PROCEEDINGS
of the International Seminar

The Council of Rector of Indonesian State University (CRISU) and The Council of University President of Thailand (CUPT)

"EXPLORING RESEARCH POTENTIALS"

Editors:
A. Muslim (Indonesia); Siti Herlinda (Indonesia); Nurly Gofar (Malaysia);
Melanie Boursnell (Australia); K.T. Tantrakarnapa (Thailand);
Judhiastuty Februhartanty (Indonesia); Misnaniarti (Indonesia);
Najmah (Indonesia); Suci Destriatania (Indonesia)

Published by Sriwijaya University
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The Council of Rector of Indonesian State University (CRISU)
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FOREWORD

Dear special guests:

Minister for National Education, Ambassadors of Thailand for Indonesia, Ambassadors of Indonesia for Thailand, all delegates from The Council of Rector of Indonesian State University (CRISU) and The Council of University President of Thailand (CUPT), Government of South Sumatra and Palembang City, and all The 6th CRISU-CUPT Conference, International Seminar and Exhibition participants

On behalf of the Sriwijaya University as Host University, I would like to extend my warmest welcome to all of the participant of The 6th CRISU-CUPT Conference, International Seminar and Exhibition, held on 20th-22nd October 2011 at Sriwijaya University Palembang with the join theme “Exploring Research Potentials”.

There will be many challenges and opportunities in higher education in the Asean Community in the next decade. This is, therefore, considerable significant will arise from the The 6th CRISU-CUPT Conference, International Seminar and Exhibition. The previous five CRISU-CUPT conferences have been significantly deepening the relationships and come up with very fruitfull discussion in various subjects of collaboration and cooperation, for example, global warming, global mobility, academic interaction and cross-fertilization. The 5th conference was held in Chiang Mai, Thailand on July 7th-9th 2010 and appointed Sriwijaya University as a host for the 6th conference.

The 6th CRISO-CUPT conference will include many agenda, with not only include the meeting of the President Forum, the Dean Forum, and the Student Forum, but also will include international Seminar and Exhibition. This conference, therefore, might come up with more fruitfull conclusion and deepest commitment among participants.

With regard to considerable conference agenda, we greatly appreciate any support and sponsorship derived from any governmental as well as private institutions for the success of the conference. Great appreciation is also handed to organizing committe of the conference for any voluntarily effort that bring to the success of the conference.

The 6th CRISU-CUPT Conference, International Seminar and Exhibition is being attended by about 600 participants. I hope you enjoy the beauty of Palembang City as one of the oldest city in Indonesia which is 1318 years old, established during the glory of the vast Sriwijaya Kingdom. The city also have variety of interesting culture and places.

Palembang, October 2011
Chairperson,

Prof. Dr. Badia Perizade, M.B.A
Rector of Sriwijaya University

# TABLE OF CONTENTS

## Foreword

### Table of Contents

### Papers of Keynote Speakers:

1. Mental Illness In Australia (Dr. Melanie Boursnell, University of Newcastle Australia)  
2. Chemical Toxicology towards humans health and EHIA (Environmental Health Impact Assessment) in Thailand (Prof. Kraitch Tantrakarnapa, Faculty of Public Health, Mahidol University, Thailand)
3. Nutrition transition in Indonesia (DR. Ir. Judhiastuti Februhartanty, M Sc, SEAMEO RECFON Indonesia, Indonesia University)
4. Cancer : Genetic And Environmental Causes And Risk Factors (Prof. Dato' Dr. M.S. Lye, University Putra Malaysia)
5. Accelerating Diversification In Food Consumption Based on Indigenous Resources as An Alternative Action To Support Food Security In Indonesia (Prof. Dr. Rindit Pamdayun, M.P. Sriwijaya University, Indonesia)

### Papers of Presenters:

**A. Food Security**

1. Diversity, Domination, and Distribution Of Rice Stem Borer Species and its Interaction with Egg Parasitoids in Various Land Typology in Jambi (Wilyus Siti Herlinda, Chandra Irsan, Yulia Pujiasutti: Agriculture Faculty of Jambi University, Faculty of Agriculture, Sriwijaya Universit)

2. Land Suitability for Elaeis Guineensis Jacq Plantation in South Sumatra, Indonesia (M. Edi Armando, M.A. Adzemi, Elisa Wildayana, M.S. Imanudin, S.J. Priatna and Giano: Faculty of Agriculture, Sriwijaya University, South Sumatra, Indonesia, Faculty of Agrotechnology and Food Science (FASIM), UMT Terengganu, Malaysia, Forestry Delineation Agency, Department of Forestry, Indonesia)

3. From Economic Valuation to Policy Making in Forest Conversion for Elaeis Guineensis Jacq Plantation (Elisa Wildayana, M. Edi Armando and M.A. Adzemi: Faculty of Agriculture, Sriwijaya University, Indonesia, Faculty of Agrotechnology and Food Science (FASIM), UMT Terengganu, Malaysia)

4. Floating Agriculture Model from Bamboo for Rice Cultivation on Swamp Land At South Sumatra (Siti Masreah Bernas, Siti Nurul A.F. and Agung Maulana: Soil Science Program Study and Low Land Management Field, Agricultural Faculty, Sriwijaya University)

5. The Responsiveness of Jambi Rice Acreage to Price and Production Costs (Edison: Faculty of Agriculture, Jambi University, Indonesia)

*Proceedings of the International Seminar, Palembang 20-22 October 2011*
6. Wage Rigidity Analysis as an Indicator of Agricultural and Non Agricultural Labor Market Distortions In Indonesia: Error Correction Model (ECM) Approach (Dewi Adriani, Andy Mulyana, Amarni Minha, Nurlina Tarmizi) Faculty of Agriculture, Sriwijaya University, Indonesia)

7. Predator Aphis gossypii on Vegetable at Low Land areas in South Sumatera (Khodijah, Haperidah Nunilahwati, Dewi Medalima: Faculty of Agriculture, Sriwijaya University, Indonesia)

8. Population and Attack of Liriomyza Sativae (Diptera: Agromyzidae) and Its Interaction with Parasitoid on Tomato Cropping in Lowland of South Sumatera (Siti Herlianda, M. Yunos Umar, Yulia Pujiatmini, and Rosdah Thalib, Chandra Irsan: Plant Pest and Disease Department, Faculty of Agriculture, Sriwijaya University)

9. Integration of Palm Fruit Plantation And Cattle; Potential System to Improve Cattle Production (Armina Fariani, Arfan Abrar and Gatot Muslim: Animal Science Department, Faculty of Agriculture, Sriwijaya University)

10. Application of Penicilliun spp. Produced in Waste Materials to Control Root Rot Diseases Caused by Sclerotium rolfsii Sacc. on Chili (A. Muslim; Sari Eka Permata; Harman Hamidson: Program Study Agroecotechnology, Faculty of Agriculture, Sriwijaya University)

11. Purification and Characterization Collagenase from Bacillus licheniformis F11.4 (Ace Bachaki, Maggy T. Sukartono, Sukarno, Dahrl Syah, Azis B.Sitanggang, Siswati Setiyahadi, and Friedhelm Meinhardt: 1Department of Fisheries Product Technology, Faculty of Agriculture Sriwijaya University, 2Faculty of Agricultural Technology Bogor Agricultural University, 3Agency for the Assessment and Application of Technology, Republic of Indonesia, 4Institute for Molecular Microbiology and Biotechnology, University of Munster Germany)

12. Biological Reproduction Menochilus Sexmaculatus (F.) Predator Chili (Aphis Gossypii Glover) From Central Vegetable At Low Land Areas In South Sumatera (Haperidah Nunilahwati, Dewi Medalima, dan Khodijah: Agriculture Faculty of Sriwijaya University, Indonesia)

13. Competitiveness and Minimum Regional Price of Arenga Palm Sugar; Case Study of Small Palm Sugar Industries in Rejang Lebong Regency, Bengkulu Province (Ketut Sukiyono, Bambang Sumantiri, Nusril And Evanila Silvia: Department of agricultural socio – economics, Faculty of Agriculture, Bengkulu University)

14. Plant Clinic: Driving Farmers Profit Partners (Chandra Irsan, Suwandi, A. Muslim, Siti Herlianda: Department of Plant Pests and Diseases, Faculty of Agriculture, Sriwijaya University)

15. The Role of Biotechnology In Overcoming the World Food Crisis (Suranto: Department of Biology, Faculty of Natural Sciences and Mathematic-UNS-Solo)

16. The Impact of Innovation Acceleration of Paddy Commodities at Irrigation Agroecosystem In Musi Rawas Regency (Yanter Hutapea and Tumarlan Thamrin: South Sumatra Assessment Institute for Agricultural Technology, Indonesia)

17. Performance of Several High Lines of Tolerant Rice to Iron Toxicity in Tidal Swamp Area in South Sumatra
   (Tumarlan Thantrin, Rudy Soehendi, Waluyo dan Syahri: South Sumatra Assessment Institute for Agricultural Technology, Indonesia)

18. Performance of Submergence Tolerant Rice in South Sumatra to Anticipate the Impact of Climate Change
   (Tumarlan Thantrin, Imelda SM, Waluyo dan Syahri: South Sumatra Assessment Institute for Agricultural Technology, Indonesia)

19. The Dynamics of Iron (Fe) Solubility As a Result of Sulphate Acid Soil Reclamation and the Way to Control
   (NP. Sri Ratmini\(^1\); dan Arifin Fahmi: South Sumatra Assessment Institute for Agricultural Technology, Indonesia)

   (NP. Sri Ratmini dan Herwenita: South Sumatra Assessment Institute for Agricultural Technology, Indonesia)

21. Study of Erosion on Different Types of Land Use in the Region Upstream Watershed Area (Das) Komering South Sumatra
   (Satria Jaya Priatna\(^1\); M. Edi Armanto\(^1\); Dinar DA. Putranto\(^1\); Edward Saleh\(^1\), Robiyanto IIS\(^1\); Niken Suhesti\(^1\) and S.N Aidil Fitril\(^1\): Faculty of Agriculture, Sriwijaya University, South Sumatra, Faculty of Engineering, Sriwijaya University, South Sumatra, Indonesia Indonesia)

B. Environmental and Climate Change

22. Study of Palm Empty Fruit Bunches Processing Technology As Saccharide Source For Friendly Environment Surfactant (Joni Karman: Assessment Institute for Agricultural Technology in South Sumatera)

23. Assessment of Pb Content of Motor Vehicle Emissions of Origin On Soil And Plant In Island Village Semambu Kui 22 Highways Indralaya – Palembang (A. Napoleon, Dwi Probowati S, Marji Putranto: Faculty of Agriculture Sriwijaya University)

24. Using The Forest Zone Through The Low Carbon Development for The Welfare of the Orround Forest Society (Using the Forest Zone through the Low Carbon Development for the Welfare of the Orround Forest Society (Najib Asmani: Agriculture Faculty and Graduate Post Program Sriwijaya University, Palembang, Indonesia)

25. Run off, Erosion, and Yield of the Sweet Corn (Zea mays var. sicicharata) as result of Sheep Manure Application and Terracing (Ruaraita Ramadhahina Kawaty: Faculty Agriculture Tridinanti University, Indonesia)

26. Stilbenes from The Heardwoodof Morus Nigra and their Cytotoxicity (Ferlinahayati\(^1\); Enis H. Hakimi\(^2\); Yana M. Syah\(^2\); Lia D. Juliawaty\(^2\); Jalifah Latip\(^1\): Department of Chemistry, Faculty of Mathematics and Natural Sciences, Sriwijaya University, Natural Product Research Group, Department of Chemistry, Institut Teknologi Bandung, School of Chemical Science & Food Technology,}

27. Responses of Several Tropical Plant Species to Polluted Air Condition in the City
(E.S. Hadi and Dian Augustinia: Department of Agroecotechnology Faculty of Agriculture Srijaya University)

28. Freshwater Fish Diversity in Pulo Kerto Musi River, Palembang-South Sumatra: A Preliminary Results
(Hilda Zulkifli, Doni Setiawan and Indra Yustian: Department of Biology, Faculty of Science, Srijaya University)

29. Vegetational Structure and Composition in Pulo Kerto Island, Musi River-Palembang, South Sumatra
(Indra Yustian dan Hilda Zulkifli: Department of Biology, Faculty of Science, Srijaya University)

30. Climate Change, Environment and Plant Diseases Development
(Nurhayat i: Department of Plant Pest and Disease, Agriculture Faculty, Srijaya University)

31. Biophysical Characteristics of Tailings Deposition Area and Its Contribution to Vegetation Growth
(Yuunita Windusari1, Robiyanto Hendro Susanto2, Zulkifli Dahlan, Wisnu Susetyo3, And Indra Yustian1: Doctoral student of Environmental Science and Lecture of Mathematic and Sciences Faculty of Srijaya University, 2 Lecture of Environmental Sciences Programme, Srijaya University and Supervisor commission, 3Senior Advisor PT Freeport Indonesia and Supervisor commission)

32. Biodegradation of Petroleum Hydrocarbon by Single and Consortium of Hydrocarbonoclastic Bacteria From Petroleum Polluted Mangrove Areas
(Hary Widjajanti1, Iswandi Anas2, Nuni Gofar2, Moh Rasyid Ridho: Agricultural Science of the Graduate Program of Srijaya University)

C. Energy, Education and Others

33. Temperature and Relative Humidity Gains of "Teko Bersayap" Model Solar Dryer
(a Research Note)
(Yuwan, Bosman Sidebang and Evanila Silvia: Department of Agricultural Technology, Faculty of Agriculture, University of Bengkulu)

34. Proposes of Implementation of Sustainable Subgrade on Highway Construction in South Sumatera By Using Coal Combustion Products (CCPs) as Stabilizer
(Achmad Fauzi1, Usama Juniansyah Fauzi2, Wan Mohd Nazmi2: 1, 2 The Faculty of Civil Engineering and Earth Resources, University of Malaysia Pahang, Malaysia, 2Faculty of Civil and Environmental Engineering, Institut Teknologi Bandung, Indonesia)

35. Green Pavement by Using High Density Polyethylene Modified Asphalt as Aggregate Replacement by, Faculty of Civil Engineering and Earth Resources, University Malaysia Pahang
(Wan Mohd Nazmi and Wan Abdul Rahman Wan Rohaya Wan Idris, and Achmad Fauzi Abdul Wahab: Faculty of Civil Engineering and Earth Resources, Universiti Malaysia Pahang, Lebuhraya Tun Razak, Gambang, Kuantan, Pahang, Malaysia)

36. Social Benefit of Coal Mining Activity (Syahidin Zakir and Restu Juniah)
   1Dept. Public Administration Faculty Social and Political Sciences, Sriwijaya University, 2Environmental Science Program University of Indonesia

37. Behavior of Connection Rotations Composite Steel Beam with Partial Strength Using Trapezoid Web Profiled
   (Anis Siggaff, Mahmood Md. Tahir, and Arizo Sulaiman: Civil Engineering Department, Faculty of Engineering, Sriwijaya University, 2Steel Technology Centre, Faculty of Civil Engineering, University Teknologi Malaysia, 3Faculty Of Civil Engineering, Universiti Teknologi Malaysia)

38. Chemical Compound from Endophytic Fungi of Medicinal Plant Used in Treatment Of Gout (Elfita, Munarni, Munawar: Faculty of Mathematics and Natural Sciences, Sriwijaya University)

39. 3-OXO Friedelin Compound from the Stem Bark of Manggu Leuweung (Garcinia cornea)
   (Munarni, Elfita, Handi: Department of Chemistry, Faculty of Mathematics and Natural Science, Sriwijaya University, Indralaya, South Sumatera, Indonesia)

40. Antioxidant Flavonoids from Tunjak Langit (Helminthostacys Zypalonica)
    (Fitria, Munarni dan Eliza: Department of Chemistry, University of Sriwijaya)

41. The Industry Characteristic and Managers View: their Influence On Employment Relations In The Indonesian Hospitality Industry (Explorations From Three Case Studies)
    (Hendragunawan S., Thayf, John Lever: Hasanuddin University, Indonesia)

42. Competitiveness of Management State-Owned Enterprises (Soes) Telecommunications
    (Kesi Widjajanti: Faculty of Economic Semarang University, Semarang, Indonesia)

43. Prospects and Challenges of The Introduction of Open Educational Resources in Indonesia (Daryono, Udang Kusmawan, Olivia Idrus)

44. Research Collaboration on Quality Assurance for Open and Distance Learning in Asia
    (Endang Nugraheni, Aminudin Zuhairi: Universitas Terbuka, Indonesia)

45. Fast Ship Serving Makassar, South Sulawesi to Majene, West Sulawesi
    (Muhammad Alham Djabbar and Andi Haris Muhammad: Ocean Engineering Study program, Department of Naval Architecture, Faculty of Engineering, Hasanuddin University, Makassar, Indonesia)

D. Public Health and Medical Science

46. Pesticides Exposure and Liver Dysfunction on Childbearing-Age Women in Kersana Sub District, Brebes Regency
    (Arum Siwiendrayanti, Public Health Department, Sport Science Faculty, Semarang State University)

47. Factors Related to The Occurrence of Low Back Pain Complaints On Employee Section of Corporate Customer Care Center (C4), PT Telekomunikasi Indonesia. (Yuli Amran, M. Fadri Hansenz, Juniari Tri Syafitri, State Islamic University Syarif Hidayatullah Jakarta)  

48. Relation of Work Risk Factors with Musculoskeletal Disorders (MSDs) Complaints of Gold Miner Workers In Subdistrict Cilegret-Banten on 2010 (Yuli Amran, Ratihana Nadra Alkaff, Endang Bukhori, State Islamic University Syarif Hidayatullah Jakarta)  

49. Effect of Rehydration Solutions on Fatigue Among Women Workers (Mardiana, Public Health Department, Sport Science Faculty, Semarang State University)  

50. The Association between Risk Factors, RULA Score, and Musculoskeletal Symptom among Workers in a Printing Manufacturing Company, Malaysia (MC Foong, 1Mohd Yusof, 1B Mohd Raife, and 1AA Ahmad  

1Department of Community Health, Faculty Medicine and Health Sciences, University Putra Malaysia., )  

51. Productive Work Time Lost Because Of Employee Smoking Behaviour in Wood Industry in Jepara District Central Java (Nurjanah1, Zahroh Shaluhiyah2, Bagoes Widjanarko2 : 1Master Student of Health Promotion Program of Diponegoro University, lecturer of Health Faculty of Dian Nuswantoro University, Semarang. 2Lecturer of Health Promotion Program of Diponegoro University, Semarang)  

52. Water Quality and Water Borne Disease at The Lowland Ecosystem in Banyuasin (Dianita Ekawati7, Tan Malaka2, Robiyanto2, M.T. Kamaluddin3, Dwi Setiaawan4, Amar Muntaha1  

1Department of Public Health, STIK Bina Husada, Palembang 30131, Indonesia  
2Medicine Faculty of Sriwijaya University  
3Agriculture Faculty of Sriwijaya University  
4Veterinary Medicine Faculty of Sriwijaya University)  

53. Measuring Escherichia Coli in Foods And Beverages Towards Certification of Cafeteria In Campus (Dewi Susanna1, Yvonne M. Indrawan1, Zakianis1, Tris Eryando1, Lassie Fitria1, Kartika A Dimarsie1, Aria Kusuma1,  

1Faculty of Public Health, Indonesia University, 2Doctoral Student of Public Health Science, Indonesia University, Indonesia)  

54. Pesticide, Adverse, and Safe Handling to Woman of Child Bearing Age (WCA) in Agriculture Area (Imelda Gemaal Puha : Faculty of Public Health, Sriwijaya University, Indonesia)  

55. Comparative Analysis of Occupational Safety and Health Risk Management Program at University of Indonesia and National University of Singapore (Anita Camelia, Faculty of Public Health, University of Sriwijaya, Indonesia)  

56. Analysis of Levels of lead (Pb) in semen and sperm motility at the Laboratory of Medical Biology Faculty of Medicine, University of Sriwijaya Palembang (Nani Sari Murni1, Tan Malaka2, dan M. Zulkarnain3 : STIK Bina Husada,  

2Faculty Medicine Of Sriwijaya University)
57. The Correlation of the Use of PPE (Personal Protective Equipment With Respiratory Disorders of Wood Furniture Workers In Kecamatan Indralaya and Kecamatan Indralaya Utara 2011 (Herliahwati, Christine Sihaloho : Nursing Science Study Program, Faculty of Medicine, Sriwijaya University, Indonesia)

58. Value of Children as Determinants Parenting Nutrition on The Environment Vulnerable Sociocultural Nutrition (Village Pecuk, District Mijen, Demak Regency, Central Java) (Oktia Woro Kasmini H. Department of Public Health Sciences FIK UNNES Semarang)

59. Analysis of Rhodamine B in Cookie of Traditional Food Type (Study at Pasar Tjungg Jember Regency) (Khoiron, Astri Rizky Vitantina, Ratayu Sri Pujialti, Department of Environmental Health and Occupational Health & Safety Faculty of Public Health, University of Jember)

60. Determinant Factor of Anemia Status Among Vegetarian Female Adolescent In Badung District of Bali Province (Putu Widarini, School of Public Health Udayana University)

61. Diet, nutrition and the prevention of cervical cancer (Ciptaningtyas, R, State Islamic University Syurif Hidayatullah Jakarta)

62. The Correlation Between Macro Nutrient Consumption and Physical Activities With Overweight Among Children In Elementary School (Study at Al-Furqan Elementary School, Jember Regency) (Leersia Yusi Ratnowati, Sulistiyan, Dwinda Prianto, Public Health Faculty, Jember University)

63. Correlation of family participant with nutrition status of children under five years old in peguyangan village work area paskesmas iii of north denpasar (Ni Ketut Sutiari, Ni Luh Sudiasil, I Gusti Agung Ayu Mahayuningsih, School of Public Health, Faculty of Medicine, Udayana University)

64. Does Birthweight Related With Chronic Diseases In Adult Life? (Sucr Destriatania : Faculty of Public Health, University of Sriwijaya, Indonesia)

65. Experience Breastfeeding Mother On Teens At Work Area Health Center Payaranan Year 2011 (Bina Melvia Girsang, Faculty of Medicine, Nursing Science Study Program UNSRI)

66. The Effect of Maternal Nutrition Anemia towards Low Birth Weight (Rini Mutahar, Misnaniarti, Fatmalina Febry : Faculty of Public Health, Sriwijaya University, Indonesia)

67. Relationship Unhealthy Snack Habits with Diarrhea Incidence In Elementary School Children (Fatmalina Febry, Najmah, Indah Purnama Sari : Faculty of Public Health, Sriwijaya University, Indonesia)

68. Relationship Between Age and Lifestyle with Prevalence Hypertension in Poly
    Medicine Moehammad Hossin Hospital Palembang of the Year 2011
    (Nilson Sitornus, Desti Wuliatusti, Health Polytechnic of Palembang Nursing
    Program)

69. Determinants Pulmonary Tuberculosis Incident in District Banyumas Multilevel
    Modelling Approach
    (Rismata Kesuma, Kamaruddin, Ngudiantoro, Ibrahim Eddy, Tjej Yan
    Suryadi, Departement of Public Health, STIKES Darul Ma'arif Al Hossen Baturaja,
    Indonesia)

70. Enabling Factors of Doing Pap Smear Test among Women at Age ≥ 35 Years in
    Denpasar Who Diagnosed Cervical Cancer At Sanglah Hospital 2011
    (Ni Luh Putu Suariyani, Regina Chrystanie Weling : School of Public Health,
    Faculty of Medicine, Udayana University)

71. The Difference of Urinary Excretion Iodine (UEI) Increase between Primary School
    Children With and Without Ascariasis After Administration of Oral Iodized Capsule
    (Galuh Nita Prameswari, Public Health Department, Sport Science Faculty,
    Semarang State University)

72. Analysis of Determinants of Tuberculosis In The Workers at PT. Perkebunan
    Nusantara XII (Persero) of Jember Regency
    (Anita Devi Prahastuti Sujoso, Ria Nuri Estu Karisma, Irna Prasetyowati,
    Department of Environmental Health and Occupational Safety Health, Faculty of
    Public Health, University of Jember)

73. Risk Factors of Lymphoma at dr. Soebandi Hospital of Jember District- East
    (Ni'mal Baroya, Pudjo Wahjudi, Annisa Reyopause, Public Health Faculty,
    Jember University, Jember)

74. Hip Structure Associated with Hip Fracture in Women: Data From the Geelong
    Osteoporosis Study (GOS) Data Analysis- Geelong, Australia
    (Margaret Henry¹, Najmah², L. Gurrin², J.Pasco³
    ¹ Department of Clinical and Biomedical Sciences, The University of Melbourne,
    Australia. ² Faculty of Public Health, Sriwijaya University, Kampus Unsri
    Indralaya, Ogan Ilir, Sumatera Selatan, Indonesia. ³ School of Population Health,
    The University of Melbourne, Australia)

75. The Study of Diabetes Mellitus Risk Factors in Bangka Belitung
    (Titi Sari Renovati, Anisyah, Amar Muntaha, Dianita Elawati, Vera Susanti,
    Environmental Health Laboratory Agency and Disease Control,
    Palembang, Indonesia)

76. Association of Knowledge, Perception, and Source of Information about Hiv Aids
    With Attitudes From Indonesian People To People Living With Hiv Aids (PLHA)
    (Analysis Of SDKI 2007), Indonesia, 2010
    (Yeni, Najmah, Rini Mutahar : Faculty of Public Health, Sriwijaya University,
    Indonesia)

77. Identification of Covert Patients With Filariasis and Epidemiologic Study of
    Filariasis in Sub-District of Tangkano, Muna Regency, Province of Southeastern
    Sulawesi in 2009
    (Ramadhan Toepu, Devi Sivittri Efendi : Public Health Department of
78. Characteristics among Injecting Drug Users: Accessing and Not Accessing Needle and Syringe Program in Palembang, South Sumatera (Najmah Faculty of Public Health, Sriwijaya University, Indonesia) 599

79. The Use of Salivary A-Amylase And Stress-Related Symptoms Questionnaires as Indicator For Psychological Distress Among Breast Cancer Survivors (Yong, H.W., Zubaidah, J.O., Saidi, M., Zalikah, M.S., Yong, H.Y. and Zailina. H: Universiti Putra Malaysia, Selangor, Malaysia) 605

80. Self-Concept in Sexual Behavior of Campus Chicken’s (Ayam Kampus) In Senarang (Eti Rimawati. Health Faculty Universitas Dian Nuswantoro) 619

81. The Sexual Relation Scripts of Premarital Sexual Intercourse among University Students In Bandar Lampung (Roro Rukmini Windi Perdani: Faculty of Medicine, University of Lampung, Lampung Province, Indonesia) 626

82. Development of Posyandu Information System for Supporting Surveillance of Maternal and Child Health (Case Study at Manisrejo Urban Village Taman District in Madura City, East Java Province) (Afu Khoiri, Public Health Faculty, University of Jember) 635

83. Health Financing Reform as a Result of Decentralization Policy in Bali (Putu Ayu Indrayati, Pande Putu Jamara, School of Public Health Medicine Faculty of Udayana University) 641

84. The Relationship between Marketing Mix and University Student Interest in Choosing Public Health Science Study Program Faculty of Medicine Andalas University 2011 (Isniati, Syahril, Vonicha Regia, Faculty of Medicine, Andalas University) 647

85. Healthy Behavior-Based Development Model to a Free Larvae Aedes Aegypti by Environmental Health Education In The Eastern District Padang (Nizwardi Azkha, Rizanda Machmud: Faculty of Medicine, Universitas Andalas, Padang, Indonesia) 658

86. Health Care Seeking Behaviour of Community and Tb Patients, And Capability of Nonformal Health Services Provider in Tjnung Bintang Subdistrict, Indonesia (Nurul Ismay, Agus Setyo Widodo, Darman Zayadan, Ferzal Masra, Haris Kadarusman, Bachi Alijahbana) 1Faculty of Medicine Lampung University, 2Health Office Lampung Province, 3Health Institute Umitra Lampung, 4Faculty of Medicine Padjadjaran University) 670

87. Influence of Life Skills on Sexual Behavior in Adolescent at Seberang Ulu Area of Palembang (Iche Andriyani Liberty, Nur Alam Fajar, Elvi Sunarsih: Faculty of Public Health, Sriwijaya University, Indonesia) 677


89. The Development Study of ‘Desa Siaga’ In Ogan Ilir District (Misnaniarti, Asmaripta Ainy, Nur Alimi Fajar : Faculty of Public Health, Sriwijaya University, Indonesia)

90. Injection Drug Users (IDU) Behavior Toward Methadone Maintenance Therapy Program At Erunaldi Bahar Hospital 2010 (Tri Novia Kumasasari Faculty of Public Health, Sriwijaya University, Indonesia)

91. The Experience of Parents Who Have Temper Tantrums Toddler (Arie Kusumaningrum, Chodijah Abdul Qudus, Elia Yulia Fitri : School of Nursing Science, Faculty of Medicine, Sriwijaya University, Indonesia)

92. Factors that Influence the Behavior of Male Adolescence Smokers at Junior High School Kramat Jakarta (Cicilia Nony, Budi Sulistyowati, Wuryastuti : School of Health Science, Sint Carolus)

93. Stratification of Public Health Services For Elderly at Urban and Rural Areas in Indonesia (Ari Istiani, Rusliant and Sachriani : Home Economics Department, Jakarta State University, Indonesia)

Summary Seminar

Name and Address of Presenter International Seminar

APPLICATION OF *Penicillium* spp. PRODUCED IN WASTE MATERIALS TO CONTROL NECK ROOT ROT DISEASES CAUSED BY *Sclerotium rolfsii* Sacc. ON CHILI

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ABSTRACT

The research was conducted to know the ability of *Penicillium* spp. grown in various substrates to suppress neck root rot disease caused by *Sclerotium rolfsii* on Chili. The research was arranged in Randomized Completely Design with 11 treatments and 3 replications as consisted a control, *Penicillium* spp. isolates P8 and P10 produced in combination of substrates of tapico dregs+bunch of palm oil; coconut dregs+bunch of palm oil; wheat dregs+bunch of sawdust; coconut dregs+bunch of sawdust; and Yeast extract+sucrose+agquades. The result showed that application of *Penicillium* spp. effectively reduced the neck root rot disease caused by *S. rolfsii* on chili. Seedlings treated with *Penicillium* spp. grown by various substrates significantly (0.05%) reduced disease severity ranged from 61.01-94.91%. Based on the results, *Penicillium* spp has potential as biocontrol agent against *Sclerotium rolfsii* on Chili.

Key words: Biocontrol, *Penicillium* sp.; *Sclerotium rolfsii*; Chili

INTRODUCTION

Neck root rot diseases caused by *Sclerotium rolfsii* Sacc is one of the most destructive and economically damaging diseases of chili. Species of *S. rolfsii* as soil-borne pathogen cause a variety of diseases on many different type of plant such as paddy (Purwanti et al., 1997), green bean alfalfa, peanut, and bean (Caresini, 1999); papaya and corn (Uchida, 2007). Prayoga (2007) reported that, the percentage of diseases incidence caused by *S. Rolfsii* on Chili in Pemulutan Sub-District, District of Ogan Ilir was 2.6%. Other survey conducted by Akbar (2007) reported that the percentage of diseases incidence caused by *S. Rolfsii* on Chili in Pangkalan Balai Sub-District, District of Banyuasin ranged from 28% - 80%.

Soil microorganisms are ideal for use as biocontrol agents against soil-borne disease. Previous research demonstrated that *Penicillium oxalicum* spp as soil inhabitants effectively reduced Fusarium wilt of tomato caused by *Fusarium oxysporum* f.sp. *lycopersici* through induced resistance (De Cal A., et al., 1995; De Cal, A., et al., 1997; De Cal, A., et al., 2000). Koike et al. (1997) reported that *Penicillium* spp beside could reduced antracnose disease caused by *Colletotrichum orbiculare* and bacterial leaf blight caused by *Pseudomonas syringae pv. Lachrymans* on cucumber, it also could increase plant growth.

The objective of this research was to evaluate *Penicillium* spp produced in waste materials for control of neck root rot disease caused by *Sclerotium rolfsii* on chili.


70
MATERIALS AND METHODS

Fungi

Biocontrol agent used in this study was *Penicillium* spp. (isolates P8 and P10) as the Plant Growth Promoting Fungi (PGPF) isolated from rhizosphere of chili plant cultivated in low land area. *Sclerotium rolfsii* Sacc was obtained from an infected chili plant was used as the pathogen.

Plant

All chili seeds were surface-sterilized with 1% hydrochloric acid for 15 min and rinsed three times in sterile distilled water before sowing.

Inoculum Preparation

For inoculums of *Penicillium* spp isolates: For solid inoculum, each isolates of *Penicillium* spp (isolate P8 and P10) was cultured on potato dextrose agar (PDA) for 3 days at 25°C in the dark. Five mycelial discs (5 mm) of the isolates cut from the edges of three-day old cultures were added to 100 g moist autoclaved combinations of various substrates (1:1, dry various substrates/distilled water, w/v) contained in a 500 ml Erlenmeyer flask e.i: 1). tapioca dregs+bran+bunch of palm oil (TBP); 2). coconut dregs+bran+bunch of palm oil (CBP); 3). tapioca dregs+bran+sawdust (TBS); 4). coconut dregs+bran+sawdust (CBS); 5). Yeast extract+sucrose+aquadest (YSA). The cultures were incubated in the dark for 10 days at 25°C and shaken regularly to aid even colonization. The infested media substrates were air-dried for 7 days and stored at 4°C until used. While for liquid media, each isolates of *Penicillium* spp (isolate P8 and P10) was cultured on potato dextrose agar (PDA) for 3 days at 25°C in the dark. Two-Three mycelial disks (5 mm) of the isolates cut from the edges of three-day old cultures were added to liquid media contain 15 g yeast extract and 20 g sukraca per liter distilled water. The cultures were incubated in the dark for 5 days at 25°C at stasis condition. The conidia were harvested by filter the culture and then used for this study.

For inoculums of pathogen, *Sclerotium rolfsii*. The procedures was prepared similar to solid inoculums of *Penicillium* spp. described above, except the media substrates used for pathogen was bran+corn+rice-straw with comparison 4:3:1 for each material, respectively.

Assay of *Penicillium* spp produced by various substrates for control

The inoculums of *Penicillium* spp. were pulverized in a blender for about 30 sec. (1 to 2 mm particle size) and mixed (1.5%, w/w) with sterilized potting medium (soil+kompos). The liquid inoculums were applied to potting medium in the concentration 10^5 conidia/g potting medium. Small polybags were filled with approximately 20 g with potting medium amended with inoculums *Penicillium* spp. One surface-sterilized chili seed was sown in each small polybag. The seedlings were allowed to grow for 21 days. The treated seedlings with *Penicillium* spp. were transferred to polibag (20x15 cm) which filled with potting soil. The inoculums of pathogen was then inoculated in soil surround the seedlings (1 g pathogen inoculums per seedling). The seedlings were kept in greenhouse to allow their grow for the next 14 days. The seedlings not treated with *Penicillium* spp and challenged with *S. rolfsii* were set up as control. Treatments were replicated 3 times and each replicate consists of 5 plants.

Disease severity based on the foliar symptom was assessed using a scale of 0 to 4; 0 = healthy; 1 = 0-25% yellowing; 2 = 25-50% yellowing; 3 = 50-75% yellowing; 4 = >75% yellowing.

or dead plant. The percentage of disease severity in each replication within the treatment was calculated using the formula:

\[
K = \frac{\sum(nxy)}{ZxN} \times 100\%
\]

Where:
- \(K\) = Disease Severity (%)
- \(n\) = number of seedlings infected by pathogen in each scale
- \(v\) = Diseases scale (0-4)
- \(Z\) = The highest of disease scale
- \(N\) = total seedlings

Data analysis

The experiments were carried out in randomized completely design. Treatments means obtained for percentage disease severity were compared using honest significant difference (HSD) at \(P = 0.05\) and \(P = 0.01\).

RESULTS AND DISCUSSION

On the whole, all application of *Penicillium* spp produced in waste materials significantly reduced disease severity compared to control. Reduction of disease severity by *Penicillium* spp isolates, however, differed depending on *Penicillium* isolates and kinds of waste materials used for inoculums production. However, Statistically, there are no significantly different among treatments with *Penicillium* spp (Table 1). Seedlings treated with *Penicillium* spp provided the reduction of disease severity was ranged from 61%-94.91%. The highest reduction against disease severity was provided in the treatment TBP P10 (94.91%), followed by TBS P8 and CBP P8 (88.13%), while the lowest was performed by YSA P10 (61.01%) and YSA P8 (69.49%).

Table 1. The effect of treatment with *Penicillium* spp. produced by various substrates against disease severity of neck root rot of chili caused by *Sclerotium rolfsii* Sacc a)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Disease severity (%)</th>
<th>HSD (0.05)</th>
<th>Reduction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>100</td>
<td>a)</td>
<td></td>
</tr>
<tr>
<td>YSA P10</td>
<td>38.34</td>
<td>b</td>
<td>61.01</td>
</tr>
<tr>
<td>YSA P8</td>
<td>30.00</td>
<td>b</td>
<td>69.49</td>
</tr>
<tr>
<td>CBP P10</td>
<td>28.34</td>
<td>b</td>
<td>71.18</td>
</tr>
<tr>
<td>TBS P10</td>
<td>20.00</td>
<td>b</td>
<td>79.66</td>
</tr>
<tr>
<td>CBS P10</td>
<td>18.34</td>
<td>b</td>
<td>81.35</td>
</tr>
<tr>
<td>CBS P8</td>
<td>16.67</td>
<td>b</td>
<td>83.04</td>
</tr>
<tr>
<td>TBP P8</td>
<td>13.34</td>
<td>b</td>
<td>86.43</td>
</tr>
<tr>
<td>TBS P8</td>
<td>11.67</td>
<td>b</td>
<td>88.13</td>
</tr>
<tr>
<td>CBP P8</td>
<td>11.67</td>
<td>b</td>
<td>88.13</td>
</tr>
<tr>
<td>TBP P10</td>
<td>5.00</td>
<td>b</td>
<td>94.91</td>
</tr>
</tbody>
</table>

a) Data were taken 8 days after inoculation of pathogen

b) Mean of 3 replication with 5 plants per replication. Values followed by the same letter in each column do not differ significantly (\(P = 0.05\)) according to Honest significant different test. Data were analyzed after transformation to arc sin √x.
In these study, all treatments using *Penicillium* produced in waste materials were effective in reducing disease of neck root rot disease on chili caused by *S. rolfsii* Sacc. under greenhouse condition (Table 1). This study support previous result conducted by some researchers who demonstrated that that *Penicillium* spp effectively reduced Fusarium wilt of tomato caused by *Fusarium oxysporum* f.sp. *lycopersici* (De Cal, A. et al. 1995); bacterial angular leaf spot caused *Pseudomonas syringae* pv. *tachyrhynca* and Fusarium wilt caused by *Fusarium oxysporum* f.sp. *cucumerinum* on cucumber through induced systemic resistance by the increasing lignin accumulation, sapsopride generation and chemiluminescence activity (Kolk et al. 2001). The biocontrol ability of *Penicillium* spp against neck root rot of chili obtained in this study holds a great possibility for their use as protective agents against Sclerotium diseases.

Its application as a waste materials (tapioca dregs; coconut dregs; bran; bunch of palm oil; sawdust) medium preparation that serves as a food base probably contributed to their successful establishment. It suggested that when the antagonists were introduced into small pot for preparing chili seedlings, it became establish in the rhizosphere and root area before transplanting in pathogen-infested soil. This ability might trigger host defense reaction, which was then transferred to the whole root or might be stem and leaf against neck root rot of chili De Cal et al. (1997) reported that, tomato plants treated with *Penicillium oxalicum* reduced disease severity of fusarium wilt of tomato when the antagonist and pathogen were inoculated in different points of tomato roots. Biles and Martyn (1989) observed that, prior inoculation of watermelon root with avirulent *Fusarium oxysporum* f.sp. *nivium* induced resistance in both local and systemic, in that induced watermelon plants were protected from both fusarium wilt and anthracnose. Muslim et al. (2003a,b,c) reported that prior treatment of seedlings with *Hypovirulent Bimucolate Rhizoctonia* (HBNR) in paper pot during seedling stage before transplanting into bigger pot contained pathogen-infested soil, effectively reduced Fusarium diseases of tomato and spinach. The mechanisms of biological control of the Fusarium diseases using HBNR might be related to competition for colonization site or nutrient and induced resistance.

The effectiveness of *Penicillium* spp produced in waste materials against neck root rot of chili were also might be related to the contain of the waste materials used as medium were plenty of nutrient which increase its growth. Pareira (2008) reported that tapioca dregs and bran contain protein and carbohydrate,. Furthermore, Wahyono (2007) reported that bunch of palm oil contain nutrient such as nitrogen (0.4 %), P₂O₅ (0.029 – 0.05 %), and K₂O (0.15 – 0.2 %). Widastoety (2008) reported that coconut dregs contain essensial nutrients such as K, P, Ca, Mg and N. It also contain organic material, ash, Fecin, hexoselulosa, selulosa, pentosa and lignin.

CONCLUSION

The conclusion of this study is seedlings treated with *Penicillium* spp produced in waste materials based on tapioca dregs; coconut dregs; bran; bunch of palm oil; and sawdust, effectively reduced disease severity of neck root rot of chili caused by *Sclerotium rolfsii* Sacc. ranged from 61%-94.91%.

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Proceedings of the International Seminar, Palembang 20-22 October 2011 73


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