# WORKSHOP
# *CERATOCYSTIS* IN TROPICAL HARDWOOD PLANTATIONS
**Yogyakarta – Riau, February 15th – 18th 2016**

## 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.

*Ceratocystis*is is considered an extreme threat, causing mortality of up to 20% in *Acacia* plantations in Vietnam and, combined with *Ganoderma* root rot has reduced productivity on infected sites by 15 $m^3$/ha/year. Effective disease management is critical to the economic viability of plantations in SE Asia. Switching from acacia to eucalypt is not a universal solution because eucalypts have their own pest and disease challenges and site type limitations and require higher levels of management inputs (vegetation management and fertilizer application) to achieve satisfactory growth rates than do acacias.The workshop will review the current state of knowledge about *Ceratocystis* diseases in forest trees with a special focus on tropical hardwood plantations.

The aim of this workshop is to update the status of *Ceratocystis* disease in hardwood plantation in SE Asia, and link the management strategies between interested events/parties. This will be achieved by addressing the following main topics :

- Disease Impact
- Taxonomy
- Host-pathogen Interaction

- Host Resistance
- Dissemination and Biosecurity
- Silvicultural Management

Speakers at the workshop consist of 2 keynote speakers from Brazil and South Africa, and 21 presenters from Australia, Indonesia, Malaysia, United States of America and Vietnam.

We wish to acknowledge all our invited speakers, presenters, and all participants for contributing to this workshop. We also thank the Australian Centre for International Agricultural Research (ACIAR), University of Tasmania, the Centre for Forest Biotechnology and Tree Improvement (CFBTI), the Vietnamese Academy of Forest Sciences (VAFS), the University of the Sunshine Coast, the University of Gadjah Mada (UGM), Riau Andalan Pulp and Paper (PT. RAPP-RGE), PT. Arara Abadi-Sinarmas Forestry, PT. Musi Hutan Persada (MHP) and the International Union of Forest Research Organisations (IUFRO) for their support.

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## TENTATIVE PROGRAM

| DAY 1 | MONDAY, 15 February 2016 |
|---|---|
| 11.00 – 12.45 | Registration and Lunch |
| 12.45 – 13.05 | **Dr. Henry Bastaman (Director-General of FOERDIA)**<br>Opening address |
| **Session 1** : | **Disease Impact (part 1), chaired by Dr. Anto Rimbawanto** |
| 13.05 – 13.20 | **Dr. Irsyal Yasman (APHI)**<br>'The challenges of developing plantation forest in Indonesia' |
| 13.20 – 13.30 | **Assoc. Prof. Caroline Mohammed (University of Tasmania, and CSIRO)**<br>Background to ACIAR Project |
| 13.30 – 14.20 | Keynote Speaker<br>**Prof. Acelino Couto Alfenas (Universidade Federal de Viçosa)**<br>'Impacts and control of *Ceratocystis* wilt caused by *Ceratocystis fimbriata* on different crops in Brazil' |
| 14.20 – 14.40 | **Dr. Suwandi (Sriwijaya University)**<br>*'Ceratocystis* wilt of Lansium tree: New disease and threat to Duku fruit production in Indonesia' |
| 14.40 – 15.00 | **Rahman Gilang Pratama (Gadjah Mada University)**<br>'Spatial and temporal distribution of stem-canker diseases on *Acacia decurrens* at Gunung Merapi National Park, Yogyakarta, Indonesia' |
| 15.00 – 15.15 | Afternoon tea |

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| **Session 2** | : | **Silvicultural Management, chaired by Dr. Abdul Gafur** |
| 15.15 – 15.35 | | **Dr. Chris Beadle (CSIRO)**<br>'Silviculture of *Acacia* species' |
| 15.35 – 15.50 | | **Dr. Tran Lam Dong (VAFS)**<br>'Dealing with diseases from a silvicultural perspective in Vietnam' |
| 15.50 – 16.10 | | Discussion |
| **Session 3** | : | **Taxonomy, chaired by Dr. Morag Glen** |
| 16.10 – 17.10 | | Keynote Speaker<br>**Dr. Irene Barnes (University of Pretoria)**<br>'*Ceratocystis manginecans* causing *Acacia mangium* canker and wilt: taxonomy, biology and population genetics' |
| 17.10 – 17.30 | | Discussion |
| 17.30 | | Close |
| 18.30 | | Workshop dinner at Bale Raos |
| **DAY 2** | | **TUESDAY, 16 February 2016** |
| **Session 4** | : | **Disease Impact (part 2), chaired by Prof. Pham Quang Thu** |
| 08.30 – 09.15 | | **J.B. Friday, Lisa Keith, Flint Hughes and Phil Cannon (USDA Forest Service)**<br>'*Ceratocystis* wilt of 'Ōhi'a (Rapid 'Ōhi'a Death): A new disease in Hawai'i (by skype) |
| **Session 5** | : | **Host-pathogen Interactions, chaired by Prof. Pham Quang Thu** |
| 09.15 – 09.35 | | To be advised. |
| 09.35 – 09.50 | | **Dr. Trang Tran Thanh (VAFS)** |

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| | | 'Chemical responses to *Ceratocystis* in *Acacia'* (by skype) |
| **Session 6** | **:** | **Resistance, chaired by Dr. Chris Beadle** |
| 09.50 – 10.10 | | **Dr. Chris Harwood (CSIRO)**<br>'The background to *Acacia* genetics and breeding' (by skype) |
| 10.10 – 10.30 | | **Dr. Jeremy Brawner (University of the Sunshine Coast)**<br>'Evaluating *Ceratocystis acaciivora* symptom expression in *Acacia mangium* breeding populations and clonal seed orchards' |
| 10.30 – 11.00 | | Morning tea |
| 11.00 – 11.20 | | **Dr. Arif Nirsatmanto (CFBTI)**<br>'Screening trials to develop *Ceratocystis* resistant breeds of *Acacia* in Indonesia: Summarizing the research plan' |
| 11.20 – 11.40 | | **Prof. Wickneswari Ratnam (Universiti Kebangsaan Malaysia)**<br>'SNP diversity and implications for disease resistance breeding in *Acacia mangium* and *Acacia auriculiformis'* |
| 11.40 – 12.00 | | **Dr. Abdul Gafur (RAPP)**<br>'Other *Acacia* species as a source of resistance to *Ceratocystis'* |
| 12.00 – 12.20 | | **Aswardi Nasution (RAPP and University of Tasmania)**<br>'Developing a rapid screening protocol for resistance to *Ceratocystis'* |

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| 12.20 – 12.40 | | **Riassalma Rizkatiwi (Gadjah Mada University)**<br>'Initial response of some *Acacia mangium* provenances to *Ceratocystis* sp. in the nursery and field' |
| 12.40 – 13.00 | | Discussion |
| 13.00 – 14.00 | | Lunch |
| **Session 7** | : | **Dissemination and Biosecurity, chaired by Assoc. Prof. Caroline Mohammed** |
| 14.00 – 14.20 | | **Prof. Pham Quang Thu (VAFS)**<br>'*Ceratocystis* wilt - a serious threat to *Acacia* plantations in Vietnam' |
| 14.20 – 14.40 | | **Dr. Abdul Gafur (RAPP)**<br>Field Trip Introduction |
| 14.40 – 15.00 | | **Dr. Meitini Wahyuni Proborini (Udayana University)**<br>'*Ceratocystis* as a fungal parasite on wooden statues' |
| 15.00 – 15.20 | | Discussion |
| 15.20 – 15.50 | | Afternoon tea |
| **Session 8** | : | **Other Potential Management Strategies, chaired by Dr. Istiana Prihatini** |
| 15.50 – 16.10 | | **Dr. Budi Tjahjono (Arara Abadi, Sinarmas Forestry)**<br>'*Ceratocystis* disease incidence in *Acacia* plantation of Sinarmas Forestry and its management' |
| 16.10 – 16.20 | | **Aswardi Nasution (RAPP and University of Tasmania)**<br>'Endophytic bacteria as potential biological control agents' |

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| 16.20 – 16.40 | | To be advised. |
| 16.40 – 17.00 | | Discussion |
| **Session 9** | : | **Eucalyptus Rust Workshop, chaired by Dr. Budi Tjahjono** |
| 17.00 – 18.00 | | **Prof. Acelino Couto Alfenas (Universidade Federal de Viçosa)** |
| | | Presentation by followed by general discussion |
| 18.00 | | Workshop close |
| 20.00 | | **ACIAR FST 2014-068 Project meeting, chaired by Assoc. Prof. Caroline Mohammed** |
| **DAY 3** | | **WEDNESDAY, 17 February 2016** |
| **Session** | : | **Field Trip to Riau** |
| | | Meeting point at Adisutjipto airport Yogyakarta |
| 10.30 – 12.30 | | Flight to Pekanbaru |
| 12.30 | | Arrival at the SSQ II airport – Pekanbaru |
| 12.30 – 13.30 | | Lunch |
| 13.30 – 15.00 | | Travel to RAPP Complex |
| 15.00 – 16.00 | | Unigraha Hotel check in |
| 16.00 – 17.30 | | RGE Technology Center tour |
| 19.00 | | Dinner at Unigraha Hotel |
| **DAY 4** | | **THURSDAY, 18 February 2016** |
| 06.00 – 07.00 | | Breakfast at Unigraha Hotel |
| 07.00 – 10.30 | | Travel to trial sites in Baserah from Unigraha Hotel |
| 10.30 – 12.00 | | Visit to trial site 1 and site 2 |
| | | 'Level of *Ceratocystis* natural infection in some *Acacia mangium* materials' |
| 12.00 – 13.00 | | Lunch in Baserah |

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|---|---|
| 13.00 – 14.00 | Travel to trial site in Teso |
| 14.00 – 15.00 | Trial visit |
| | 'Effect of singling on *Acacia mangium* survival' |
| 15.00 – 17.30 | Travel to Pekanbaru |
| 17.30 – 18.00 | Hotel check in at Pekanbaru |
| 19.00 | Dinner |

KEYNOTE SPEAKER

# KEYNOTE SPEAKER

# IMPACTS AND CONTROL OF *CERATOCYSTIS* WILT CAUSED BY *Ceratocystis fimbriata* ON DIFFERENT CROPS IN BRAZIL

Acelino Couto Alfenas
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## Abstract

*Ceratocystis fimbriata* Ellis & Halsted was first reported causing black rot of sweet potato, (*Ipomoea batatas* L.), in 1890 in New Jersey, EUA. Its taxonomy remained confused for about 50 years, in which the fungus received different names as *Sphaeronaema*, *Ceratostomella*, *Ophiostoma*, *Endoconidiophora* and finally *Ceratocystis fimbriata*. Currently, phylogenetic analysis indicates that there at least five species complexes within the genus *Ceratocystis*: i) *C. fimbriata* complex; ii) *C. paradoxa* complex; iii) *C. coerulescens* complex; iv) *C. moniliformis* complex; and v) C. *thielaviopsis* complex, containing soil borne asexual spores, but not important in forestry. The *C. fimbriata* complex is divided in four clades: African, Asian, North American and Latin American clades. In the Latin America clade are included: *C. fimbriata sensu stricto*, *C. platani*, and *C. cacaofunesta*. In the past few years, several studies have

been conducted on the *C. fimbriata* complex, involving physiological specialization, morphological characterization, inter-sterility between isolates and phylogenetic analyses. Some of these studies culminated in the description of new species. Most studies conducted in Brazil have shown that isolates from different hosts belong to only one species and that several new species described based on ITS sequences are in reality genotypes of *C. fimbriata*. In Brazil, *C. fimbriata* was first reported on *Crotalaria juncea* in the regions of Campinas and Tietê, in the state of São Paulo in 1935. In 1938, symptoms of drought and death were observed in mango trees in Recife, state of Pernambuco, and the disease named as "Mal do Recife", was attributed to *Diplodia recifensis*. In the 1960s, the disease was attributed to *C. fimbriata*. Four decades later, *Ceratocystis* wilt was reported on eucalypts in the South of Bahia and subsequently on other hosts including gmelina, fig, colocasia, teak, rubber tree, andiroba (*Carapa guianensis*), and kiwi fruit. In my talk, I will emphasize the disease impacts and its control on different host species in Brazil.

# *Ceratocystis manginecans* CAUSING *Acacia mangium* CANKER AND WILT: TAXONOMY, BIOLOGY AND POPULATION GENETICS

Irene Barnes and Mike Wingfield
*Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, South Africa.*
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## Abstract

The devastating canker and wilt disease of *Acacia mangium* was first discovered in Riau by Wingfield in 2005, and the causal pathogen subsequently described as *Ceratocystis acaciivora* by Tarigan et al. (2011). This name was later reduced to synonymy with *Ceratocystis manginecans* (Fourie et al. 2015), the cause of a canker/wilt disease of mango and various leguminous trees in the Middle East (Van Wyk et al. 2007; Al Adawi et al. 2006; 2013b). *Ceratocystis manginecans* resides in *Ceratocystis sensu stricto* as defined by de Beer et al. (2014), typified by the tuber rot pathogen of sweet potato, *Ceratocystis fimbriata sensu stricto*. While there is debate regarding the best name to apply for the *A. mangium* pathogen, from a phylogenetic standpoint, it is very different to the sweet potato fungus and other species in *C. fimbriata sensu lato*. From a pathology perspective, this is the

most important issue to consider. Unlike the sweet potato fungus, the *A. mangium* fungus is never a root pathogen. Rather, it infects trees exclusively through wounds arising from pruning and animal damage. Our research has shown that it does not infect sweet potato and likewise, the sweet potato fungus does not infect *A. mangium*. When *A. mangium* is inoculated with *C. manginecans*, even when well-established in plantations, trees die very rapidly. We have also shown that the pathogen does not cause serious disease on *A. auriculiformis* and *A. crassicarpa* and that inoculated *Eucalyptus* spp. are resistant to infection (Redzuan Rauf, unpublished). Our population genetic studies, using microsatellite markers (Barnes et al. 2001; Fourie, unpublished), show that *C. manginecans* was most likely introduced into Oman and Pakistan and that a single clone is responsible for the devastating losses in those countries, on all host trees that it infects (Al Adawi et al. 2013b; 2014). This clone has been effectively spread via the wood-boring scolytid insect vector, *Hypocryphalus mangifera*, that is native to areas such as India where Mango also has its origin (Al Adawi et al. 2013a). In contrast, our large populations of isolates from Indonesia, Vietnam and Malaysia display high levels of diversity with those from Vietnam being the most genetically diverse (authors and collaborators unpublished). This suggests an Asian origin for the fungus. Together with a large number of colleagues, mostly also involved in this

ACIAR Project, very large numbers of trees have been inoculated with *C. manginecans*. This work (Brawner et al. 2015 and unpublished) has shown that some resistance to the pathogen can be found in *A. mangium*. This represents a substantial opportunity for the future. Similarly, there is also good evidence to show that selected *A. mangium* x *A. auriculiformis* hybrid clones will not be damaged by the disease. Ongoing studies include those relating to disease resistance in *Acacia* spp. as well as those relating to the infection biology of the pathogen, particularly concerning its insect vectors.

**Acknowledgements**
This work forms part of the graduate studies of numerous students including Ali Al Adawi, Arista Fourie, FeiFei Liu and Redzuan Rauf. We are also grateful to collaborators including Jeremy Brawner, Mahadir Lapammu, Yani Yaparudin (and her colleagues), David Boden, Pham Thu, Martin Tarigan, Abdul Gafur and Ali Al Adawi for their valuable contributions.

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ORAL SESSION

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# THE CHALLENGES OF DEVELOPING PLANTATION FOREST IN INDONESIA[1)]

Irsyal Yasman[2)]

[1)]*Paper presented at Scientific workshop on* Ceratocystis *in hardwood plantation, February 15-16, 2016, Yogyakarta, Indonesia;*
[2)]*Vice chairman of the Association of Indonesian Forest Concessionaires (APHI)*

## Abstract

The Association of Indonesian Forest Concessionaires (APHI) is a business organization that includes more than 77% of forest concessions in Indonesia. The association consists of 274 members of natural forest concessions and 154 members of plantations forest concessions in Indonesia. APHI member covers almost 7.5 million ha of plantations out of 10.3 million ha plantation area issued by the government. The last five years plantations reach an area of 2.12 million ha, or about 423,000 ha per year. An average timber production from plantation reach 21.5 million $m^3$ per year. An estimated 65% of the area of forest plantations in Indonesia to plant fast growing species such as *Acacia mangium*, *Eucalyptus pellita* and *Gmelina arborea*, for industrial pulp and paper. Whereas the species of *Tectona grandis* (Teak), *Antocephalus* spp. (Jabon), *Paraserianthes*

*falcataria* (Sengon), and *Dipterocarps* (Meranti) were planted for construction timber, panels and furniture.

Fast-growing species like *Acacia crassicarpa* are also planted in peat swamp forest by applying the technology of cultivation of peatlands through drying and setting the water table. The challenge of developing plantation forest in Indonesia is pests and diseases appear at the second rotation onward for most of the fast growing species. Forest pests in plantation is recorded in the form of stem borers, termites and pest animals such as monkeys and orangutan. A very serious disease in plantations in Indonesia are a variety of diseases such as the heart rot, white root fungus, *Ganoderma*, *Ceratocystis*, and various diseases in the nursery. The risks of developing the disease might arise due to inappropriate land preparation and introducing of new species in the peatlands plantation. It is recommended to do collaborative research in pest and diseases due to high risk of fast growing plantation in Indonesia.

**Keywords :** forest plantation, pest and diseases, *Acacia* spp.

# *CERATOCYSTIS* WILT OF LANSIUM TREE: NEW DISEASE AND THREAT TO DUKU FRUIT PRODUCTION IN INDONESIA

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## Abstract

Defoliation, dieback and massive mortality of Lansium tree (*Lansium domesticum* Corr.) in Ogan Komering Ulu (OKU) Regency, South Sumatera Province was first noticed in 2014 and caused substantial economic losses to duku fruit production. The disease prevailed along Ogan riverside on the trees after being scratched by a squirrel. A *Ceratocystis* species was consistently recovered from the infected branches, stem and taproot. Koch's postulates were fulfilled by inoculating the mycelial plug of fungus onto the stem and braches of Lansium tree. The morphology of teleomorph and anamorph were similar to that of *C. acaciivora*. Blast searches of *ITS* and *TUB* region in GenBank indicated that isolates are grouped within the *C. fimbriata sensu lato* species complex.

# SPATIAL-TEMPORAL DISTRIBUTION OF STEM CANKERS DISEASES ON *Acacia decurrens* AT GUNUNG MERAPI NATIONAL PARK, YOGYAKARTA, INDONESIA

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## Abstract

The aims of the research are 1) To evaluate the change of incidence and intensity of stem rot symptoms and 2) To recognize the spatial-temporal distribution pattern of stem rot symptom at Gunung Merapi National Park. The systematic sampling were used to collect data. Line plot made with 175 $m^2$ and consisted of three sub plot with each size 25 $m^2$. First plot was determined by the distance 10 m from the edge of boundary area. The distance between sub plot was also 10 m, while the distance between plot was 20 m. Data were collected once in two months from February to August 2014, then the data collection were : wounds number on the stem and presence or absence of sap on the wound, which observed at lower stem (0-1/3 as), middle stem (>1/3-2/3), upper stem (2/3-3/3); the distance of each tree were asymptomatic with symptomatic trees nearby and initial

inoculum; and monthly secondary climate data from *Badan Meteorologi Klimatologi dan Geofisika* (BMKG), Yogyakarta. The results showed that the incidence of stem rot symptoms were increase gradually. The stem rot incidence were increased from February (79.3%), April (89.7%), June (96.6%), and August (98.3%). However, severity of stem rot symptom in the observation area were low, less than 25%. Based on the vertical distribution of the symptoms were indicated that the wound spread from lower stem to middle and upper stem respectively. However, horizontal spread of the symptoms were extended on 0-2 m distance, and showed logistic pattern.

**Keywords :** *Acacia decurrens*, GMNP, stem rot symptoms, *Ceratocystis*, spatial distribution

# *CERATOCYSTIS* WILT OF ʻŌHIʻA (RAPID ʻŌHIʻA DEATH): A NEW DISEASE IN HAWAIʻI

J. B. Friday[1)], Lisa Keith[2)], Flint Hughes[3)], Phil Cannon[4)]
[1)]*UH Mānoa CTAHR Cooperative Extension Service;*
[2)]*USDA ARS Pacific Basin Agriculture Research Center;*
[3)]*USDA Forest Service Institute of Pacific Islands Forestry;*
[4)]*USDA Forest Service, Vallejo, California*

## Abstract

In 2011 there were a few reports of some ohi'a (*Metrosidero spolymorpha*) trees dying rapidly on a couple of hectares of land in the SE corner of the Big Island of Hawaiʻi. By 2014 the affected area had spread to 3,000 ha and by 2016 the affected area has spread to 23,000 ha. Ohi'a comprises 80% of the native forest of Hawaiʻi and provides much beauty and is the basis for much of the wildlife, water and other benefits that forests bring to a land area.

There is a lot of interest in how this pathogen was able to move about so quickly and kill so many trees, so there have been many studies on the etiology of the disease. Possible pathways that are being studied include movement in logs, in the soil, from insect frass, on vehicles as well as live insects. Management strategies are also being developed for this disease. Preventing movement of infected trees or tree parts

around the state was adopted immediately and may have kept the disease from moving to other islands in the state. Sanitary measures including decontaminating chainsaws and vehicles have been adopted and public education has been a high priority, including a website to keep the public informed. http://www2.ctahr.hawaii.edu/forestry/disease/ohia_wilt.html#article. Work on the biology and population genetics of this strain of *Ceratocystis fimbriata* is ongoing. Aleurioconidia are particularly difficult to kill and two distinct strains have been found in association with Ōhi‘a in Hawai‘i.

# EVALUATING *Ceratocystis acaciivora* SYMPTOM EXPRESSION IN *Acacia mangium* BREEDING POPULATIONS AND CLONAL SEED ORCHARDS

Jeremy Brawner[1)], Yani Japarudin[2)], Mahadir Lapammu[2)], Redzuan Rauf[2)], David Boden[3)] and Mike Wingfield[4)]

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[4)]*Forestry and Agricultural Biotechnological Institute (FABI), University of Pretoria, 0001 Pretoria, South Africa*

## Abstract

A dramatic rise in the incidence of a serious canker and wilt disease of *Acacia mangium* has led to the replacement of thousands of hectares of plantation forests in eastern Sabah. A disease screening program was initiated to evaluate levels of disease resistance and tolerance to the causative fungus, *Ceratocystis acaciivora*, in an *A. mangium* breeding population. Resistance was evaluated as the presence or absence of external symptoms in two open-pollinated progeny trials. In addition, tolerance was evaluated in one of these trials by measuring the size of lesions produced

following a controlled inoculation with *C. acaciivora*. Heritability estimates were low to moderate for growth traits but were close to zero for the range of traits used to evaluate *Ceratocystis* resistance and tolerance. Nevertheless, significant differences were found among the three sources (families from the local land race, Queensland and Papua New Guinea origins, all selected in progeny trials in Sabah) and among populations within these sources for many of the traits used to assess damage by the pathogen. Significant differences in lesion length among sources were also evident, however no differences among populations within sources were found. In a subsequent study undertaken in a clonal seed orchard, 5 ramets of 100 clones were inoculated and just 10 of these clones demonstrated little damage with at least 4 of the ramets remaining relatively healthy and surviving 3 months after inoculation. Results of these studies indicate little additive genetic variation for tolerance will make development of resistant breeds challenging and a low incidence of resistance will require screening of large populations to identify sufficient material for deployment.

**Keywords** : *Acacia mangium*, *Ceratocystis*, resistance, tolerance, genetic parameters

# SCREENING TRIALS TO DEVELOP *CERATOCYSTIS* RESISTANT BREEDS OF *ACACIA* IN INDONESIA: SUMMARIZING THE RESEARCH PLAN (A PART OF OBJECTIVE 2 IN ACIAR PROJECT FST/2014/068)

Arif Nirsatmanto[1)], Anto Rimbawanto[1)] and Jeremy Brawner[2)]

[1)]*Center for Forest Biotechnology and Tree Improvement, Yogyakarta, Indonesia;*

[2)]*Forest Industries Research Centre, University of the Sunshine Coast, Queensland, Australia*

## Abstract

Objective 2 of the ACIAR project FST/2014/068 is to reduce the impact of *Ceratocystis* and one of the activities focuses on rapid screening of germplasm for the deployment of disease resistant acacia. The research is initiated with large screening trials of *Acacia* species of commercial interest in Indonesia. The screening trial strategy encompasses five components: 1) the use of clonally replicated material, 2) preliminary screening through artificial inoculation, 3) propagation of resistant clones, 4) verification of resistance via the establishment of demonstration blocks, 5) further confirmation via the establishment of host age trials. The target species for these trials are *Acacia mangium* and *A.*

*crassicarpa*. Genetic material from five unrelated populations of each species will be used in the screening trials and will become base populations for the further development of resistant breeds. The screening trials will be established in three locations in Sumatra region under collaboration with three partner companies: PT Arara Abadi-Sinarmas Forestry (Riau), PT. RAPP (Riau) and PT MHP (South Sumatra). The expected output of the screening trial includes resistant or tolerant *Acacia* breeds as well as a robust and repeatable screening protocol for *Ceratocystis* resistant individuals.

**Keywords:** *Acacia mangium*, *Acacia crassicarpa*, *Ceratocystis*, screening trial, ACIAR project

# SNP DIVERSITY AND IMPLICATIONS FOR DISEASE RESISTANCE BREEDING IN *Acacia mangium* AND *Acacia auriculiformis*[1)]

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[1)]*Paper presented at Scientific workshop on* Ceratocystis *in hardwood plantation, February 15-16, 2016, Yogyakarta, Indonesia;*
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## Abstract

Single nucleotide polymorphism (SNP) markers have become popular due to their usefulness in various applications such as diversity analysis, fingerprinting, QTL and association mapping, marker-assisted selection, and candidate gene discovery for important traits. Disease is a hazard to plants in their natural habitat, and domesticated plants for aesthetic or commercial production. *Acacia mangium* and *A. Auriculiformis*, widely planted in Southeast Asia for pulp and timber industry, have progressively demonstrated vulnerability to various diseases. In this paper, we report the genetic diversity of natural populations of both species using SNP markers, and identify genes and protein families in the *Acacia* SNP set that are related to pathogenesis and disease

resistance. We successfully genotyped 73 (96%) and 59 (98%) SNP loci for *A. mangium* and *A. auriculiformis* respectively on SequenomMassARRAYiPLEX platform. The mean expected ($H_e$) and observed ($H_o$) heterozygosity in *A. auriculiformis* were 0.246 and 0.236, and in *A. mangium* were 0.267 and 0.251. Inbreeding values ($F_{IS}$) ranged from 0.135 to 0.537 at 11 SNP loci in *A. auriculiformis,* and ranged from 0.014 to 0.738 at 43 SNP loci in *A. mangium.* Population differentiation ($G_{ST}$) ranged from 0.034 to 0.552 in *A. auriculiformis* and ranged from 0.025 to 0.630 in *A. mangium* populations. Cluster analysis using neighbour-joining method showed that populations cluster according to their respective origin in both species. We found 37 genes and protein families that might play a role in disease resistance when comparing the SNP sequence of *A. mangium* and *A. auriculiformis* to the genome of model legume species *Medicago truncatula*. The implications of SNP diversity for disease resistance breeding for the species are discussed.

**Keywords:** SNP diversity, disease resistance, *Acacia*

WORKSHOP
*CERATOCYSTIS* IN TROPICAL HARDWOOD PLANTATIONS
Yogyakarta – Riau, February 15th – 18th 2016

# OTHER *ACACIA* SPECIES AS A SOURCE OF RESISTANCE TO *CERATOCYSTIS*

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## Abstract

***Ceratocystis acaciivora***, the causal agent of wilt, canker, and dieback on *Acacia mangium*, is currently one of the major diseases in commercial *A. mangium* plantations in Indonesia. Use of resistant genotypes is considered the most feasible control method in most plantation forests. Unfortunately, data on resistance in *Acacia* species to this pathogen are not sufficiently available. The aim of this study was to identify resistance sources in different *Acacia* species including *A. mangium*, *A. auriculiformis*, *A. crassicarpa*, *A. aulacocarpa* and hybrid of *A. mangium* x *A. auriculiformis* to *Acacia* wilt caused by *C. acaciivora*. The most virulent isolate collected from previous studies was used to inoculate container-grown, 12-week-old rooted cuttings of each *Acacia* species. Prior to inoculation, wounds (2 x 3 mm) were made on the stems of the rooted cutting using a laboratory blade and an agar disc taken from an actively growing colony on 2% MEA, with the

mycelium facing downwards, was placed in the wound, then covered with Parafilm. For control, plants were inoculated with sterile MEA plugs. The plants were evaluated weekly for 30 days for length of xylem discoloration, plant wilting and mortality. Wilting symptom started to appear 2 weeks after inoculation in all species except *A. crassicarpa* and *A. auriculiformis*. Based on this study, it is obvious that among the different *Acacia* species tested, *A. crassicarpa* and *A. auriculiformis* are promising sources of resistance.

**Keywords:** *Acacia aulalocarpa*, *A. auriculiformis*, *A. crassicarpa*, *Acacia mangium*, resistance, wilt

# INITIAL RESPONSE OF SOME *Acacia mangium* PROVENANCES TO *Ceratocystis* sp. IN THE NURSERY AND FIELD

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## Abstract

The aim of this study were to evaluate an initial response of some *Acacia mangium* provenance to *Ceratocystis* sp. infection in the nursery and in the field. The *A. mangium* provenances were originated from Papua New Guinea (Kini, Wipim, Oriomo, Bensbach, Gubam); Meraoke, Indonesia (Mutting); and Queensland (Claudie River and Pascoe River). The seedlings were artificially inoculated with *Ceratocystis* sp., isolated from decayed stem of *Acacia mangium* stand in the field. The study was conducted in shade house using *Completely Randomized Design* (CRD), both in the nursery and in the field. Data observation under the shade house were tissue and cellular response also callus growth, while data observation in the field was height, diameter and wilting symptom intensity due to *Ceratocystis* sp.

Based on percentage of dark brown color on the wound, mycelia spread in the vascular tissue, and disease intensity in the field, provenances Kini, Wipin and Oriomo from Papua New Guinea were potentially resistant to *Ceratocystis* sp. up to four months trees in the field.

**Keywords :** *Acacia mangium*, *Ceratocystis* sp., potentially resistant

# *CERATOCYSTIS* WILT - A SERIOUS THREAT TO *ACACIA* PLANTATIONS IN VIETNAM

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## Abstract

Acacia species including *A. mangium*, *A. auriculiformis*, Acacia hybrids are commonly planted in Vietnam, where they are utilized for the production of pulp, MDF boards and saw logs. Recently, plantations of *Acacia* and *Eucalyptus* in Vietnam have experienced a serious disease problem with up to 20% mortality in many locations. The aim of this study was to consider the incidence of this disease and to better understand its causal agent. Wilt, crown die-back symptoms as well as discoloured wood were common features on 3-4 year-old trees. Forty-one *Ceratocystis* isolates obtained from discoloured wood specimens of *Acacia* plantations collected

in 15 provinces were examined based on morphology and DNA sequence data. Based on a comparison of sequence data generated from 24 isolates using three gene regions, it was clear that a *Ceratocystis* sp. in the *Ceratocystis fimbriata* complex causes this disease, but the species-level identification remains contentious. The most closely related species are *C. acaciivora* and *C. maginecans*. This *Ceratocystis* disease is the most serious problem to *Acacia* spp. plantations and emerging issue to *Eucalyptus* plantations in Vietnam. The epidemiology and control methods of the disease are discussed in this study.

**Key words :** *Acacia*, *Eucalyptus*, *Ceratocystis* sp. and wilt

# FIELD TRIP INTRODUCTION:

## -) LEVEL OF *CERATOCYSTIS* NATURAL INFECTION IN SOME *Acacia mangium* MATERIALS

## -) EFFECT OF SINGLING ON *Acacia mangium* SURVIVAL

Hengki Marantika, Slamet Untung, Marthin Tarigan, Sabar T.H. Siregar, Muhammad Yuliarto, Abdul Gafur, Ching Yong Wong, and Mukesh Sharma
*AAA Group R&D, P.T. Riau Andalan Pulp and Paper*
*Pangkalan Kerinci 28300, Indonesia*

## Abstract

The field trip has been arranged to observe commercial plantation forest facilities located in the Province of Riau. It will mainly consist of two major programs, RGE Technology Center tours (February 17) and *Ceratocystis*-related trial visits (February 18). While RGE Technology Center is based in Pangkalan Kerinci, the trial sites include Teso and Baserah Estates. Two trials with different topics are the primary destinations in this field trip. The first one is "Level of *Ceratocystis* natural infection in some *Acacia mangium* materials" (in compartments J060 and I027, Baserah), the

other is "Effect of singling on *Acacia mangium* survival" (in compartment F006, Teso East). To summarize the preliminary results of the two trials at two years, variation in the level of *Ceratocystis* infection within the *A. mangium* materials tested was observed. In the meantime, artificial wounding did not seem to affect the level of infection. More data are yet to be collected before a comprehensive conclusion is made.

**Keywords:** dieback, disease infection, variation, wilt, wounding

# *CERATOCYSTIS* AS FUNGAL PARASITIC ON WOODEN STATUE

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## Abstrak

Kerajinan patung sebagai salah satu ikon pariwisata di Bali dan merupakan komoditas ekonomi yang penting bagi masyarakat. Salah satu problem pada kerajinan patung adalah penyimpanan karena banyak produk-produk setengah jadi atau sudah siap jual terkontaminasi oleh jamur, hal ini kemungkinan terkait dengan kelembaban udara yang tinggi.
Penelitian eksplorasi secara kualitatif dilakukan di daerah yang dikenal luas sebagai kawasan penghasil kerajinan patung kayu di Bali, Gianyar dan Ubud Bali. Hasil penelitian menemukan lima jenis jamur kontaminan yaitu *Aspergillus niger*, *Cladosporium* sp., *Ceratocystis* sp., *Botrytis* sp. dan *Curvularia* sp.

**Keywords :** patung kayu, *Ceratosystis* sp., *Curvularia* sp., Bali

**Abstract**

Wooden craft statue is one of the iconic souvenirs of Bali and it is also an important economical commodity of local communities livelihood. One critical constraint of industrial wooden-craft statue is fungus contamination on either stored-unfinished or finished statues. It might be related to high air-humidity. Qualitative observation was carried out at recognized region of wooden craft statue producer in Bali i.e. Gianyar and Ubud. This study found five frequent species of fungi i.e. *Aspergillus niger*, *Cladosporium* sp., *Ceratocystis* sp., *Botrytis* sp. and *Curvularia* sp.

**Keywords :** craft wooden statue, *Ceratosystis* sp., *Curvularia* sp., Bali

# *CERATOCYSTIS* DISEASE INCIDENCE IN *ACACIA* PLANTATION OF SINARMAS FORESTRY AND ITS MANAGEMENT

Budi Tjahjono, Heru Indrayadi , Fadjar Sagitarianto, Bayo Alhusaeri, Fahrizawati
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## Abstract

*Ceratocystis* disease is becoming a major concern in the *Acacia mangium* plantation of Sinarmas Forestry in Sumatera and Kalimantan Region. Some observations were conducted in the plantation as well as R&D trial plots to determine the incidence level of *Ceratocystis* disease and to find the disease management options. The sample trees were taken from plantation compartments using transects method, and from R&D trial plots. Singling operation in the plantation do more harm to the trees and increase the incidence of *Ceratocystis* disease. The wounds caused by mechanical damage and animal (monkey, squirrel, rat, stem borer) attack easily followed by *Ceratocystis* infection. Therefore, minimizing wounding and singling operation as well as reducing pest damages can be considered as preventive action in the management of *Ceratocystis* disease. Fungicide (i.e.

carbendazim) and some endophytic microbes (i.e. *Trichoderma* sp.) showed potential as other components in the integrated management of this disease. There are some differences in the level of disease among the species and the hybrids of A. *mangium* x *A. auriculiformis*. Planting suitable alternative resistant species other than *A. mangium*, especially *Eucalyptus pellita* and *A. crassicarpa* can be important options for *Ceratocystis* disease management in Sumatera and Kalimantan.

# FIELD TRIP PROGRAM

Field Trip

## International workshop on *Ceratocystis* in hardwood plantation

PT RAPP
Riau, Indonesia

February 18, 2016

# WORKSHOP
# *CERATOCYSTIS* IN TROPICAL HARDWOOD PLANTATIONS
**Yogyakarta – Riau, February 15th – 18th 2016**

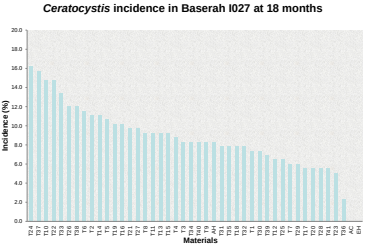
## Program

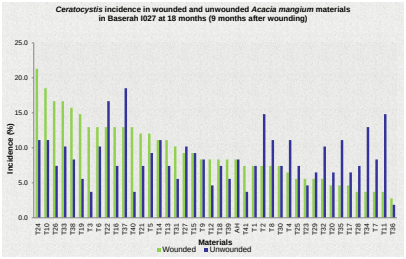
| Date/Day | Time | Activity | Location |
|---|---|---|---|
| February 17 Wednesday | 12:30 | Arrival at the SSQ II Airport | Pekanbaru |
| | 12:30-13:30 | Lunch | Pekanbaru |
| | 13:30-15:00 | Travel to RAPP Complex | Pekanbaru |
| | 15:00-16:00 | Uniqraha Hotel check in | Kerinci |
| | 16:00-17:30 | RGE Technology Center tour | Kerinci |
| | 19:00 | Dinner at Uniqraha Hotel | Kerinci |
| February 18 Thursday | 06:00-07:00 | Breakfast at Uniqraha Hotel | Kerinci |
| | 07:00-10:30 | Travel to trial sites in Baserah from Uniqraha Hotel | Kerinci |
| | 10:30-12:00 | Trial visit | Baserah |
| | 12:00-13:00 | Lunch | Baserah |
| | 13:00-14:00 | Travel to trial site in Teso | Baserah |
| | 14:00-15:00 | Trial visit | Teso |
| | 15:00-17:30 | Travel to Pekanbaru | Teso |
| | 17:30-18:00 | Hotel check in | Pekanbaru |
| | 19:00 | Dinner | Pekanbaru |
| February 19 Friday | AM/PM | Good bye everyone, you all have a safe and nice flight home!!! | Pekanbaru |

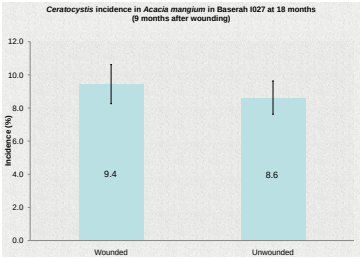
## Level of natural infection of *Ceratocystis* in some *Acacia mangium* materials

**Site 1**

| | |
|---|---|
| **Trial Code** | : **AMFPD119** |
| **Objective** | : To evaluate variation in tolerance/susceptibility in *A. mangium* materials to *Ceratocystis* natural infection |
| **Location** | : I027 Baserah |
| **Area** | : 7.5 ha |
| **Rotation** | : 4 |
| **Material** | : 41 AM + 1 AH + 1 AC + 1 EH materials |
| **Established** | : May 2014 |
| **Duration** | : 5 years |
| **Trial Design** | : RCBD |
| **Replication** | : 6 |
| **Plot size** | : 6 trees x 6 trees |
| **Spacing** | : 3 m x 2 m (initial stocking 1667 trees per ha) |



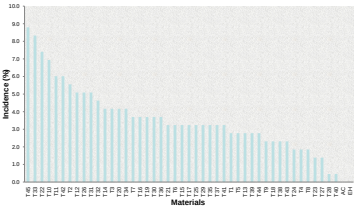



## Level of natural infection of *Ceratocystis* in some *Acacia mangium* materials

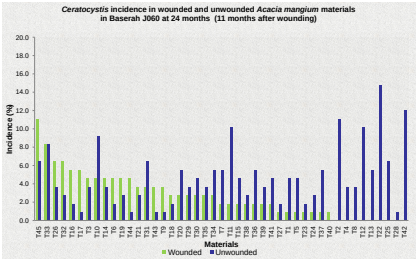
**Site 2**

| | |
|---|---|
| **Trial Code** | **: AMFPD103** |
| **Objective** | : To evaluate variation in tolerance/susceptibility in *A. mangium* materials to *Ceratocystis* natural infection |
| **Location** | : J060 Baserah |
| **Area** | : 6.5 ha |
| **Rotation** | : 4 |
| **Material** | : 45 AM + 1 AC + 1 EH materials |
| **Established** | : January 2014 |
| **Duration** | : 5 years |
| **Trial Design** | : RCBD |
| **Replication** | : 6 |
| **Plot size** | : 6 trees x 6 trees |
| **Spacing** | : 3 m x 2 m (initial stocking 1667 trees per ha) |

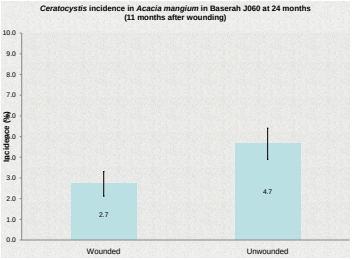
***Ceratocytis*** **incidence in Baserah J060 at 24 months**




***Ceratocystis*** **incidence in wounded and unwounded *Acacia mangium* materials in Baserah J060 at 24 months (11 months after wounding)**




***Ceratocystis*** **incidence in *Acacia mangium* in Baserah J060 at 24 months (11 months after wounding)**



## Effect of Singling on *Acacia mangium* Survival at Age 2 Years

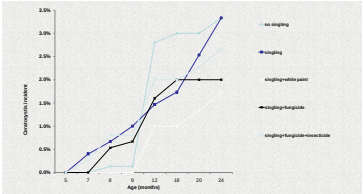
### Background

- *A. mangium* mortality loss to *Ceratocystis* is also considered to be related to man-made wound such as singling/pruning
- Singling is needed to improve stem quality (stem form and growth) although only 30-40% of trees only need singling
- Mortality due to *Ceratocystis* in unsingled stand also happens
- If singling still needs to be done, what treatments can be done to reduce mortality after singling?

### Methods

- Trial Code : AMTND40
- Design : RCBD; 5 replicates
- Location : Teso East F006
- Planted : March 2013
- Established : September 2013 (at age 6 months)
- Genetic : FAM0016 (Cuttings; CP material)
- Plot size : 10 x 15 trees
- Spacing : 3 x 2 m
- Treatments : No singling
  Singling (no scar treatment)
  Singling + white oil paint
  Singling + fungicide (Bavistin)
  Singling + fungicide (Bavistin)+ insecticide (Confidor)

### *Ceratocystis* (based on total count)




### Interim Conclusion

- Difficult to directly correlate singling and *Ceratocystis* incidence
- Plot variation may reflect the variation of inoculum
- However, scar treatment with oil paint (which cover the wound) may reduce or slow down the infection until 2 years
- Application of fungicide or insecticide to singling scar is not effective

# CURRICULUM VITAE

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- **EDUCATION AND TRAINING**
  - Federal University of Viçosa (Brazil) - B.S. in Forestry - 1974.
  - Federal University of Viçosa (Brazil) – Plant Pathology, M.Sc. - 1978.
    Title of the dissertation – 'Identification of three species of *Cylindrocladium* isolated from leaf spots of *Eucalyptus*'
  - University of Toronto (Canada) – Plant Pathology, Ph.D. - 1983.
    Title of thesis – 'Virulence and isoenzymes patterns of *Cryphonectria cubensis* (Bruner) Hodges, causal agent of the *Eucalyptus* canker'

➢ **SUMMARY OF SCIENTIFIC PRODUCTION**

- Papers published in Referee Journals (239)
- Papers published in Proceedings and Symposia (565)
- Books and book chapters
  - ✓ Books (11)
  - ✓ Chapters (33)
- Registered patent and patent requests (3)

➢ **REGISTERED PATENT**

RIZOLYTUS "Bioproduct for rooting induction of cuttings, and mnicuttings of *Eucalyptus* and biological control of *Eucalyptus* diseases in nursery" Trademark registration No. 002852 PTO. August 6, 2002.

➢ **PATENT REQUESTS**

- Rooting process (Addition). 2002. Patent: Innovation n.INPI PI0203326-7, August 6, 2002 (Deposit), August 7, 2002 (Review) August 8, 2002 (Grant).
- Rooting of *Eucalyptus* mediated by rhizobacteria. 2001. Patent: Innovation. INPI No. PI011400-5, "Process of rooting ...". 12 Feb. 2001 (Deposit), 13 of fev. 2001 (Review); 13 of fev. 2001 (Grant).

➢ **HONOURS**

- 2014 Honorable Mention: Identificação de bactérias associadas a mancha-bacteriana do eucalipto no Brasil", Universidade Federal de Viçosa – SIA.
- 2013 Honorable Mention: Genetic diversity of *Ralstonia solanacearum* infecting Eucalyptus spp. in Brazil by ERIC-PCR technique**,** Universidade Federal de Viçosa.

- 2011 Honoured Professor – Honour lecture for Forestry Undergraduates, July 2011, Universidade Federal de Viçosa.
- 2011 Guiding the student teacher Patricia Machado da Silva, 1st place in category Post-Graduate Masters and Doctoral Award at the 10th Golden Blue Furnas, State of Minas Gerais.
- 2007 Cover picture of Revista Brasileira de Fitopatologia - 2007, Brazilian Society of Plant Pathology.
- 2005 FAO expert in Forest Pathology in Uruguay.
- 2005 Cover picture of Revista Brasileira de Fitopatologia - 2005, Brazilian Society of Plant Pathology.
- 2004 Diploma of Merit 2004 Forestry, State Forestry Institute – IEF.
- 2004 Cover picture of Revista Brasileira de Fitopatologia - 2004, Brazilian Society of Plant Pathology.
- 2004 Peter Henry Rolfs Gold Medal for Merit in Research 2004, Universidade Federal de Viçosa.
- 2001 Honoured Professor – Honour lecture for Forestry Undergraduates, March 2001, Universidade Federal de Viçosa.
- 2001 Honoured Professor - Graduates Forestry, August 2001, Universidade Federal de Viçosa.
- 2000 Honorable Mention: Ppr-1 - The first gene for rust (*Puccinia psidii*) resistance, mapped in *Eucalyptus grandis*. Universidade Federal de Viçosa, 2004 to 6/12/2000. Viçosa, MG. X Symposium, Federal University of Viçosa.
- 1998 Cover picture of the Plant Disease, vol.82, No. 7, 1998, Plant Disease.
- 1996 1A CNPq Research Fellow, process 522625/96 1993-up to now.

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species and populations of tree pathogens. She has been involved in developing microsatellite markers used to gain insights into the routes of invasion and pathways of movement of some of the world's most serious forest pathogens. Dr Barnes completed her MSc in the department of Microbiology and Plant Pathology at the University of Pretoria and obtained a Ph.D. degree in Genetics in 2009. She has been the recipient of the Young Plant Pathologist Award from the Southern African Society for Plant Pathology, and recently received the exceptional young academic achievement award from the University of Pretoria.

# WORKSHOP
# *CERATOCYSTIS* IN TROPICAL HARDWOOD PLANTATIONS
**Yogyakarta – Riau, February 15th – 18th 2016**

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