PREFACE

Plantation forests of Australian hardwood species in South-East Asia now exceed 7M ha. The viability of these plantations is increasingly threatened by diseases and pests. Biological control is a sustainable option for the management of these pests and diseases.

The aim of this workshop is to explore the myths and realities of biological control in forestry and how to critically evaluate success or failure in hardwood plantation in SE Asia. An additional benefit is to establish and nurture professional networks among forest health workers in SE Asia and beyond. This workshop will address the following main topics:

- Principles of biological control
- Biological control of insect pests
- Biological control of diseases
- Case studies focusing on forestry pests and diseases

Speakers at the workshop include 2 keynote speakers from Switzerland and Australia, and 14 presenters from Australia, Indonesia, Vietnam, Fiji and Thailand. All workshop participants will have the opportunity to contribute to a simulation exercise

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Biotechnology and Tree Improvement (CFBTI), the Vietnamese Academy of Forest Sciences (VAFS), the University of the Sunshine Coast (USC), the University of Gadjah Mada (UGM), Riau Andalan Pulp and Paper (PT. RAPP-RGE), PT. Arara Abadi-Sinarmas Forestry and PT. Musi Hutan Persada (MHP).

INVITED SPEAKERS

Marc Kenis is a forest entomologist with 28 years' expertise in biological control, invasive insects ecology, impact and management and risk analysis. He is presently leading the Invasion Ecology and Risk Assessment Section at CABI in Switzerland and chairman of the IUFRO Working Party 7.03.13 "Biological Control of Forest Pests and Pathogens". He is presently involved, with the FAO, in the development of a guide on biological control against forest pests.

Simon Lawson is Associate Professor in Forest Health at the University of the Sunshine Coast (USC), and Director of the Biological Control of Eucalypt Pests Alliance (BiCEP). His research focus is on developing effective, sustainable management methods for insect pests of plantation forests, with an emphasis on biological control. He also has strong interests in forest biosecurity, population modelling, and in international collaborations in forest health and biosecurity.

ORGANISING COMMITTEE

Morag Glen, Anto Rimbawanto, Nur Hidayati, Desy Puspitasari, Abdul Azis, M. Nurdin Aswandi

SCIENTIFIC COMMITTEE

Caroline Mohammed, Morag Glen, and Simon Lawson

WORKSHOP PROGRAM

DAY 1	Friday, 21/7/2017	
Time	**Title**	**Presenter**
08.30 am	Welcome and opening of workshop	Anto Rimbawanto
08.40 am	Introduction to the concepts of biological control	Simon Lawson
09.00 am	The biological control of fungal pathogens	Caroline Mohammed
09.30 am	Biological control strategies against insect pests – by introduction against invasive insects	Marc Kenis
10.00 am	**Break**	
10.30 am	Flow chart for biological control introducing regulatory processes	Simon Lawson, Caroline Mohammed
11.00 am	Simulation exercise for ALL participants – exotic bronze bug	Facilitators: Simon Lawson, Caroline Mohammed, Morag Glen, Marc Kenis
12.00 pm	**Break for Friday prayers**	
02.00 pm	Overview of current program with Gall wasp	Simon Lawson
02.30 - 02.45 pm	Parasitism of leaf roller *Strepsicrates* (Lepidoptera: Tortricidae) infesting *Eucalyptus* (Myrtaceae) commercial plantations in Riau, Indonesia	Wagner de Souza Tavares
02.45 - 03.00 pm	Biological control of termites in Fiji	Salaseini Bureni
03.00 - 03.15 pm	Teak insect pests and biological control in Thailand	Tipakhon Phusakhon
03.15 -	Using endophytes to protect plants	Pham Quang Thu

03.30 pm	against pathogens and insect pests	
03.30 - 04.00 pm	Insect threats and their control – the role for biological control	Panel discussion with industry representatives Chair by Simon Lawson

DAY 2	Saturday, 22/7/2017	
Time	**Title**	**Presenter**
08.30 am	Development of biological control against *Ganoderma* root rot in Indonesian hardwood plantations	Morag Glen
08.45 am	Isolation, identification and antagonism testing of potential biological control agents against root rot caused by *Ganoderma philippii*	Desy Puspitasari
09.00 am	Pot trials of candidate biological control agents against root rot in *Acacia mangium* and *Eucalyptus pellita* caused by *Ganoderma philippii*	Heru Indrayadi
09.15 am	Field trial of candidate BCAs using wood-block inoculum	Abdul Gafur
09.30 am	Methods for producing oidia of *Phlebiopsis* sp. and *Cerrena* sp. as potential biological agents against *Ganoderma philippii*	Desy Puspitasari
09.45 am	Colonization of Eucalyptus and Acacia stumps by candidate biological control agents	Nur Hidayati/Husna Nurrohmah
10.00 am	Development of methods for detection of stump colonization by candidate biological control agents	Istiana Prihatini

10.15 am	Biocontrol of white root rot of hardwood trees using herbaceous plants	S. Suwandi and Chandra Irsan
10.30 am	Endophytic bacteria from *Acacia mangium* and their potential antagonism against *Ceratocystis* sp.	Aswardi Nasution
10.45 am	**Break**	
10.45 – 11.00 am	Potential use of endophytic fungi to enhance the quality of *Acacia crassicarpa* seedlings infected with *Fusarium* sp.	Heru Indrayadi
11.15 am	*Uromycladium falcatarium*, the *Falcataria mollucana* rust fungus, – potential biocontrol agent or biosecurity threat for some Leguminosae in the Pacific islands	Sri Rahayu
11.30 am	Fungal threats to forest trees in SE Asia and possibilities for biocontrol	Round table discussion – all participants
12.00 pm	Conservation and augmentation of insect biocontrol in an IPM system	Marc Kenis
12.30 pm	Summary and close of workshop	Su See Lee (ex FRIM and Vice President IUFRO)

ABSTRACTS

(in chronological order)

Introduction to the concepts of biological control

Simon Lawson[1] and Caroline Mohammed[2]

[1]Forest Health at the University of the Sunshine Coast (USC); [2]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia

This talk presents an overview of biocontrol, outlining the what, why, how and when of biological control, using examples specific to plantation forestry to highlight these concepts. We emphasise the need to consider biological control as one part of an integrated pest management system (IPM), i.e. biological control is not a 'silver bullet' and needs to be used in conjunction with other complementary methods, such as silviculture and tree breeding. We touch on what organisms function as biocontrol agents (BCAs), where you might find BCAs, mechanisms underpinning both biocontrol action and application. Finally, the question "What makes a good biocontrol agent?" will be considered.

The biological control of fungal pathogens

Caroline Mohammed

School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia

Fungal pathogens are among the most important factors that cause losses in agriculture, horticulture and forestry. Due to tree mortality, growth rates of *Acacia mangium* in Sumatra in those areas impacted by fungal diseases (*Ganoderma* and *Ceratocystis*) have been reduced to less than15 m3/ha/yr, while non-impacted areas have growth rates in the range of 22 to 35 m3/ha/yr. This talk focuses on the biocontrol agents of fungal plant diseases, their modes of action, application strategies, current status and future outlooks especially in forestry.

Biological control strategies – by introduction against invasive insects

Marc Kenis

Invasion Ecology and Risk Assessment Section, CABI, Delémont, Switzerland

Biological control is defined as the manipulation of living organisms (natural enemies) to control other living organisms (pests). Natural enemies can be used in various ways. This presentation will introduce the first approach, called “classical biological control” (CBC) and defined as the introduction of a natural enemy of exotic origin to control a pest, usually also exotic, aiming at permanent control of the pest. CBC has been carried out widely over a variety of target organisms, but most commonly against insects, using parasitoids and predators and, occasionally, pathogens. All stages of a biological control programme will be described and we will answer questions most commonly asked regarding CBC against insect pests, with particular emphasis on tree pests. The presentation will cover the following topics: rates of successes in CBC among different systems, different target insect groups and different agents; temporal trends in CBC practices and successes; economic and environmental benefits; risks and ways to mitigate the risks; the potential use of CBC against native pests; accidental successes through the adoption of the invasive pests by native natural enemies or accidentally introduced agents; and prospects and constraints for the practice of CBC in the future.

Flow chart for biological control introducing regulatory processes

Simon Lawson[1] and Caroline Mohammed[2]

[1]Forest Health at the University of the Sunshine Coast (USC); [2]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia

Here we present an outline of the process of a classical biological control program, from initial discovery of a new exotic pest to its eventual successful control, and the regulatory environment in which this happens. Each country will have its own regulatory framework, but must at a minimum conform to ISPM-3 "Guidelines for the export, shipment, import and release of biological control agents and other beneficial organisms". A recent example of how this process has worked in a forestry classical biological control program in southeast Asia will be used to illustrate this process in practice.

Simulation exercise for ALL participants – exotic bronze bug

Simon Lawson[1], Caroline Mohammed[2], Morag Glen[2], and Marc Kenis[3]

[1]Forest Health at the University of the Sunshine Coast (USC); [2]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia; [3]Invasion Ecology and Risk Assessment Section, CABI, Delémont, Switzerland

This session focuses on a practical interactive exercise in decision making following the introduction of a new exotic insect pest into a plantation estate. A series of decision points will be outlined for group discussion and this will reinforce the concepts of classical biological control and its regulatory environment as outlined in previous talks.

Overview of current program with Gall wasp

Simon Lawson

Forest Health at the University of the Sunshine Coast (USC)

The gall wasp *Leptocybe invasa* is a major global pest of eucalyptus plantations in temperate and tropical regions. It was first discovered in Israel in 2000 and had invaded every continent in which eucalypts are grown within 10 years. The wasp induces galls on the midribs, petioles and stems of young eucalypts and can severely impact on growth and form of susceptible trees. In most situations, chemical control is not feasible and while genetic resistance has been identified, these genotypes are often not available to smaller growers. Therefore *L. invasa* was a good candidate for biological control. Here we review the biological control of *Leptocybe invasa* globally, with particular emphasis on the situation in southeast Asia.

Parasitism of leaf roller *Strepsicrates* (Lepidoptera: Tortricidae) infesting *Eucalyptus* (Myrtaceae) commercial plantations in Riau, Indonesia

Wagner de Souza Tavares, Marthin Tarigan, Abdul Gafur, Wong Ching Yong, Mukesh Sharma

AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia

The leaf roller *Strepsicrates* sp. (Lepidoptera: Tortricidae) causes serious damage in young *Eucalyptus* (Myrtaceae) plantations in Riau, Indonesia. However, leaf rollers are attacked by parasitoids that have the potential to be reared and released in a biological control program. Around 800 leaf roller larvae of different ages were collected from May to December 2016 in several compartments planted with hybrid *Eucalyptus* in Riau, Indonesia and brought to the laboratory until emergence of leaf roller adults or parasitoids. Parasitoids and hyperparasitoids emerging from leaf roller puparia in were recovered from samples and identified. Emergence rate of leaf roller and parasitism rate (in %) were evaluated. Parasitoids recovered were the hymenopterans *Pristomerus* sp. (Ichneumonidae), *Zosteragathis* sp. (Braconidae), a non-identified braconid species, and a tachinid (Diptera: Tachinidae) species. Hyperparasitoids collected were two perilampid wasp species. *Zosteragathis* sp. parasitizes first instar leaf roller larvae. Tachinid flies were the more abundant parasitoids with six individuals collected, followed by *Zosteragathis* sp. with two individuals (one male and one female). Four and one individuals of the perilampid species were recovered, respectively. Emergence rate of leaf roller adults collected as larvae was 92% in the laboratory. Parasitism rate was 1.88%, with all parasitoids collected classified as solitary endoparasitoids, and these belong to small groups of insects poorly studied. The high diversity of parasitoids that emerged from leaf roller pupae indicates the possibility of development of a biological control program for this pest in eucalypt plantations.

Key words: parasitoid, Perilampidae, *Pristomerus*, Tachinidae, *Zosteragathis*

Biological control of termites in Fiji

Salaseini Bureni

P.O.Box 2218
Government Building, Suva, Fiji Islands

Biological control is a sustainable management option as it is environmentally friendly and cost effective and does not involve the use of chemical controls. In forests, injured and suppressed trees are more vulnerable to pests and diseases, especially trees that no longer have the ability to withstand normal environmental fluctuations, such as drought or waterlogging. These conditions allow plantation forests to be more susceptible to pest and disease infestation. In Fiji there is considerable potential for the use of the hyphomycete fungus, *Metarhizium anisopliae* (Metschn) as a biopesticide for the control of termites (*Neotermes* spp.). This fungus is a soil organism able to infect and kill a wide range of insects. It can be applied at termite feeding sites and the colony is then killed by the fungus. Termites are social insects and live in colonies and they can spread this disease in and around the colony, causing an epidemic leading to the destruction of the colony. This fungus and a nematode, *Heterorhabditis* sp., were assessed against *Neotermes rainbowi* infesting coconut palms. Both proved to be highly effective in completely eradicating the infestation of this termite. During an outbreak of the termite *Coptotermes gestroi* in Fiji, these controls were not used as the Government through the Biosecurity Authority of Fiji (BAF) opted to use chemical controls. The main concern was that this termite species has underground colonies, therefore limiting the scope for use of these controls.

Teak insect pests and biological control in Thailand

Tipakhon Phusakhon and Decha Wiwatwitaya

Department of Forest Biology, Faculty of Forestry, Kasetsart University, 10900 Bangkok, Thailand

Teak (*Tectona grandis* L.f.) is a large hardwood tree in the family Lamiaceae. Teak has beautifully patterned yellow to dark brown heartwood and is an important economic resource for Thailand. Teak has many uses, including for building and furniture, for example in cabinets, beds and tables, and is also exported for making yachts. Teak has been a very popular timber historically, and in the future it is predicted to be even more popular because of its very beautiful texture and color, trends in wood utilization and good carbon storage capability. Teak had been logged from natural forests for more than 200 years under concessions to private companies before cancellation of these concessions by the Thai government in 1989. Teak plantations have since greatly increased in northern Thailand, to respond to the growth in teak demand and utilization in the future.

Large areas of pure, even-aged teak plantations are susceptible to up to 72 species of insect pests, which can be classified into six types, including trunk and branch borers, leaf feeders, shoot borers, inflorescence and fruit destroyers, sap-suckers, and root feeders. In Thailand, there are few research programs about biological control and other methods for prevention and management of insect pests of teak. Insect pests that severely damage teak stands include: (1) Teak defoliator (*Hyblaea puera* Cramer; Lepidoptera: Hyblaeidae), which severely and rapidly damages the leaves of teak during dry weather early in the rainy season. If not controlled, growth losses in 5-year and 3-year old saplings amount to 44 and 70%, respectively. This pest can be managed using the bacterial and fungal biopesticides Bt (*Bacillus thuringiensis*), and *Metarhizium* sp., and with the predators *Eocanthecona furcellata* (Hemiptera: Pentatomidae) and *Sycanus collaris* (Hemiptera: Reduviidae); (2) Teak canker grub (*Acalolepta cervinus* (Hope); Coleoptera: Cerambycidae) damages the bole and forms a lesion around the trunks of

saplings and trees by eating the cambium at 1-2 metres above ground. Saplings 1-3 years old are at risk of breaking. This pest is managed by mechanical destructive methods, using hands-on techniques; (3) Teak Beehole Borer (*Xyleutes ceramicus* Walker; Lepidoptera: Cossidae) permanently damages stems by larval feeding in the tree trunk, and this damage increases cumulatively every year. Some teak trunks contain many borer holes and attack on trees can last more than 30 years, which decreases the quality and price of teak wood by 20-30%. Control is achieved through mechanical destruction, by light trapping, using a sex pheromone and by other biological methods including generalist predators such as *E. furcellata*, *Sycanus collaris*, ants (Hymenoptera: Formicidae) and mantids (Mantodea: Mantidae), the biopesticides Bt and *Metarhizium* sp. and the nematode (*Steinernema siamkayai* n. sp.). Even though there are biological control methods available to manage insect pest damage, it is not effective enough to prevent pest problems. However, it is expected that in the future there will continue to be outbreaks of insect pests in teak plantations, so there is a need to apply and use methods to prevent and eliminate teak pests. These should include the steps to plan, manage and maintain plantations well. Biological control is also an important method for plantation management and prevention and elimination of insect pests, and it does not have negative effects on the environment in teak plantations or surrounding forests.

Keywords: Teak, Insect pests, Biological control

Using endophytes to protect plants against pathogens and insect pests

Pham Quang Thu, Vu Van Dinh and Le Van Binh, Dang Nhu Quynh

Forest Protection Research Centre, Vietnamese Academy of Forest Sciences, Duc Thang, Bac Tu Liem, Hanoi, Vietnam

Acacia mangium, A. auriculiformis and Acacia hybrids and eucalypts (*Eucalyptus camaldulensis*, *E. urophylla* and hybrids) are the main plantation species in Vietnam. Recently, acacia and eucalypt plantations have experienced serious disease and insect pest problems. The most important pests are Ceratocystis wilt disease (*Ceratocystis manginecans*), Phytophthora root rot (*Phytophtora cinnamomi*) eucalypt leaf blight (*Calonectria* spp.)and eucalypt gall wasp (*Leptocybe invasa*). These pests are causing large scale economic losses. Controlling these pests with chemicals is difficult, costly and polluting to the environment. Endophytes reside in most healthy plants. Some endophytes isolated from acacia and eucalypt are beneficial to host plants, providing growth promotion, disease and insect pest prevention and nitrogen fixation. Two endophytic bacterial strains were isolated and identified as *Bacillus subtilis* and *B. amyloliquefaciens*. Inoculation of acacia seedlings with the bacterial endophytes increased growth and induced disease resistance compared to uninoculated control seedlings. To manage the blue gum chalcid*, Eucalyptus camaldulensis* saplings were inoculated with a conidial solution (1.0 x 10^6 CFU/ml) of *Beauveria bassiana*. Inoculated seedlings showed good growth and reduced incidence and severity of damage caused by the wasp. Therefore, it is widely suggested that microbial agents as endophytes are the potential biological pesticides to protect plants.

Key words: *Acacia, Eucalyptus,* endophytes, *Bauveria brassiana,* plant protection

Development of biological control against *Ganoderma* root rot in Indonesian hardwood plantations

Morag Glen[1], Desy Puspitasari[2], Nur Hidayati[2], Siti Husna Nurrohmah[2], Istiana Prihatini[2], Abdul Gafur[3], Heru Indrayadi[4], Anto Rimbawanto[2], Caroline Mohammed[1]

[1]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia; [2]Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia; [3]AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia; [4]Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau

Root rot is a particularly intractable problem in plantation forestry or other perennial horticulture systems and development of an effective biological control program can be a lengthy process. Our program for biological control of *Ganoderma* root rot is based on a system that was successfully developed for control of *Heterobasidion annosum* in softwoods in the UK and Europe. By following the strategy that has evolved from the two or more decades of research that has been invested into developing the *Phlebiopsis gigantea* biological control, we hope to reduce the time needed to develop an effective biological control system for *Ganoderma* root rot. Many steps in the process still need optimisation, beginning with the selection of candidate BCA isolates, through inoculum production, colonisation and demonstration of effectiveness in disease reduction. A team of researchers at the Centre for Biotechnology and Tree Improvement and the University of Tasmania have worked with staff at three of Indonesia's tree plantation companies to adapt this system to the current problem. This overview will establish the context for the following presentations.

Isolation, identification and antagonism testing of potential biological control agents against root rot caused by *Ganoderma philippii*

Desy Puspitasari[1], Luciasih Agustini[2], Morag Glen[3], Anto Rimbawanto[1], Sri Rahayu[4], Arif Wibowo[5] and Caroline Mohammed[3]

[1]Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia; [2] Pusat Penelitian dan Pengembangan Hutan, FOERDIA, Bogor, Indonesia; [3]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia; [4]Faculty of Forestry, UGM, Yogyakarta, Indonesia; [5]Faculty of Agriculture, UGM, Yogyakarta, Indonesia

Ganoderma philippii is an aggressive root pathogen that spreads from infected trees to adjacent healthy trees. Inoculum in the form of diseased roots increases with each successive rotation. Reduction of inoculum is a primary strategy for biological control of root rot caused by *Ganoderma philippii.* To this end, isolates of wood-degrading basidiomycetes were obtained and identified by sequence analysis of the rDNA internal transcribed spacer (ITS) regions. Candidate BCAs were selected on the basis of close relationships to commercially successful biological control agents. These and additional wood-degrading basidiomycetes were tested *in vitro* for aggressiveness against the root rot pathogens *G. philippii* and *Phellinus noxius.*

Pot trials of candidate biological control agents against root rot in *Acacia mangium* and *Eucalyptus pellita* caused by *Ganoderma philippii*

Heru Indrayadi[1], Desy Puspitasari[2], Abdul Gafur[3], Anto Rimbawanto[2], Morag Glen[4] and Caroline Mohammed[4]

[1]Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau; [2]Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia; [3]AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia; [4]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia

Fungal isolates with demonstrated *in vitro* antagonism against *G. philippii* and *Phellinus noxius* were tested in pot trials. Wood-blocks colonised with the pathogen were placed alongside wood-blocks colonised with candidate biological control agents in potting soil prior to planting of seedlings. Plants were maintained for 18 months with symptoms recorded weekly. A low level of root rot disease was observed in the trial, so that tree survival did not effectively discriminate among candidate BCAs. At the end of the trial, re-isolation was attempted from inoculum blocks of both pathogen and BCA. Cultures of *Cerrena* and *Phlebiopsis* were re-isolated from the woodblock inoculum, but neither *G. philippii* nor *P. noxius* were re-isolated from pathogen inoculum that was adjacent to these two candidate BCAs. Overgrowth of pathogen inoculum by *Phlebiopsis* and *Cerrena* was evident visually and by re-isolation. In addition, mycelium of *Phlebiopsis* sp. appeared to colonise the rhizosphere.

Field trial of candidate BCAs using wood-block inoculum

Abdul Gafur[1], Marthin Tarigan[1], Desy Puspitasari[2], Anto Rimbawanto[2], Morag Glen[3], Caroline Mohammed[3]

[1]AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia; [2]Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia; [3]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia

A field trial to assess the efficacy of several candidate biological control agents was established on a 6 ha compartment in Riau province. Treatments consisted of eight candidate biological control agents (BCAs) and one control. The candidate BCAs, including two isolates of *Phlebiopsis,* two of *Cerrena,* two of *Phlebia* and two commercially confidential isolates, were applied as woodblocks colonised with BCA mycelium, one woodblock applied to each planting hole. Plot sizes are 25 trees in a 5 x 5 configuration with a single buffer row and 20 replicates for each treatment. Annual surveys are being undertaken to assess the level of root rot in each plot as well as tree height and dbh measurements. At two years after planting, differences in tree survival are not significant though survival is highest in plots treated with Cr1 (*Cerrena* sp.), WA033 (commercially confidential) and Pb11 (*Phlebiopsis* sp.). Survival was lowest in plots treated with Pl3 (*Phlebia* sp.), WFA068 (commercially confidential) and Cr7 (*Cerrena* sp.). Significant differences were also lacking in height and dbh, though treatments with low survival (Cr7 and control) had the greatest height and dbh measurements. Patchy distribution of *Ganoderma philippii* inoculum increases the difficulty of showing significant differences in results.

Methods for producing oidia of *Phlebiopsis* sp. and *Cerrena* sp. as potential biological agents against *Ganoderma philippii*

Desy Puspitasari[1], Morag Glen[2], Istiana Prihatini[1], Nur Hidayati[1], Anto Rimbawanto[1], Siti H. Nurrohmah[1], Caroline L. Mohammed[2]

[1]*Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia;*
[2]*School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia*

Phlebiopsis sp. (a closely relative of *P. gigantea*) and *Cerrena* sp. are wood-rotting Basidiomycotina. They are potential biological control agents (BCA) for *Ganoderma philippii* in *Acacia mangium* plantation based on their in vitro capability to inhibit the mycelial growth of *G. philippii*. Isolates of these two fungal species produce asexual spores known as oidia. These oidia can be used in suspension to facilitate BCA application in the field, as is done for *Phlebiopsis gigantea,* in biological control systems against root rot caused by *Heterobasidion annosum* in European forests. Experiments were conducted to increase oidia production, including different growth substrates and varying inoculum methods. A range of agar media were tested, but growth on small, sterilized wood blocks gave a higher oidia production than any of the agar media tested. Single oidial isolates from the same parent culture varied greatly in levels of oidia production. Inoculation of wood blocks or petri dishes by oidial suspension resulted in higher oidia production than inoculation with an agar plug of mycelium. Oidia in aqueous suspension have a short period of viability, particularly at ambient temperatures. Further research into methods for preservation and storage of oidia is ongoing.

Keywords: oidia production, biological control, root rot disease

Colonization of Eucalyptus and Acacia stumps by candidate biological control agents

Nur Hidayati[1], Morag Glen[2], Siti H. Nurrohmah[1], Heru Indrayadi[3], Abdul Gafur[4], Istiana Prihatini[1], Desy Puspitasari[1], Anto Rimbawanto[1], Caroline L. Mohammed[2]

[1]Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia; [2]School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia; [3]Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau; [4]AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia

Root rot caused by *Ganoderma philippii* has a major impact on profitability of Indonesian *Acacia mangium* plantations. Effective, economically viable control methods are needed. Biological control has proven effective in reducing root rot caused by *Heterobasidion* spp. in European pine and spruce stands. Stumps are inoculated with *Phlebiopsis gigantea*, a wood-rotting Basidiomycete which can be applied as oidia (asexual spores) to freshly cut stumps to prevent colonization by the pathogen. This method is being tested for applicability to the *Acacia/Ganoderma* system. Two candidate biological control agents (BCAs) are species of *Phlebiopsis* (a close relative of *P. gigantea)* and *Cerrena* sp. Oidia from several isolates of each species have been tested for their ability to colonise freshly cut stumps. Field trials were established in Riau province, Sumatra. Oidia were applied as aqueous suspensions within 2 hours of felling trees. After 6 months, a 12 cm slice was removed from the top of each stump and tested for BCA presence at 3 different depths – 5 mm, 50 mm and 100 mm. Using a combination of culturing and PCR techniques, we were able to confirm the colonization of eucalyptus and Acacia stumps by both candidate BCA species. In some stumps, the candidate BCA had colonized to a depth greater than 100 mm. Ongoing trials are investigating the timing and extent of root colonization by BCAs. A trial to establish whether disease levels are reduced following oidia application to stumps is also in progress.

Keywords: biological control, oidia, root rot disease

Development of methods for detection of stump colonization by candidate biological control agents

Istiana Prihatini[1], Morag Glen[2], Nur Hidayati[1], Anto Rimbawanto[1], Siti H. Nurrohmah[1], Desy Puspitasari[1], Caroline L. Mohammed[2]

[1]*Centre for Forest Biotechnology and Tree Improvement, FOERDIA, Yogyakarta, Indonesia;*
[2]*School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia;*

Candidates for biological control of root rot caused by *Ganoderma philippii* in *Acacia mangium* include *Phlebiopsis* sp. (a close relative of *P. gigantea*) and *Cerrena* sp. These are wood-rotting Basidiomycotina which can be applied as oidia (asexual spores) to freshly cut stumps to prevent colonization by the pathogen. Field trials are underway to determine their effectiveness in disease reduction and to optimize application methods. Both of these research activities need a rapid and reliable method for determining the extent of stump colonization by the candidate BCA. In the *Heterobasidion annosum/Phlebiopsis gigantea* system, on which the current approach is based, stump sections can be incubated until characteristic features of the pathogen and BCA appear on the wood surface. These features are not produced in the *A. mangium* system, so alternative methods of determining stump colonization are needed. Isolation and identification of the fungal isolates is time-consuming when large numbers of samples need to be processed. We used a PCR-based approach, using species-specific primers to detect DNA from the two BCA species. These PCRs can be used on DNA extracted directly from wood, but sample size for DNA extraction is limited, so alternative approaches were also tested. These included (i) incubating small fragments of stump on agar media and sampling the mycelium that grew, without sub-culturing onto fresh media, and (ii) incubating larger slices of stump samples in paper bags at ambient conditions until mycelia appeared on the wood surface, then picking off the mycelium for DNA extraction and PCR testing. Using these techniques, we were able to confirm the colonization of eucalyptus and Acacia stumps by both candidate BCA species.
Keywords: PCR detection, biological control, root rot disease

Biocontrol of white root rot of hardwood trees using herbaceous plants

S. Suwandi and Chandra Irsan

Department of Plant Protection, Faculty of Agriculture, Sriwijaya University
Jl. Palembang-Prabumulih Km.32 Indralaya Palembang, Indonesia

White root rot caused by *Rigidoporus microporus* is a major disease of tropical hardwood trees that difficult to control. A single individual fungus may survive for more than 40 years and infect multiple host species, as revealed by somatic incompatibility. The fungus produces a pseudosclerotia that is resistant to chemicals and antagonists (biodegradation). Some herbaceous perennial plants have been tested for their biocontrol potential against white root rot of rubber tree. Mycelia of *R. microporus* exhibited an abnormal colonization after exposure to a filter-sterilized root exudate of herbaceous plants in an *in vitro* assay. Inoculum potential and infection of *R. microporus* could be suppressed after burying for 3 months in soil planted with herbaceous plants in a pot test. Herbaceous plants [arrowroot (*Marantha arundinacea*), java curcumin (*Curcuma xanthorrhiza*), sansevieria (*Sansevieria fasciata*), mallaca galangal (*Alpinia malaccensis*), greater galangal (*Alpinia galanga*), indian shot (*Canna indica*), curcumin (*Curcuma longa*), wild taro (*Colocasia esculenta*), and water yam (*Dioscorea alata*)] could partially suppress rhizomorph formation in soil, shortened inoculum viability, reduced rhizomorph length, and prevented colonization and infection on taproot of rubber seedling. In a field trial, growing of herbaceous plants around the infected stump for two years could enhance the stump decay. Herbaceous plants (4-10 plants) planted around the rubber trees could also suppress infection of *R. microporus* without negative effect on tree growth. These results highlight the potential benefit of herbaceous perennial plants for long-term biocontrol of white root rot pathogen in hardwood plantation.

Endophytic bacteria from *Acacia mangium* and their potential antagonism against *Ceratocystis* sp.

Aswardi Nasution[1,2], Morag Glen[1], Katherine Evans[1], Abdul Gafur[2] and Caroline Mohammed[1]

[1]*School of Land and Food, University of Tasmania, Hobart, Tasmania 7001, Australia;* [2]*AAA Group R&D, P.T. Riau Andalan Pulp and Paper, Pangkalan Kerinci 28300, Riau – Indonesia*

The diversity of bacterial endophytes from roots, stems and phyllodes of *Acacia mangium* in Indonesian forest plantation was studied. Bacteria were isolated from trees between one and five years old. Isolates were identified by 16S rDNA sequences analysis. In total, 278 isolates of endophytic bacteria were obtained, with the majority isolated from roots and young acacia trees. The sequence analysis showed that endophytic bacteria grouped into five clusters; Fermicutes, Alphaproteobacteria, Betaproteobacteria, Gammaproteobacteria and Actinobacteria. Fermicutes was the predominant group with 66.2% of all isolates. Among 25 genera which were isolated succesfully, *Bacillus* and *Burkholderia* were the most frequently isolated genera of endophytic bacteria in *Acacia mangium*. Species of several of the general isolated in this study have previously demonstrated potential as biological control agents (BCAs) and/or plant growth promoters. In-vitro assays of the 278 culturable endophytic bacteria resulted in 157 isolates which exhibited strong growth and nutrient competition (more than 70% of inhibiting ability) against *Ceratocystis fimbriata.* Non-contact tests showed that nine isolates produced metabolites which inhibited growth of *C. fimbriata*. The phylogenetic analysis of 16S rDNA from the isolates which produced antifungal compounds placed these endophytic bacteria in the genera *Staphylococcus, Paenibacillus, Lysinibacillus, Pantoea, Ralstonia, Cupriavidus* and *Ochrobactrum*. Methods for inoculating Acacia seedlings with these endophytic bacteria are currently being investigated. This will allow future studies into the potential of these endophytic bacteria to control *Ceratocystis* infection in field conditions.

Potential use of endophytic fungi to enhance the quality of *Acacia crassicarpa* seedlings infected with *Fusarium* sp.

Heru Indrayadi[1], Nur Malinda[2], Yusup Kurniawan[1], Fahrizawati[2], Leonardo S. S. Oliveira[1] and Budi Tjahyono[1]

[1] Plant Protection, R&D Sinarmas Forestry; [2] State Islamic University of Sultan Syarif Kasim Riau; Corresponding author: heruindrayadi@gmail.com

Acacia crassicarpa is a hardwood species with good properties for pulp production and has been widely planted in Indonesia. The significant increase in *A. crassicarpa* production comes with increasing concerns about pest and disease occurrence. *Fusarium* sp. is an economically important pathogen of *A. crassicarpa,* causing damping-off in seedlings and leading to high mortality in the nursery. Endophytic fungi such as *Trichoderma* sp. and *Gliocladium* sp. play an important role in assisting plant growth as well as suppressing disease attack. Thus, the aim of this study was to determine the effectiveness of *Trichoderma* sp. and *Gliocladium* sp. for increasing survival rate and growth of *Acacia crassicarpa* seedlings in the nursery. Isolates of *Trichoderma* sp. (E022, E033) and *Gliocladium* sp. (E027, E029, E009) and a mixture containing all isolates were tested on seedlings. The control consisted of no application of antagonistic agent. Seeds were sown into media contaminated with *Fusarium* then sprayed with spore suspensions at 1 x 10^6 spores/mL (10 mL per seed). The experiment was conducted in a randomized complete block (RCB) design with 3 blocks and 48 replicates seedlings per each block for each treatment. Application of *Trichoderma* sp. and *Gliocladium* sp. increased plant survival, diameter, height and numbers of leaves at 75 days after inoculation by using antagonist organisms. Treatment with isolates E022 (*Trichoderma* sp.) and E027 (*Gliocladium* sp.) were significantly different than control and showed, respectively, increased survival rates of 17.9% and 11.9%, plant height by 39.8% and 35.9%, plant diameter 13.5% and 10.6%, and number of leaves 54.4% and 60.2%, respectively. Based on the results found herein, the isolate E022 of *Trichoderma* sp. showed the best potential for application in *A. crassicarpa* nurseries.

Uromycladium falcatarium, the *Falcataria mollucana* rust fungus – potential biocontrol agent or biosecurity threat for some leguminosae in the Pacific Islands ?

Sri Rahayu[1] and Kenneth Puliafico[2]

[1]*Faculty of Forestry, University of Gadjah Mada, Yogyakarta, Inodnesia;* [2]*USFS Hawaii, USA*

Uromycladium falcatarium is a rust fungus causing extremely damaging disease on sengon (*Falcataria moluccana*) in Indonesia. The rust gall fungus has spread across the commercial sengon plantations of Indonesia, Malaysia and the Philippines causing economically significant damage to timber production. The fungus causes growing tissues of sengon trees to form large twisted knots which can lead to the death of young trees and is damaging to trees of all ages. On the other, *F. mollucana* is an extremely invasive weed in the entire Pacific Islands and must be suppressed or intensively controlled in order to prevent ecosystem changes. A series of experiments were conducted with this fungus on a suite of Fabaceae species from Hawaii. Plants were exposed to fungal spores under laboratory, greenhouse and natural field conditions. Although several non-target test plants showed signs of initial infection in the laboratory there was no disease progression in any plants other than the target *F. moluccana*. Galls and spores were only formed on the target plants in deliberately inoculated pathogen greenhouse conditions. Field tests are still ongoing but have not shown any signs of non-target impact. The results of these experiments suggest that this or other isolates of *U. falcatarium* may be suitable candidates for further research as potential biological control agents. We also discuss how our study can help address concerns about the biosecurity threats posed by *Uromycladium* species.

Keywords : Bio control, *Uromycladium falcatarium*, Fabaceae, Pacific islands

Conservation and augmentation of insect biocontrol in an IPM system

Marc Kenis

Invasion Ecology and Risk Assessment Section, CABI, Delémont, Switzerland

This presentation will cover two other approaches of biological control against insect pests, with particular focus on tree pests: (1) "Biological control by conservation" includes all the methods conserving or enhancing the efficiency of natural enemies (parasitoids, predators, nematodes, pathogens) already present in the system. This can include cultural practices favouring natural enemies, rational use of pesticides that are less harmful to natural enemies, etc. (2) "Biological control by augmentation" is the augmentation of the density of natural enemies by regular releases. Releases can be inoculative (i.e. inoculation at the beginning of the season of a small number of biological control agents that will reproduce), or inundative (i.e. mass releases for a single and immediate control). For these two approaches, we will provide examples of applications against tree pests worldwide as well as prospects and constraints. We will also explain how these approaches are most successful when integrated into IPM systems.

LIST OF PARTICIPANTS

Abdul Gafur
RGE Fiber Research and Development, Town Site I, PT. RAPP Complex, Pangkalan Kerinci 28300, Indonesia
abdul_gafur@aprilasia.com

Abdul Azis
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia

Anto Rimbawanto
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia
rimba@indo.net.id

Aswardi Nasution
RGE Fiber Research and Development, Town Site I, PT. RAPP Complex, Pangkalan Kerinci 28300, Indonesia

Awit Suwito
Pusat Penelitian Biologi, Indonesian Institute of Sciences, Bogor, Indonesia

Budi Tjahjono
Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau, Indonesia
budi_tjahjono@sinarmasforestry.com

Caroline Mohammed
Tasmanian Institute of Agriculture, University of Tasmania, Tasmania, Australia
caro.mohammed@utas.edu.au

Chandra Irsan
Laboratory of Plant Pathology, Faculty of Agriculture, Sriwijaya University, South Sumatra, Indonesia
chandra.irsan@gmail.com

Chhang Phourin
Department of Forestry and Community Forestry, Forestry Administration, Ministry of Agriculture, Forestry and Fishery, Cambodia
c_phourin@yahoo.com

Dao Ngoc QUANG
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
daongocquang@vafs.gov.vn

Desy Puspitasari
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia
diesy_puspita19@yahoo.com

Hem Chanrithy
Department of Forestry and Community Forestry, Ministry of Agriculture, Forestry and Fishery, Cambodia
ken_piseth@yahoo.com

Heru Indrayadi
Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau, Indonesia
Heru.Indrayadi@sinarmasforestry.com

Istiana Prihatini
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia
istiana.prihatini@biotifor.or.id

Jeremy Brawner
Forest Industries Research Centre, University of the Sunshine Coast, Maroochydore, Qld 4558, Australia
jeremybrawner@gmail.com

Leonardo S.S. Oliviera
Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau, Indonesia

Luciasih Agustini
Pusat Penelitian dan Pengembangan Hutan, FOERDIA, Ministry of Environment and Forestry, Bogor, Indonesia

Marc Kenis
Invasion Ecology and Risk Assessment Section, CABI, Delémont, Switzerland
www.cabi.org/about-cabi/staff/marc-kenis

Marius Duplessis
Plant Protection Section, R&D PT. Arara Abadi, Sinarmas Forestry, Riau, Indonesia

Marthin Tarigan
RGE Fiber Research and Development, Town Site I, PT. RAPP Complex, Pangkalan Kerinci 28300, Indonesia
marthin_tarigan@aprilasia.com

Morag Glen
University of Tasmania, Tasmania, Australia
Morag.Glen@utas.edu.au

Ms Mayouly Hongsathilath
Provincial Department of Agriculture and Forestry, Lao
ly_bt@yahoo.com

Muhamad Nurhuda Nugraha
PT. Fajar Surya Swadaya, East Kalimantan, Indonesia
mnurhudanugraha@gmail.com

Muhammad Nurdin Asfandi
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia

Nghiem Chi
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
nghiem.chi@vafs.gov.vn

Nguyen Duc Kien
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
nguyen.duc.kien@vafs.gov.vn

Nguyen Manh Ha
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam

Nuriyanto
PT. Fajar Surya Swadaya, East Kalimantan, Indonesia
nuriyanto@forestindo.com

Nur Hidayati
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia
inunghidayati@yahoo.com

Penpitcha Choosa-nga
Kasetsart University, Thailand
Penpitcha_jamja@hotmail.com

Pham Quang Thu
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
phamquangthu@vafs.gov.vn

Piseth Ken
Department of Forest Plantation and Private Forest, Cambodia

Purnamila Sulistyawati
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia

Ragil SB. Irianto
Pusat Penelitian dan Pengembangan Hutan, FOERDIA, Ministry of Environment and Forestry, Bogor, Indonesia

Salaseini Bureni
P.O.Box 2218 Government Building, Government Building, Fiji Island
amasibalavu99@gmail.com

Seva Oktarina
PT. Musi Hutan Persada, South Sumatra, Indonesia

Simon Lawson
Forest Health at the University of the Sunshine Coast (USC)
SLawson@usc.edu.au

Siti Husna Nurrohmah
Centre for Forest Biotechnology and Tree Improvement Research (CFBTI), Yogyakarta, Indonesia
siti_husna_n@yahoo.com

Sri Rahayu
Faculty of Forestry, Gadjah Mada University, Yogyakarta, Indonesia
tatarahayu@yahoo.com

Su See Lee
Private consultant, Malaysia
leesusee08@gmail.com

Suwandi Saleh
Laboratory of Plant Pathology, Faculty of Agriculture, Sriwijaya University, South Sumatra, Indonesia
suwandi.saleh@gmail.com

Tharnrat Kaewgrajang
Kasetsart University, Lardyaow, Chatuehak, Bangkok 10900, Thailand
ffortrk@ku.ac.th

Thet Paing Phyo
National Resources Development Company, Myanmar
kothetlay96927@gmail.com

Miss Tipakhon Phusakhon
Kasetsart University, Lardyaow, Chatuehak, Bangkok 10900, Thailand
tipakhon.ph@gmail.com

Tomy Listyanto
Faculty of Forestry, Gadjah Mada University, Yogyakarta, Indonesia

Tran Lam Dong
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
tranlamdong@gmail.com

Trang Tran Thanh
Vietnamese Academy of Forest Sciences, Hanoi, Vietnam
Trang.TranThanh@utas.edu.au

Tuvou Lusiana Nayara
P.O.Box 2218 Government Building, Government Building, Fiji Island

U ZAW WIN
National Resources Development Company, Myanmar
naywunmyat1@gmail.com

Wagner de Souza Tavares
RGE Fiber Research and Development, Town Site I, PT. RAPP Complex, Pangkalan Kerinci 28300, Indonesia

Wai Wai Than
Forest Research Institute, Myanmar
waiwaikyaw2007@googlemail.com

Yulminarty
Universitas Riau, Indonesia
yulminarty@yahoo.com

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Australian Center for International Agricultural Research

Center of Forest Biotechnology and Tree Improvement (CFBTI)

University of Tasmania

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