of Tree Condition and Disease Expression on trees at trial initiation at City Forest and Etterby indicated the presence of the fungus in the 2013 -2014 growing season. At the first assessment, Tree Condition was minor at 4.5 and 8.0% at City Forest and Etterby respectively, and the mean of Disease Expression, 1.5 and 3.1% at City Forest and Etterby. Wattle rust was not detected until the second assessment at Commondale. The fungicides were applied on four occasions, the first two and last two applications following on from each other, but separated by ca. 28 days with no application. This break was recommended due to the spraying of the same fungicide within each treatment plot on consecutive occasions, resulting in the potential development of resistance. This application strategy, together with the use of fungicides from different modes of action, either individually or as a mixture, reduced rates of application (not tested in these trials), is used to prevent the development of resistance by the targeted fungus to the active ingredients (Peever and Milgroom 1995). The presence of wattle rust at City Forest and Etterby, combined with this break in fungicide application, allowed for the re-infection of all the trees within these two trials. This in part may explain the reduced tree growth recorded at these two sites when compared to Commondale, as well as the similar differences in tree growth between the Control and where fungicides were applied (Rest Combined). If wattle rust had been excluded in the treated plots over the whole growing season, a greater increase in growth could have been expected than the 39.7 and 19.7 % (using Biomass Index) recorded at City Forest and Etterby. This is

in comparison to Commondale, where the 21.7% increase in Biomass Index was the result of infection only occurring within the last three months of tree growth.

Even though symptoms of wattle rust were much lower, and developed later in the season at Commondale than the other two sites, the benefits of control from inception are apparent (**Figure 2**). In general, this supports the principle of applying the fungicides tested as a preventative treatment, rather than as corrective spray. The importance of applying fungicides during periods of active wattle rust growth is clearly illustrated by the re-infection of trees at City Forests and Etterby during the 28-day period where no fungicides were applied. Combining knowledge of wattle rust epidemiology, with environmental conditions conducive to its growth will assist with the timing of when to initiate preventative applications. Having fungicides from different groups will also allow for a continuation of spraying during periods of high fungus infection, reducing the possibility of any negative impacts.

2.4. Conclusion

Wattle rust had a significant and negative impact on tree growth, irrespective of site and/or previous infection. All fungicides tested and at all the rates applied, proved effective for control. The optimum rate selected, timing and frequency of application will be based on a combination of prevention of resistance as well as optimization of growth. For the most effective control of wattle rust, fungicides should be applied as a preventative, rather than corrective measure. However, in cases of severe infestation, a corrective application could also be used to aid the management of wattle rust.

Future work will need to focus on reducing costs associated with the overall number of times that applications need to be made over a growing season. This may be done through either extending the period of efficacy beyond the recommended 28 days, and or by linking the timing of application to periods of active infestation. For the latter to occur, a better understanding of seasonal growth of wattle rust is needed such that the timing of spraying be timed so as to prevent infection.

Chapter 3: Use of adjuvants and fungicide application timing for the control of wattle rust (*Uromycladium acaciae*) in *Acacia mearnsii* plantations, South Africa

Abstract

Acacia mearnsii (black wattle) plantations in South African cover approximately 110 000 ha. Uromycladium acaciae (wattle rust) has spread over Limpopo to the Western Cape of South Africa. The disease of black wattle causes reductions in growth and mortality from severe infections. In October 2015 a trial was initiated in southern KwaZulu-Natal to determine the effectiveness varied application schedules and adjuvants of fungicide for the management of wattle rust. Various treatments were laid out in a factorial design and consisted of two adjuvants application schedules (42 or 56 days between application) and four adjuvants (none; poly-1-p-menthene; borax + orange oil; poly-1-p-menthene & borax + orange oil). Three additional treatments were included where one was a control with no fungicides applied. The other two additional treatments had fungicides applied according to the recommended 28-day schedule (one application commencing in October and the other, November). Wattle rust had a significant impact upon Groundline Diameter and Biomass Index but not Height. All of the adjuvants and application schedules were effective in managing wattle rust. The most effective fungicide application used will therefore be based upon cost and in a manner that will reduce the likelihood of acquired resistance developing in wattle rust populations.

3.1. Introduction

Acacia mearnsii De Wild. (black wattle) plantations in South African cover approximately 110 000 ha of which 78% belong to commercial farmers, 18% to three corporate companies and 4% to small-scale growers (Chan et al 2015). Wood and bark are both utilized from black wattle plantations for various products. The wood is used primarily in pulp milling for its desired pulping properties. The majority of the wood produced in South Africa is shipped to Japan due to the economic advantages conveyed by the high wood fiber density and pulp yield. In 2013, 800 000 bone dry metric tonnes of black wattle wood chips were exported from South Africa (Chan et al 2015). The bark from black wattle is primarily used as a source of vegetable tannin extract for the tanning of leather products as well as for adhesives. Forty five thousand tonnes of bark extract are produced on average per year from black wattle plantations in South Africa (Chan et al 2015). Charcoal and firewood are also significant products from black wattle plantations, although they are generally produced as secondary products with ca. 200 000 tonnes produced in 2012-13 (Chan et al 2015, DAFF 2015). Black wattle, in conjunction with various *Pinus* and *Eucalyptus* species, is a significant species for smaller timber growers as reliance on multiple species provides a means of flexibility and reduced susceptibility to market price fluctuations (as compared to reliance on production from a single species).

The land available for plantation forestry is limited in South Africa due to forestry being in competition with other land use activities. Planting and water use permits are therefor issued on a limited basis by the South African government (Govender 2007). Optimal production of black wattle on the available land base is therefore necessary to satisfy the market demands and established industries which are dependent on black wattle (Dunlop and MacLennan 2002). To achieve this, mitigating abiotic and biotic risks is one component which can aid in achieving optimum yields. Drought, hail, snow and frost are abiotic risks to black wattle which can largely be avoided by not planting black wattle in areas where these risks occur (Davidson 1989; Sherry 1971). A number of pests and diseases also occur within wattle stands and cause reduced growth and mortalities. Wattle bagworm (Kotochalia junodi Heylaerts), brown wattle mirid (Lygidolon laevigatum Reut.) and various lappet moths of the Lasiocampidae family are common pests of black wattle in South Africa (Dunlop and MacLennan 2002). Common diseases of black wattle include Ceratocystis albifundus Wingfield, De Beer & Morris and various Phythoptora and Botrytosphaeriaceae species (Roux and Wingfield 1997). Research has been conducted for the management of these pest and diseases and has been successful in reducing losses in black wattle plantations.

The most recent disease of black wattle is a rust fungus identified as *Uromycladium acacia* (Cooke) P. Syd. & Syd. 1914 (wattle rust) (McTaggart et al. 2015). Since the detection of wattle rust in 2012, wattle rust has spread throughout the entire wattle growing region of KwaZulu-Natal. It has also been recorded from Limpopo to the Western Cape (McTaggart et al 2015). The disease affects trees of all age classes and is of significant concern to black wattle growers as it causes

reduced growth as well as mortality where severe infections occur. In determining methods to reduce damage caused by the disease, fungicides have been screened for the purpose of managing wattle rust (Little and Payn 2016). However, the products tested in the screening trials recommend re-application every 28 days. Thus, a number of fungicide applications would occur each growing season if the recommended application period were to be adhered to. South African forestry aims to implement practices that are environmentally, socially and economically sustainable. To achieve this, South Africa's forest industry subscribes to the principles of environmental, social and economic responsibility set out by the Forestry Stewardship Council (FCS). According to Forestry South Africa (FSA 2012), approximately 85% of South Africa's plantations are FSC certified. FSC has guidelines regarding the use of pesticide use in forestry (FSC 2005). Reducing the use of pesticide, although not explicitly stated in the guidelines for pesticide use by FSC, would contribute to the appropriate use of pesticides in forestry.

An important consideration when using pesticides is to avoid the development of acquired resistance of the target organism to the pesticide. Managing the development of acquired resistance to fungicides in wattle rust populations is necessary to ensure the long-term utility of fungicides used for controlling the disease. There are a number of recommendations by the Fungicide Resistance Action Committee (FRAC) that can be used to avoid the development of resistance to fungicides (Brent and Holloman 2007). Avoiding repetitive use of a fungicide (from the same fungicide group or mode of action), mixing or alternating fungicides (from separate fungicide groups or modes of action), limiting the number and timing of treatment applications, avoiding eradicant use, maintaining recommended dose rates by the manufacturer and integrating fungicide use with non-pesticide methods are strategies that can be used to reduce the occurrence of resistant strains of fungal pathogens developing (Brent and Holloman 2007). In addition, using multiple strategies in an integrated pest management plan also reduces the likelihood of resistance developing. The fungicides screened for the management of wattle rust all (methyl(α E)-2-[[6-(2-cyanophenoxy)-4-pyrimidinyl]oxy]- α contain azoxystrobin (methoxymethylene), which is considered a fungicide with a high risk for the development of resistance (FRAC, 2016). Using methods to avoid fungicide resistance is therefore essential to ensure that those fungicides containing

azoxystrobin remain effective for managing wattle rust for as long as possible. Limiting the number of applications is one particular resistance management strategy that may not only aid in maintaining long term use of fungicides but also has the potential to reduce costs and non-target damage associated with using fungicides to manage wattle rust. Monitoring the development of resistance is also required to provide information on whether lack of disease control is a result of resistance, or due to other causes and to determine whether resistance management strategies are effective (Brent and Holloman 2007).

Pesticide use may also be improved through the use of an adjuvant due to the additional effects that adjuvants may convey. Improved coverage, absorption, persistence on foliage, improved pesticide translocation and increased efficacy may arise from the use of adjuvants (eg Thompson et al. 1996; Maschhoff et al. 2000; Hart et al. 1992; Young and Hart 1998). Logically, increased pesticide effectiveness may provide greater levels of pest and disease control.

To determine whether the use of adjuvants and reduced fungicide applications would be effective in managing wattle rust, a trial was established at Harding in southern KwaZulu-Natal in October 2015. Two adjuvants (poly-1-p-menthene; borax (sodium tetraboratedecahydrate) + orange oil) and three different application timings (28/42/56 days between application) were tested with azoxystrobin (methyl(α E)-2-[[6-(2-cyanophenoxy)-4-pyrimidinyl]oxy]- α -(methoxy methylene)benzeneacetate),+difenoconazole(1-[[2-[2-chloro-4-(4-chlorophenoxy) phenyl]-4-methyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole).

3.2. Materials and Methods

A site on Sheepwalk farm in Harding in southern KwaZulu-Natal was located in the wattle growing region of South Africa on an existing stand of black wattle (**Table 6**). The site was selected such that it fell within areas where wattle rust was known to occur. The site was also selected so that the trees were between 0.5 and 1.0 m in Height at the time of trial initiation. This would allow for the continued manual spraying of the fungicides onto the foliage of the trees through the growing season when using a knapsack sprayer. In addition, the trees would be in their exponential growth phase, the period during which any negative/positive treatment impacts (if any) would most likely be expressed.

Table 6. Site characteristics for an *Acacia mearnsii* trial initiated in October 2015 in the southern KwaZulu-Natal for testing of adjuvants and application schedules for the control of wattle rust.

Region	Magisterial district	Harding		
.	Plantation/trial name	Sheepwalk		
Latitude Longitude		30° 37' 27.7" S 29° 49' 53" E		
Altitude (m a.s.l.)		957		
MAP (mm)		898		
MAT (°C)		16.4		
Aspect		Undulating, steep, South- facing slope		
Selected soil	Soil form	Inanda 1200		
physical and pesticide	Soil depth (m)	1.2		
properties	Soil texture	Clay		
Spacing (m) Stems per hectar	e (sph)	2 x 3 1 666		
Seed lot/orchard		PSO-10		
Date planted		01/02/2015		
Drought risk	>850 mm	45.1		
(%)*	<650 mm	13.7		
	Climate zone	WT2 (warm temperate)		
Potential productivity*	Growing conditions for species planted	Optimum		
	Site index (age 5)	15		

*Smith et al. 2005

The trial design consisted of a 2 x 4 factorial with three additional treatments, replicated three times and laid out as a randomized complete blocks design (Cochran and Cox 1968). The factorial combination consisted of application schedules (42 and 56 days between application) and four adjuvant combinations (none; poly-1-p-menthene; borax + orange oil; poly-1-p-menthene & borax + orange oil) (**Table 7**). The three additional treatments consisted of one control with no fungicides applied. The other two additional treatments had fungicides applied according to the recommended 28-day schedule (one commencing in October and the other, November). Each treatment plot consisted of a single line of 22 trees, with the inner 20 trees being measured (2 buffer trees at each end). In addition, there were single lines of non-treated trees on either side of the treated rows so as to act as a buffer between adjacent plots.

The fungicides were sprayed onto the foliage to run-off using a 16 L knapsack sprayer, fitted with an air-induction, twin-flat-fan TeeJet Turbo TwinJet[®] AITTJ60 nozzle. This nozzle was selected due to its ability for good canopy penetration and cover. The nozzle is also recommended for the broadcast application of fungicides where good drift control and coarse droplets are required (TeeJet Technologies 2015). Pressure was regulated to 1.5 KPa, resulting in a spraying volume of 669 L ha⁻¹. The rate of fungicide applied was 1 L ha⁻¹ assuming a spray volume of 1000 L water ha⁻¹. The dates when the fungicides were applied, together with the climatic conditions on the day of spraying were recorded (**Table 8**). Standard silvicultural practices were implemented in the trial. Weeding, implemented to remove competition from unwanted vegetation, was implemented in all but four of the plots in the trial. This was unintentional and the un-weeded plots were included in the analysis, and run as a covariate.

3.2.1. Assessments

Tree Height and Groundline Diameter were taken on three occasions, at trail initiation, three months after trial initiation and one month after the final fungicide application (Table 8). Biomass Index was calculated as Gld² x Ht and provides a good index of overall tree performance in young trees (Eccles et al. 1997). Tree Condition was quantified through the visual estimation of a combination of disease symptoms. A modified Braun Blanquet method (Kent and Coker 1996) for the estimation of the area of each tree affected by wattle rust was used, whereby increasing values are assigned, based on increasing cover (Table 9). This method of overall assessment of Tree Condition was selected as cover estimates are not biased by tree size. As the visual expression of any fungal pathogen varies according to the type, stage and severity of infection, a number of visible symptoms were scored so as to quantify Disease Expression. These were adapted from symptoms as described by Dick (2009) and included the scoring of trees for teliospore masses (brown pustules) on the leaves, deformed pinnules or pinnae and stem lesions (teliospore masses on the main stem, or branches) according to the volume of tree affected, where 0, 1, 2 and 3 = 0, 1-25%, 26-50% and 3 = +50%affected respectively.

Table 7. Treatments, including adjuvant-related information, tested for the control of wattle rust in an *Acacia mearnsii* trial initiated in October 2015 in the southern KwaZulu-Natal.

Treatments	Fungicide Trade Name	Fungicide Active Ingredient (g L ⁻¹)	Adjuvant Trade Name ¹	Adjuvant Active Ingredient (g L ⁻¹)	Rate of adjuvant ha ⁻¹ assuming spray volume 1000 L ha ⁻¹	Application Period	Spray Starting Date
1	Amistar Top [®]	azoxystrobin (200 g L ⁻¹)	-	_	_	42	30/10/2015
2	Amstar rop	difenoconazole (125 g L ⁻¹)				56	30/10/2015
3	Amistor Top [®]	azoxystrobin (200 g L ⁻¹)	Nu Film D [®]	poly 1 p monthone (975 g 1^{-1})	1250 ml	42	30/10/2015
4	Amistal Top	difenoconazole (125 g L ⁻¹)	NU-FIIII F	poly-1-p-mentitiene (875 g L)	1230 111	56	30/10/2015
5	Amistar Top [®]	azoxystrobin (200 g L ⁻¹)	Orocorb [®]	borax (10 g L ⁻¹)	208 ml	42	30/10/2015
6	Amistal Top	difenoconazole (125 g L ⁻¹)	Olosoid	orange oil (50 g L ⁻¹)	206111	56	30/10/2015
7	. .	azoxystrobin (200 g l ⁻¹)	Nu-Film P [®]	poly-1-p-menthene (875 g L ⁻¹)	1250 ml	42	30/10/2015
8	Amistar Top"	difenoconazole (125 g L^{-1})	Orosorb [®]	borax (10 g L ⁻¹) orange oil (50 g L ⁻¹)	208 ml	56	30/10/2015
	-	Additional Treatments	s (Controls)				
9	Control	-	-	-	-	-	-
10	Control	-	-	-	-	-	-
11	Amistor Ton [®]	azoxystrobin (200 g L^{-1})				28 days	02/10/2015
12	Amisiai Top	difenoconazole (125 g L ⁻¹)	-	-	-	28 days	30/10/2015

Table 8. Sequence of events in terms of assessment dates and the application of treatments in an Acacia meansii trial initiated inOctober 2015 in southern KwaZulu-Natal.

Trial Sheepwalk										
Assessment dates 02/10/2015				08/01	/2016		29/03/2016			
Tree age when assessed (days) 243					33	34		415		
Application data		1 st	2 nd	3 rd		4 th	5 th	6 th	7 th	8 th
Application da	le	2/10/2015	30/10/2015	16/11/20	15 29/11/2015 24/12/2015 18		18/1/2016	10/2/2016	19/2/2016	
	Time of spraying (hrs)	14:42 - 16:57	10:28 – 11:41	15:40 - 17	7:10	08:40 - 10:55	10:48 – 13:50	12:04 – 15:11	08:57 – 10:43	09:07 – 09:51
Conditions at	Temperature shade (°C)	22.6	30.0	20.1		24.5	33.5	24.9	24.7	28.8
application	Relative humidity (%)	74.0	60.3	78.4		71.1	60.7	71.1	70.9	72.1
	Wind speed (m s ⁻¹)	9.7	3.0	9.4		4.7	7.6	4.5	1.9	3.1

As the trees at showed signs of the presence of wattle rust disease from the 2015 growing season, the third assessment of Tree Condition and Disease Expression was calculated relative to the first assessment.

Table 9. *Acacia mearnsii* "Tree Condition" quantified through the visual estimation of a combination of defoliation, discolouration and disease symptoms. A modified Braun Blanquet method (Kent and Coker, 1996) for the estimation of the area of each tree affected was used, whereby increasing values were assigned, based on increasing cover affected.

Tree Condition score	Percentage area affected	Median value used for analyses
1	rare: 1-2 leaflets (less than 5 %)	1
2	few: 3 - 4 leaflets (less than 5 %)	2
3	many: 5 - 10 leaflets (less than 5 %)	3
4	abundant: > 10 leaflets (less than 5 %)	4
5	5 -12 %	8.75
6	12.5 - 25 %	18.75
7	25 - 50 %	37.5
8	50 - 75 %	62.5
9	75 -100 %	87.5

3.2.2. Analysis

Plot means from the final assessment date were analysed as a 2 x 4 factorial with three additional treatments using Statistica for Windows (Dell 2015). Prior to the analysis, the data were checked to ensure that the assumptions for a valid ANOVA were not violated (**Table 10**). Before any comparison between treatments, an *F*-test was carried out to determine the overall significance of the differences between all treatment means within the experiment. Only if the *F*-value was significant, were treatment differences further investigated using least significant differences (*Isd's*). The first measurement was used in addition to the presence of weeds as co-variates for subsequent measures. These were significant as co-variates and therefore were included in the final analysis so as to stabilize the variance. Changes in Tree Condition and Disease Expression were calculated relative to initial assessments (and expressed as a percentage) and are displayed using descriptive graphing techniques (**Figures 3c** and **2**).

Table 10. Summary of analyses of variance showing means squares for selected tree variates at the final assessment date (415 days after planting) for an *Acacia mearnsii* trial initiated in October 2016 in southern KwaZulu-Natal for testing of adjuvants and application timing for the control of wattle rust. The first assessment (243 days after planting) and un-weeded plots are included as covariates.

Source of variation	d.f.	<i>Ht</i> (m)	Gld (cm)	Bĺ¹
Rep	2	0.423**	0.803*	558.011**
control.rest of treatments	3	0.084	0.84*	323.513**
control.adjuvant	3	0.0434	0.155	52.369
control.application timing	1	0.001	0.005	7.578
control.adjuvant.application timing	3	0.043	0.022	5.669
Weeds covariate	(1)	0.0002	1.496**	454.478**
Measurement 1 covariate	(1)	1.051**	3.423**	2116.962**
Residual	29 (27)	0.063	0.184	57.785
Total	42			
Grand Mean		2.46	3.88	64.96
Standard error of the difference		0.25	0.43	7.60
(control.adjuvant.application timing)				
Coefficient of variation (units)	10.22	11.05	11.70	
Levene's test for homogeneity of variar	nce,	1.87 ^{\$}	1.75 ^{ns}	1.89 ^{\$}
Shapiro-Wilk test for Normality		0.97 ^{ns}	0.98 ^{ns}	0.98 ^{ns}

Note: ^{\$}, * and ** indicates significance at *F*-prob <0.10, 0.05 and 0.01; and ^{ns}, non-significance. ¹Biomass Index was square root transformed

3.3. Results and Discussion

A site was selected in the black wattle commercial growing region of southern KwaZulu-Natal to test the effectiveness of different application schedules of fungicides, as well as the use of adjuvants for the management of wattle rust. By the end of the trial period, tree growth, in terms of Groundline Diameter and Biomass Index, was negatively impacted by rust (**Table 10**, **Figure 3**). Height was not as affected by rust infection in either the sprayed treatments or controls. No significant difference was detected between the application schedules nor between the adjuvants tested. However, spraying, which commenced in October on a 28-day schedule, had a lower growth response, albeit not significant, for both Groundline Diameter and Biomass Index in comparison to the other treatments (**Figure 3**). Lowered growth in the 28-day October treatment may be due to phytotoxicity, caused by over application of fungicide for the level of wattle rust observed.

However, any phytotoxicity recorded was difficult to differentiate from the Disease Expression symptoms recorded (**Figure 4**). Phytotoxicity, as a result of fungicide use, was also suspected to have occurred in a trial by Little and Payn (2015) where one of the trials (Commondale) had spraying commence before wattle rust was observed in the trial. In both instances, the application of fungicides had greater benefits than the effect of phytotoxicity. All of the fungicide application schedules and adjuvant mixtures were effective in controlling wattle rust. Although the fungicides were applied to trees already infected by rust, they were effective as a corrective treatment, as supported by Little and Payn (2016).



Figure 3. Groundline Diameter (a), Biomass Index (square root transformed) (b) and relative Tree Condition (c) for an *Acacia mearnsii* trial initiated in October 2015 in southern KwaZulu-Natal for testing fungicide application schedules and adjuvants for the management of wattle rust. Letters on bars (figures 3a and 3b) indicate significance at p< 0.05. Standard Error bars are included on Figure 3c.

Tree Condition, analyzed at the third measurement relative to the first, was also reduced due to the application of fungicides in untreated plots (**Figure 4**). Tree Condition in untreated controls (1.8%) was notably lower in relation to a previous trial conducted on the use of fungicides for the management of wattle rust (Little and Payn 2016). This is likely as a result of the difference in climatic conditions, particularly rainfall, experienced between the two sets of trials. Rust fungi epidemiology is known to be associated with climatic conditions, such as moisture, temperature, wind etc. (Agrios 2005). Relative Disease Expression was reduced among all treatments, including controls (**Figure 4**). Reduced rainfall is likely to also have caused the lowered Disease Expression.



Figure 4. The relative occurrence for an *Acacia mearnsii* trial initiated in October 2015 in southern KwaZulu-Natal for testing fungicide application schedules and adjuvants for the management of wattle rust. Treatments means are shown as solid bars, the 95% confidence levels by the boxes, and the standard deviation by the bars.

Replications in the trial were located across the slope to reduce variation associated with the slope of the trial location. Replicates (Rep) were highly significant (*F*-prob< 0.01) and accounted for the highest portion of the variation for all tree growth variates (**Table 10**). This may be attributed to variations in soil depth down the slope profile.

Application of fungicides need to be timed according to climatic conditions associated with rust growth, as was indicated by the lowered growth response due to the 28-day application schedule commencing in October. This needs to occur in conjunction with the optimum number applications for managing the development of acquired resistance and that will not result in phytotoxicity.

3.4. Conclusion

Tree Groundline Diameter and Biomass Index were negatively impacted by wattle rust, irrespective of previous infection. Height was not significantly impacted by wattle rust, likely due to the low incidence of wattle rust observed. All of the adjuvants and fungicide application schedules tested were effective for the management of wattle rust. Phytotoxicity may arise from excessive applications when wattle rust infection is low. Adjuvants and application schedules will need to be selected according to the most cost effective strategy as well as in a manner as to avoid the resistance developing. Monitoring the effectiveness of fungicide applications in field is essential for monitoring acquired resistance.

Fungicides currently registered for the control of wattle rustcontain azoxystrobin, difenoconazole and cyproconazole. As cyproconazole use is listed as highly hazardous by the FSC, azoxystrobin and difenoconazole are currently the only fungicides registered for the management of wattle rust. Alternative fungicides from different groups/modes of action need to be tested for their effectiveness in managing wattle rust, to avoid resistance developing to the two currently registered fungicides. This will allow a section of fungicides that can be used in combination and/or alteration which will reduce the likelihood of resistance developing in wattle rust. Combining multiple methods of managing wattle rust, such as planting resistant/tolerant varieties of black

wattle with the use of various fungicides will also contribute to reducing the likelihood of resistance.

Testing a number of additional fungicides from different fungicide groups/modes of action to those tested for the management of wattle rust needs to be conducted. This will provide a selection of fungicides that can be used in mixtures and/or alteration to avoid acquired resistance developing in wattle rust populations. Due to the influence of climatic variables on wattle rust occurrence, further trials to test the use of adjuvants and altered timing is recommended under different climatic conditions to understand the influence weather has on treatments.

Chapter 4: Use of Regression Trees to link *Uromycladium acaciae* symptoms to *Acacia mearnsii* growth variates

Abstract

From 2012/13 Uromycladium acaciae (wattle rust) has spread throughout the Acacia mearnsii (black wattle) growing area of South Africa. The newly emerged disease affects trees of all age classes and causes growth reductions and mortalities with severe infestations. Fungicides have been tested and have been found to be effective for managing the disease. Timing of fungicide application is necessary for optimal use of these fungicides. Fungicide applications could potentially be linked to the emergence of different wattle rust symptoms to optimize fungicide use. Wattle rust symptoms were analysed from the untreated control plots of two trials, one in the KwaZulu-Natal midlands and one in southern KwaZulu-Natal, to determine whether wattle rust Disease Expression could be linked to black wattle tree growth. Regression trees were used for the analysis, as linear and multiple regression techniques would be unsuitable for the type of data collected. Regression trees were overfitted and attempts at testing the robustness of the model by cross-validation were unsuccessful. No individual symptom emerged as a significant predictor of tree growth, indicating that fungicide application should take place with the onset of any of the wattle rust symptoms tested.

4.1. Introduction

South Africa has ca. 110 000 ha planted to *Acacia mearnsii* De Wild. (black wattle) (Chan et al. 2015). The species is grown primarily for its bark tannin extracts and wood (Griffin et al. 2011). Currently 85% of the revenue from the species is obtained from the timber, and 15% from the bark (Chan et al. 2015). South Africa produces ca. 45 000 tonnes of bark extract annually and in 2015, exported 800 000 bone dry metric tonnes (BDMT) of black wattle wood chips (Chan et al. 2015). Of the area planted to black wattle, 78% (86 000 ha) is owned by 600 private commercial growers who rely on the species as one of their primary timber crops. Different species (from three genera: *Pinus, Eucalyptus* and *Acacia*) are planted by these private growers so that they are

less susceptible to negative market forces, with the dual income (bark and timber) generated from black wattle enabling them to achieve this objective. The preservation and continuation of the black wattle industry in South Africa is thus important, and when challenges arise that negatively impact on the sustainability of this industry, these risks need to be adequately managed.

Black wattle productivity is influenced by a number of pests and diseases, the most recent of which is Uromycladium acacia (wattle rust) which has emerged in South Africa since 2012/13. The disease has been documented from Limpopo to the Western Cape. Although fungicide screening trials were initiated to provide short-term control of the disease (Little and Payn, 2016), determining when management intervention is required (fungicides or otherwise) is necessary for the cost-effective, long-term management of wattle rust. Management intervention can be scheduled based upon a combination of four main indicators, namely climatic, economic, biological or epidemiological. Climatic indicators refer to changes in temperature, humidity and precipitation, such as the timing of insecticide application to coincide with insect degree days (Murray 2008). Economic indicators can be based upon levels at which the cost of intervention is exceeded by the loss that would occur without intervention, often termed 'economic injury level' (Stern et al. 1959; Norris et al. 2003; Radcliffe et al. 2009). Biological indicators are linked to the life cycles of both the pest or disease and its host (Agrios 2005). For example, certain stages of the host's life cycle may be linked to an increase in pest or disease abundance, such as the commencement of flowering, and can act as queues for the implementation of control measures. Intervention in pest and disease life cycles, while varied and complex, are often linked to stages at which the pest or disease is vulnerable to management inputs. Epidemiological indicators are determined so as to coincide with the pest or disease population reaching a significant enough level (on a local or regional scale) to warrant intervention. These indicators are critical to determine if a management strategy is to be timed correctly for optimum effectiveness.

To determine if wattle rust symptoms could be linked to tree growth performance (and hence be used as an indicator for management intervention), a number of disease and tree growth variates were recorded and analysed in four trials in KwaZulu-Natal and Mpumalanga.

4.2. Materials and Methods

Trials were established on four contrasting sites, City Forestry and Etterby (KwaZulu-Natal Midlands), Sheepwalk (southern KwaZulu-Natal) and Commondale (SE Mpumalanga), located in the wattle growing regions of South Africa on existing stands of *A. mearnsii* (**Table 11**). Of these, four trials were implemented from which two were selected for more detailed disease symptom analysis, as part of a research initiative to determine the effectiveness of fungicides for managing wattle rust.

Table 11. Site characteristics and assessment dates for two *Acacia mearnsii* trials initiated in October/November 2014/15 in southern KwaZulu-Natal and Midlands for testing of fungicides for the control of wattle rust.

Pagion	Magisterial district	Р	ietermaritzbur	g		Harding		
Region	Plantation/trial name		City Forestry			Sheepwalk		
Latitude		2	29° 34' 52.69" S	6		30° 37' 27.7" S	;	
Longitude		3	30° 19' 52.28" E	Ē	29° 49' 53" E			
Altitude (m a.s.l	.)		1 075		957			
MAP (mm)			904			898		
MAT (°C)			15.8			16.4		
Aspect		uniform,	gentle ENE-fac	ing slope	undulating,	steep, South-f	acing slope	
Selected soil	Soil form		Inanda 1100		Inanda 1200			
physical and	Soil depth (m)		0.50		1.2			
pesticide	Soil toxturo	Clay				Clay		
properties	Solitexture				City			
Spacing (m)		3 x 1.8				2 x 3		
Stems per hecta	are (sph)	1 852 sph			1 666			
Seed lot/orchard	d	PSO-10			PSO-10			
Date planted			05/03/2014		01/02/2015			
Drought risk	>850 mm		45.1			45.1		
(%)	<650 mm		13.7			13.7		
	Climate zone		cool temperate		N	warm temperate	e	
Potential	Growing conditions for		Ontimum			Ontimum		
productivity	species planted	Optimum				Optimum		
	Site index (age 5)	15			15			
Assessment dates		24/10/2014	09/12/2014	11/03/2015	02/10/2015	08/01/2016	29/03/2016	
Tree age when a	assessed (days)	233	279	371	243	334	415	

The control plots from two trials were selected for further regression tree analysis, as within these two trials there was the uninhibited growth of wattle rust over the monitoring period (**Figure 5**) (Little and Payn, 2016).

4.2.1. Assessments

Tree Height (Ht in m) and Groundline Diameter (Gld in cm) were measured on three occasions, at trail initiation and then at one month after the second and fourth application of the fungicides (**Table 11**). Biomass Index was calculated as Gld² x Ht and provides a good index of overall tree performance in young trees (Eccles et al. 1997). Biomass Index received a square root transformation prior to the analysis to normalize the variance caused by squaring the data. For use as possible co-variates when analysing individual tree data, all trees were scored for the presence of blanking (replanting of dead seedlings), double stems, multiple leaders, browsing, dead tops or any physical damage to the base of the stems from manual weeding operations. Tree Condition was quantified through the visual estimation of a combination of defoliation, discolouration and Disease Expression. A modified Braun Blanquet method (Kent and Coker 1996) for the estimation of the area of each tree affected was used, whereby increasing values are assigned, based on increasing cover (**Table 12**). This method of overall assessment of Tree Condition was selected as cover estimates are not biased (influenced) by tree size.

As the visual expression of any fungal pathogen varies according to the type, stage and severity of infection, a number of symptoms were scored so as to quantify Disease Expression. These were adapted from symptoms as described by Dick (2009) and included the scoring of trees for teliospore masses (brown pustules) on the leaves, deformed pinnules or pinnae and stem lesions (teliospore masses on the main stem, or branches) according to the volume of tree affected, where 0, 1, 2 and 3 = 0, 1-25%, 26-50% and +50% affected respectively. In addition, spermagoinia were recorded in the Sheepwalk trial.

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Table 12. Acacia mearnsii "Tree Condition" quantified through the visual estimation of a combination of defoliation, discolouration and Disease Expression. A modified Braun Blanquet method (Kent and Coker, 1996) for the estimation of the area of each tree affected was used, whereby increasing values were assigned, based on increasing cover affected.

Tree Condition	Percentage area affected	Median value used
score		for analyses
1	rare: 1-2 leaflets (less than 5 %)	1
2	few: 3 - 4 leaflets (less than 5 %)	2
3	many: 5 - 10 leaflets (less than 5 %)	3
4	abundant: > 10 leaflets (less than 5 %)	4
5	5 -12 %	8.75
6	12.5 - 25 %	18.75
7	25 - 50 %	37.5
8	50 - 75 %	62.5
9	75 -100 %	87.5

4.2.2. Analysis

To determine the link (if any) between Disease Expression and tree growth, regression trees were used to determine their relationship and relative importance using Statistica for Windows (StatSoft Incorporated 2013). Simple linear and multiple regression techniques would be inappropriate for the analysis due to the predictor variables (Disease Expression) being categorical and unbalanced, and therefore regression trees (Brieman et al. 1984) were used. In addition, co-linearity occurred among the different wattle rust symptoms. Regression trees display the associations between each symptom and proxy for Groundline Diameter, Height and Biomass Index (Figure 6). Each regression tree was developed and partitioned according to the impact of that specific symptom on tree growth. Each node of the regression tree displays the mean, variance (var) and number of data points (n) of the data associated with that node (predicted by the symptom). Where each regression tree node splits, the node displays either an increase in growth with a decrease in symptom severity, or a decrease in growth as a consequence of increased symptom severity. Due to the expression of wattle rust from the second measurement date onwards, the response variable of tree growth difference (period between second and third measurement) was

used within the regression trees. For the development of the regression trees, the response data from Control plots was used as no fungicides were applied in these plots, and therefor Disease Expression developed without hindrance.



Figure 5. The relative occurrence of Disease Expression for three *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga for testing of fungicides for the control of wattle rust. Treatments means are shown as solid bars, the 95% confidence levels by the boxes, and the standard deviation by the bars. Abbreviations used are TM (teliospore masses), PR (powdery

residues), DP (deformed pinnules), DL (deformed leaflets), CL (curling leaflets) and SL (stem lesions).



Figure 6. The relative occurrence of Disease Expression for one *Acacia mearnsii* trial initiated in October 2015 in southern KwaZulu-Natal for testing of adjuvants and altered fungicide applications for the control of wattle rust. Treatments means are shown as solid bars, the 95% confidence levels by the boxes, and the standard deviation by the bars.

Only data from City Forestry and Sheepwalk were considered for the development of regression trees due to the low and delayed expression of wattle rust at Commondale, and the severity of wattle rust at Etterby.



Figure 7: Regression tree for Groundline Diameter (a) and Height (b) for one *Acacia mearnsii* trial initiated in October/November 2014 in the KwaZulu-Natal Midlands for testing of fungicides for the control of wattle rust.

4.3. Results and Discussion

Disease Expression recorded at City Forestry, Commondale and Etterby (**Figure 5**) was notably greater than that recorded at Sheepwalk (**Figure 6**). In addition, Disease Expression was greater in untreated controls than treatments for both trials (**Figures 5 and 6**). This is most likely due to differences in climate for the periods in which the two series of trials were conducted, with an average rainfall of 76 mm from October 2014 to March 2015 and 66 mm from October 2015 to March 2016 (SAWS 2016). The higher rainfall at City Forests over which the trial was conducted was more conducive for the development and expression of wattle rust.

Regression trees were developed for both City Forestry and Sheepwalk. However, these were overfitted due to the model containing too much random error and therefore in need validation. Overfitting can be reduced through pruning, crossvalidation and v-fold cross-validation (Statsoft 2015). Using v-fold cross-validation to assess the robustness of the regression trees led to the regression trees being reduced to a single node in all but two cases, with no symptom emerging as significant from any other. Attempts to test the robustness of the regression trees by building a model using City Forestry data, and cross-validating using Sheepwalk data (and vice versa) were unsuccessful. Two regression trees which were robust enough to predict growth (after using v-fold cross-validation) were for Groundline Diameter and Height, assessed as a relative increase in growth (**Figure 7**) for City Forestry data. Powdery Residues emerged as a significant predictor of Groundline Diameter growth reduction and Deformed Pinnules emerged as a significant predictor of Height growth reduction. However, this result was not replicated using Sheepwalk data.

A number of possibilities may explain the failure of the regression tree models to distinguish between the various Disease Expression symptoms:

- The data used for the building of the regression tree models may be insufficient for both data sets (n<112), which provides limited data from which to develop a model.
- The categories used to score the symptoms may have been too broad to distinguish between the symptoms.
- The relatively low abundance of Disease Expression recorded at Sheepwalk (Figure 6), in relation to those observed at City Foresty (Figure 5), may also explain why no individual symptom emerged as significant (from any other) when fitting regression trees using Sheepwalk data.
- Symptoms may also not be distinguishable from each other, as none may be a significant predictor of growth reduction when compared with each other.

4.4. Conclusion

No wattle rust symptoms emerged as a significant predictor of tree growth when using the two data sets used for the development of regression trees. The scoring method attempted to distinguish the individual symptoms and their severity to differentiate if any symptoms could be used as a predictor for tree growth. Although the method distinguished individual symptoms, the categories used to assess individual symptoms may have been too broad, and hence led to the lack of normal distribution in the data. The frequency of observations, primarily used in the trials for testing differences in tree growth, may have been too infrequent when used to differentiate Disease Expression development over time. A greater number of disease severity categories, coupled with more frequent observations, may give rise to more revealing results. In addition, it may not be possible to differentiate individual symptoms based upon their impact on tree growth, due to multi-collinearity occurring between the symptoms. There may also be no direct link between individual symptoms and tree growth.

CHAPTER 5: Economics of fungicide application schedules for the control of wattle rust (*Uromycladium acaciae*) in *Acacia mearnsii* plantations, South Africa

Abstract

Acacia mearnsii (black wattle) plantations in South African cover approximately 110 000 ha. Uromycladium acaciae (wattle rust) has spread over Limpopo to the Western Cape of South Africa. The disease of black wattle causes reductions in growth and mortalities in severe infections. In October 2014 six trials were initiated in Mpumalanga and KwaZulu-Natal to determine the effectiveness of fungicides, varied application schedules, and adjuvants for the management of wattle rust. Relative growth for Biomass Index was compared to untreated controls to obtain comparisons within and between sites. Costs versus benefit were compared using a two-way table to determine the most optimum treatment. The largest portion of treatment costs was attributed to the cost of fungicide. No treatment was found to be optimal for the recommended rate of application. The use of adjuvants increased the cost of treatment, without additional benefit in growth. Control of wattle rust is beneficial, although costly if over-applied. Rotation-end data is required to determine whether fungicide use is economical for managing wattle rust over an extended period of time.

5.1. Introduction

Acacia mearnsii De Wild (black wattle) is a commercially grown species in South Africa, with black wattle plantations covering ca. 110 000 ha within South Africa (Chan et al. 2015). The species is of significant commercial value, particularly to private timber growers who maintain 78% (86 000 ha) of the black wattle grown area (Chan et al. 2015). The species is utilized for its bark and wood. The bark is used primarily for its tannin extracts, which are used for leather production and the wood is used for woodchip exports due to the high price obtained (Chan et al. 2015). Charcoal and firewood are also produced as secondary products.

Black wattle has a number of pests and diseases which may cause growth losses if left unmanaged. *Kotochalia junodi* Heylaerts, *Lygidolon laevigatum* Reut and

various lappet moths of the Lasiocampidaie family are common insect pests of black wattle (Dunlop and MacLennan 2002). Notable diseases include Ceratocyctis albifundus Wingfield, De Beer and Morris and various *Phytophthora* and *Botrosphaeriacia* species (Roux and Wingfield 1997). The most recent significant disease of black wattle is Uromycladium acaciae De Wild (wattle rust), which has caused growth reductions and mortalities in black wattle plantations (Little and Payn 2016). Due to the recent occurrence of wattle rust (from 2012), management strategies are still being developed for its control. As part of this process, selected fungicides were tested for their potential for managing wattle rust (Little and Payn 2016). Decisions on the use fungicides cannot be based solely on the effectiveness of the fungicides in managing the disease. Rather the determination of treatment costs relative to growth benefits is important as this will take into consideration the various levels of pests and diseases that can be tolerated before the cost of managing them exceeds the benefit gained (Norris et al. 2003). For example, the economic injury level (Stone and Pedigo 1972) is a threshold which can be used to establish when to manage a pest or disease (Norris et al. 2003), where the cost of managing a pest or disease is required for determining the economic injury level.

Rotation-end data comparing black wattle growth with and without the application of fungicides is required to fully understand the economics of fungicide use. Due to the long term nature of rotation-end research and the current need for interim management recommendations relating to wattle rust, the costs associated with fungicide from shortterm trials may provide an indication upon which management decisions can be based.

Data obtained from six fungicide trials for the control of wattle rust were obtained to compare the costs associated with different fungicide application schedules. All trials contained fungicide application treatments, as well as an untreated control. Growth reductions at the end of one growing-season were compared to treatment costs to determine the effectiveness of the application of various fungicides, albeit with different application regimes.

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5.2. Materials and Methods

Six trials were conducted as part of a wattle industry research initiative (Wattle Rust Working Group) to determine the effectiveness of fungicides for the management of wattle rust (Table 13). Three trials were initiated to screen potential fungicides and were located in the KwaZulu-Natal Midlands and southern Mpumalanga. Azoxystrobin $(methyl(\alpha E)-2-[[6-(2-cyanophenoxy)-4-pyrimidinyl]oxy]-\alpha-(methoxymethylene)$ benzeneacetate) and difenoconazole (1-[[2-[2-chloro-4-(4-chlorophenoxy)phenyl]-4methyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole) were applied in all trials. cyproconazole1(α -(4-chlorophenyl)- α -(1-cyclopropylethyl)-1H-1,2,4-triazole-1-ethanol) was applied in one trial. Two adjuvants (poly-1-p-menthene; borax (sodium tetraboratedecahydrate) + orange oil) were applied in one trial. One trial, located in southern KwaZulu-Natal, was initiated to determine the optimum timing of application (if any), and the potential for fungicide + adjuvants for prolonging the period before reapplication was required. Two wattle rust exclusion trials, in which wattle rust was either controlled or not were located in the KwaZulu-Natal Midlands to assess the impact of wattle rust on tree growth. The six trials covered a range in terms of site productivities (and hence growth), and were located in areas in which wattle rust was known to occur.

Fungicides were applied at varying rates and application schedules using a spray volume of between 500 - 1000 L water ha⁻¹ (**Tables 14 and 15**). The timing and application rates of fungicide applied on each occasion was recorded. The cost of fungicides were: azoxystrobin + cyproconazole R895.20 L⁻¹; azoxystrobin + difenoconazole R785.30 L⁻¹; and azoxystrobin R664.00 L⁻¹. The costs of the adjuvants tested were: borax (sodium tetraboratedecahydrate) + orange oil R87.00; and poly-1-p-menthene R208.00 (**Tables 16, 17 and 18**). One man-day ha⁻¹ at R 138.34 ha⁻¹ was assumed for fungicide application using a knap-sack sprayer (excluding cost of fungicide and adjuvant). The costs used are provided for the purpose of comparing principles, and the actual prices of fungicides and adjuvants may differ according to supplier and formulation of the active ingredients.

Table 13. Site characteristics for six *Acacia mearnsii* trials initiated in 2014/2015 in the KwaZulu-Natal/SE Mpumalanga for testing of fungicides, rates of application, adjuvants and application schedules for the control of wattle rust.

Desien	Magisterial district	Harding	Pietermaritzburg	Richmond	Paulpietersburg	Hilton
Region	Plantation/trial name	Sheepwalk	City Forestry	Etterby	Commondale	Hilton College
Latitude Longitude		30° 37' 27.7" S 29° 49' 53" E	29° 34' 52.69" S 30° 19' 52.28" E	29° 50' 30.17" S 30° 11' 24.33" E	27° 18' 26.78" S 30° 46' 13.75" E	29° 28' 29.58'' S 30° 18' 50.01'' E
Altitude (m a.s	s.l.)	957	1 075	1 094	1 100	780
MAP (mm)		898	904	968	908	1117
MAT (°C)		16.4	15.8	16.8	18.1	17.5
Aspect		Undulating, steep, South- facing slope	uniform, gentle ENE-facing slope	uniform, moderate NW- facing slope	uniform, gentle S- facing slope	Uniform,gentle South West facing slope
Selected soil	Soil form Inanda 1200		Inanda 1100	Griffon 2100	Clovelly	Inanda 1200
physical and pesticide	Soil depth (m)	1.2	0.50	0.80	1.00	-
properties	Soil texture	Clay	Clay	Clay/Loam	Clay	Sandy/Clay
Spacing (m) Stems per hec	tare (sph)	2 x 3 1 666	3 x 1.8 1 852 sph	3 x 1.8 1 852 sph	3 x 1.8 1 852 sph	3 x 1.5 2222 sph
Date planted		01/02/2015	05/03/2014	02/12/2013	05/02/2014	23/10/2014
Drought risk	>850 mm	45.1	45.1	74.9	70.9	-
(%)*	<650 mm	13.7	13.7	5.0	3.6	-
	Climate zone	WT2 (warm temperate)	CT8 (cool temperate)	WT3 (warm temperate)	WT8 (warm temperate)	WT6 (warm- temperate)
Potential productivity*	Growing conditions for species planted	Optimum	Optimum	Optimum	Optimum	Optimum
	Site index (age 5)	15	15	18	20	-

*Smith et al. 2005
Trial	Treatments	Fungicide trade name ¹	Fungicide Active ingredient (g L ⁻¹)	Rate of application of fungicide ha ⁻¹ assuming a spray volume of 1000 L ha ⁻¹	Adjuvant Trade Name ¹	Adjuvant Active Ingredient (g L ⁻¹)	Rate of application of adjuvant ha ⁻¹ assuming spray volume 1000 L ha ⁻¹	Application Period
City Forestry, Commondale, Etterby	1 2 3	Ortiva [®]	azoxystrobin (strobilurin) (250 g L ⁻¹)	500 ml (0.5%) 1000 ml (1.0%) 2000 ml (2.0%)	-	-	-	28
	4 5 6	Amistar Top [®]	azoxystrobin (strobilurin) (200 g L ⁻¹) difenoconazole (triazole) (125 g L ⁻¹)	500 ml (0.5%) 1000 ml (1.0%) 2000 ml (2.0%)	-	-	-	28
	7 8 9	Amistar Xtra [®]	azoxystrobin (strobilurin) (200 g L ⁻¹) cyproconazole (triazole) (80 g L ⁻¹)	500 ml (0.5%) 1000 ml (1.0%) 2000 ml (2.0%)	-	-	-	28
	10 11	Control Control	-	-	-	-	-	-
	1 2 3		azoxystrobin (strobilurin) (200 g L ⁻¹) difenoconazole (triazole) (125 g L ⁻¹)		- Nu Film D [®]	-	- 1250 ml	42 56 42
	4 5 6	Amistar Top [®]		1000 ml (1.0%)	Orosorb [®]	borax (10 g L $^{-1}$) orange oil (50 g L $^{-1}$)	208 ml	56 42 56
Sheepwalk	7 8				Nu-Film P [®] Orosorb [®]	poly-1-p-menthene (875 g L ⁻¹) borax (10 g L ⁻¹)	1250 ml 208 ml	42 56
	9 10	Control Control	-	-	-	orange oll (50 g L) - -	-	
	11 12	Amistar Top [®]	azoxystrobin (strobilurin) (200 g L ⁻¹) difenoconazole (triazole) (125 g L ⁻¹)	1000 ml (1.0%)	-	-	-	28
Etterby, Hilton	1 2	Amistar Top [®]	azoxystrobin (strobilurin) (200 g L ⁻¹) difenoconazole (triazole) (125 g L ⁻¹)	500 ml (0.5%)	-	-	-	28

Table 14. Treatments, including fungicide-related information, tested for the control of wattle rust in six *Acacia mearnsii* trials initiated in October/November 2014 in the KwaZulu-Natal Midlands, southern KwaZulu-Natal and SE Mpumulanga.

Table 15. Sequence of events in terms of assessments dates and the application of fungicides in six Acacia meansii trials initiated

 in October/November 2014 in the KwaZulu-Natal Midlands and SE Mpumulanga.

Trial	City Forestry					Etterby			Commondale				
Assessment dates	24/10/201	4 09/	12/2014	11/03/2015	24/1	0/2014	10/12/2014	11/0	3/2015	22/11/2014	12/01	2015	03/04/2015
Tree age when assessed	233		279	371	3	326	373	2	464	290	34	1	422
Funciaida anniaction data	1 st	2 nd	3 rd	4 th	1 st	2	nd	3 rd	4 th	1 st	2 nd	3 rd	4 th
Fungicide application date	23/10/2014	20/11/2014	13/01/2015	03/02/20	15 23/10/2	014 20/11	/2014 13/	01/2015 0	4/02/2015	22/11/2014	10/12/2014	05/02/2015	09/03/2015
Days between re- application	0	28	54	21	0	2	8	54	22	0	18	57	32
Trial						Sheepwalk							
Assessment dates	02/10/2015					08/01/2016			29/03/2016				
Tree age when assessed (days)	243						334			415			
For visida and testion data	1 st		2 nd	:	3 rd	4 th		5 th		6 th	7 th		8 th
Fungicide application date	2/10/201	15	30/10/2015	16/1	1/2015	29/11/201	5	24/12/2015	1	8/1/2016	10/2/2016	;	19/2/2016
Trial							Etterby						
Assessment dates	з	30/10/2014		30/09/2015 23/02/2016			10/05/2016		04/10/2016				
Tree age when assessed		07		342		488				565 743			
Fundicide application date	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th	8 th	9 th	10 th	11 th	12 th	13 th
	30/10/2014	17/12/2014	30/01/2015	20/03/2015	29/04/2015	02/07/2015	07/08/2015	27/08/2015	22/09/20	15 19/10/2015	19/11/2015	07/12/2015	27/01/2016
Days between re- application	0	48	44	49	40	64	36	20	26	27	31	18	51
Trial	Hilton College												
Assessment dates	2	27/10/2014		03/02/2015			29/02/2016			19/05/2016		29/09/2016	
Tree age when assessed		04		103			494			574		707	
Fungicide application date	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th	8 th	9 th	10 th	11 th	12 th	13 th
	30/10/2014	17/12/2014	30/01/2015	13/03/2015	28/04/2015	02/07/2015	11/08/2015	27/08/2015	23/09/20	015 23/10/2015	18/11/2015	08/12/2015	28/01/2016
Days between re- application	0	48	44	42	46	65	40	16	27	30	26	20	51

Table 16. Cost of treatments for three *Acacia mearnsii* trial initiated in October/November 2014 in the KwaZulu-Natal Midlands and southern Mpumalanga for the screening of fungicides for the management of wattle rust.

Site	Applications	Fungicide Trade Name ¹	Fungicide Active Ingredient (g L ⁻¹)	Relative growth (BI %)	Total Rand ha ⁻¹ as	suming a spray volu	ıme of 1000 L ha ⁻¹
					0.5x rate	1x rate	2x rate
		Ortiva ^{®1}	Azoxystrobin (250 g L ⁻¹)	39.3	1881.36	3209.36	5865.36
City Forestry	4	Amistar Extra®	Azoxystrobin (200 g L ⁻¹) Cyproconazole (80 g L ⁻¹)		2123.96	3694.56	6835.76
		Amistar Top®	Azoxystrobin (200 g L ⁻¹) Difenoconazole (125 g L ⁻¹)		2343.76	4134.16	7714.96
		Oriva [®]	Azoxystrobin (250 g L ⁻¹)		1881.36	3209.36	5865.36
Commondale	4	Amistar Extra®	Azoxystrobin (200 g L ⁻¹) Cyproconazole (80 g L ⁻¹) 21.6	2123.96	3694.56	6835.76	
		Amistar Top [®]	Azoxystrobin (200 g L ⁻¹) Difenoconazole (125 g L ⁻¹)		2343.76	4134.16	7714.96
		Ortiva®	Azoxystrobin (250 g L⁻¹)		1881.36	3209.36	5865.36
Etterby	4	Amistar Extra®	Azoxystrobin (200 g L ⁻¹) Cyproconazole (80 g L ⁻¹)	16.3	2123.96	3694.56	6835.76
		Amistar Top®	Azoxystrobin (200 g L ⁻¹) Difenoconazole (125 g L ⁻¹)	-	2343.76	4134.16	7714.96

Table 17. Cost of treatments for an *Acacia mearnsii* trial initiated in October 2016 to test fungicides for the management of wattle rust. The trial was located at Sheepwalk Farm in southern KwaZulu-Natal. All fungicide applications contained Amistar Top[®] (Azoxystrobin (200 g L⁻¹) Difenoconazole (125 g L⁻¹)) at a cost of R 895.2 L⁻¹. Treatment related information, climate and tree growth assessments are listed in detail in Chapter 3.

Applications	Adjuvant Trade Name ¹	Adjuvant Active Ingredient (g L ⁻¹)	Rate of adjuvant ha ⁻¹ assuming spray volume 1000 L ha ⁻¹	Adjuvant Rand L ⁻¹	Relative growth (BI %)	Total Rand ha ⁻¹ assuming a spray volume of 1000 L ha ⁻¹
6					12.5	6201.24
4	-	-	-	-	30.4	4134.16
3					23.8	3100.62
	Orosorb [®]	borax (10 g L ⁻¹) orange oil (50 g L ⁻¹)	208 ml	87.0		4482.16
1	Nu-Film P [®]	poly-1-p-menthene (875 g L ⁻¹)	1250 ml	208.0		4966.16
4	Orosorb [®] + Nu-Film P [®]	poly-1-p-menthene (875 g L ⁻¹) borax (10 g L ⁻¹) orange oil (50 g L ⁻¹)	1250 ml 208 ml	295.0	23.8	5314.16
3	Orosorb	borax (10 g L $^{-1}$) orange oil (50 g L $^{-1}$)	208 ml	87.0		3361.62
	Nu-Film P [®]	poly-1-p-menthene (875 g L ⁻¹)	1250 ml	208.0	23.8	3724.62
	$Orosorb^{\texttt{®}}$ + Nu-Film $P^{\texttt{®}}$	poly-1-p-menthene (875 g L ⁻¹) borax (10 g L ⁻¹) orange oil (50 g L ⁻¹)	1250 ml 208 ml	295.0	23.0	3985.62

Table 18. Cost of treatments for two *Acacia mearnsii* exclusion trials initiated in October 2014 to assess the impact of wattle rust on tree growth. Trials were located at Etterby and Hilton College in the KwaZulu-Natal Midlands. All fungicide applications contained Amistar Top[®] (Azoxystrobin (200 g L⁻¹) Difenoconazole (125 g L⁻¹)) at R 895.2 L⁻¹.

Site	Applications	Fungicide Trade Name ¹	Fungicide Active Ingredient (g L ⁻¹)	Relative growth (BI %)	Total Rand ha ⁻¹ assuming a spray volume of 1000 L ha ⁻¹
Etterby	13	Amistar Top [®]	Azoxystrobin (200 g L ⁻¹) Difenoconazole (125 g L ⁻¹)	41.2	7617.22
Hilton College	13	Amistar Top [®]	Azoxystrobin (200 g L ⁻¹) Difenoconazole (125 g L ⁻¹)	38.3	7617.22

In all trials, the tree Height (Ht in m) and Groundline Diameter (Gld in cm) were measured at regular intervals. From these measures the Biomass Index (BI) was determined where BI = Gld² x Ht. Biomass Index proved to be a good measure for the detection of treatment difference related to the presence of wattle rust (Little and Payn 2015). As the trials were all imposed on young seedlings, mostly without disease symptoms, the BI of subsequent measurements was calculated relative to the initial measurement. The relative BI (expressed as a percentage) for each fungicide treatment within each trial was then calculated relative to untreated controls. This measure (relative BI) would also allow for a comparison across sites. Costs were determined on a treatment bases per trial. These were then partitioned into six equal classes of R 1 500 to cover the range of costs (less than R 1 500 to greater than R 7 500). These classes were arbitrarily chosen so as to illustrate cost-related principles. A two-way table was used to compare growth and cost differences between treatments across all six trials.

5.3. Results and Discussion

5.3.1. Growth

Differences in growth observed between and within trials were a function of site productivities, time of planting (tree age), different treatments and levels of wattle rust (**Table 19**). Despite this, the application of fungicides in all trials resulted in significant improvements in BI growth of between 12.5% and 41.2%, and at a cost between R 1 881 and R 7 617. The effects of site productivity, tree age and wattle rust expression resulted in four applications of azoxystrobin + cyproconazole, azoxystrobin + difenoconazole and azoxystrobin having BI growth between 16 – 25% as well BI growth greater than 32%.

5.3.2. Costs

Control plots did not incur any costs, but tree growth was less than when fungicide was applied. There were no observable differences in tree growth between treatments which contained adjuvants and those which did not (**Table 19**).

Table 19. Cost-benefit of six *Acacia mearnsii* trials initiated in 2014/2015 to assess the impact of wattle rust on tree growth. Trials were located at Commondale in Mpumalanga, at City Forestry, Etterby and Hilton College in the KwaZulu-Natal Midlands and at Sheepwalk in southern KwaZulu-Natal. Abbreviations used are AD (azoxystrobin + difenoconazole), AC (azoxystrobin + cyproconazole), A (azoxystrobin), ^B (borax + orange oil) and ^P (poly-1-p-menthene). Numbers in brackets indicate the total number of applications for each treatment. Superscript numbers indicate rates other than 1x the recommended rate of application. Shaded blocks indicate optimum treatments, with the darkest shade indicating the most optimal.

			Relative improvement in Biomass Index vs control (%)									
		0 – 8	8 – 16	16 – 25	25 – 32	32<						
a ⁻¹)	<1500	none	-	-		-						
Cost per treatment (Randh	1500 – 3000	-	-	A ^{0.5} (4), AC ^{0.5} (4), AD ^{0.5} (4)	-	A ^{0.5} (4), AC ^{0.5} (4), AD ^{0.5} (4)						
	3000 – 4500	-	-	A(4), AC(4), AD(4), AD ^B (4), AD ^B (3), AD ^P (3), AD ^{BP} (3)	AD(3)	A(4), AC(4), AD(4)						
	4500 – 6000	-	-	A ² (4), AD ^P (4), AD ^{BP} (4)	-	A ² (4)						
	6000 – 7500	-	AD(6)	AC ² (4)	AD(4)	AC ² (4)						
	7500<	-	-	AD ² (4)	-	AD ² (4), AD(13)						

The use of adjuvants increased the cost of treatments in which they were used (**Table 19**). The largest growth response among the treatments was at the City Forestry trial (BI growth relative to control growth 39%) with four applications of fungicide, without adjuvants, and at Etterby and Hilton trial which had 13 applications of fungicide, without adjuvants (BI growth relative to control 41% and 38% respectively). However, the 13 applications achieved a growth response at a greater cost than treatments with fewer applications (**Table 19**). The greatest percentage of the cost of treatments is attributed to fungicides, averaging 76% of the total cost of treatments. Costs were reduced when fungicide applications were less frequent and when reduced rates were applied (**Table 19**).

5.3.3. Cost:Benefit (2-way table)

The optimum scenario for fungicide use would be the greatest reduction of wattle rust at the lowest cost of treatment. Costs are directly linked to the total number of applications, fungicide and rates applied, and the addition of adjuvants. These costs are additive, therefore the simplest regime (which would most likely also be the cheapest) consists of the least number of applications, lowest fungicide cost, reduced rate of application without the need for adjuvants, provided there is no reduction in growth. The largest growth response for the lowest cost was four applications of azoxystrobin, four applications of azoxystrobin + cyproconazole and four applications of azoxystrobin + difenoconazole at 0.5x the recommended rate of application. Although the reduced rate indicated optimal cost versus benefit, no optimal treatment existed at the recommended rate of application (**Table 19**). Cost of treatments may vary and be reduced based upon method of application and fungicide formulation used. The cost benefit analysis is based upon short term trials. To fully assess the economics of the use of fungicides for managing wattle rust, information will be required for the impact of treatments carried through to rotationend. This will determine whether the use of fungicides can be offset by the difference in yield between treated and untreated black wattle when infected by wattle rust.

5.4. Conclusion

Results from six trials were compared to determine costs associated with using fungicide for the management of wattle rust. Differences between site, climatic conditions and tree age led to varying growth responses. Control of wattle rust using a registered fungicide is beneficial, although costly if over-applied. The largest portion of the cost of treatments was attributed to the cost of the fungicide. The use of adjuvants did not affect growth of wattle trees, but did increase the cost of treatments. Although no treatment was found to be optimal for both cost and growth at the recommended rate of application, alternative formulations of fungicide (not yet tested) may reduce costs. The optimum treatment selected will be a combination of as few applications as possible at the required rate of application. This will be ideally applied preventatively, before wattle rust can affect tree growth, and timed according to climatic and epidemiological cues to obtain maximum benefit from treatment. Rotation-end data is required to determine whether fungicide use is economical for managing wattle rust over an extended period of time. The current fungicides are mostly contact in terms of mode of action, and need to reapplied, both within one season and over the rotation. Costs of multiple applications accumulate and hence this needs to be taken into consideration with long-term data. Systemic fungicides may help reduce costs provided the efficacy duration is longer, therefore extending the period between re-applications. Tree age and wattle rust may interact differently in terms of tree growth and Disease Expression (older trees may be less effected by wattle rust). Management using fungicides may then need to be adjusted accordingly.

6. SYNTHESIS AND CONCLUSION

6.1. Summary of major findings

To address the current lack of fungicides available (and knowledge around their application) for the management of wattle rust, a series of trials were implemented. The objectives were to screen fungicides for their potential use and determine the rates at which they are best applied, extend periods between the reapplication of fungicide through understanding the role of adjuvants and application timing, linking of wattle rust Disease Expression to tree growth to aid with the timing of application, and the cost:benefits associated with fungicide use.

Wattle rust had a negative impact on tree growth in the trials conducted for the screening of fungicides as well as the trial conducted for refining fungicide use. Height was not significantly affected by wattle rust in the trials for refining fungicide use, as a result of the relatively low level of wattle rust recorded. Site qualities, tree age and wattle rust severity had an impact on tree growth and response to treatments across all sites.

The fungicides screened for their effectiveness in managing wattle rust were found to be effective at the three rates at which they were applied, and are best applied preventatively. The use of adjuvants did not provide any additional benefit above fungicide alone, but increased the costs associated with the use of fungicides. Altered timing of the recommended 28-day application to 42 and 56 days were not found to be significantly different. Although commencing treatment in October on a 28-day schedule resulted in over application of fungicides in the trial for refining fungicide use. This resulted in phytotoxicity occurring, which had also been observed in the fungicide screening trials.

When using regression trees to determine a link between tree growth and Disease Expression, no symptom emerged as a significant predictor of tree growth. This may be attributed to the scoring system used, the limited data from which to build regression trees, the relatively low abundance of wattle rust symptoms at the trial for refining fungicide use or there may not be any individual symptom which can predict tree growth. Costs of fungicide use depend upon the number of applications, the cost and rate of application of fungicides, and use and cost of adjuvants. The largest portion of the costs of fungicide use for each application was attributed to the cost of fungicides themselves. Control of wattle rust using fungicide is beneficial but costly if over applied. No optimal treatment at the recommended rate of application was found, using the two-way method of analysing cost-benefit.

6.2. Significance of study

Research was necessary to establish whether fungicides are effective for managing wattle rust, and may serve as a management tool. The data from the fungicide screening trial enabled the registration of fungicides for use on wattle rust. Fungicides were found to be effective for managing wattle rust and hence can be used by black wattle growers. Determining the use of adjuvants and altered timing of fungicide application was an essential step towards optimising fungicide use. To determine the cost versus benefit of fungicide use, comparisons were made using different treatments conducted across all trials. These cost comparisons can be used by wattle growers to make decisions on whether the use of fungicide is economical or not. Although no link was found between wattle rust symptoms and tree growth, the application of fungicide is best applied at the onset of symptoms. Black wattle growers can benefit from the studies conducted and apply the results to manage wattle rust.

6.3. Limitations of study

The fungicides tested are from a limited number of fungicide groups (and hence modes of action). This is a risk for acquired resistance, which may develop in wattle rust populations with repetitive use of the same fungicides. Only one trial was conducted to test altered timing of fungicide applications and the use of adjuvants and fungicide. This trial also had low wattle rust expression (in relation to the trials conducted to screen fungicides). Two trials were initially established but one established on Etterby plantation in the KwaZulu-Natal Midlands was severely browsed by goats and therefore had to be abandoned. It is not known whether altered timing of fungicide applications and the use of adjuvants may yield different results under different environmental and site conditions (and disease severity) as

their loss of one trial prevented a cross site comparison among the treatments. Limited data was available to use to predict tree growth from individual wattle rust symptoms. The data was also categorical and unbalanced and therefore limited the number of statistical methods which could be used for analysis. Experiments designed to provide data for this purpose may yield better results, rather than using data from trials designed for other purposes. The economics of fungicide use are based upon short term trials. Rotation-end data is required to determine whether fungicide use is economical for managing wattle rust over an extended period of time, and what repeated applications over a longer time period may yield. Tree age may also elicit different responses to fungicide use were conducted for screening fungicides and for refinement of fungicide use were conducted in trees less than 500 days old. Trees of greater age may not only respond differently to fungicide use, but may also be impacted by wattle rust differently.

6.4. Implications for integrated pest management

Effective pest management programs rely upon a number of methods to manage a pest or disease. This reduces the likelihood of acquired resistance developing among the pest or disease population. Currently, only fungicides are available for the management of wattle rust, although alternative management methods are being researched. This is due to the reduced length of time required to determine fungicide management options when compared to alternatives. Once alternatives have been investigated, fungicide use can then be incorporated into an IPM for wattle rust. Linking fungicide applications to the biology, epidemiology and climatic factors associated with wattle rust is critical to ensure optimal use of fungicides. Fungicide use needs to be monitored and wattle rust populations observed to limit the likelihood of acquired resistance developing. Fungicides also need to be applied in a manner that reduces the likelihood of acquired resistance developing.

6.5. Future research possibilities

A need exists for fungicides to be tested from fungicide groups other than those screened for the management of wattle rust. This will provide a range of fungicides that, when used alternately or in combination, will reduce the likelihood of acquired resistance developing in wattle rust populations. Testing adjuvants and altered timing under different climatic conditions may yield different results to the fungicide refinement trial conducted. A repeat of the trial testing adjuvants and altered fungicide timing is recommended under climatic conditions more conducive to the development of wattle rust. Whether individual symptoms can be used to predict tree growth is still unknown. Predicting tree growth from wattle rust symptoms may be achieved by monitoring tree growth and wattle rust symptom development on a more regular time scale than the two-monthly period of observation in the fungicide trials. Long term research comparing rotation-end yield of treated versus untreated wattle tree growth will provide greater understanding of the economics of fungicide use. Linking fungicide applications to climate and epidemiological cues will assist in optimising fungicide use. Therefore, research into understanding the link between wattle rust life cycle, epidemiology and weather conditions will aid in determining an optimal fungicide application strategy. Research into fungicide application on trees of varying age classes may uncover whether fungicide use is necessary on different tree ages and how wattle rust impacts on trees of different ages.

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