The composition of ant species on banana plants with Banana Bunchy-top Virus (BBTV) symptoms in West Sumatra, Indonesia.

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ABSTRACT. A brief study on ant species on banana plants with Banana Bunchy-top Virus (BBTV) symptoms was conducted by direct collection in four regencies and one city of West Sumatra Province in Indonesia. During the sampling we found 39 banana plants with BBTV symptoms, of which36 were occupied by insects, and of these 35 plants contained ants. A total of 24 species of ants, belonging to 16 genera, were collected. Myrmicinae was the subfamily with the highest number of species (11), followed by Formicinae (seven) and Dolichoderinae (six). *Tetramorium* and *Technomyrmex* were the genera with the most species (four). *Dolichoderus thoracicus* was found most frequently among samples, accounting for16% of species occurrences, followed by *Tapinoma melanocephalum* and *Paratrechina longicornis* (11 and 10% respectively). Seventeen species of ants were found associated with aphids of which six showed a statistically-significant association, while seven ant species were not observed with trophobionts. Species that were found more often associated with aphids were found more frequently during the study.

Keywords: ants; composition; banana; BBTV; West Sumatra; Indonesia.

INTRODUCTION

The number and composition of ant species in an area can indicate the health of an ecosystem and provide insight into the presence of other organisms, since many ant species have obligate interactions with plants and other animals (Alonso & Agosti 2000). Ants are perhaps the most common and dominant animal mutualists in terrestrial environments (Ness *et al.* 2010). The ant is one of the ideal model organisms for measuring and monitoring biodiversity because ants are numerically dominant in ecological systems and are ecologically significant species that function as predators or symbionts of plants and other organisms. Ants are relatively easy to collect in a standardised way, reasonably diverse at sites, identifiable and also suitable indicators of environmental conditions (Peck *et al.* 1998; Herwina & Nakamura 2007).

Bananas and plantains represent the largest fruit crops globally in terms of production and trade. The total world production in 2006 was 113 million metric tons (Mt) (Daniells *et al.* 2011). Banana (*Musa* spp.) is the most important fruit in Indonesia (Damarjati 2000). This fruit has been selected by the government of Indonesia through the Indonesian Agricultural Agency for Research and Development (IAARD) as one of the most important food and cash crops to support national food security and economic development programmes. The crop is a staple,

contributes to a health-promoting nutritious diet, and is the first solid food/fruit fed to babies in Indonesia. Bananas are cultivated over extensive areas in the country (about 2400 km²), providing millions of job opportunities and contributing 53 % of all fruit exports (about 23 million USD) from the country (Nasir *et al.* 2005; Nasir 2010).

Pests and diseases are major constraints on banana production in Indonesia. At least four pests and diseases seriously affect banana plantations in the country, namely Bacterial Wilt, Fusarium Wilt and Sigatoka Leaf Spot. In West Sumatra (in Solok, Tanah Datar, Agam, Limapuluh Kota, Pasaman and Pariaman regencies) a high incidence of Sigatoka Leaf Spot has been reported, as well as Fusarium diseases (except in Pariaman, where Sigatoka was only at moderate levels) (Damarjati 2000). Sigatoka Leaf Spot is a black leaf-streak disease caused by the ascomycetous fungus Mycosphaerella fijiensis Morelet and the weevil borer Cosmopolites sordidus (Germar), a native of Malaysia and Indonesia (Mourichon et al. 1997). Currently in West Sumatra, banana plantations are threatened seriously by Banana Bunchy-top Virus (BBTV), together with wilt pathogens such as Fusarium (Nasir et al. 1999) and the Banana Blood-disease bacterium (Nasir 2010).

Banana plants infected with BBTV show a significant reduction in petiole length and spacing, canopy spread and height, leaf area, pseudostem diameter and chlorophyll content (Hooks et al. 2008). Suckers infected by BBTV are usually severely stunted, with leaves that do not expand normally and remain at the top of the pseudostem. Infected banana plants usually will not fruit or will have abnormal fruit with irregular "morse-code" streaking (Nelson 2004) (Fig. 2b). BBTV is transmitted vegetatively, through tissue culture and by the aphid vector Pentalonia nigronervosa Coquerel (Banana Aphid). No mechanical transmission has been reported (Diekmann & Putter 1996). Pentalonia nigronervosa is found whenever banana is grown and has become a significant pest of banana due to its ability to transmit BBTV (Ploetz et al. 2003; Nelson et al. 2006; Hooks et al. 2009). Banana Aphid populations are often tended by ant species (Nelson et al. 2006) since they produce large quantities of honeydew. Information about ant species on banana plants in Indonesia is still scanty, especially that about ants on bananas infected by BBTV. This work aimed to study the composition of ant species on banana with BBTV symptoms and their association with aphids, as there is no such information on it in West Sumatra to date.

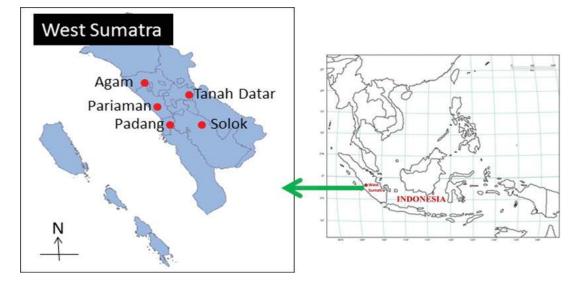


Fig. 1. The locations where ants were collected from banana with BBTV symptoms in West Sumatra.

Parameters	Solok	Pariaman	Agam	Tanah Datar	Padang
Total number of banana plants with BBTV symptoms	16	8	1	6	8
Total number of banana plants with ants	15	6	1	5	8
Total number of ant species on banana plants	17	8	3	12	9
Total number of ant individuals on banana plants	478	99	10	123	178
Temperaturerange during the study(°C)	27-31	30-32	25-29	29-30	29-32
Humidity range during the study (%)	52-62	56-60	56-68	59-66	52-59
Elevation range (m above sea level)	400-800	32-102	1019	378-393	28-128

Table 1: Total number of banana plants with BBTV symptoms, banana samples with ants, number of species and individuals of ants collected, temperature range during the study, relative-humidity range during the study and elevation range at each sampling location in West Sumatra.

MATERIALS AND METHODS

Ants were collected from September to October 2010 (the rainy season) in four regencies and one city (Solok, Pariaman, Agam, Tanah Datar and Padang: hereafter we refer to regencies and city as sampling locations) of West Sumatra Province (1°00'S, 100°30'E, Fig. 1). The altitudes of the locations range from 28 to 1019 m above sea level, temperature-range during the study was about 25 to 32°C (Table 1) and rainfall amounts to about 2121 mm per year (Environmental Impact Management Agency of West Sumatra 2012). We collected ants from bananas infected by BBTV at the five locations, at which bananas were available almost everywhere in small and traditional plantations, gardens or backyards. The infected bananas (Solok 16 plants, Pariaman 8, Agam 1, Tanah Datar 6, Padang 8) were inspected all over for insects, especially ants, for about 15 minutes per plant (Table 5). All foraging ants were collected (as well as other insects) directly from all parts of infected banana plants (leaf, petiole, pseudostem etc.) using forceps and aspirators, then stored in vials in100% ethanol.

Ants were sorted to genus and morphospecies level at the Animal Taxonomy Laboratory of the Department of Biology of Andalas University, following Bolton (1994) and then identification of ant to species level was conducted under the supervision of ant taxonomists at the Graduate School of Science and Engineering, Kagoshima University, Japan (see Acknowledgements). The ant specimens are housed in the Museum of Biology Department of Andalas University, Indonesia. The proportion of each ant species among all banana samples was described by relative frequency (frequency of one species divided by frequency of all species in all banana samples: a measure of the proportion of species/tree occurrences accounted for by that species). We used Estimate S 7.5 (Colwell 2005) for the calculation of rarefaction curves of observed and estimated number of ant species. Fisher's Exact tests were used to test the significance of the association of ant species with aphids, using unassociated species as a comparison, and Spearman Rank correlation was used to check the correlation of ant frequency and association with aphids. For certain species we counted the numbers of aphids in the trophobiotic associations.

RESULTS

During the study we found 39 banana plants with BBTV symptoms. Among them 36 banana plants were occupied by insects, and of these 35 (97 %) contained ants. Forty percent of bananas with BBTV symptoms that were occupied by ants were also attended by aphids (Table 1, Appendix). A total of 24 ant species, belonging to three subfamilies, 11 tribes and 16 genera, were collected (Table 2). Myrmicinae was the subfamily with the highest number of species (11) followed by Formicinae (seven) and Dolichoderinae (six).

Table 2: List of subfamilies, tribes and species of ants collected on banana with BBTV symptoms in West Sumatra (N=total number of individuals;SOCs=Species Occurrences;M = mean number of individuals per occurrence; RF= Relative Frequency).

Subfamily, tribe and species	Ν	SOCs	Μ	RF (%)
DOLICHODERINAE				
Dolichoderini				
Dolichoderus thoracicus (Smith, 1960)	154	14	11.0	15.9
Tapinoma melanocephalum (Fabricius, 1793)	147	10	14.7	11.3
Technomyrmex albipes (Smith, 1961)	1	1	1.0	1.1
Technomyrmex horni Forel, 1912	4	1	4.0	1.1
Technomyrmex kraepelini Forel, 1905	34	5	6.8	5.7
Technomyrmex vitiensis Mann, 1921	203	4	50.8	4.5
FORMICINAE				
Camponotini				
Camponotus (Tanaemyrmex) sp.	1	1	1.0	1.1
Camponotus sp. 1	1	1	1.0	1.1
Polyrhachis laevissima Smith, 1958	1	1	1.0	1.1
Lasiini				
<i>Nylanderia</i> sp.	47	6	7.8	6.8
Paratrechina longicornis (Latreille, 1802)	67	9	7.4	10.2
Oecophyllini				
Oecophylla smaragdina (Fabricius, 1775)	1	1	1.0	1.1
Plagiolepidini				
Anoplolepis gracilipes (Smith, 1857)	1	1	1.0	1.1
MYRMICINAE				
Crematogastrini				
Crematogaster (Orthocrema) sp.	7	3	2.3	3.4
Formixocenini				
Cardiocondyla wroughtonii (Forel, 1980)	21	2	10.5	2.3
Metaponini				
<i>Vollenhovia</i> sp.	1	1	1.0	1.1
Pheidolini				
Pheidole sp. 1	1	1	1.0	1.1
Pheidole sp. 2	65	7	9.3	7.9
Solenopsidini				
Monomorium floricola (Jerdon, 1981)	25	4	6.3	4.5
Solenopsis geminata (Fabricius, 1804)	47	7	6.7	7.9
Tetramoriini				
Tetramorium smithi Mayr,1979	2	2	1.0	2.3
Tetramorium kheperra (Bolton, 1976)	1	1	1.0	1.1
Tetramorium pacificum Mayr, 1870	24	2	12.0	2.3
Tetramorium bicarinatum (Nylander, 1846)	32	3	10.7	3.4
= 24 species				

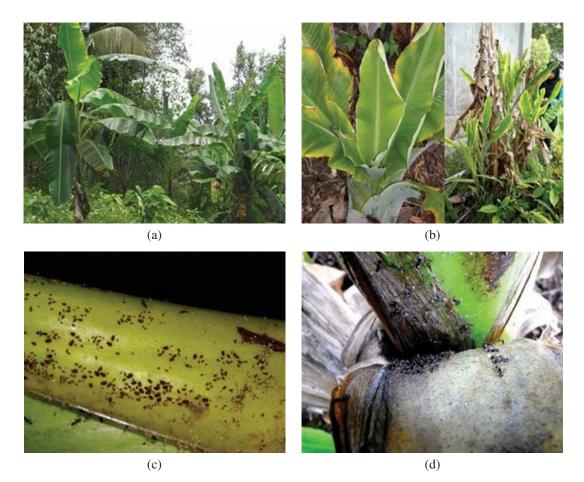


Fig. 2. Bananas and ants: (a) Traditional banana plantation without BBTV symptoms; (b) Banana with BBTV symptoms, including significant reduction in height and leaves that do not expand normally; (c) Aphids and *Technomyrmex* ants on leaf petiole of banana with BBTV symptoms; and (d) *Dolichoderus thoracicus* foraging on petiole of banana with BBTV symptoms.



Fig. 3. Photograph of ants on bananas with BBTV symptom (a) *Dolichoderus thoracicus*, the most frequent ant among bananas with BBTV symptoms in West Sumatra; (b) *Technomyrmex vitiensis*, the most locally abundant ant during the study.

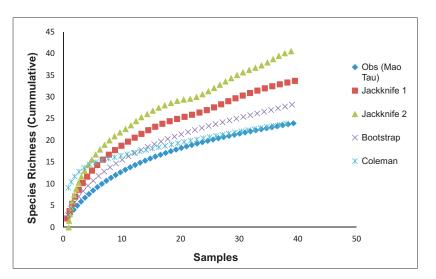


Fig.4. Observed species (Mao Tau), Jackknife 1, Jackknife 2, Bootstrap richness estimator and Coleman Rarefaction of ants collected on banana with BBTV symptoms in West Sumatra.

Table 3: Ant species collected on bananas with BBTV symptoms and the percentage of observations when they were with aphids. Given are the species names, number of total observations, percentage of observations with aphids and the P- value of the Fisher's exact test. We tested the hypothesis that the respective species differed in their association with trophobionts from the seven species that were never observed tending aphids. * marks an almost-significant result.

Species	Total Observations	Percentage of observations with aphids	Fisher's exact test P value
Dolichoderus thoracicus	14	50	< 0.01
Tapinoma melanocephalum	10	60	< 0.01
Tetramorium bicarinatum	3	67	< 0.01
Pheidole sp. 2	7	43	< 0.05
Solenopsis geminata	7	43	< 0.05
Tetramorium pacificum	2	100	< 0.05
Technomyrmex vitiensis	4	50	0.057 n.s*
Paratrechina longicornis	9	33	0.074 n.s.
Camponotus (Tanaemyrmex) sp.	1	100	0.083 n.s.
Camponotus sp. 1	1	100	0.083 n.s.
Technomyrmex albipes	1	100	0.083 n.s.
Technomyrmex horni	1	100	0.083 n.s.
Tetramorium kheperra	1	100	0.083 n.s.
Nylanderia sp.	6	33	0.110 n.s
Cardiocondyla wroughtonii	2	50	0.154 n.s.
Crematogaster (Orthocrema) sp.	3	33	0.214 n.s.
Technomyrmex kraepelini	5	20	0.313 n.s.
Monomorium floricola	4	0	not tested
Tetramorium smithi	2	0	not tested
Anoplolepis gracilipes	1	0	not tested
Oecophylla smaragdina	1	0	not tested
Pheidole sp. 1	1	0	not tested
Polyrhachis laevissima	1	0	not tested
Vollenhovia sp.	1	0	not tested

Tetramorium (Myrmicinae) and *Technomyrmex* (Dolichoderinae) were the most speciose genera (with four species respectively). *Dolichoderus thoracicus* was the species found most frequently among banana samples, with a relative frequency of 15.9 % followed by *T. melanocephalum* and *P. longicornis* (11.3 % and 10.2 % respectively).

Technomyrmex vitiensis was the most locally-abundant ant in this study (with a mean of 50.8 individuals per occurrence) but was low in relative frequency (4.5 %) (Table 2).

Several estimators were used to predict the total number of ant species expected to be present in the sampling area (Fig. 4). The Bootstrap estimator predicted 31.7 species, while Jackknife 1 and 2 predicted 33.7 and 40.5 respectively.

Of 24 species of ants collected on banana plants with BBTV symptoms, 17 species were associated with aphids while seven, with a total of 11 observations, were not associated with trophobionts (Table 3). Six species differed significantly in the strength of their aphid association from the group of ants not found with aphids (Fisher's Exact tests, $P \le 0.05$, see Table 3). For five ant species we counted the number of aphids that they tended (Table 4). T. vitiensis and Pheidole sp. 2 with four and seven observations had the highest mean number of aphids. Among ant species there was a positive correlation between the percentage of occurrences associated with aphids and the frequency at bananas (Spearman Rank correlation N = 24, R = 0.729, T = 5.00, P < 0.001).

Table 4: Five ant species with the number of aphids they tended. Given is the ant species name, the number of observations, the mean number of aphids counted at the observations and their standard deviation.

Species	Number of observations	Mean number of aphids	SD	
Dolichoderus thoracicus	14	30.6	15.1	
Tapinoma melanocephalum	10	69.0	87.8	
Paratrechina longicornis	9	21.3	18.0	
Pheidole sp. 2	7	433.3	468.2	
Technomyrmex vitiensis	4	572.0	799.0	

Table 5: Total number of species and individuals of ants, total number of species and individuals of all insects collected and cultivars of each sample of banana with BBTV symptoms from each sampling location in West Sumatra (BS = Banana Sample; Sa = Total number of ant species; Si = Total number of species of all insects; Na = Total number of individuals of ants; Ni = Total number of individuals of all insects; N = Total number of individuals of samples visited by ants/other insects; * = also attended by Homopterans).

Location	Date (2010)	BS	Sa	Si	Na	Ni	Cultivar
Solok	23 Sep	1	8	11	52	58	Sirandah
		2	5	9	54	60	Ambon Hijau
		3	1	6	6	979	Ambon Hijau*
		4	1	2	1	95	Sirandah*
		5	1	4	7	10	Ambon Hijau
		6	1	2	178	1315	Kepok*
		7	4	8	74	313	Raja Serai*
		8					Kepok
	24 Sep	9	1	7	24	90	Ambon Hijau*
		10	2	3	17	18	Ambon Hijau

Location	Date (2010)	BS	Sa	Si	Na	Ni	Cultivar
		11	1	4	11	63	Sirandah
		12	2	3	3	4	Mas
		13	1	2	7	9	Ambon Hijau
		14	2	3	14	18	Lidi
		15	2	5	6	9	Ambon Hijau*
	6 Oct	31	4	5	24	121	Sirandah*
Pariaman	26 Sep	16					Ambon Hijau
		17	3	4	19	20	Ambon Hijau
		18	2	6	17	62	Jantan*
		19	5	6	22	35	Rotan*
		20	2	3	21	23	Manis
		21					Jantan
		22	1	4	17	48	Jantan
		23	1	3	3	5	Jantan
Agam	27 Sep	24	3	5	10	17	Mas*
Tanah Datar	6 Oct	25		1		1	Lidi
		26	3	6	25	83	Ambon Hijau*
		27	3	5	3	6	MasakSehari
		28	4	9	42	85	MasakSehari*
		29	5	7	36	48	Ambon Hijau*
		30	2	9	17	58	Godok*
Padang	18 Oct	32	3	5	11	13	Ambon Hijau
		33	2	3	36	38	Ambon Hijau
		34	2	2	58	58	Ambon Hijau
		35	3	4	15	16	Manis
	19 Oct	36	3	3	9	9	Manis
		37	1	2	19	20	Manis
		38	2	2	16	16	Manis
		39	2	2	14	14	Manis
Ν					888	3837	
S					35	36	

DISCUSSION

The tending of homopterans by ants is well known. The majority of homopteran-tending ant species occur in the subfamilies Dolichoderinae, Formicinae and Myrmicinae, although some Ectatomminae also obtain significant nutrition through such interaction (Schultz & McGlynn 2000). In this study we found ants from the Dolichoderinae, Formicinae and Myrmicinae but no Ectatomminae foraging on bananas with BBTV symptoms. Myrmicinae had the highest number of species, as found in many previous ant studies (Ito *et al.* 2001; Herwina & Nakamura 2007;

Abera et al. 2007; Brühl & Eltz 2010; Pfeiffer et al. 2011). The three ant species most frequently collected on bananas with BBTV symptoms in this study (D. thoracicus, T. melanocephalum and P. longicornis) were also collected as arboreal ants of oil palm Elaeis spp. plantations in Borneo and Peninsular Malaysia and characterised as invasive, alien and tramp species (Pfeiffer et al. 2008). In West Sumatra, D. thoracicus are very abundant in fruit orchards and cacao Theobroma cacao L. plantations (Herwina unpublished). These species are general scavengers and also tend Hemiptera to collect honeydew. They nest either in the soil or arboreally (Shattuck 1999). In Australia T. melanocephalum are general scavengers but have a preference for honeydew and often tend aphids or coccids (Shattuck 1999). Paratrechina longicornis is a widespread introduced tramp species in many tropical countries, forming large colonies in open soil, under rocks or other objects (Shattuck 1999) and has also been collected as a ground-dwelling forest ant in oil palm plantation in Sabah, Malaysian Borneo (Brühl & Eltz 2010).

Among the banana samples infected by BBTV we found aphids (Hemiptera: Aphididae) on 14 bananas (40 %) and among 24 ant species collected, 17 (71%) were found together with aphids. The most significant aphid associations based on these observations were for *D. thoracicus*, T. melanocephalum and T. bicarinatum, followed by Pheidole sp. 2, S. geminata and T. pacificum (Table 3). Technomyrmex vitiensis, the most locally-abundant ant in this study where it occurred, was found in four banana samples of which two were also occupied by a large number of aphids. Most individuals of this species were found in only one banana plant, together with more than a thousand individuals of aphids (Appendix, Table 3), giving it the highest mean number of aphids during the observations (Table 4). This species is reported to be a general scavenger, foraging on the ground, low vegetation and trees (Shattuck 1999).

Three *Pheidole* species were reported as important predators of banana weevils in Uganda (Abera *et al.* 2007). These ants are general predators and scavengers and also feed on seeds. We found two species of *Pheidole* in this study (Table 2) of which one, *Pheidole* sp. 2, had a positive association with aphids (Table

3, Table 4).

Anoplolepis gracilipes (the Long-legged Ant) has been reported as a dominant banana pest in Hawaii (Constantinides & McHugh 2003) and elsewhere. This species was the second most abundant ant collected by pitfall traps in Bogor Botanical Garden (Herwina & Nakamura 2007) and the most frequent ground-dwelling ant in oil-palm plantations of Borneo (Brühl & Eltz 2010). Anoplolepis gracilipes was also the commonest invasive ant in forest and cacao agroforests of Central Sulawesi (Bos et al. 2008). Shattuck (1999) wrote that nests of this species are primarily in the soil, but may be arboreal as well. They are general predators on a range of arthropods and are known to tend Hemiptera to collect honeydew. This species was found as only a single individual in this study together with other eight singleton species (Table 2), and no aphids were associated (Table 3).

Only a single *Oecophylla smaragdina* queen and her eggs were found in this study. This is interesting since in Indonesia *O. smaragdina* is frequently encountered on perennial fruit trees such as Mango *Mangifera indica* L. and Rambutan *Nephelium lappaceum* L. as a very active predatory ant (Herwina unpublished.). The queen of *O. smaragdina* found in this study was about to start a new colony on banana. *Oecophylla* is predatory and also tends homopterans (Brown 2000), but no aphids were found during the sampling of this species.

Comparing the number of ant species between this study and study of ants in a banana farming system in Uganda (Abera *et al.* 2007) is not simple since the methodologies were different (direct collection of ants from banana plants was used in this study while in Uganda pitfall traps and bait traps were used), but we found nine genera (23 %) in common.

Although the sampling could be considered sufficient because the number of observed species in this study was more than half of the estimated species total (see Chao &Lee 1992), the accumulation curves showed that ant species saturation was not yet reached. A larger number of samples would be more revealing.

This is the first report about ants on banana plantations in Indonesia, and suggests ant communities could have a significant influence on pest populations. Species that were found more often associated with aphids were found more frequently at the bananas than species without aphids. Future study is needed to learn more about the function of ants as well as aphids and other insects foraging on bananas to deal with the spread of BBTV on banana-growing areas and plantations in West Sumatra. Comparative study of ants between healthy banana and banana infected by BBTV is also suggested.

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