ISOLATION OF ENDOPHYTE MICROORGANISM of Acacia mangium

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ISOLATION OF ENDOPHYTE MICROORGANISM of *Acacia mangium*

By

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DEDICATION

For my beloved family:

Hajar Binti Mokhtar;

Umi Kalsom Binti Mokhtar,

Abdul Latif Bin Taimah,

Razali Bin Mokhtar,

To all my friends, especially Nur Aqilah Binti Kamal, Noorsaleha Binti Abdul Nasir, and Nuramirah Binti Mohd Nazim.

Thank you for your encouragements supports

And the sacrifices that you have given.

Last but not least,

Thank you for everything. May Allah Bless All of us.

ABSTRACT

Recent years *Acacia mangium* plantations were attacked by serious wilt disease and cause the destruction of the plantation. Owner of the plantation should take this matter seriously and find the best treatment to overcome this wilt disease. This study was conducted to find a biological control especially endophyte microorganisms to overcome the pathogen of wilt disease. To perform this study, healthy seedling of *Acacia mangium* and its gummosis were collected randomly at Sultan Idris Shah Forest Education Centre and brought to the Plant Pathology Laboratory, Faculty of Forestry. The fresh sample was then cultured into Potato Dextrose Agar (PDA). After a few days of incubation, a microorganism that was grown on PDA were subcultured into special media respectively. In total 76 microorganisms that have been isolated were shortlisted and categorized into fungi and bacteria with similar morphology. This consist of 12 fungi isolates and 11 bacteria isolates. To test their capability, the antagonistic test was done between isolated endophyte microorganisms against *Ceratocystis fimbriata* and several selected pathogen. This test was performed by the dual culture method. From this activity, all the 12 fungi showed positive inhibition against *Ceratocystis* fungus. The fungus that showed the highest inhibition is the fungus isolated from gummosis which is D(4) fungus with 64.58% inhibition. The lowest inhibition was the fungus isolated from root nodule which is HS/N3(C)1 fungus which is 12.50% inhibition. Meanwhile, for endophyte bacteria, only 2 from 11 bacteria showed positive inhibition against *Ceratocystis* fungus. The bacteria were HS/R2(A)2 isolated from primary root and have 8.33% inhibition and HS/N2(A)1 is isolated from root nodule with 2.08% inhibition.

ABSTRAK

Pada tahun kebelakangan ini, ladang *Acacia mangium* telah diserang oleh penyakit layu yang serius dan menyebabkan kemusnahan dan kerugian pada perladangan *Acacia mangium* ini. Pemilik ladang perlu mengambil berat perkara ini dan mencari rawatan yang terbaik untuk mengatasi penyakit layu ini. Oleh itu, kajian ini dijalankan untuk melihat kawalan biologi terutamanya endofit mikroorganisma untuk memerangi patogen penyakit layu. Anak benih *Acacia mangium* yang sihat dan gamosis yang telah dikumpulkan secara rawak yang diambil di Pusat Pendidikan Hutan Sultan Idris Shah dan telah dibawa ke dalam Makmal Patologi Tumbuhan, Fakulti Perhutanan. Sampel segar ini kemudiannya di kultur ke Potato Dextrose Agar (PDA). Selepas beberapa hari pengeraman, mikroorganisma yang ditanam di atas PDA telah disubkulturkan ke dalam media khas masing-masing. Dalam jumlah 76 mikroorganisma yang telah diasingkan dan disenarai pendek dan dikategorikan kepada kulat dan bakteria dengan morfologi yang serupa. Daripada 12 pencilan kulat dan 11 pencilan bakteria satu ujian antagonistik telah dijalankan bagi menguji keupayaan mereka merencat pertumbuhan kulat *Ceratocystis* dan beberapa patogen yang dipilih. Ujian ini dilakukan dengan kaedah pertumbuhan dual. Dari aktiviti ini, kesemua 12 kulat telah menunjukkan perencatan positif, kulat yang menunjukkan perencatan tertinggi iaitu sebanyak 64.58% adalah kulat yang diasingkan daripada gamosis iaitu kulat C(2) dan perencatan yang paling rendah 12.50% adalah kulat yang diasingkan daripada nodul akar iaitu kulat HS/N3(C)1. Sementara itu, bagi bakteria endofit hanya 2 daripada 11 bakteria telah menunjukkan perencatan positif apabila dilawankan dengan kulat *Ceratocystis fimbriata*. Bakteria yang tertinggi nilai perencatannya ialah bakteria HS/R2(A)2 sebanyak 8.33% iaitu bacteria yang diasingkan daripada akar utama. Bakteria kedua yang menunjukkan positif perencatan ialah bakteria yang diasingkan daripada nodul akar iaitu bacteria HS/N2(A)1 sebanyak 2.08%.

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APPROVAL SHEET

I certify that this research project report entitled "Isolation of Endophyte Microorganism of *Acacia Mangium*" by Julia Binti Abdul Latif has been examined and approved as a partial fulfillment of the requirements for the Degree of Bachelor of Forestry Science in the Faculty of Forestry, Univeristi Putra Malaysia.

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TABLE OF CONTENTS

DEDICATION	I
ABSTRACT	ii
ABSTRAK	iii
AKNOWLEDGEMENTS	iv
APPROVAL SHEET	v
LIST OF TABLES	vii
LIST OF FIGURES	viii
LIST OF ABBREVIATIONS	ix

CHAPTER

1 INTRODUCTION

1.1 Background	1
1.2 Problem Statement	3
1.3 Objective	5

2 LITERATURE REVIEW

2.1 *Acacia mangium*	6
2.2 Endophyte microorganism	8
2.3 *Ceratocystis* sp.	11
2.4 *Pestalotiopsis* sp.	13
2.5 *Rigidoporus* sp.	14
2.6 *Fusarium* sp.	15

3 METHODS AND MATERIALS

3.1 Plant materials and design of experiments	
3.1.1 Collecting sample	17
3.1.2 Isolation of Endophytes Microorganisms	17
3.1.3 Subculturing of endophyte microorganism	18
3.1.4 Antagonistic test Activity	19

4 RESULT AND DISCUSSION

4.1 Isolation of endophytic microorganism	21
4.2 Distribution of endophytic microorganisms on healthy *A. mangium*	21
4.3 Antagonistic activity of isolated Endophytic microorganisms	24
4.4 Antagonism test performance	36
4.5 Analysis variance of Antagonistic activity	40

5 CONCLUSION AND RECOMMENDATION 46

REFERENCES 48

APPENDICES

APPENDIX A 53
APPENDIC B 58

LIST OF TABLES

TABLE		PAGE
4.1	Distribution of endophytes microorganisms isolate from healthy *A. mangium*	19
4.2	Total endophytes microorganism after short list based on morphology	20
4.3	Isolated endophytes microorganisms from selected *A. mangium* parts on PDA and NA medium.	21
4.4	Dual culture plate assay between endophytes fungi against four pathogenic fungi obtain from FRIM	34
4.5	Dual culture plate assay between endophytes bacteria against four pathogenic fungi obtain from FRIM	36
4.6	Comparison of antagonistic index of endophytes microorganism against *Ceratocystis*, *Pestalotiopsis*, *Rigidoporus* and *Fusarium*.	39
4.7	Multiple Mean Comparisons between *Ceratocystis, Pestalotiopsis*, *Rigidoporus* and *Fusarium*.	40
4.8	Multiple comparision in terms of homogeneity between *Ceratocystis*, *Fusarium*, *Pestalotiopsis* and *Rigidoporus*.	42

LIST OF FIGURES

FIGURE		PAGE
4.1	Antagonism Index of endophytes fungi source from primary root, against *Ceratocystis sp.*, *Pestalotiopsis sp., Rigidoporus sp.* and *Fusarium sp.*	23
4.2	Antagonism Index of endophytes fungi source from root nodule, against *Ceratocystis sp.*, *Pestalotiopsis sp., Rigidoporus sp.* and *Fusarium sp.*	24
4.3	Antagonism Index of endophytes fungi source from gummosis, against *Ceratocystis sp.*, *Pestalotiopsis sp., Rigidoporus sp.* and *Fusarium sp.*	25
4.4	Antagonism Index of endophytic bacteria source from primary root against *Ceratocystis sp.*, *Pestalotiopsis sp., Rigidoporus sp.* and *Fusarium sp.*	28
4.5	Antagonism Index of endophytic bacteria source from root nodule against *Ceratocystis sp.*, *Pestalotiopsis sp., Rigidoporus sp.* and *Fusarium sp.*	29
4.6	Comparison of endophyte fungi source and inhibition towards *Ceratocystis sp., Pestalotiopsis sp.*, *Rigidoporus sp.* and *Fusarium sp*	31
4.7	Comparison of endophyte bacteria source and inhibition towards *Ceratocystis sp*, *Pestalotiopsis sp.*, *Rigidoporus sp*. and *Fusarium sp*	32

LIST OF ABBREVIATIONS

PDA	Potato Dextrose Agar
FRIM	Forest Research Institute of Malaya
SISFEC	Sultan Idris Shah Forest Education Center
PIRG	Percentage of Inhibition of Radial Growth
UPM	Universiti Putra Malaysia

CHAPTER 1

INTRODUCTION

1.1 Background

1.1.1 Forest Plantation

Plantation forestry is of growing importance in many countries include Peninsular Malaysia, Sabah and Sarawak. In 1982, Malaysian was introduce with a plantation programmed known as the Compensatory Forest Plantation Program (CFPP) that initiated by the Forestry Department in Peninsular Malaysia. The CFPP was planned to establish about 188000 ha of forest plantation using *Acacia mangium*, *Gmelina arborea*, and *Paraserianthes falcataria* as the main species. These chosen species is expected to produce yield of general utility of saw logs at the rate 210 meter cube per hectare at the age of fifteen years old. In fact, commercial forest plantation is the way for the timber based industries in Malaysia which aim to increase their production in future to meet the government's target of national export value earning amounting is RM53 billion by 2020 (Lim et al., 2011).

Most of the forest plantation trees grown in eastern and southern Africa are established from seed, commonly imported from South Africa or Australia. Based on record, the area of forest plantation for *Acacia mangium* in Malaysia is about 310,000 ha. *Acacia mangium* is one of several promising fast-growing tree species in plantations on Peninsular Malaysia, Sabah, and Indonesia. It has also shown a high survival rate in poorer soils.

Recent experiments have demonstrated basic conditions for the successful introduction of *Acacia mangium* in tropical regions, as well as the application of appropriate silvicultural treatments. For climate change mitigation, *Acacia mangium* plantations are expected to provide efficient and profitable (Matsumura, 2011). According to FAO (2006) world forest statistics, deforestation has become a serious global problem. Degradation of forest resource cause impact in the loss of structure, diversity, function, and other qualitative features is also an increasingly salient issue, especially in tropical region (Potter, 2006).

1.2 Problem Statement

The wilt disease caused by *Ceratocystis fimbriata* fungi is threat to *Acacia mangium* plantation around the Peninsular Malaysia, Sabah and Sarawak. This problem is possibly could seriously give big damage to forest plantation sector because the disease has the potential to spread and threat to other agricultural sectors. When this happen the growth rates of *Acacia mangium* that can produce good quality of saw log became reduced and affecting future timber yield.

Since then, no treatment has been developed that efficiently controls the disease. Therefore, the development of an alternative method such as bio-control is desirable. Thus, to prevent the spread of *Ceratocystis fimbriata* fungus and to reduce the use of pesticides to control the wilt diseases, studies of complementary or alternative methods, especially bio-control, have sparked great interest in silviculture.

This study will focused on isolation of endophyte microorganism in healthy *Acacia mangium* against *Ceratocystis* and another selected pathogens which are *Pestalotiopsis*, *Rigidoporus*, and *Fusarium*. Isolation of endophyte microorganism is one way to find inhibitor that could be can fight against *Ceratocystis fimbriata* that cause wilt disease to *Acacia mangium*. As exampale, there a study on spruce budworm *Choristoneura fumiferana* that attacked conifers. The result showed that endophyte *Phialocephala scopiformis* that lives in *Picea glauca* (Pinaceace) can produce chemical compound against the pathogen *Choristoneura fumiferana* (Rohlfs and Churchill, 2011; Larkin et al., 2012). So it is believed that Acacia mangium

has its own endophytic microorganism that secrete chemical compound against *Ceratocystis fimbriata* and selected pathogens that attack this species.

1.3 Objective

1. To determine the distribution of endophyte microorganism on healthy *Acacia mangium*.
2. To assess the inhibition between endophytes microorganism and *Ceratocystis fimbriata* and several selected pathogens in dual culture method.

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