

Proceedings

International Seminar on Agro-tourism Development (ISAD 2011)

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Contents

Table of Contents

	Committees Preface	
	Sponsors	
	Keynote Speaker	4
	Agro-tourism Development Policy in Yogyakarta Special Region, Indonesia. Sultan of Yogyakarta.	1
	Plenary Speakers	
1	Policy and Program of Agro-tourism Development in Indonesia. Syukur Iwantoro. Head of Agro-tourism Commissions, Ministry of Agriculture Indonesia.	2
2	The Role of Education in Developing Best Service Management in Agro-tourism Ahmad Shuib. Institute of Agricultural and Food Policy Studies, Putra Infoport, Universiti Putra Malaysia.	3
3	Development and Problem of Green Tourism in Rural Japan: A Possible Model for Green (Agro) in Bantul Regency, Yogyakarta Special Region. Haruo Kuroyanagi. Jogakuen Sugiyama University, Japan.	12
4	Developing Agro-tourism Region Based on Local Resources for Sustainable Development (A Study in Bantul Regency, Yogyakarta Special Region Province). Siti Syamsiar. UPN "Veteran" Yogyakarta.	18
5	Harnessing Agri-Tourism Opportunities through Public and Private Partnerships. Jesusa D. Ortuoste. Sultan Kudarat State University Philippines.	25
6	Business Strategy for Developing Sustainable Agro-tourism. Marc Vanacht. AG Business Consultants, USA.	27
7	Agro-tourism Development for Community Empowerment. M. Reza. Taman Buah Mekarsari, Indonesia.	32
8	Ecotourism Program in Oil Palm Plantation. Barakala, SSi., MEEM. PT. Bisma Dharma Kencana, Indonesia	37
	Category : Oral Presentation	
	Services	
9	Visitors Satisfaction Analysis on Salak Pondoh Agrotourism in Turi, Sleman Yogyakarta. Budiarto.	43
	Socio-culture	
10	Access to Land and Changes in Agrarian Structure. Mustapit.	54

11	Sinamay Women Weavers in Argao and Carmen, Central Philippines. Wilma C. Giango, Rebecca dC. Manalastas, Adora A. Villaganas, Cynthia Z. Sy, Carmelina C. Aguirre, Elvisa O. Basubas, Arwin Z. Visitacion.	65
12	The Women Trisikad Drivers of Cebu City, Central Philippines: Their Fantasies and Aspirations. Rebecca dC. Manalastas, Ponciano C. Bontia, Adora A. Villaganas, Wilma C. Giango, Cynthia Z. Sy, Carmelina C. Aguirre, Elvisa O. Basubas.	68
13	The Role of Natural Resources and Socio-Cultural Aspects as The Basic Components in The Tourism Village Development in Administrative Territory of Yogyakarta (DIY) Province. Budi Widayanto, Heni Handri Utami.	72
14	Community Development Based on Tourism Village: An Exploratory Review of Alternative Development Approaches. Eko Murdiyanto.	82
	Economics and business	
15	The Flourishing Small-Scale Entrepreneurial Activities in Station 3, Barangay Manok-Manok, Boracay, Aklan, Western Philippines. Marde T. Ponce, Rodolfo B. Burgos, Rebecca Dc. Manalastas, Adoracion A. Lawas, Adora A. Villaganas, Wilma C. Giango, Cynthia Z. Sy, Carmelita C. Aguirre, Arwin Z. Visitacion.	90
16	The Role of Women in Small-Scale Informal Trade Centers of Handicraft Industry in Cebu, Philippines. Adora A. Villaganas, Victor D. Villaganas, Rebecca dC. Manalastas, Wilma C. Giango, Cynthia Z. Sy, Carmelina C. Aguirre, Elvisa O. Basubas.	94
17	Competitiveness Acceleration Strategy on Agribusiness "Three-Five Model".	98
18	Strategic Analysis on Development of Value Chain for Ecotourism Village Gamplong Sleman Yogyakarta. Dwi Aulia Puspitaningrum.	105
19	Assessment of Women in Secondhand Retail (Ukay-Ukay) Trade: in Cebu, Central Philippines. Ponciano C. Bontia, Rebecca dC. Manalastas, Adora A. Villaganas, Wilma C. Giango, Cynthia Z. Sy, Carmelina C. Aguirre, Elvisa O. Basubas, Arwin Z. Visitacion.	111
20	An Exploratory Analysis of Economic Viability of Maize Farming in the Province of East Nusa Tenggara. Yusuf, Masniah, and Jangkung Handoyo Mulyo.	115
21	Impact of Agropolitan Development on Socio Economics of Farmers in Sleman Agro-tourism District, Yogyakarta. Antik Suprihanti.	128
22	The Assessment of Supply Chain Management on Tangerine cv Pontianak in West Kalimantan, Indonesia. Supriyanto, A, A. Musyafak, L. Zamzami.	141
23	The Flourishing Bakasi (<i>gymnothoraxrichardsoni</i>) Livelihood in Cordova, Cebu, Philippines. Cynthia Z. Sy, Wilma C. Giango, Rebecca Dc. Manalastas, Adora A. Villaganas, Carmelina C. Aguirre, Elvisa O. Basubas, Arwin Z. Visitacion, Marde T. Ponce, Nerissa Bontia.	153
24	Feasibility Study of Dragon Fruit-Based Agrotourism Development in Yogyakarta Province. Agus Santosa.	157

25	The Influence of Tea Plucking Technology Implementation to The Product, Job Opportunity for Woman and Study of Agro-tourism at PT. Perkebunan Nusantara VII (Persero) Pagar Alam Business Unit, South Sumatera Province. Siti Hamidah, Juarini.	166
	a selfa fa pere una di conce di concentratione	
	Regional development	
26	Agro Tourism and Rural Development in Sri Lanka (With Special Reference to Nuwara Eliya District). Wijitapure Wimalaratana, S.M.P Senanayke.	174
27	Agro-tourism Development in Timor Tengah Selatan: Limitations and Advantages. Dina Viktoria Sinlae.	189
28	Opportunities of Agro-tourism Development in Nepal. Dhruba Raj Bhattarai, Shanta Pokhrel.	201
29	Agro-tourism Planning in Kedungkayang, Wonolelo, Sawangan, Magelang. Lis Noer Aini, Bambang Heri Isnawan, Arif Suwatno.	208
30	Development of Agro-tourism at The Orchard Mangunan at Bantul Yogyakarta Special Region Province. M. Nurcholis.	216
31	Agro-tourism Rehabilitation by using Local Resources as an Effort of the Community Welfare Improvement at the Southern Slope of Mount Merapi, Yogyakarta, Indonesia. Alif Waluyo, Sari Virgawati.	227
32	Community-Based Agri-ecotourism in Kulon Progo and Bantul Regency. Teguh Kismantoroadji.	234
33	Potentials of the Tourist Village of Karangtengah Imogiri Bantul. Indah Widowati.	241
	Land management	
34	Water Utilization Efficiency Against the Growth and Yield of Rice (<i>Oryza sativa</i>) in Watershed Yeh Ho Tabanan-Bali. IGK. Dana Arsana, Djoko Prajitno, Abdul Syukur, Heru Hendrayana.	248
35	Substitution of Ammonium Sulphate Fertilizer to Increase The Productivity of Sugarcane Cultivation on Low-Land and Up-Land for Supporting The Development of Agro-tourism Economically. Nurhidayati, Abdul Basit Sunawan.	258
36	Agro-tourism Base of Good Agriculture Practices. Endah Budi Irawati.	270
37	Development of Agriculture and Tourism on The Land of Sand, Opportunities and Challenges. Muryanti.	278
38	Influence of Aggregate Stability on Infiltration. Subroto Padmosudarso.	285
39	Soil Characteristic That is Overgrown with Mangroves Api-Api (<i>Avicennia sp</i>) and Nipah (<i>Nypa fruticans</i>) in Estuary Area, Cijulang, Ciamis, West Java. Dyah Arbiwati, Ajeng Angguni P, Partoyo.	295
40	Determination of Classification Land Suitability for Tourism Areas in Jonggol, Bogor District. S. Setyo Wardoyo.	301
41	The Essential of Precision Agriculture to Minimize The Environmental Impact of Agro-tourism Development. Sari Virgawati, Sri Sumarsih, Herwin Lukito.	306

The Decrease of Water Quality in Lake Rawa Pening The City of Semarang. Eko Amiadji Julianto, Lanjar Sudarto, Lalu Agus Wirawan. Potential and Development of Coastal Sandy Land for Agriculture and Tourism. Didi Saidi, Tuti Setyaningrum, A.Z. Purwono Budi Santoso. Processing of agricultural products 44 Forest Conservation and Food Security Based on Local Food Resources of Iles-Iles (Amorphophallus muelleri Blume) in Supporting Ecotourism. Sumarwoto and Budiadi. Agronomy 45 Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. 46 An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodijoi, Sri Sumarsih. 47 Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bail. I Ketut Kariada, I. G. K. Dana. 48 Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Molit Eko Poerwanto. 49 Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. 50 The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. 51 Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. 52 Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. 53 Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. 54 Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. 55 The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. 56 Quality Improvement Phalaenopsis amabiliis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih.			
Potential and Development of Coastal Sandy Land for Agriculture and Tourism. Didi Saidi, Tuti Setyaningrum, A.Z. Purwono Budi Santoso. Processing of agricultural products Forest Conservation and Food Security Based on Local Food Resources of Iles-Iles (Amorphophallus muelleri Blume) in Supporting Ecotourism. Sumarwoto and Budiadi. Agronomy 45 Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. 46 An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. 47 Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. 48 Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. 49 Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. 50 The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. 51 Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. 52 Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. 53 Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoo. 54 Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism Ottavia S. Padmini. 55 The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. 66 Quality Improvement Phalaenopsis amabilis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. 67 Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-touri	42	The Decrease of Water Quality in Lake Rawa Pening The City of Semarang. Eko Amiadji Julianto, Lanjar Sudarto, Lalu Agus Wirawan.	315
Forest Conservation and Food Security Based on Local Food Resources of Iles-Iles (Armorphophallus muelleri Blume) in Supporting Ecotourism. Sumarwoto and Budiadi. Agronomy Agronomy Agronomy Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih.	43	Potential and Development of Coastal Sandy Land for Agriculture and Tourism.	322
Forest Conservation and Food Security Based on Local Food Resources of Iles-Iles (Armorphophallus muelleri Blume) in Supporting Ecotourism. Sumarwoto and Budiadi. Agronomy Agronomy Agronomy Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih.		Processing of agricultural products	
 Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	44	Forest Conservation and Food Security Based on Local Food Resources of Iles-Iles (Amorphophallus muelleri Blume) in Supporting Ecotourism.	331
 Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi. An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 			
An Inventory of Pests and Disease Attacking Jatropa at Potorono Village, Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih.		Agronomy	
Yogyakarta, Indonesia to Support The Development of Agro-tourism. R.R. Rukmowati Brotodjojo, Sri Sumarsih. Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	45	Agro-tourism Potential and Sustainable Agriculture in Lampung. Rusdi Evizal, Fembriarti Erry Prasmatiwi.	339
 Integrated Farming Systems to Support Agro-tourism at Kerta Village, Payangan Subdistrict of Gianyar-Bali. I Ketut Kariada, I. G. K. Dana. Management of CVPD by Controlling Diaphorina citri for Developing Agrotourism on Citrus. Mofit Eko Poerwanto. Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	46	Yogyakarta, Indonesia to Support The Development of Agro-tourism.	349
Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	47	Interested Forming Systems to Support Agro-tourism at Kerta Village,	359
 Environmentally Friendly Rice Production Increased By Plant Growth Promoting Rhizobacteria to Develop Agro-tourism. Oktavia S. Padmini. The Potential of Plant Tissue Culture in The Agro-tourism Development. Endah Wahyurini. Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	48	Management of CVPD by Controlling <i>Diaphorina citri</i> for Developing Agrotourism on Citrus. Mofit Eko Poerwanto.	372
 Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	49	Environmentally Friendly Rice Production Increased By Plant Growth Promoting	383
Alasmalang, Banyumas, Central Java. Sakhidin. Performance of Three Dahlia Cultivars with Respect to Foliar Fertilizer Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	50	Endah Wahyurini.	393
 Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti. Aloe Vera Organic Cultivation for Supporting Tourism Industry in Yogyakarta. Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	51	Sustainable Durian Production for Rural Agro-tourism Development in Alasmalang, Banyumas, Central Java. Sakhidin.	402
 Ellen Rosyelina Sasmita, Supono Budi Sutoto. Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	52	Applications to Support Agro-tourism Around Mount Merapi. Heti Herastuti.	406
Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini. The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana. Quality Improvement Phalaenopsis amabillis Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of Diaphorina citri Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	53	Ellen Rosyelina Sasmita, Supono Budi Sutoto.	411
 Sugeng Priyanto, Maryana. Quality Improvement <i>Phalaenopsis amabillis</i> Bl. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of <i>Diaphorina citri</i> Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism 	54	Increasing Rice Production By Legumes Substituting on Crop Rotation and Organic Fertilizer to Develop Agro-tourism. Oktavia S. Padmini.	420
Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih. Oviposition Preference Determination of <i>Diaphorina citri</i> Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	55	The Harvesting Moment is an Agro-tourism on The Winged Bean Cultivation. Sugeng Priyanto, Maryana.	429
Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism	56	Quality Improvement <i>Phalaenopsis amabillis</i> BI. to Beauty of Agro-tourism Area. Retno Suryati, H. Mustadjab HK, Ika Septiningsih.	432
on older mane, commission	57	Oviposition Preference Determination of <i>Diaphorina citri</i> Kuwayama to The Symtomatic and Asymtomatic Citrus Plant of CVPD for Supporting Agro-tourism on Citrus. Mofit Eko Poerwanto, Chimayatus Solichah.	440
The Effect of Gibberellin Concentration and Composition of Media on The Growth of Kawista. Aisyah Fatwa Sari, Lagiman, Ami Suryawati.	58	The Effect of Gibberellin Concentration and Composition of Media on The Growth of Kawista. Aisyah Fatwa Sari, Lagiman, Ami Suryawati.	448

59	Assessment Types of Extracts and Length of Soaking Time on the Growth of Pepper (Piper nigrum L.) Plant Cutting. Darban Haryanto, Suwitno, Daisy Prapto Sriyani.	454
60	Callus Regeneration Post Gamma Ray Irradiation for Producing Seeds That Were Expected Resistant to Fusarium Wilt Disease to Support Agro-tourism. Ari Wijayani, Mofit Eko Poerwanto.	460
	Other topics related to Agro-tourism	
61	Increasing Household Food Security through The Role of Traditional Local Staple Food. Rita Hanafie.	466
62	Strategy Analysis of Creative Industries Development in Bantul District, Province of Yogyakarta. Dyah Rachmawati, Ahmad Muhsin.	473
63	Agriculture Biodiversity to Support Food Sovereignty. Lagiman.	482
	Category : Poster	
64	Pine for Forest Conservation to Support Agro-tourism. Rina Srilestari.	489
65	Selection of Soybean Variety for Leaf-Use Only and Its Characteristics Survey. Yong Duk Kim, In Kwan Song, Kwang Ju Lee, Young Taek Yang, Sung Taek Kim, Won Young Han, Bong Chan Kim.	494
66	The Influence of Jasmine Bush Stalks and Plant Growth Regulators on Growth and Development of Young Plants to Supply The Demand of Flowers as a Support for Agro-tourism. Rati Riyati, Sugeng Priyanto, Istu Ragil Murni.	505
67	Pakel Diversity in Sleman Regency for Agro-tourism Development in Administrative Territory of Yogyakarta (DIY). Basuki, Suyanto Zaenal Arifin.	512
68	Restructuring of Kinahrejo Area Based on Agro-Ecotourism After The Eruption of Merapi Using Ornamental Plants. Ari Wijayani, Irhas Effendi, Gunawan Nusanto, Hendri Gusaptono, Susilastuti, Eko Amiadji.	522
69	Natural Laboratory as a Society Learning Center. Bargumono, Subroto Padmosudarso.	530
70	Evaluation of Grading Tool on <i>Arumanis</i> Mango Plantations. Wahyunindyawati, Sri Harwanti.	541

MANAGEMENT OF CVPD BY CONTROLLING Diaphorina citri FOR DEVELOPING AGRO-TOURISM ON CITRUS

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ABSTRACT

Indonesia is the world citrus producer with more than two hundreds varieties of citrus. The varieties are locally specific which are potential for agro-tourism. Most of citrus orchards have been destroyed by Citrus vein phloem degeneration (CVPD). CVPD is the most impediment disease in citrus production in the world. It mainly vectored by Diaphorina citri Kuwayama. The disease also caused some of Indonesian indigenous citrus varieties endangered. Intensive application of insecticides was ineffective and also costly. Comprehensive strategy for managing CVPD is being conducted and evaluated by the integration of planting disease free seedling, and controlling disease vector (D. citri). The controlling strategy consists of guava intercropping, alternative host sanitation, planting area isolation, and mineral oil application. The strategy would decrease the infection of CVPD by reducing the inoculums titer, eliminating contacts between disease inoculums with the vector, and decreasing infective vector population. Conservation of Indonesian locally specific citrus varieties will develop agro-tourism on citrus

KEYWORDS: Citrus, CVPD, D. citri, guava intercropping, mineral oils

INTRODUCTION

Indonesia is currently the second world citrus producer after China with more than 210 varieties of citrus. Most of them are unique and locally specific. They have been supporting tourism and potentially for developing agro-tourism on citrus. The citrus production in 1995 was 143,059 ton, and it decreased to 91,469 ton in 1996. The production increased into 696,422 ton in 1997 and decreased sharply into 449, 552 ton in 1999. Recently, citrus production in Indonesia has developed rapidly. In 2007, the production increased to 2,625,884 ton. In 2008, the production decreased again into 2,467,632 ton and in 2010 were 2,032,665 ton. It was due to the orchard extension (BPS, 2011). Fluctuation on citrus production was mainly caused by Citrus vein phloem degeneration (CVPD). The disease caused 50 million citrus trees lost both in South Asia and South East Asia CVPD was first reported in Indonesia in 1964 (Semangun, 1991). In Indonesia the most susceptible cultivated form of citrus to CVPD is C. reticulata Blanco (Rutales: Rutaceae). Severe epidemics of the disease occurred in the 1960s, especially in Java and Sumatera where at least 3 million trees were destroyed. It was informed that the disease has destroyed 9 million out of 42.8 million total citrus-trees in Indonesia in 1990. The economic loss was about 35 billion rupiah per year (Nurhadi, 1991). Many highly productive plantations where yields were 20 ton per ha decreased sharply into 8.6-15 ton per ha per annum (Irawan et al., 2003). A citrus rehabilitation program based on the eradication of infected trees and the production and use of ISAD, Yogyakarta, INDONESIA, December 6-8, 2011 372

disease-free bud wood by shoot-tip grafting was initiated in the mid 1980s. However, the citrus trees in Indonesia have not been free of CVPD (Bove *et al.*, 2006). CVPD continues to be the major impediment to citriculture in Indonesia.

CITRUS

Citrus is considered originated from Southeast Asia between India and China and southwards through Malesia (Dugo & Di Giacomo, 2002). Citrus was spread from the Far East to Middle East, Mediterranean, and eventually America. The genus Citrus belong to the Plantae Kingdom, Spermatophyta Division, Angiospermae Sub division, Dicotyledoneae Class, Rutales Order, the Rutaceae family. There were six genus belong to the Rutaceae family: 1) Citrus; 2) Microcitrus; 3) Fortunella; 4) Poncirus; 5) Cymenia, dan 6) Eremocitrus (Martasari & Hardiyanto, 2003). Recent work suggests that the genus Citrus comprises about 25 species (Mabberley, 2004). They are: C. medica, C. australis, C. australasica, C. garrawayi, C. glauca, C. gracilis, C. inodora, C. neocaledonica, C. oxanthera, C. sp. (Oxanthera fragrans), C. undulata, C. polyandra (syn. Clymenia), C. warburgiana, C. wintersii (syn. Microcitrus papuana), C. cavaleriei (C. ichangensis), C. japonica – kumquats (Fortunella), C. reticulata, C. trifoliata (syn. Poncirus), C. maxima – pomelo, C. amblycarpa (a possible hybrid), C. halimii, C. hystrix – kaffir lime (C. macroptera), C. warburgiana, C. swinglei, C. latipes.

Some citrus in Indonesia which are developed, such as *C. reticulata*: SoE mandarin from NTT; Batu 55 mandarin from East Java; *C. sinensis*: Madu siem, Maga siem and Beras Sitepu from Medan; Pontianak siem from West Kalimantan; Pacitan sweet orange from East Java; Waturejo sweet orange from Central Java; *C. maxima*: Nambangan Pummelo, Sri Nyonya Magetan from East Java; *C. aurantifolia* and *C. hystrix*: "Nipis"; and *C. medica*: "Lemon" (Agisimanto & Sutarto, 2007).

CVPD

CVPD, also recognised as Huanglongbing or greening disease, was firstly found and produced a serious problem in the Chaozhou/Shantou region of Guangdong in the mid 1930s (Lin, 1956). The disease epidemic was found in some Asian countries (Tirtawidjaja et al., 1965; Garnier & Bové, 2000; Weinert et al., 2004), and in the Arabian Peninsula (Bové & Garnier, 1984). It was also found in Africa, Mauritius and Réunion (Garnier et al., 1996), United States of America (Florida and Louisiana), Cuba (Martínez et al., 2008), and Brazil (Teixeira et al., 2005). The disease caused 50 million citrus trees lost both in South Asia and South East Asia (Gonzales, 1987).

At the initial stage trees develop mottle leaves in one or few branches similar to that caused by the lack of zinc (Dwiastuti et al., 2003). Leaves become smaller and bunchy. In most cases, the disease appeared localized on the tree. In severe cases, all cultivars of C. reticulata suffered from dieback of branches, mottling or yellowing of leaves were pronounced and extensive: the leaves become reduced in size, grow in an upright position and become vein corking. Trees will suffer from premature yellowing of fruit, undersized fruits that fall before maturity, browning of the stem and dieback exhibited by the branches. The fruits become bitter and sour and the sugar concentration decreases. In subtropical climates, CVPD induces heavy leaf shedding at the beginning of the winter season (Gonzales, 1987). CVPD agent is a fastidious or unculturable Gram-negative bacterium with the proposed name 'Liberibacter asiaticus' and 'Liberibacter africanus' for Asian and African types, respectively (Nakashima et al., 1998). The hosts of CVPD agent are not only citrus, but also Catharanthus roseus ISAD, Yogyakarta, INDONESIA, December 6-8, 2011

(periwinkle), Cuscuta campestris and Murraya exotica also. The latent period in citrus plants before symptoms is expressed ranges from four months to one year or more. Once the citrus plant has been infected, it will die in two to four years (Su & Huang, 1990). There were 27 species and hybrid of Rutacea genus which could become the host of CVPD i.e. Atalantia, Citrus (including Fortunella and Poncirus), Murraya (M. paniculata) in Citreae, Bergera (B. koenigii or Murraya koenigii), and Clausena in Clausenaeae. The most devastated commercial citrus is C. medica, followed by C. reticulata (Yang et al., 2006).

CVPD is not transmitted by seeds, but is naturally transmitted through vegetative propagation and by insect vector (Subandiyah et al., 2000). 'Candidatus L. africanus' is transmitted by insect vector of Tryoza erytreae while 'Candidatus L. asiaticus' and 'Candidatus L. americanus' are transmitted by D. citri (Bove, 2006). The pathogens have been detected in haemolymph and salivary glands of T. erytreae and D. citri (Xu et al., 1988).

VECTOR OF CVPD (Diaphorina citri)

D. citri Kuwayama (Homoptera: Psyllidae) in Asia was spread in the Arabian Peninsula (Saudi Arabia and Yemen), and from Afghanistan through the Indian Subcontinent, Southeast Asia and East Asia (the Ryukyu Archipelago, Okinawa and Kyushu in Japan, in China, particularly Taiwan and the coastal provinces of Guangxi, Guangdong, Fujian and Zhejiang), the Philippines and through the Indonesian archipelago to north eastern Papua New Guinea (Bellis et al., 2005). In America it was distributed in Hawaii, Guam, Alabama, California, Florida, Georgia, Louisiana, Mississippi, South Carolina, Texas, Commonwealth of Puerto Rico, Bahamas, Cayman Islands, Cuba, Jamaica, Dominican Republic, Guadeloupe, Venezuela, Brazil, Paraguay, Uruguay and Argentina; the Mascarenes (Mauritius and Réunion) (Halbert & Manjunath, 2004).

Female psyllids lay eggs only in the growing tips on the new shoots of the host plant of young host plants, preferring flush growth < 6 mm in length. Numbers of eggs laid on a flush decline rapidly as the length of flush increases, and when individual leaves attain lengths > 10 mm, and flushes 50 mm (Leong, 2006). The population density of eggs and nymphs is entirely dependent on the availability and abundance of new plant shoots (Tsai et al., 2002). Most eggs are laid within 14 days of new growth commencing (Lin et al., 1973). They suck phloem sap from juicy leaves or petioles, and excrete white pellets or threads. It takes 15 days to complete the five nymphal instar stages and adults can live for up 6 to 9 months (Xie et al., 1989). Flight is limited to 0.5-2 km when searching for a host; dispersal over 90 to 470 km is, or may be, possible in strong winds, such as those associated with cyclones (Halbert et al., 2008).

Only adult psyllids and the 4-5th instar nymphs are able to acquire the pathogen (Capoor et al., 1974; Xu et al., 1988). However, Hung et al. (2004), using PCR, detected the pathogen in second to fifth instar nymphs and adult D. citri. After feeding on a diseased plant for 30 minutes or longer, the pathogens remain latent inside the vector from between 3-20 days (Chavan, 2004). After acquisition from an infected plant, the pathogen multiplied first in the midgut epithelial cells of D. citri, and then penetrated through the basal lamina and entered into the haemocoel, where it multiplied again, before invading haemocytes, nerves tissue, fat body, trachea, muscle and eventually the salivary gland (Jiang, 2005). Once the psyllid vector acquires the pathogens, it can transmit them throughout its life span. However, it can not transfer the virulence to its progeny via eggs (Xu et al., 1991; Hung et al., 2004). The disease may shorten the age

of female, decrease fecundity, survivorship of second instars nymph, and the intrinsic rate of increase; and increase the ratio of male/female (Poerwanto, 2000; Jiang, 2005).

CONTROL STRATEGY OF D. citri

Integrated strategy for controlling CVPD and conserving endangered citrus varieties can be developed by mineral oil application, guava intercropping, alternative host sanitation, and planting area isolation. It also contributed in reducing highly poisoned pesticides contamination in ecosystem, especially in the area of citrus orchards.

Oils application

HMOs and AMOs (Horticultural and Agricultural Mineral Oils) are highly refined mineral oils derived from crude petroleum oils. They are paraffinic (\geq 60% of carbon atoms occur in chains). HMOs are called narrow-range petroleum spray oils and AMOs are called broad-range petroleum spray oils. Common median nCy values of HMOs are nC21 and nC23. For AMOs they are nC23, nC24 and nC25 (Agnello, 2002; Beattie and Hardy, 2005; Beattie, 2009). Those mineral oils are less phytotoxic to the plant and do not induce any carcinogenic effect to human (Beattie, 2010; personal communication)

Application of oils for citrus pests is ranged from 0.4% to 0.5% sprays at 5-14 day intervals within flush cycles (beginning when the first buds in each cycle begin to open) to control a range of pests and diseases, particularly *D. citri* (Poerwanto *et al.*, 2008, 2010) and citrus leafminer, simultaneously. Application of mineral oils to citrus mandarin leaves reduced 56.7% - 61.3% proportions of psyllids attracted to host plant volatiles (Poerwanto *et al.*, 2008). *D. citri* gravid female rejected to lay their eggs on plant treated with mineral oils (Rae *et al.*, 1997). Responses of adult *D. citri* to mineral oil deposits are olfactory and related to detection by antennal receptors of oil volatiles and/or plant volatiles. This outcome indicates that application of the oils to the mandarin leaves may have: (a) suppressed release of attractant host plant volatiles; (b) masked attractant host plant volatiles; (c) led to the release of repellent volatiles from leaves; and/or (d) to adults being repelled by oil volatiles (Poerwanto *et al.*, 2008).

Guava interplanting

Based on observations by Vietnamese ACIAR researcher team, it is suggested that citrus groves interplanted with guava trees citrus was free from *D. citri* invasion and low incidence of HLB-infected trees (Beattie *et al.*, 2006). Certain volatile compounds in guava leaf could be developed as repellents for the management of citrus psylla. Guava fruits and leaves produce a wide range of volatile compounds, such as sesquiterpenes (Sagrero-Nieves *et al.*, 1994; Ogunwande *et al.*, 2003), aldehydes and alcohols (Idstein & Schreier, 1985; Begum *et al.*, 2002, 2004; Soares *et al.*, 2007). Some of these aldehydes and alcohols are the so called 'green leaf volatiles' that have been shown to have repellent effects on insects (Jang & Light, 1991).

The present study revealed that the repellent action of guava against citrus psylla is dose-dependent, with very low doses having little effect on citrus psylla. This result indicates that to control citrus HLB by interplanting guava trees in citrus groves, sufficient numbers of guava trees are needed to keep the dosage of volatile compounds emitted from guava at an effective level in the entire grove (Zaka, et al. 2010). In China, observations revealed that even in the presence of guava trees scattered inside or around the groves, citrus psylla populations are high. This could be due to the fact that the guava trees present did not release enough active volatile compounds (Beattie et al.,

ISAD, Yogyakarta, INDONESIA, December 6-8, 2011

2006). In Indonesia, ACIAR researcher team interplanted guava among the young citrus plants. The ratio of guava: citrus population is 1:8 up to the next 6 months, there was no confirmed with PCR) and 0.4 imago of D. citri with 0.3 nymph colony and 0.1 egg 200 citrus trees of 3-6 years old (Pustika et al. 2008). In Vietnam, it was suggested that (Beattie et al., 2006).

Alternative host sanitation

D. citri colonies are also found on ornamental plants, such as orange jasmine (Murraya paniculate and Murraya exotica), and Bergera koenegii all year round (Tsai et al., 2002). Four of 16 species of weeds in citrus orchards are able to be the alternative host. There were Alternathera philoeroides of grass group, Amaranthus spinosus, Ludwigia perrenis, and Boerhavia erecta of broadleaf weeds (Hardiastuti & Poerwanto, 2011).

D, citri was able to survive a maximum of eight days with the mean longevity of 5.91 \pm 0.251 days on the broadleaf weed B. erecta, but was not able to complete one life cycle. It was significantly different to other weeds, A. spinosus, A. philoeroides, and L. perrenis which were survived for 3.57, 0.20, and 0.20 days respectively. The ability to survive in non host plants indicated the existence of nutrients content in the weed that resembles its host plant nutrient content or the existence of certain substances that could stimulate D. citri to stay for feeding (Hardiastuti & Poerwanto, 2011). The substances could be a various compounds of alcohol and aldehyde from the leaves of which were specific and volatile used by insect to find its host plant (Visser, 1986). However, the type and number of nutrient content was not as complete as in the host plant for surviving and completing its life cycle. Similar results were also obtained by Sudiono and Purnomo (2008) in Gemini virus isect vectors (Bemecia tabaci), and Hardiastono (2001) on Peanut Stripe Virus (PStV). B. tabaci were able to live on broadleaf weed Ageratum conyzoides. The weeds also serve as a source of inoculum since they were able to be infected by Gemini virus. While Amaranthus spinosus, Bidens pilosa, Crotalaria incana, Glycine max and Physalis angulata could be as alternative hosts and source of inoculums for PStV infection.

The implications of those survivorships of *D. citri* in ornamental plants and some weed species is the presence of alternative host for *D. citri* when citrus plant is not available as a food source, either because there are no plants or when plants are treated with pesticides. The existence of alternative host will cause the population of *D. citri* is available throughout the season and serve as initial population for the next generation population. The role of *D. citri* as a propagative vector of CVPD pathogen will also make the disease inoculums always available throughout the season in field (Hardiastuti and Poerwanto, 2011). This will further complicate the control measures against CVPD on citrus plant, since the disease transmission is highly dependent on the availability of disease inoculums and insect vector population in the field (Chen, 1998).

Planting area isolation

It had been reported that plant odours (volatile) detection and processing plays an important role in host plant selection by herbivorous insects (Moran and Brown, 1973; Todd et al., 1990; Appleton et al., 2004; Bichao et al., 2005). The volatile information was received by insect's central nervous system at a fine-scale spatio-temporal

resolution (Bruce et al., 2005). Odours vary substantially, depending on species, cultivar, growing conditions, age and plant parts (Takabayashi et al., 1994). Establishing new citrus orchard in the isolated area will delay the D. citri infestation and CVPD infection. It takes longer time for citrus plant odours to be detected by D. citri and attract its colony to infest.

Initial colony of *D. citri* was found 20 months after planting at new citrus orchard in an isolated area, far from the established citrus orchard and was surrounded by paddy's field. Five months more was needed for the colony of *D. citri* to colonized 100% citrus plants in one block. The colonization of *D.citri* in other blocks started from one plant spread to other plants at the same block before migrating to other nearby blocks (Poerwanto, 2010). The spread was very slow because the flight ability of psyllid was very low. Aubert (1990) and Tsai *et al.* (2002) reported that wind was needed by psyllid in spreading in long distance (0.5 – 4 Km).

CONCLUSIONS

Management of CVPD disease should be comprehensively conducted to conserve local citrus varieties for developing agro-tourism on citrus. It should be started by seeking new area for establishing citrus orchard. The area should be far enough from citrus orchard and free from alternative hosts of CVPD and its vector. Second step is interplanting guava among citrus plants followed by using mineral oils when the vector colony was found. Last but not least is keeping the orchard always clean of weeds which could be used as alternative host by vector.

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