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Review Article

Hylastes ater (Curculionidae: Scolytinae) Affecting *Pinus radiata* Seedling Establishment in New Zealand

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The introduced pine bark beetle *Hylastes ater* has been present in New Zealand for around 100 years. The beetle has been a minor pest on pines. Research was undertaken to control the pest in the 1950s–1970s, with a biological control agent introduced with limited success. Following a reasonably long period with minimal research attention, renewed interest in developing a better understanding of the pest status was initiated in the mid to late 1990s. Subsequently, a significant amount of research was undertaken, with a number of studies exploring the role of this pest of exotic forests in New Zealand. These studies ranged from attempting to quantify damage to seedlings, evaluate the role of the beetle in vectoring sapstain fungi, explore options for management, and evaluate the potential for chemical and biological control. From these studies, a number of findings were made that are relevant to the New Zealand exotic forest industry and shed new light onto the role of secondary bark beetles globally.

1. Introduction

The introduced pine bark beetle, *Hylastes ater* (Paykull) (Curculionidae: Scolytinae), is a pest of reestablished *Pinus radiata* D. Don forests in New Zealand. First recorded in New Zealand in 1929 [1], it has become a problem in second and third rotation forests where it breeds under the bark of stumps and other similar logging waste (log sections). Both adults and larvae feed on the phloem. Adults lay eggs in galleries, and larvae may take up to 300 days to develop to maturity. Subsequent emergence of adults from stumps is not necessarily immediate, and some adults continue to feed for longer periods [2]. Emerging adults feed on seedlings that have been planted in the immediate area. This maturation feeding characteristically involves the adult beetle eating the bark around the root collar of a seedling below the ground. In severe cases, seedlings may be completely ring barked and will die. Beetle feeding also commonly causes considerable sublethal damage, and feeding wounds may serve as a point of entry for soil-borne pathogens.

Despite initial concerns, historically *H. ater* was not regarded as a significant forest establishment pest in New Zealand. More recently, surveys have indicated that attacks on *P. radiata* seedlings by *H. ater* may be more common than previously documented and not evenly distributed across forest estates [3]. *Hylastes ater* usually attacks seedlings within the first year after planting [3]. Consequently, mortality surveys that are undertaken much later may fail to detect dead seedlings that are difficult to see, or death is attributed to other causes. In cases where dead seedlings are observed, they must be removed from the soil for inspection around the root collar region to confirm feeding damage by *H. ater* as a potential cause of mortality. Forest establishment practices currently focus on lowering initial stocking rates and planting higher quality (and more expensive) seedling material. This means that low amounts of damage by *H. ater* may be more significant to forest establishment operations than previously experienced [3].

2. Damage of Pine Seedlings by *H. ater*

In New Zealand, large areas of mature *P. radiata* forest are harvested all year round. The resulting stumps create a continuous supply of breeding habitat allowing *H. ater* and/or

other beetle populations to continuously persist at epidemic levels compared with a natural forest environment [4, 5]. Adults emerge from stumps following larval development and may begin maturation feeding on seedlings that were planted following harvesting operations [3].

In New Zealand, *H. ater* does not build up high populations in all areas. Surveys where live and dead seedlings were destructively sampled in 60 compartments in the central North Island showed that seedling mortality due to severe *H. ater* damage in most compartments was less than 5% [3]. However, seedling mortality was occasionally higher (up to 30%). These surveys revealed relatively few dead seedlings without evidence of severe feeding damage (i.e., root collar region completely ring barked), suggesting that seedling mortality due to severe *H. ater* damage was more likely than other factors (e.g., drought, poor planting). These other factors may have contributed to seedling death. There was evidence of some seedling attack by *H. ater* in most compartments. Sublethal damage by *H. ater* was identified by destructively sampling live seedlings along transects and was observed to be greater than 30% in half of all the compartments sampled over the two-year period (and was over 80% in some areas) [3]. Seedlings were occasionally found to have survived severe attacks (multiple feeding attempts or complete girdling of the stem) by *H. ater*. When such seedlings were inspected at a later date (i.e., one year after damage had occurred), they were found to be alive and "growing well", suggesting that if mortality did not occur subsequent to the *H. ater* feeding event, recovery was likely. Overall, this study suggested that considerable amounts of feeding damage might have previously been undetected as surveys were undertaken to identify areas of seedling mortality, and dead trees were often not removed from the ground for inspection. It is unlikely that in such surveys live trees were destructively removed.

The time trees were harvested was identified to be an important factor in determining whether seedlings are likely to be attacked by *H. ater* [3]. Sites harvested during autumn and planted the following winter were at the greatest risk, and the risk of damage decreased with increasing time between harvesting and planting. Sites harvested prior to spring and planted the following winter were seldom found to contain seedlings that were attacked by *H. ater*.

The relationship between harvesting history and the likelihood of seedling damage was related to the life history of *H. ater*, including flight activity and competition for brood sites by other bark beetles. *Hylurgus ligniperda* (Fabricius) (Curculionidae: Scolytinae) was found to be the dominant species in stumps during summer months [6]. *Hylurgus ligniperda* was first discovered in New Zealand in 1974 and, like *H. ater,* breeds in the stumps and logs of *Pinus* spp., and is found throughout New Zealand, but is not a threat to seedlings [7, 8]. Sites harvested during spring to late summer were colonised predominantly by *H. ligniperda* suggesting that this species is able to outcompete *H. ater* during this period. While *H. ater* has a spring flight, sites harvested in late summer-autumn contained the largest populations of *H. ater,* relative to *H. ligniperda.* The subsequent *H. ater* populations resulting from these autumnal colonisation events emerge during the following late spring/summer and attack seedlings [6].

Experimentation in forest establishment practices by some forestry companies in New Zealand resulted in areas being replanted outside of the traditional winter replanting times using containerised tree stocks. Essentially, this means the planting season was extended so re-establishment could, in theory, occur year round. In reality, planting was not undertaken during the driest months of summer. The implications of this replanting approach, with regard to seedling damage by *H. ater,* was not fully assessed, but, in theory, trees could be planted immediately (within a month) following harvesting. While harvested land was traditionally left "fallow" for extended periods to allow weeds to germinate and be controlled, this practice was being challenged by at least one forestry company during the early 2000s in an attempt to minimise the period between harvesting events. Consequently, areas previously considered of low risk due to the emergence of *H. ater* populations before planting occurred may be more at risk if populations of *H. ater* larvae are present in stumps when seedlings are planted.

3. The Relationship between *H. ater* and Sapstain Fungi

Bark beetles are known worldwide as vectors of fungi, largely due to interactions between aggressive bark beetles and fungi. These fungi were thought to play an important role in the tree killing by bark beetles (e.g., members of the genus *Dendroctonus*) [9–13]. Six and Wingfield [13] have recently challenged this view and have presented several arguments against a pathogenic role by these fungi. Firstly, tree-killing bark beetles do kill trees in the absence of virulent pathogens. Secondly, the growth of fungi follows beetles colonisation and is relatively slow until colonisation by beetles has resulted in tree heath deteriorating beyond the point where the tree might survive. Thirdly, virulent fungi are found to be associated with bark beetles that do not typically kill trees, and many tree-killing beetles carry weak or nonpathogenic fungi. Finally, most bark beetles do not kill trees and still carry fungi similar fungi to their tree killing relatives, which indicates that fungi play important roles other than killing trees. Instead, Six and Wingfield [13] suggest that fungal phytopathogenicity may be important for fungi that exhibit this characteristic to compete with other fungi and/or survive in living trees.

Staining fungi are a significant economic concern to the *P. radiata* forest industry [14–17], due to the high susceptibility of *P. radiata* wood to staining [18]. While some saprophytic, pathogenic, and endophytic fungi cause sapstain in wood, it is generally the saprophytic fungi that invade timber after the tree has been harvested [19]. The staining effect only becomes evident when conditions are favourable for fungal growth. In New Zealand, this is normally after harvesting when the sapwood dries and the aerobic sapstain fungi are able to grow in the wood cells. In some instances, wood may have to be discarded prior to processing [15, 19].

Sapstaining fungi are commonly recorded from less aggressive bark beetles. In particular, the fungal species of *Leptographium*, *Graphium,* and *Ophiostoma* have been found on *H. ater* in Britain, South Africa, and Australia [18, 20, 21]. In New Zealand, *Leptographium* sp., *L. lundbergii* Largerburg and Melin, and *Ophiostoma ips* (Rumb.) Nannf. were isolated from *H. ater* [22–24]. The presence of *L. procerum* (Kendrick) Wingfield and *O. huntii* (Robins-Jeff.) DeHoog and Scheffe in New Zealand is likely to be due to its introduction with either *H. ater* or *H. ligniperda* [18, 25]. *Ophiostoma ips* is commonly associated with bark beetles [26–29]. Species of *Hylastes* are known vectors of fungal root diseases in other parts of the world [30–32]. In these cases, *Hylastes* adults attack the roots of stressed or diseased adult trees and vector root disease fungi [30–32].

Reay et al. [24, 34] described a strong relationship between the sublethal attack of *P. radiata* seedlings by *H. ater* and invasion by sapstain fungi. The presence of sapstain fungi was found to increase as severity of damage increased. Half of severely attacked seedlings were found to contain sapstain fungi, indicating the potential for large numbers of seedlings throughout forests to be infected [24, 34]. The sapstain fungi were from the Ophiostomataceae [15, 29]. Most frequently isolated were *O. huntii* and *O. galeiforme* (Bakshi) Math-Käärik [24, 34]. *Ophiostoma huntii* has been isolated from many parts of the world [25] and has been associated with several species of bark beetles, including *H. ater* [25, 26] and *H. porculus* Erichson, and may be an important species in red pine decline [10]. *Ophiostoma galeiforme* is a European species, which has been found with *Hylurgops palliatus* (Gyllehan) on larch in Scotland [35, 36] and *Hylastes cunicularius* (Erichson) in Sweden [37]. Mathiesen-Käärik [37] describes *O. galeiformis* as a "secondary" staining fungus. *Ophiostoma galeiforme* may have been introduced into New Zealand with *H. ater* [24]. The remaining sapstain species isolated from seedlings by Reay et al. [24, 34] are commonly found in New Zealand pine plantations [15, 17].

Fortunately, the fate of fungi following feeding damage appears to be limited. When areas of damaged seedlings were revisited three years following planting, Reay et al. [38] failed to isolate any sapstain fungi species from the previously damaged trees that were sampled. However, *Sphaeropsis sapinea* (Fr.) Dyko and B. Sutton (which was not isolated from seedlings in initial sampling following seedling damage) were isolated from 10–16% of seedlings at the three-year after beetle attack sampling. *Sphaeropsis sapinea* is an important opportunistic fungal pathogen of *P. radiata* (and other conifers) in New Zealand. While there was a possibility that colonisation by the bark beetle vectored fungi may have had some influence on the health, growth, and long-term fate of the trees, this was not investigated [38].

Hylastes ater has been suggested as the mechanism by which a number of species of fungi have been introduced into New Zealand. Therefore, it is possible that future introductions of *H. ater* (or other bark beetles) may establish new fungal species (or other organisms). If new fungal pathogens were introduced into New Zealand by other means, there is potential for *H. ater* to vector these throughout forests. Therefore, continued treatment of bark beetles as biosecurity threats to New Zealand is imperative, despite the establishment of several species.

4. Molecular Characterisation of *Hylastes ater* and Associated Species

Hylastes ater is currently the only example of the genus found in New Zealand, but other Coleoptera can colonise similar environments. *Hylurgus ligniperda* is found under the bark of pine stumps, often with *H. ater.* Another bark beetle beetle, *Pachycotes peregrinus,* (Chapuis) (Scolytinae) and a native pinhole borer, *Platypus apicalis* White (Platypodinae) are also found in pine stumps. *Hylastes ater* may be confused with *P. peregrinus* [2] by inexperienced forest management personnel and is morphologically similar to closely related European species, such as *H. brunneus* Erichson. As biosecurity incursions are a constant threat to New Zealand exotic plantation forestry, identification of new occurrences of bark beetles is important. Larval stages are difficult to identify with morphological characteristics, so we investigated molecular identification of available species. This preliminary data is not intended as a full phylogenetic analysis, but rather to provide, through GenBank, reference sequences for each species for future researchers.

4.1. Methods. A number of individuals of *H. ater*, *H. ligniperda*, *P. peregrinus, Treptoplatypus caviceps* (Broun) (Platypodinae), *Platypus apicalis,* and *P. gracilis* Broun (Platypodinae) were collected from various sites throughout New Zealand (Table 1). In addition, specimens of *H. brunneus*, *H. cunicularius*, *Hylobius abietis* L. (Molytinae), and *Austroplatypus incompertus* (Schedl) (Platypodinae) were obtained from outside New Zealand and were included (Table 1). Sampling and collection of beetles were not systematic. Using DNA extracted from the heads, elytra, and legs (DNeasy Plant Kit, Qiagen), PCR was used to amplify the terminal region of the 28S rRNA domain 2 region was performed using the primers 28S-F (5′-AGAGAGAGTTCAAGAGTACGTG-3′) and 28S-R (5′-TTGGTCCGTGTTTCAAGACGGG-3′) [39]. Amplifications were carried out using 30 cycles of 15 sec at 98°C, 30 sec at 48°C, 40 sec at 72°C. PCR products were cleaned using an Eppendorf Perfect Prep Gel Cleanup Kit and sequenced directly (AWCGS Sequencing Facility, Massey University, New Zealand). Resulting sequences were aligned and compared using Bayesian inference (Figure 1). Sequences were aligned using ClustalX [40]. Phylogenetic analysis using Bayesian inference was conducted using MRBAYES version 3.1.2 [41, 42]. Models of nucleotide substitution were selected using the Akaike Information Criterion (see [43]) in MrModelTest v2 [44] implemented in PAUP*4.0b10 [45]. The model selected was GTR + G, which is a general time reversible model [46, 47] with a gamma-shaped rate variation across sites and a proportion of invariable sites. Two runs of four chains saving trees every 100 generations were conducted. After 1,000,000 generations, the two runs had converged close to the same value (determined by when

Table 1: Beetle isolates and GenBank sequence data used in this study.

Isolate	Species	Location, date, collected by	GenBank number
Hylg 1	*Hylurgus ligniperda*	Auckland, NZ. 2007 Reay	JN544556
Hylg2	*Hylurgus ligniperda*	Auckland, NZ. 2007 Reay	JN544555
Hylg3	*Hylurgus ligniperda*	Auckland, NZ. 2007 Reay	JN544554
Hyls96	*Hylastes ater*	Canterbury, 2004 Reay	JN544548
Hyls99	*Hylastes ater*	New South Wales, Australia 2004 Reay, D Kent	JN544549
UK1	*Hylastes brunneus*	Galway, Ireland 2005 Reay, Walsh	JN544550
UK12	*Hylastes brunneus*	Galway, Ireland 2005 Reay, Walsh	JN544551
UK8	*Hylastes cunicularius*	Northumberland, England 2005 Reay, Glare	JN544552
UK9	*Hylastes cunicularius*	Northumberland, England 2005 Reay, Glare	JN544553
UK6	*Hylobius abietis*	Galway, Ireland 2005 Reay, Walsh	JN544547
Pla90	*Austroplatypus incompertus*	New South Wales, Australia 2004 D Kent	JN544546
Platy1	*Platypus apicalis*	Auckland, NZ. 2007 Reay	JN544557
Platy2	*Platypus apicalis*	Auckland, NZ. 2007 Reay	JN544558
Platy3	*Platypus apicalis*	Auckland, NZ. 2007 Reay	JN544559
Platy4	*Platypus gracilis*	Canterbury, NZ. 2007 Reay	JN544561
Platy5	*Platypus gracilis*	Canterbury, NZ. 2007 Reay	JN544562
Platy6	*Platypus gracilis*	Canterbury, NZ. 2007 Reay	JN544563
Platy7	*Treptoplatypus caviceps*	Canterbury, NZ. 2007 Reay	JN544568
Platy8	*Treptoplatypus caviceps*	Canterbury, NZ. 2007 Reay	JN544569
Platy10	*Platypus gracilis*	Westland, NZ. 2007 Reay	JN544564
Platy20	*Treptoplatypus caviceps*	Westland, NZ. 2007 Reay	JN544570
Pla47	*Platypus gracilis*	Canterbury, NZ. 2007 Reay	JN544560
Pac6	*Pacyhcotes perigrinius*	Dunedin,NZ. 2002 S Reay	JN544566
Pac7	*Pacyhcotes perigrinius*	Tokoroa, NZ. 2004 Reay	JN544567
Pac8	*Pacyhcotes perigrinius*	Tokoroa, NZ. 2004 Reay	JN544565

the standard deviation of split frequencies fell below 0.005) and the first 25% of trees were discarded as burn-in. The consensus tree, with the posterior probabilities for each split and mean branch lengths, was visualised using Treeview 1.6.6 [48].

4.2. Molecular Identification Using 28S rRNA. Using this short sequence of the 28S rRNA, it was possible to distinguish between all species (Figure 1). As stated above, this is not a phylogenetic study and Figure 1 is provided simply for visual reference of the separations seen between species using this DNA segment. Clear separation was achieved between Platypodinae and Scolytinae, as would be expected, but also between the species of Scolytinae. The three *Hylastes* species were separated into a group with the related species, *H. ligniperda*.

The results of this analysis show the potential for the 28S region of RNA to be used for the identification of Curculionidae and may be a useful biosecurity tool, particularly if larvae of the Curculionidae are intercepted at the border.

5. Mitigating Impacts of *H. ater*

Early efforts to reduce the impact of *H. ater* in New Zealand included importation and release of three species of predatory *Rhizophagus* beetles, as no native predators were known [2]. These were originally imported in 1933 from Britain and released but did not establish. Further importations and release from Europe of natural enemies were made in 1975 and 1976. A parasitic wasp, *Rhopalicus tutele* (Walker) (Pteromalidae), and the predatory beetle, *Thanasimus formicarius* L. (Cleridae), were released but had little impact.

Following the work of Reay and Walsh [3, 49], management practices that could reduce likelihood of attack were recommended. As discussed above, high-risk sites could be planted later in the season in spring/early summer (rather than during winter) when late instar larvae are present allowing little time for seedlings to establish and grow prior to beetle emergence and may result in seedlings being more vulnerable to damage.

In New Zealand, chemical insecticides are rarely used in plantation areas to control *H. ater*. A carbosulfan insecticide was shown to protect seedlings from damage by *H. ater* but is not currently in operational use [49].

6. The Potential Role of Biocontrol of *Hylastes ater* Using Insect-Pathogenic Fungi

Currently, site management is the only economically viable option for minimising impacts to regenerative plantings due to *H. ater* damage in commercial operations. This results

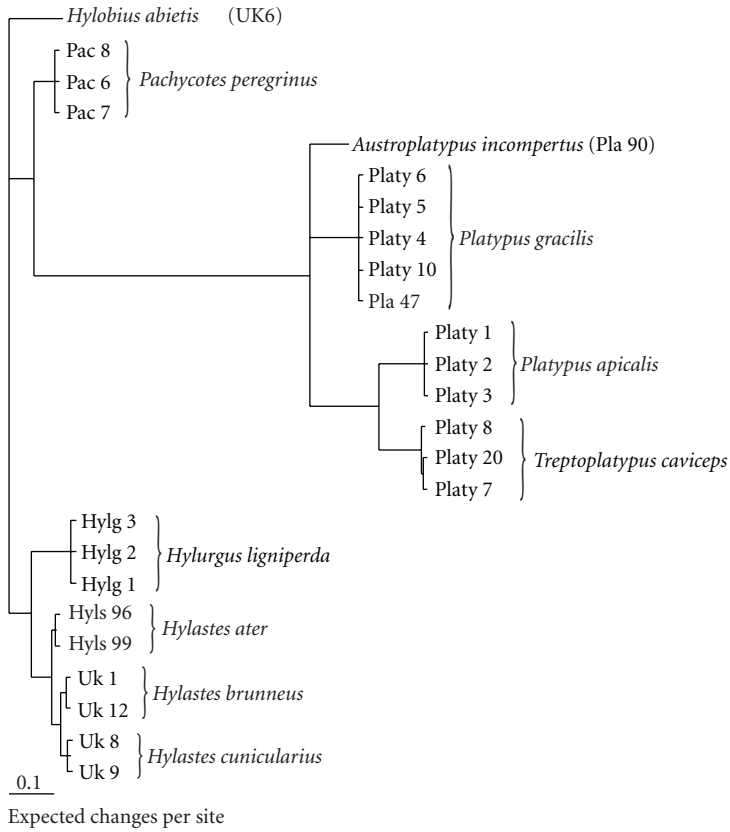

Figure 1: Representation of the species divergence using comparison of partial 28S rDNA sequences.

in land out of production and open to colonisation by weeds and erosion for a significant period. In addition, the increased use of containerised cuttings (in addition to bare root stock) has meant that planting seasons are extended, resulting in more sites at higher risk from *H. ater*. Alternative management options that would protect and promote the overall health and establishment of pine seedlings while reducing pest threat would benefit the forest industry. Moreover, improved control options are needed for use against any new species incursions. Biosecurity incursions are a constant threat to New Zealand plantation forests.

Entomopathogenic microbes have been developed as commercially available biopesticides for some pests. For example, the bacterium *Bacillus thuringiensis* Berl. has been used as the active agent in numerous biopesticides used in forestry for control of lepidopteron pests. Over a number of years, we have been investigating entomopathogens of *H. ater* in New Zealand and the potential for developing biopesticides.

Entomopathogenic fungi are important mortality factors in bark beetle populations, although the natural infection rate and impact on beetle populations is estimated to be relatively low [50]. Fungi in the genus *Beauveria* (Balsamo) Vuillemin are the most common species reported attacking bark beetles [51]. This genus contains a number of species, all of which are pathogenic to arthropods, including insects and Acari [52, 53], and occupy diverse habitats above and below ground [54–56].

Beauveria caledonica Bissett and Widden was isolated from *H. ater* and *H. ligniperda* in New Zealand and subsequently shown to be pathogenic to these two species in laboratory bioassays [57]. Previous to this, *B. caledonica* was not known to be pathogenic to insects. In the UK and Ireland, *B. caledonica* was isolated by concentrating on the major forestry pest, the large pine weevil, *H. abietis* [57]. *Hylobius abietis* is a serious pest of spruce and pine plantation trees, with an average of 33% and up to 100% of new plantings being killed per annum when untreated in some regions [58].

A survey of *Beauveria* spp. in substrates (soil, stumps, bark and grass from insect galleries) associated with bark beetles in *P. radiata* cutover forests was undertaken to identify what fungal isolates might be present in these forest systems [59]. *Beauveria* spp. were commonly isolated from all substrates sampled and were recovered from all but one of the six sites surveyed. However, there was considerable variation within and between sites in the relative prevalence of the fungi across all substrates and within substrate types, and three species of fungi were isolated (*B. caledonica*, *B. bassiana* (Balsamo) Vuillemin and *B. malawiensis* Rehner and Aquino de Muro) [59]. *Beauveria caledonica* was isolated from all substrates in this study, including beetle and larval cadavers. It was not isolated from live insects [59]. In total,

13 *Beauveria* isolates representing the three species recovered were selected and found to be pathogenic to both *H. ater* and *H. ligniperda* in laboratory bioassays [59]. Thus, in spite of the lack of *B. bassiana*-infected cadavers recovered in the field, the fungus is clearly able to infect and kill both bark beetle species and has been demonstrated for other bark beetles in the laboratory [60–65]. However, no epizootics have been reported in field populations. This may simply be due to natural inoculum levels being too low to initiate an epizootic, or due to inhibition of the fungi in the field. *Hylastes ater* was found to be less susceptible to all of the isolates tested than *H. ligniperda*, although the reasons for this are unclear. However, it is likely to have implications for any control programme using fungal entomopathogenic fungi.

While entomopathogenic fungi from *Beauveria* are predominant, fungi from other genera have been recovered from *H. ater* cadavers [66]. These include *Metarhizium flavoviride* var. *pemphigi* Driver and Milner and *Hirsutella guignardii* (Maheu) Samson, Rombach and Seifert. While some *Metarhizium anisopliae* (Metsch.) Sorokin. isolates are known to be pathogenic to bark beetles in laboratory bioassays, *Metarhizium* spp. do not appear to have been previously isolated from field-collected specimens [51]. Similarly for *H. guignardii*, this record is therefore not only a first for *H. ater* in New Zealand but may be the first record from a bark beetle [66]. Recovery of the fungi from the cuticles of bark beetle adults clearly demonstrates the capacity for insect-mediated movement of the fungi in a pine forest [59]. The recent research into entomopathogenic fungi represents recent attempts to obtain new ways to mitigating the impacts of *H. ater* in New Zealand. Differences in the natural prevalence of different species suggests that some isolates may be better suited as biocontrol agents as they persist in the environment better, while the high levels of inoculum detected in frass indicate that virulent, environmentally competent isolates must be selected and formulation and application technologies to efficiently target specific stages of the pest developed to effectively utilize these pathogens in bark beetle management.

The use of entomopathogenic fungi as biopesticides has been considered (e.g., [59]), but our estimates of production costs of *Beauveria* spp. could be prohibitively expensive for broadcast application against an occasional pest in pine plantations. This, coupled with the difficulty of applying fungi to larvae and adults in cryptic habitats, makes use of biopesticides unlikely without a major application development. Interestingly, further investigation of entomopathogenic fungi in New Zealand pine plantations found that *B. bassiana* also exists as an endophyte in some trees. A survey by Reay et al. [67] found that *B. bassiana* could be recovered from needle samples, as well as roots and seed from approximately 15% of 125 trees sampled over the country. Further research has demonstrated that the fungus can be established as an endophyte in seedlings M. Brownbridge et al. (then pers comm). *Beauveria bassiana* has been found as an endophyte of a number of plant species around the world, and the presence of the fungus has been shown to impact feeding in some insects [68]. The fungus, as an endophyte, may also offer protection against phytopathogens [69]. We are currently researching the potential of endophytic *Beauveria* in New Zealand pines as a method to reduce bark beetle populations. Endophytic entomopathogenic fungi would provide cost-effective methods to inoculate trees against bark beetles as we have shown seedlings can be infected with the fungus and *Beauveria* is carried in seed [67].

7. Conclusion

Hylastes ater may not have been considered an important pest of pine plantations in New Zealand during the early years of establishment in New Zealand, but more recently has been acknowledged as a pest of new second-generation plantings.

Investigation of biological control options has included predators, parasites, and entomopathogenic microbes. No introduced predator or parasite has yet had an impact on the populations of the beetles. Entomopathogenic fungi, especially *Beauveria* spp., are common in *H. ater* populations in New Zealand, but development as biopesticides is unlikely to be successful for *H. ater* due to the cost of any product and application to the cryptic environments being difficult. The discovery that the fungi can exist in pines as endophytes may, however, hold some promise for a cheaper method to use the entomopathogenic fungi in *H. ater* control.

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