PROCEEDING

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(ICBS BIO-UGM 2013)

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Biological Approach for Sustainable Development
of Tropical Biodiversity for Human Prosperity"

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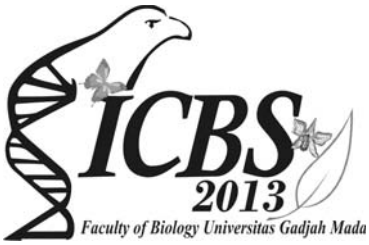

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WELCOMING SPEECH FROM CHAIR PERSON OF THE ORGANIZING COMMITTEE

Distinguished guest,
Rector Universitas Gadjah Mada, Prof. Dr. Pratikno, M.Sc., Keynote Speaker, invited speaker, participants, sponsors, Ladies and Gentlemen,

Good Morning and May God bless all of us.

It gives me great pleasure to extend to you all a very warm welcome to the **International conference on Biological Science: Advances in Biological Science: Biological Approach for Sustainable Development of Tropical Biodiversity for Human Prosperity** (ICBS Bio-UGM 2013), here in Yogyakarta.

Ladies and Gentlemen, It is gratifying to note that the conference is designed to improve awareness on environmental sustainability, in order to understand the right policy regarding bio-conservation action. To increase the consciousness and understanding on the potential, economic value and sustainable management of tropical biodiversity through biotechnology and bioengineering application and To strengthen international scientific network of biological and biology-related scientists to share and exchange progress in various fields of biological research.

No matter how much we can do by ourselves on the institutional and national level, it is never enough. International level of collaboration work would be the best answer. Therefore I wish that this event which is attended by distinguish speaker and attendants from Japan, Sweden, Australia, Korea, Malaysia, and Indonesia, would be a great opportunity for us to established scientific collaboration between scientist internationally.

Hereby, on the behalf of Organizing committee I acknowledge Prof. dr. Ali Ghufron Mukti M.Sc.,Ph.D (Universitas Gadjah Mada/Ministry of Health) as a Keynote speaker, and also for the following invited speakers, Prof Yasumasa Bessho (NAIST, Japan), Prof. Woo Je-Tae (Chubu University, Japan), Prof. Choi Byeong-Dae (Gyeong Sang National University, Korea), Assoc. Prof. Eva Albers (Chalmers University of Technology, Sweden), Prof. Manshor Mashoor (USM, Malaysia), Dr. Fransisco Lopez Ruiz (Curtin University, Australia) and Dr. Tjut Sugandawaty Djohan, M.Sc. (Universitas Gadjah Mada, Indonesia) for willingness to share their valuable knowledge and scientific information.

To make this conference happen, I would like to gratefully acknowledge to the valuable contributions from personal and institutional sponsorships including Universitas Gadjah Mada, Bank Negara Indonesia (BNI), CIDES, PT. Fajar Mas Murni Semarang, Miconos Transdata Nusantara, P.T. Roche, Drs. Heri Susanto, MM., Dr. Suwarno Hadisusanto, and Dra. Lucia Rina.

I would like also to take this opportunity to express my sincere thanks to Dean and Vice Dean of Biology Faculty, Universitas Gadjah Mada, for giving us opportunity and support to organize this conference. Heartfelt thank is delivered to steering committee, academic reviewer, Organizing Committee, for all of participation and hard works. All of them have been working since the beginning of the planning stage and they are still here today for all of us.

P-PD-04

IN VITRO SELECTION ON FUSARIC ACID OF *Vanilla planifolia* PLANTLETS FOR OBTAINING A CULTIVAR, WHICH RESISTANT TO *Fusarium oxysporum* f. sp. *vanillae*

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ABSTRACT

The most production constrain on *Vanilla planifolia* Andrews plantation recently has been caused by foot rot disease that later influence in decreasing the yield product. This disease is caused by *Fusarium oxysporum* f. sp*. vanillae* (*Fov*). So far, the disease has not been successfully prohibited although some experiments had been conducted. The use of foot rot resistant cultivar has been introduced, which expected has high yield as one alternative method for controlling this disease. A resistant vanilla plantlet to *Fov* has been initiated by *in vitro* selection on MS medium containing fusaric acid (FA) on selective concentration. The purpose of research were to study and determine: 1) The proper combination of 2,4-D and NAA for callus initiation and shoot development from shoot tip explants, along with proper concentration of BAP for shoots initiation from nodal explants; 2) The FA concentration of plantlet selection tolerant to steady growth; 3) The proper concentration of FA for *in vitro* selection for suppressing the *Fov*. Results showed that: 1) the proper concentration of both combination between 2,4-D and NAA is about 2,0 mg/L and 10 mg/L respectively for callus initiation and shoot development from shoot tip explants; also the BA concentration is 1,0 mg/L for shoot initiation from a nodal explants; 2) the FA tolerant concentration for plantlet selection with vanilla steady growth is between 90 ppm-110 ppm; 3) the 110 ppm of FA was effective for suppressing the *Fov* compared to 90 ppm and 100 ppm respectively.

Key words: *Vanilla planifolia* Andrews, the vanilla foot rot disease, *Fusarium oxysporum* f.sp. *vanillae*, *in vitro*, fusaric acid

INTRODUCTION

Vanilla is one of export commodities industry in Indonesia foreign exchange. Market demand of Indonesian vanilla had been considerable because it has high levels of vanillin about 2,75%. The foot rot disease on vanilla is the most crucial disease caused by *Fusarium oxysporum* f. sp*. vanillae* (*Fov*), which is the most limitation and up to know days is not well managed yet. One an alternative way to control foot rot disease could be done by using a cultivar, which is resistant to this disease. In order to gets a new vanilla cultivar, which resistant to BBV by using an *in vitro* selection method on medium containing fusaric acid.

MATERIALS AND METHODS

Materials used for this research including: *Laminar Air Flow Cabinet* (*LAF*), *autoclave, Whatman filter paper* no 1 and 42, *syringe filter* with diameter of 0,45 µm and 0,22 µm, culture flasks 500 mL, digital Canon Ixus 951S camera. *Vanilla planifolia in vitro* Plantlet, fusaric acid from *Sigma chemical Co.*, *Naphthalene Acetic Acid* (NAA), *2,4-Dichlorophenoxy Acetic Acid* (2,4-D), *Benzine Amino Purine* (BAP), *Plant Preservative Mixture* (PPM), and MS (*Murashige & Skoog*) solid ready used medium.

Preliminary study was done on *in vitro* regenerated vanilla plantlet by using different explants, planted on the medium containing such a plant growth regulators. Whereas these 2,4-D, NAA and BAP as plant growth substances were used in this research. Vanilla plantlet was selected with fusaric acid (AF) supplementation to MS solid medium in different concentration (0, 90, 100, 110 and 120 ppm) for 12 weeks. Resistant plantlet has already proof when spores of microconidium *Fov.* had infected to plantlet didn't affected on this *in vitro* plantlet respectively and this result supported by Hadisutrisno (2004).

Qualitative information as the result on this research was consisted as narratives descriptive and supported by photographs. After that, data's were statistically analyzed by Completely Randomized Design. As quantitatively data's from each parameter measured, were compiled and statistically analyzed by analysis of variance (ANOVAs). If the result showed a significantly different, then was continued analyzed by using Duncan Multiple Range Test (DMRT) analysis with accuracy 95%.

RESULTS AND DISCUSSION

The results showed that a combination of 2,4-D 2,0 mg/L plus NAA 10 mg/L was the best callus for initiation and development of shoot from shoot tip explants and the BA 1,0 mg/L is the best for shoot initiation from a nodes explants. Vanilla plantlet regeneration was the best results obtained by using nodal explants. In one node explants which produced from 3 up to 6 shoots within 12 weeks (Figure 1). Capability in nodal explants forming a shoot arising due to axillaries *meristem* consisted in the stem, so explants easily regenerate to form a shoot (Neelannavar, 2006).

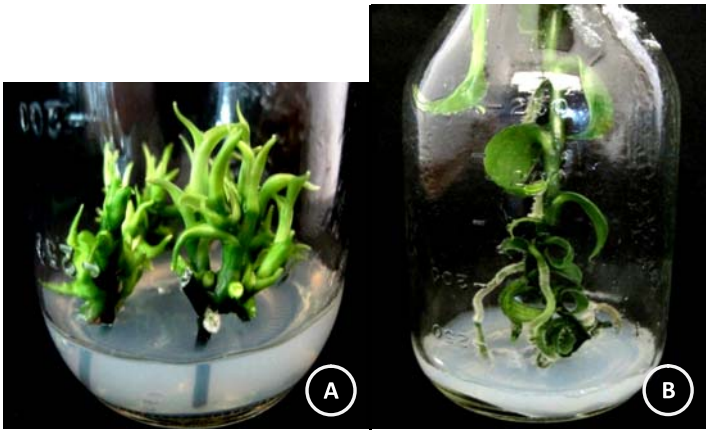

Figure 1. Plantlet regeneration from stem nodes explants *Vanilla planifolia* Andrews on MS medium + P 1 mg/L of Benzyne Amino Purin. A= 12 week of plantlet, B= 32 weeks of plantlet.

The tolerant FA concentration on plantlet selection with vanilla steady growth was between 90 ppm-110 ppm. They showed yields in plantlet vanilla at about 17,14 % (90 ppm), 12,00% (100 ppm) and 10,59 % (110 ppm), all of them insensitive to FA (Table 1) Phenotype plants insensitive toward FA may occurred as a result of peroxides enzymes on plant variants that detoxification toxins (Jayasankar *et al*., 2000; Saravanan, 2004; Svabova & Lebeda, 2005).

Table 1. Percentage of Vanilla plantlet number, which still survive on multiplication medium

Konsentrasi AF (ppm)	Percentage of Vanilla plantlet number **in** different weeks				
	I	II	III	IV	XII
0	100,00	100,00	100,00	98,89	88,89
90	27,14	25,00	25,00	23,57	17,14
100	14,00	14,00	14,00	13,00	12,00
110	12,94	12,94	12,94	12,94	10,59

By using FA concentration of 110 ppm was effective for suppressing the growth of *Fov*, by declined intensity up to 25%, compared to the concentration of 90 ppm and 100 ppm respectively. In other words by using 110 ppm fusaric acid could increased the category criteria to resistant (Table 2). This result was also supported by Agrios (2005), which showed that respond of FA selection would affect declined of pathogenic intensity in this case by Fov.

Tabel 2. Pathogenic Intensity result from defend and level vanilla at different fusaric acid concentration treatment.

treatment	Day observation							
	5		13		21		29	
	IP (%)	resistant category	IP (%)	resistant category	IP (%)	resistant category	IP (%)	resistant category
0 ppmI	62,50	sensitive	91,67	sensitive	93,75	sensitive	100,00	sensitive
90 ppm	31,25	Moderate	33.33	Moderate	41,67	Moderate	50,00	Moderate
100 ppm	33,33	Moderate	41,67	Moderate	50,00	Moderate	50,00	Moderate
110 ppm	00,00	resistant	25,00	resistant	25,00	resistant	25,00	resistant

note: IP= Pathogenic Intensity

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REFERENCES

Agrios, G.N. 2005. *Plant Pathology.* 5th Ed. Academic Press. New York. 922 p.

Hadisutrisno, B. 2004. *Taktik dan Strategi Perlindungan Tanaman Menghadapi Gangguan Penyakit Layu Fusarium*. Simposium Nasional I, 2-3 Maret 2004 di Purwokerto.

Jayasankar, S., Z. Li, and D.J. Gray. 2000. In Vitro Selection of *Vitis vinifera* 'Chardonnay' with *Elsinoe ampelina* Culture Filtrate is Accompanied by Fungal Resistance and Enhanced Secretion of Chitinase. *Planta*. 211: 200-208.

Neelannavar, V.S. 2006. *In vitro* Propagation Studies on Vanilla (*Vanilla planifolia* Andr.). Department of Horticulture College of Agriculture, Dharwad University of Agricultural. *Thesis*. Dharwad. India.

Saravanan, T., R. Bhaskaran, and M. Muthusamy. 2004. *Pseudomonas fluorescens* Induced Enzymological Changes in Banana Roots (Cv. Rasthali) against *Fusarium* Wilt Disease. *Plant Pathology Journal* 3: 72-80.

Svabova, L., and A. Lebeda. 2005. *In vitro* Selection for Improved Plant Resistance to Toxin-Producing Pathogens. *J. Phytopathol* 153: 52-64.