

Centre for Plantation Forest Research and Development

Proceedings INTERNATIONAL

Research on Plantation & Research & Research on Plantation & Research & Research

Bogor - Indonesia, 5 - 6 November 2009

PROCEEDINGS

OF

INTERNATIONAL SEMINAR RESEARCH ON PLANTATION FOREST MANAGEMENT: CHALLENGES AND OPPORTUNITIES

Editors:

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Bogor – Indonesia, November 5 - 6, 2009



MINISTRY OF FORESTRY
FORESTRY RESEARCH AND DEVELOPMENT AGENCY
CENTRE FOR PLANTATION FOREST RESEARCH AND DEVELOPMENT

PREFACE

Forest plantations contribute and are beneficial in terms of environmental, economic, as well as social. They contribute in providing raw either timber (as supply of raw material for wood-based industries) and non-timber (food, medicines, amenities, environmental services) forests product and benefits. To be positively giving impacts, forest plantations should be managed appropriately. Inappropriate management can result in many problems e.g changes in soil and water, unsustainable yield, land use conflicts, etc.

Today, the growth of population demand for more raw material, and these are greatly relied on plantation forest management. On the other hand, the great development of plantations and industries are always associated with community. Appropriate attention should also be paid for community in establishing and developing plantation forest, as the paradigm in forestry development is changing from resource-based to community-based. Meanwhile, the development of forest plantations are also strongly required to be environmentally-sound as mention in the mandate of the Kyoto Protocol, Clean Development Mechanism (CDM) as well as the concept of REDD promote utilization of forest-based environmental services, is one of example under which both large and small scale holders get opportunities to participate in plantation activities as well as contribute positively in the environmental management of plantation forests. Meanwhile, the development of forest plantations are also strongly required to be environmentally-sound.

Based on this background, an International Seminar has been organized by the Centre for Plantation Forest Research and Development, Indonesia, in Bogor, Indonesia, on November 5 – 6, 2009, with the theme of "Research on Plantation Forest Management: Challenges and Opportunities". The seminar aimed mainly to learn state of the art of R & D on plantation forest management in all parts of the world, share knowledge and experiences in R & D in relevant fields, discuss challenges and opportunities for R & D in supporting the world plantation forest development.

This proceeding is intended as documentation and publication of papers and posters presented in the seminar, which delivered in three group of topics from varied aspects of plantation forest research and development (management, industries and environment management of plantation forest). There were four invited papers, twenty eight presented papers, twenty four voluntary papers, and twenty posters' abstracts provided and published in this proceeding.

I would like to acknowledge the authors for the papers and posters contributed in this proceeding. The comments and review from the editors towards the publishing of this proceeding are also greatly appreciated. I am also grateful for the participants who had been actively involved in the seminar. Finally, it is our hope that this proceeding will be beneficial for any users and contribute towards the development of plantation forests research and development.

Bogor, November 2010

Director,

Dr. Bambang Trihartono

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REPORT BY DIRECTOR OF CENTRE FOR PLANTATION FOREST RESEARCH AND DEVELOPMENT

International Seminar Research on Plantation Forests Management : Challenges and Opportunities

IPB International Convention Centre, Botani Square, Bogor - Indonesia 5 - 6 November 2009

His Excellency, the Minister of Forestry, or the representative His Excellency, the Director General of Forestry Research and Development, Bapak Tachrir Fathoni Distinguished guests, Ladies and Gentlemen,

Good morning and welcome to this International Seminar with theme of "Research on Plantation Forests Management: Challenges and Opportunities".

Excellencies, Ladies and Gentlemen,

First of all, let us praise God Almighty, for the graciousness and mercifulness that enable us together here this morning, on the occasion of opening ceremony of International Seminar, Research on Plantation Forests Management: Challenges and Opportunities.

Excellencies, Ladies and Gentlemen,

This seminar is organized with the main objectives to learn state of the art of R & D on plantation forest management in all parts of the world, share knowledge and experiences in R & D in relevant fields, discuss challenges and opportunities for R & D in supporting the world plantation forest development.

The organizing of this seminar is part of the responsibilities of the CPFRD as an R & D governmental institution in facilitating the stakeholders who are aware of issues in plantation forests management. Through this seminar, I invite each participant to freely express their idea as well as sharing their experience in managing plantation forests, and at the end of this event, we will try to identify the key issues of plantation forest management, and derive some lessons to learned which will be useful to deal with this rapid and global plantation forest development. We will also try to see where future research should be directed in order to address the challenge of filling the gap between the supply and demand for wood and wood products and achieving sustainable forestry in the long term.

Excellencies, Ladies and Gentlemen,

I am very glad today that we have special guest from overseas. There are 3 keynote speakers and 5 invited speakers from Australia, Kenya, and Indonesia, who are confirmed to present their paper and share their idea and experience. I believe all of these speakers have world-wide reputation and huge experience in plantation forests management, and some of them might have known Indonesia quite well.

I am grateful that the Organizing Committee has received many positive responses. A hundred and fifty-five participants are registered in this seminar, which are varied in origin of institution. There are seventy participants from R&D governmental institution, eight from non-R&D governmental institution, twenty-five from private forestry companies, thirty-three from universities, and nineteen participants from overseas.

Totally the organizer has received eighty abstracts, and there will be thirty-eight papers presented and about twenty posters displayed within the two-days seminar. I hope there will be a

productive discussion during the parallel session, so at the end of the seminar each of participants can draw some lessons learnt and bring them to their institution.

I would like to take this opportunity to express our gratitude to the Excellency Mr. Tachrir Fathoni who will formally open this international seminar on behalf of the Minister of Forestry.

I would also like to express my special appreciation to Sinar Mas Group and Perum Perhutani, our partners, which has given us valuable supports in organizing this seminar. I also thank all speakers and all participants and the Organizing Committee, and everybody who make this seminar possible.

Thank you and have a productive discussion.

Bambang Tri Hartono



OPENING ADDRESS BY THE MINISTER OF FORESTRY

INTERNATIONAL SEMINAR RESEARCH ON PLANTATION FOREST: CHALLENGES AND OPPORTUNITIES BOGOR, 5 - 6 NOVEMBER 2009

Distinguished guests, participants, ladies and gentlemen
It's my pleasure to be here this morning marking the opening of the International Seminar with
theme on Research on Plantation Forest: Challenge and Opportunities. To participants from
overseas welcome you to Bogor.

Ladies and gentlemen,

According to the FAO's 2002 forest assessment, in the year 2000 around 187 million hectares plantation existed throughout the world. Although it constituted only 7 percent of the global forest area, or about 271 million hectares in 2005, but planted forests contributed a higher proportion of overall forest goods and services. The FAO estimates the global rate of new planting at 4.5 million hectares per year, with Asia accounting for 70 percent of this growth, particularly in China, Indonesia, Malaysia, and Vietnam. In terms of global plantation resources, the Asia-Pacific region is an important player.

There are some countries with major plantation which greatly supply an increasing world's wood demand, namely Pines in New Zealand, Australia, Chile and Southern United States; *Eucalypts* in South Africa, Brazil and China; and *Acacia mangium* in Indonesia. It has also recorded by FAO that five Asian countries namely China, India, Indonesia, Japan, and Thailand, are among the top ten world's plantation forest accounting for 55 percent of the global plantation forests.

Indonesia, itself, have a long tradition of regular forest plantation with *Tectona grandis* which was started in 1873 mostly in Java, and with *Pinus merkusii* in 1916 in Sumatra. After the next decades, over 9.9 million hectares plantation have been established, of which around 1 million hectares are teak in java and 1.4 million hectares are *Acacia mangium* in Sumatera and Kalimantan, with the target of degraded land dominated by imperata grasses and unproductive secondary forests. Indonesia has become the major producer of *Acacia mangium* pulp for the world market. With such a vast area of land, Indonesia could become one of the major producers of timber from plantation forest.

Ladies and gentlemen,

Let us now look at the demand side. Current level of global industrial roundwood consumption is about 1.5 billion m3 with 0.33 billion m3 share from plantation or about 20 percent of the total consumption. Based on the global scenario study this level of consumption may reach 4.1 billion m3 by 2010. This gives us a clear sign how important the role of plantation forest in the future to meet increasing demand for wood and wood products. The development of forest-based sawmills and plywood mills (1980s-1990s) and pulp and paper industries (mid 1990s) plays an important role in forest resource management. Wood supply from plantation forests will have to provide an increasingly larger share of the supply in the near future, particularly of pulplogs. Technology and marketing problems are emerging in processing logs from those plantations. The development of industrial forest plantations is focused on meeting the pulp and paper industries' needs for raw materials, and therefore the tree species are dominated by fast growers such as *Acacia* spp.

Indonesia's forest-based industry comprises a mix of non-panel forest-based industry of pulp and paper mills, and panel-based industry of sawmills, plywood mills, block board mills, chip mills

and medium-density fibreboard mills. The rapidly expanding Indonesian forest-based industries were not well supported by a long-term sustainable log supply. There were only minor efforts to increase efficiency and competitiveness. The annual log shortage is estimated to be more than 35 million cum. The technological aspects of the forest-based industry are also neglected.

Master plans are needed both for plantation forest development and for forest-based industries. In addition, plantation forests and forest-based industries are currently under initial restructuring processes.

Although the demand for a sustainable supply of industrial wood has primarily driven the importance of plantation forests, FAO has also emphasized the social and environmental dimensions of plantation forests. This is not just because they have an important role in providing economic and social benefits in eradicating poverty in developing countries, but, also important for industrialized countries where marginalized groups and indigenuous peoples have previously been excluded from the benefits of development processes.

Indonesia has experienced shortage of raw material for wood-based industries for more than five years now, where the demand of industries for logs has reached three to four times higher than the capacity of existing domestic sources both natural and plantation forests to supply. This situation on one side can be an opportunity for Indonesia to expand the existing timber plantation by reforestation of the vast area of degraded land, on another side, it can be a challenge in tackling heavy criticism particularly on social and environmental issues.

Ladies and gentlemen,

I believe we are all aware that vast development of global plantation could not be separated from the role of R & D. While developed nations may not encounter problem of funding shortage and expertise, many developing countries face a huge challenge of lacking both capacity and financial resources to undertake research that could make major contribution towards plantation development.

Nevertheless, the trends of plantation establishment will continue to grow supported by many R&D programmes that are directed towards improving the productivity of plantations. This international seminar discussing the role of R & D in plantation forest is timely and of significant importance. Therefore, speaking on a personal perspective, I look forward to this seminar, as it will has an importance in providing value and contribution towards the development of plantation forests, both for national and international level. For Indonesian competency, it is hoped that this seminar will give us information towards the improvement of the state of plantation forest management.

Overall, I hope that the exchange of experience and information will be beneficial to all participants and we all expect that the seminar will be fruitful.

With the bestow of God the Almighty I declare this seminar officially open.

On behalf of the Minister of Forestry
Director General of Forestry Research and Development Agency

Dr. Ir. Tachrir Fathoni, MSc.

CHARACTERS OF SPECIES PLEUROTOID: MORPHOLOGICAL, PHYSIOLOGICAL AND LIGNINOLYTIC STUDIES TO SUPPORT BIOINDUSTRY

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ABSTRACT

Morphological, physiological and ligninolytic characterization of pleurotoid species from Bogor district were studied. The method for isolation of pleurotoid was a tissue culture conducted, using modified MEA medium. The selected isolates were maintained and cultured on various media. To identify, the mycelium, fruiting-body, basidiospores, cystidia and other structures of the fungi were observed using a microscope stereo-monocular and scanning electron microscopy (SEM). The physiological aspects of the fungi were characterized based on the growth rate, oxidation reaction, vegetative and reproductive phase, total biomass and biological efficiency. The growth rate of the fungi was observed on media PDA, MEA, MPA with different temperatures and pH. The oxidation reaction was conducted using AAG and AAT media. The ligninolitic activity of fungi was measured based on the biodegradation of Sengon (Paraserianthes falcataria) wood sawdust substrate by the fungi. More than 30 isolates of Pleurotoid were isolated from several places in Bogor. Six isolates of those isolates were able to form fruiting bodies on media namely Pleurotus djamor EB9 and five isolates of Hohenbuehelia petaloides. Analysis cluster of isolates based on the morphological and physiological characterization showed that P. djamor EB9 was different from H. petaloides. Classification based on an ligninolytic character was different from classification based on morphological and physiological characters. P. djamor EB9 seemed to be separated from other isolates. This shows that there was different ligninolytic character among the isolates. The best isolate for bio-pulping and bio-bleaching agent was P. djamor EB9 on vegetative phase.

Keywords: Biodegradation, *Hohenbuehelia petaloides*, identification, morphological, Pleurotoid fungi, *Pleurotus djamor* EB9

I. INTRODUCTION

a. Background

A forest with its biodiversity is a very important natural resource in Indonesia. Fungi as one of them, has not optimized its potentials in Indonesia. *Pleurotus* spp. as one of the white rot fungi could also be used as edible fungi, which was able to degrade lignoselulose materials effectively and efficiently. Therefore a research to determine the degradation value of the fungi in woody species was needed.

The conventional pulping process either by way of a mechanical process and also way of chemistry also requires a real energy height (Higley & Dashek, 1998). Applying biotechnology that was by exploiting a biological process that applied mushroom decaying lignin in the process of technology bio-bleaching and bio-pulping, is one of alternatives and big breakthrough that needed to be studied (Oriaran *et al.*, 1990; Higley & Dashek, 1998).

Some species Pleurotoid was wood rotting fungi which degradation of wood substrate could become an organic material modestly through an enzymatic hydrolysis process. The organic material can be exploited by other microorganism was including flora and forestry crop (Alexopoulos et al., 1996; Dix & Webster, 1995). Species Pleurotoid like Pleurotus spp. and Hohenbuehelia spp. (Brown, 1981). One of a mushroom species decaying wood having potency to degradation of lignin is Pleurotus spp. P. ostreatus is known by a degradation of lignin is more efficiently compared to Phanerochaete chrysosporium and a big potency for a pulp industry (Kerem et al., 1992; Hadar et al., 1993). Some mushrooms which had been known as a degradation of lignin was P. chrysosporium and T. versicolor (Valmaseda et al., 1991 in Hadar et al., 1993).

Untill now research about types Pleurotoid in Indonesia is still very limited, though this mushroom has been recognized and collected and it is exploited by the public for various needs (Gunawan, 1997). Researching about the type of Pleuroroid from genus *Hohenbuehelia* in Indonesia has not existed, either morphological and also potencial.

b. Objective

The objective of this research was to study the characteristics of species the original Pleurotoid Bogor; characteristics examined included morphology, physiology and the ability of biodegradation based on the decomposition rate of the test substrate.

c. Method

This research was conducted in May 2004 to September 2006 in Bogor Agricultural University (IPB), Lembaga Ilmu Pengetahuan Indonesia (LIPI) Cibinong and the Institute of Plantation Research of Indonesia (LRPI), Bogor. Those six isolates were *Pleurotus* EB14-2, *Pleurotus* EB24, *Pleurotus* EA4, *Pleurotus* EAB7, *Pleurotus* EB6 and *Pleurotus* EB9. The identification was based on their morphological, physiological, and ligninolytical characters.

The morphological character, macroscopically and microscophically, included the visual appearance of the fruiting body, the number of stalks, the sizes of pileus and stalk, the lengths and widths of basidospora, sistidia, and basidia. The paraphine technique in making a preparats was referred to a microtom method/SPM modification (2002). The morphological observation was done by using a light microscope, lucida microscope, and scanning electron microscope (SEM). Besides microscopic and macroscopic photos, line-drawing was also conducted by a licensed painter. The terminology of macroscopic morphological characteristic description was referred to Cornér (1981 and 1994) and Brown (1981). The terminology of a fungi colony texture characteristic description was referred to Stalpers (Rayner and Boddy, 1988). The terminology of microscopic characteristic was referred to Rayner and Boddy (1988), Corner (1981 and 1994) and Brown (1981).

The isolates were characterized physiologically including the growth rate colony, oxidation reaction, total biomass, biological efficiency, vegetative and reproductive phases by using an isolate of *P. ostreatus* HO as a standard comparison. The growth rate of the fungi was observed on media PDA, MEA, MPA with temperatures of $10(\pm 1)^{\circ}$ C, $20(\pm 1)^{\circ}$ C, $29(\pm 1)^{\circ}$ C and $35(\pm 1)^{\circ}$ C, and media pH of 5, 6 and 7. The oxidation reaction was conducted using malt media with galat acids (AAG) and tannic acid (AAT). The total fresh weight, biological efficiency, vegetative and reproductive phases were observed on Sengon wood sawdust media with a weight about 400 gram.

The ligninolytic character of these six fungi isolates were measured after inoculation into Sengon wood sawdust substrate inside a plastic bag with substrate weight of about 400 gram; other additional substances were paddy scalp, gypsum, calcium and water. Observation was done on Sengon wood sawdust substrate since a vegetative phase until a reproductive phase. Samples were opened, destructed with Hammer Mill, and then dried in oven on temperature of 40° C in $4 \sim 6$ days until the water content reached 15%, and weighed about 30 gram for analyses. Analyses were done by measuring a water soluble extractive substance (TAPPI T 207 om-88 Standard), NaOH (sodium hydroxide) 1% soluble extractive substance (TAPPI T 212 om-88 Standard), alkohol-benzena soluble extractive substance (TAPPI T 204 om-88 Standard), lignin content (TAPPI T 13 os-54 Standard), holocellulose content with a method of browning (TAPPI T 211 m Standard), and a cellulose content with a Cross and Bevan method (Meulenhoff et al. 1977; TAPPI 1996).

The design of applied experiments was a completely randomized design (RAL), and at some treatment; with factorial planning and with area planning RAL. Analysis of variance data processing applied SAS9 and analysis cluster applied SPSS13.

II. RESULTS AND DISCUSSION

a. Morphological Character

The identification result of six isolates that were able to form a fruit body was five isolates of *Hohenbuehelia petaloides*, which were EB14-2 (light brown), EB24 (gray brown), EA4 (light brown), EAB7 (gray brown), and Eb6 (gray brown), and also *Pleurotus djamor* EB9 (pink). In general five isolates *H. petaloides* (EB14-2, EB24, EA4, EAB7 and EB6) had marked a macroscopic much the same except to some differences at a fresh cap diameter measure and piles color when young, but at some stage, it showed similar color and phenotype. The example was a description of *H. petaloides* EA4 (Figure 1).

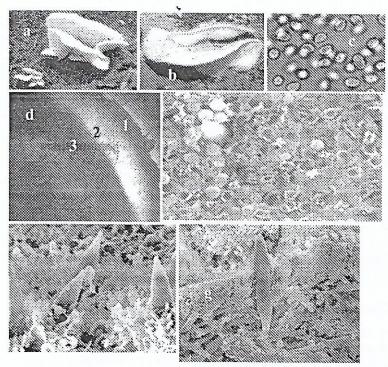


Figure 1. Vision a). Fruiting body of *H. petaloides* EB14-2 and b) EA4; c) Spora of *H. petaloides* EB14-2; d) microscopic vision of *H. petaloides* EB24: (1) pileipelis was having pigment, (2) transparent gelatine layer, (3) flesh (flesh) having a dark colour; e) pleurosistidia and basidiospore and basidia *H. petaloides* EA4 with SEM; and f) and g) basidiospore, pleurosistidia (metuloid) and mycelium in hymenium network *H. petaloides* EB6 with SEM

P. djamor EB9 has different morphology characters compared to a group of *Hohenbuehelia* sp. that was the description was hereunder, and fructification vision, spore and the hymenium seen at Figure 2.

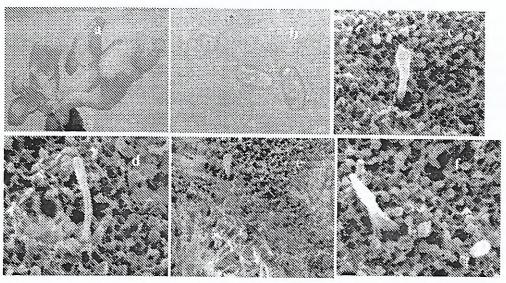


Figure 2. Vision a) Fruiting body and b) basidiospore and c), d), e) and f) basidiospore, pleurosistidia and mycelium in hymenium network *P. djamor* EB9 with SEM

The analysis result of isolate group based on morphological characters showed that *P. djamor* EB9 is different from *H. Petaloides* (Figure 3). Analysis cluster was done to character variable morphologist, which consisted of a number of handles, measure piles and handle, long and wide basidiospore, sistidia and basidia. Result of its presented at Picture.

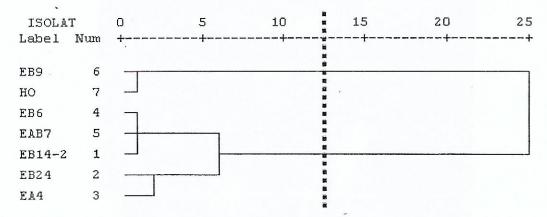


Figure 3. Dendogram based on character morphologist out of seven mushroom isolates

b. Physiological Character

Performance of a colony on optimum culture media, temperatures and pH as variables varied, showing differences in a growth rate colony of the seven isolates. The growth rate colony of five isolates of *H. Petaloides* EB14-2, EB24, EA4, EAB7 and EB6 were slow (less than 2,0 mm/days). Those of *P. djamor* EB9 was moderate (2,3~3,4 mm/days) and those of *P. ostreatus* HO was fast (6,1~7,9 mm/days). Keys of patern of *H. petaloides* were A-P-S-1.1.1.1.9.2.1.1.3.1.2, keys of patern of *P. djamor* were A-P-I-1.1.1.1.9.2.1.1.2.1.2 and keys of patern of *P. ostreatus* were A-P-I-1.1.1.1.9.2.1.1.1.1.2 (Nobles 1948).

Positive oxidation reaction on AAG and AAT media showed that all isolates including white-rot fungi. *P. djamor* EB9 and *H. Petaloides* EA4 were potentially white-rot fungi with moderatelly strong reaction on media AAG and AAT.

Reaction of oxidation. Media MEA gallic acid was added to become AAG and media MEA tannic acid was added to become AAT. All mushroom isolates showed positive reaction to tannic acid medium (AAT) and gallic acid (AAG). This was marked with the forming of light chocolate zona until dark chocolate at a medium AAT and AAG around isolate colony mycelium. The reaction of oxidation at media AAT and AAG which was strong enough owned by *P. ostreatus* HO, *P. djamor* EB9 and *H. petaloides* EA4. Reaction of oxidation at media AAT from all isolates strong enough with score +++ (moderatly strong). But at AAG, which was most uppermost was *P. ostreatus*, *P. djamor* and *H. petaloides* EA4 with score "+++" (moderatly strong), and besides at AAG has reaction of oxidation "+" (very weak). The reaction of oxidation which were positive at AAG and AAT showing all isolates which was a white decaying mushroom. Assuming that isolate *Pleurotus spp.* and *Hohenbuehelia spp.* that was tested at media AAG and AAT released enzyme ektraseluler oxidase with the happening of reaction of oxidation with gallic acid and or tannic acid.

The wild *Pleurotus* group also showed a wide variation in terms of total biomass $(34,8\sim142,4\,\text{gram})$, biological efficiency $(29,1\sim119,0\%)$, average time of vegetative phase $(14,0\sim83,0\,\text{days})$, reproductive phase $(112,5\sim199,0\,\text{days})$ and harvest frequency $(4,0\sim6,0\,\text{times})$. The vegetative phase period of *P. djamor* EB9 was the shortest $(19\,\text{days})$ compared to five isolates from genus *Hohenbuehelia* $(50\,\text{days})$.

The number of crops average of seven mushroom isolates was ranging from $2\sim6/100$ day phase reproductive. The total fresh weight and EB isolate was immeasurable that was *P. ostreatus* HO (130 grams and 110%), *P. djamor* EB9 (100 grams and 80%), and five isolate from gender *Hohenbuehelia* which had a total fresh weight between 40-24 grams and EB between 39-20%. Biological efficiency average of seven isolates could be grouped to become three groups those were *P. ostreatus* HO separated *P. djamor* EB9, and five isolates from genus *Hohenbuehelia* that stayed by one separate groups with EB which was enough low.

Analysis cluster based on physiological character is done based on the growth rate of every isolate at various media, and temperature, vegetative phase, phase reproduktif, number of crops, total fresh weight and EB. The result of analysis cluster indicated that *P. djamor* EB9 differed in cluster from five isolates *H. petaloides*. Where *P. djamor* EB9 stayed in one groups together *P. ostreatus* HO. And five isolates from *H. petaloides* stayed in a separate other cluster (Figure 4).

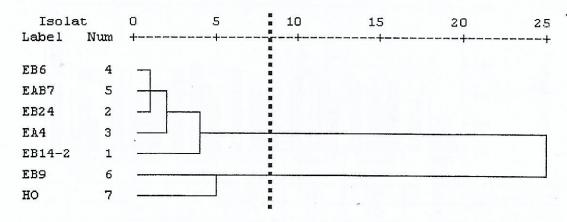
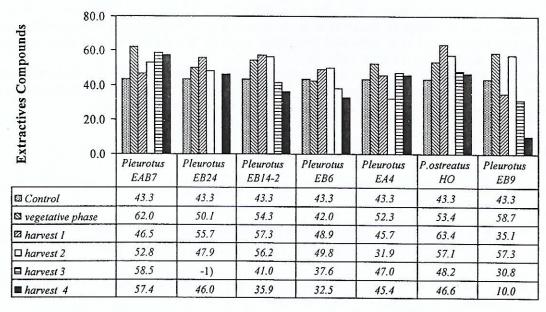


Figure 4. Dendogram based on physiological character out of seven mushroom isolates

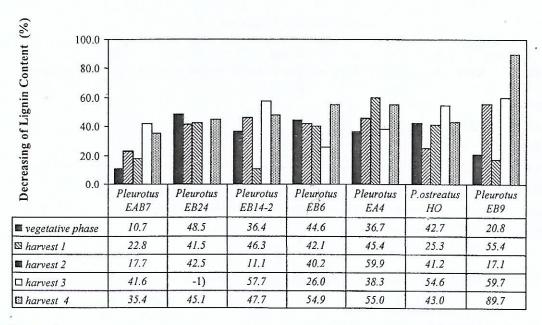
c. Ligninolityc Character

Having applied each wild *Pleurotus* group isolates resulted in increasing of extractives total compounds both in vegetative and reproductive phases (Figure 5). Each wild *Pleurotus* group isolates showed variation in decreasing the average of lignin content ($10.7 \sim 89.7\%$) and cellulose ($18.9 \sim 87.4\%$). *P. djamor* EB9 was able to decrease the highest lignin (89.7%) and cellulose (87.4%) content of substrate (Figure 6). Classification based on ligninolytic character was different from classification based on morphological and physiological characters. *P. djamor* EB9 seemed to be separated from the other isolates. This showed that there was different ligninolytic character among the isolates. The best isolate for bio-pulping and bio-bleaching agent was *P. djamor* EB9 on a vegetative phase.



1) not observed

Figure 5. Extractives total compounds of substrate having applied of each wild *Pleurotus* group isolates



1) not observed

Figure 6. Decreasing of lignin content of substrate having applied of each wild *Pleurotus* group isolates

III. CONCLUSIONS

Based on the result of characterization of morphology and physiology out of six mushroom isolates of distinguishable Bogor became two species, those are *P. djamor* EB9 having a pink colour and *H. petaloides* for five of other isolates which of were EB14-2 (light brown), EB24 (gray brown), EA4 (light brown), EAB7 (gray brown), and Eb6 (gray brown). *P. djamor* EB9 has closer affinity with *P. ostreatus* HO compared to isolates *H. petaloides*.

Grouping based on ligninolitis character differed from grouping based on character morphologies and physiological. Result of analysis cluster based on ligninolitis character showed that *P. djamor* EB9 seemed to be separated from other isolates. This showed that between isolates had ligninolitis character which were different.

From the result of this research, it was expected that *P. djamor* EB9 and *H. petaloides* EA4 are potencial as biopulping and biobleaching agent. Therefore, both the isolates needed to be checked furthermore from enzymatic activity aspect, and also to isolate the ligninase enzyme.

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