

#### ECOLOGY AND BEHAVIOR

# A Case Study on Urban Ant Fauna of Southern Kyusyu, Japan, with Notes on a New Monitoring Protocol (Insecta, Hymenoptera, Formicidae)

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Abstract Ant fauna of a green buffer belt adjacent to an industrial zone in Kagoshima City, southern Kyushu, Japan was elucidated using a standardized protocol. A total of 39 species belonging to 24 genera in five subfamilies were recorded from this park. Most speciose genera are Camponotus and Tetramorium having four species. The number of species per genus is 1.56 on average. Pheidole noda F. Smith, Monomorium chinense Santschi, Solenopsis japonica Wheeler, Tetramorium bicarinatum (Nylander) and Technomyrmex albipes (F. Smith) were most frequently encountered. Excepting Pheidole noda they are open-land and/or forest-edge inhabitants. Furthermore, Tetramorium bicarinatum and Technomyrmex albipes are tramp species. Thus, ant fauna there strongly reflects environmental conditions derived from urbanization/industrialization. Based on the present results, we propose here a new protocol monitoring ants in urban/industrial zones consisting of baiting and time unit sampling.

Key words assessment, biodiversity, Formicidae, man-made habitat, tramp species

#### Introduction

Ant activities in man-made habitats often bring negative effects to economy and public health (Beaver et al., 1986 indirectly cited from Martinez et al., 1997; Rohrbach et al., 1988; Taber, 2000) as well as positive effects (Carroll and Risch, 1990; Way and Khoo, 1992). Furthermore, introduced ant species usually establish themselves in man-made habitats, and then some of them expand to adjacent semi-natural/natural habitats and dramatically transform native biota there (Wetterer, 1998; Hoffmann et al., 1999; Vanderwoude et al., 2000). In the warmer

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part of Japan, some ant species, e.g., Monomorium pharaonis (Linnaeus), Pheidole megacephala (Fabricius) (established in the Ryukyus only), Anoplolepis gracilipes (F. Smith) (Ryukyus only), Paratrechina sakurae (Ito), Ochetellus glaber (Mayr) (= O. itoi (Forel)) and Tapinoma melanocephalum (Fabricius), are dominant in urban areas, and occasionally or permanently trespass on houses and give residents discomfort feeling (Terayama, 1986; Yamane et al., 1994; Eguchi, pers. observ.). In early 1990's, permanent populations of Linepithema humile (Mayr) were found in urban areas in western Honshu, Japan (Terayama, 2002; Touyama et al., 2003; Touyama et al., 2004). There is a possibility that the species spread over urban areas, agroecosystems and semi-natural habitats under the temperate climate and become a major threat not only to native biota but also to agroeconomy and public health.

Unfortunately, ant faunas in man-made habitats have not vet been studied well in southern Kyusyu. In the present study we elucidate features of ant fauna in a green buffer belt adjacent to an industrial zone of Kagoshima City, southern Kyushu using standardized protocols, and provide bionomics on dominant species. Urban and industrial zones, especially those under warmer climates, may act as major gateways of tramp/introduced ant species and harbors of their initial populations. The information provided here may be useful for future programs for monitoring harmful tramp/introduced ants.

## Materials and Methods

This study was conducted in "Nanatsujima" Park located in Kagoshima City, Kagoshima Prefecture (southern Kyushu), Japan (Fig. 1). This park was prepared as a green buffer belt in 1972 when "Nanatsujima" beach was reclaimed for an industrial zone, and have been opened to public since 1977. The park extends from north to south, and to the east

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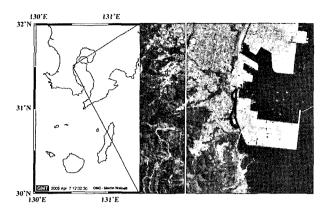


Fig. 1. Location of Nanatsujima Park. Nanatsujima Park is surrounded by a solid line on the aerial photograph (right) which was took by the Geographical Survey Institute, Japan on 12th May 2000 and is available at <a href="http://mapbrowse.gsi.go.jp/airphoto/index.html">http://mapbrowse.gsi.go.jp/airphoto/index.html</a>. The map of the southern part of Kyushu, Japan (left) is produced by an online software "Online Map Creation" available at <a href="http://www.aquarius.geomar.de/omc/make">http://www.aquarius.geomar.de/omc/make</a> map.html</a>.

faces the industrial zone and to the west a hillside partly covered with mixed woodland: 1 km in length, 12 m to 115 m in width, and 4,032 m² in area. *Prunus x yedoensis* Matsum., *Myrica rubra* Sieb. et Zucc., *Camelia japonica* L., etc. are planted. Branches of trees and the undergrowth are often pruned and cleared away, but regular pesticide/herbicide spraying are not done.

Several standardized sampling protocols for ground-dwelling ants have been proposed in recent years (Agosti *et al.*, 2000; Yamane and Fellowes, 2001), but these were primarily designed for surveying ants in forest ecosystems, with an exception of time-unit-sampling (Ogata *et al.*, 1998). In the present study we employed "Quadra-Protocol" and "30-min. Time-Unit Sampling".

## Quadra-protocol

The following four transects representing major habitats in the park were arranged for Quadra-Protocol.

Transect A, consisting of two subtransects, was set on woody habitats where *Ficus superba* Miq. var. *japonica* Miq., *Lithocarpus edulis* (Makino) and *Myrica rubra* (2 to 5 m in height) were dominant. These trees provided shade for the ground from the sun. The ground was trod down by visitors. This transect was surveyed between 22<sup>nd</sup> May and 9<sup>th</sup> June 1999.

Transect B was set in a row of *Prunus x yedoensis* trees having a mix woodland behind. The ground was wetter than that of Transect A. This transect was

surveyed between 30th June and 22nd July 1999.

Transect C, consisting of three subtransects, was set in hedges of *Rhododendron oomurasaki* Makino, *Camellia sasanqua* Thunb. and *Ilex crenata* Thunb. Leaf litter layer was relatively developed on the soft ground shaded by the hedges. This transect was surveyed between 19<sup>th</sup> July and 28<sup>th</sup> August 1999.

Transect D, consisting of three subtransects, was set on lawn patches (*Cynodon dactylon* (L.)) exposed to a lot of sunshine. This transect was surveyed between 17<sup>th</sup> September and 9<sup>th</sup> October 1999.

Ants in each transect were collected/recorded by a protocol, consisting of the following four methods (or three methods in the case of Transect D). The protocol was a little modified from the original "Quadra-Protocol" given by Yamane and Hashimoto (2001).

Honey Baiting: A series of 45 baits (small cotton pieces dampened with 20% honey/syrup solution) was set on the ground with several-meter intervals once in the daytime and once in the nighttime. Ant species attracted to each bait were recorded after around one hour (but ca. two hours for a subtransect of Transect D in the nighttime). Furthermore, in Transects A and B, a series of 45 baits was set on tree trunks (at breast height) once in the daytime and once in the nighttime.

Soil Sifting: Fifteen soil samples (20 x 20 x 10 cm in depth) were collected. All ant individuals extracted from each soil sample using a hand-sieve and a white pan were put into a single vial filled with 80% ethanol. In Transect D topsoil tightly grasped by roots of the lawn (upper 2 cm in depth) was not sifted to avoid damage to the lawn.

Litter Sifting: Leaf litter sifting was done for 30 minutes by a single person (K. Iwata) with a hand sieve and white pan between bait No. 1 and 15, between 16 and 30, and between 31 and 45 (i.e., 30-min sampling x 3 trials for each transect). Transect C had accumulated enough amount of litter for this sampling. On the other hand, for Transect A and B, litter around tree bases was sifted because litter was constantly swept out along these transects. This method was not conducted in Transect D where litter from trees was not accumulated. The basic premise of this method as well as free searching mentioned below is to collect as many ant species as possible within 30 minutes.

Free Searching: Free searching was done for 30 minutes by a single person (K. Iwata) between bait No. 1 and 15, between 16 and 30, and between 31 and 45 (i.e., 30-min sampling x 3 trials for each transect). This sampling method focused particularly on microhabitats (from a few centimeters under the ground up to the height of the collector) which were not explored by the other three methods.

#### 30-min, time-unit sampling

Eight sampling sections were arranged in the park as covering the four major habitats which were sampled by Quadra-Protocol. In each section K. Iwata and Sk. Yamane independently collected ants as many species as possible from every microhabitat (from a few centimeters under the ground up to the height of the collectors) within 30 minutes (i.e., 16 samples were obtained in total). This protocol almost follows Ogata et al. (1998). But they did not designate sampling sections in advance.

# Results

# Ant fauna of "Nanatsujima" Park: a general view

A total of 39 species belonging to 24 genera in five subfamilies were recorded from this park: Proceratiinae (1 species/1 genus), Ponerinae (5 species/3 genera), Myrmicinae (20 species/13 genera), Dolichoderinae (4 species/3 genera) and Formicinae (9 species/4 genera) (see Appendix). Most speciose genera are Camponotus and Tetramorium having 4 species. The number of species per genus is 1.56 on average. Hypoponera nubatama Terayama et Hashimoto, Temnothorax congruus (F. Smith), Tetramorium nipponense Wheeler and Crematogaster matsumurai Forel were collected exclusively by Quadra-Protocol, on the other hand Tetramorium kraepelini Forel, Vollenhovia emeryi Wheeler, Pyramica hexamera (Brown), Tapinoma sp. 3 and Ochetellus glaber (Mayr) exclusively by 30-min Time-Unit Sampling. Among the 39 species Tapinoma melanocephalum (Fabricius) was collected only in complementary samplings.

#### Results from Quadra-protocol

Thirty-three species (22 genera) were recorded from Quadra-Protocol (Table 1). Transect B was the most species- and genus-rich (24 species, 18 genera), while Transect D was the most species- and genus-poor (12 species, 10 genera). Proceratium itoi (Forel) and Tetramorium nipponense were collected from Transect A only; Odontomachus monticola Emery, Aphaenogaster famelica (F. Smith), Monomorium intrudens F. Smith, Vollenhovia benzai Terayama et Kinomura, Crematogaster matsumurai, Crematogaster vagula Wheeler and Formica havashi Terayama et Hashimoto were collected from Transect B only; and Hypoponera nubatama, Temnomyrmex congruus and Oligomyrmex sauteri Forel collected from Transect C only. There was no species collected from Transect D only.

Eighteen species (12 genera) were collected by honey baiting (Table 1). Number of species and genera attracted to honey baits was highest for Transect B (16 species belonging to 11 genera). Crematogaster vagula was collected exclusively by this method. In most trials more than 70% of 45 baits attracted ants, but only 53% attracted ants in the "Ground, Nighttime" trial on Transect A and 29% of baits attracted ants in the "Ground, Nighttime" trial on Transect D. In each trial 2-39% of baits attracted two or more species (Table 2). Most frequently attracted species were Technomyrmex albipes (F. Smith) (194 baits), Pheidole noda F. Smith (119 baits) and Tetramorium bicarinatum (Nylander) (55 baits) (Table 2). Pheidole noda and Tetramorium bicarinatum were collected in all of the 4 transects, while Technomyrmex albipes was not collected in Transect D. Formica japonica Motschoulsky was much more frequent in Transect D than in the other transects. Monomorium chinense Santschi, Solenopsis japonica Wheeler and Tetramorium tsushimae Emery were species collected exclusively from baits on the ground (Pyramica

Table 1. Diversity of ants in Nanatsuijma Park as collected by Ouadra-Protocol. Number of species collected is followed by the number of genera collected (in parentheses).

Transect	A	В	C	D	Gross	
Quadra-Protocol	20 (13)	24 (18)	20 (15)	12 (10)	33 (22)	
Honey baiting	9 (7)	16 (11)	11 (9)	8 (7)	18 (12)	
Gound, daytime	8 (7)	13 (11)	10 (8)	8 (7)	16 (12)	
Ground, nighttime	6 (4)	10 (9)	6 (6)	5 (5)	13 (10)	
Tree, daytime	3 (3)	13 (9)	-	-	14 (9)	
Tree, nighttime	3 (3)	8 (8)	-	-	9 (8)	
Soil sifting	6 (6)	8 (7)	16 (13)	8 (7)	19 (14)	
Litter sifting	11 (10)	16 (13)	14 (12)	-	24 (17)	
Free searching	18 (12)	19 (16)	13 (12)	10 (9)	24 (17)	

membranifera (Emery) was sampled only once on the ground). No species were collected only from the baits on tree trunks (Crematogaster valuga and Camponotus sp. A. were sampled only once on the tree trunk). But, when focusing on Transect A and B, Technomyrmex albipes was more frequently attracted to baits on the tree trunks than those on the ground (99 vs. 52); on the other hand, Pheidole noda was more frequently attracted to baits on the ground than those on the tree trunks (68 vs. 8). Among moderately and frequently attracted species, Monomorium chinense and Formica japonica are diural foragers, while Camponotus japonicus Mayr foraged more actively in the nighttime than in the daytime. Technomyrmex albipes and Pheidole noda foraged almost evenly in the daytime and nighttime for Transect A and B. However, for Transect C the former was more active in the daytime, while the latter was more active in the nighttime.

Nineteen species (14 genera) were collected by soil sifting (Table 1). Hypoponera nubatama was collected only by this method. Number of species and genera collected by this method was highest for Transect C (16 species belonging to 13 genera). Number of samples containing ants was 8 for Transect A, 11 for Transect B and 14 for Transect C and D; and number of species per sample was 0.67 for Transect A, 1.73 for Transect B, 3.73 for Transect C and 2.33 for Transect D. Species frequently collected were Solenopsis japonica (38 samples), Monomorium chinense (23 samples) and *Pheidole noda* (19 samples) (Table 3). Frequency of occurrence of S. japonica was especially high for Transect C and D. S. japonica (1,995 individuals) as well as M. chinense (2,066 individuals) were also numerically prominent.

Twenty-four species (17 genera) were collected by litter sifting (Table 1). Number of species per transect was 11 species for Transect A, 16 species for Transect B and 14 species for Transect C. *Temnothorax congruus* was collected exclusively by this method. The single representative worker of the species had lost legs and was weakened. It is possible that the worker was hunted by other ants outside the surveyed area.

A total of 24 species (17 genera) were collected by free searching (Table 1). Number of species per transect was 18 for Transect A, 19 for Transect B, 13 for Transect C and 10 for Transect D.

# Results from 30-min. time-unit sampling

A total of 34 species (23 genera in five subfamilies) were collected by 30-min. Time-Unit Sampling (16 trials). Number of species per sample ranged from 10 to 17 (13.2 on average). Most frequently encountered species are: *Pachycondyla chinensis* (Emery) (13

samples), Pheidole noda (15), Tetramorium bicarinatum (13), Monomorium chinense (13), Solenopsis japonica (15), Technomyrmex albipes (14), Paratrechina amia (Forel) (13), Paratrechina sakurae (Ito) (11) and Formica japonica (13). Based on these unit samples species accumulation curve and estimators of species richness (ICE, Chao 2, Jack 1, Jack 2, Bootstrap, MMMean) were calculated using the computer package EstimateS 7.0 (Colwell 2004), randomizing the data 100 times. Species richness estimated was 43.33 in ICE, 50.67 in Chao 2, 43.38 in Jack 1, 49.48 in Jack 2, 37.97 in Bootstrap and 35.83 in MMMean. The total species number recorded in the present study (39 spp.) is very close to the values of these estimators.

#### Discussion

## Features of ant fauna of Nanatsujima Park

Species richness in the park (39 species) is nearly 40% of that recorded from the mainland of Kagoshima Prefecture (Yamane et al., 1994), and almost equal to that in the Terayama Park on a hillock (ca. 300 m alt.) in the northern part of Kagoshima City (Kawahara et al., 1999). Kawahara et al. (1999) and Shimono and Yamane (2003) pointed out that forest edges and woody garden are species-rich in comparison with well-developed forests probably due to their habitat mosaic or cline which accepts both forest and open-land inhabitants. Nanatsujima Park also includes a series of artificial habitats from lawn patch to woody patch. Furthermore, it is located between mixed woodland (a possible source of forest inhabitants) and an industrial zone (a possible gateway of tramp and introduced species). These seem to be reasons why Nanatsujima Park has a rich ant fauna.

On the forest edge surveyed by Kawahara et al. (1999) forest-inhabiting species such as *Paratrechina* flavipes (F. Smith) and Crematogaster osakensis Forel were dominant. On the other hand, in Nanatsujima Park Pheidole noda, Monomorium chinense, Solenopsis japonica, Tetramorium bicarinatum and Technomyrmex albipes were frequently collected by honey baiting and/or soil sifting. Excepting Pheidole noda which primarily prefers woody habitats to dry open habitats, they are open-land and/or forest-edge inhabitants. Furthermore, Tetramorium bicarinatum and Technomyrmex albipes are tramp species (Schultz and McGlynn, 2000). Thus, despite the existence of some forest inhabitants such as Proceratium itoi, the ant fauna of Nanatsujima Park strongly reflect environmental conditions derived from urbanization/industrialization.

Ogata et al. (1998) surveyed ant faunas in two types of parks in Fukuoka City, northern Kyushu. Species most frequently found in Momochi Chuou Kouen, a newly established park (10 years old) on a reclaimed land in Hakata Bay, were Tetramorium tsushimae (8/8 samples), Tetramorium bicarinatum (8/8), Paratrechina amia [P. bourbonica (Forel) in their identification] (8/8), Lasius japonicus Santschi (7/8), Monomorium chinense (7/8), Camponotus japonicus (7/8) and Paratrechina sakurae (7/8). Those species are openland inhabitants, and according to Schultz and Mc-Glynn (2000) Tetramorium bicarinatum is a typical tramp species. On the other hand, dominant species in Minami Kouen, a relatively old park on a low hillside densely covered with higher broad-leaved trees, are Paratrechina flavipes (8/8), Aphaenogaster famelica (8/8), Crematogaster osakensis (8/8) and Pheidole noda (7/8). All of them are forest inhabitants or at least species preferring woody habitats. It is remarkable that the faunal features in Nanatsujima Park revealed by Time-Unit Sampling is more close to that in Momochi Chuou Kouen. This may be due to similar historical backgrounds shared by the two reclaimed parks (see "Materials and Methods") as well as environmental conditions common to urban areas in the temperate region. Specices richness in Momochi Chuou Kouen estimated from a species accumulation curve (Ogata et al., 1998, fig. 4) was at most 20 species, almost half of species richness estimated for Nanatsujima Park. This may be due to minor differences in climate (southern Kyushu is a little warmer than northern Kyushu) and in diversity of microhabitats (Nanatsujima Park is more woody than Momochi Chuou Kouen).

# Bionomics of dominant species in Nanatsuiima Park

Quantitative data from the present study have partly elucidated life style of ant species which are dominant in man-made habitats of southern Kyushu.

Hypoponera sauteri Onoyama is common at many places of Japan, and is also known from the Korean Peninsula and Taiwan (Japanese Ant Database Group, 2003). Because it was frequently found in soil samples from Transect C., in urban areas it can establish their permanent population in microhabitats accumulating litter and maintaining moisture. Thus, it may be relatively easily transferred by humans. Although it does not forage actively on the ground (Table 2), if it is introduced, there is a certain possibility that it becomes a competitor of native cryptic ant species.

Monomorium chinense and Solenopsis japonica are dominant in the four major habitats of the park. As ants are social insects, individuals aggregate in nesting sites. Therefore, number of individuals is not a good indicator of dominance. However biomass of the two species is undoubtedly large in open habitats of the park (Table 3). Judging from such habitat preference they have a potential being transferred by humans. Because both of the species usually recruit a huge number of workers to get foods and exclude competitors successfully (Eguchi, 2004), they may have an impact on native biota if they are introduced. Monomorium chinensis is almost diurnal, while Solenopsis japonica forages in the daytime and nighttime (Table 2).

Pheidole noda is eurychoric in the Oriental Region and primarily prefers woody habitats (Japanese Ant Database Group, 2003; Eguchi, pers. comm.). The species usually coexists with Pheidole indica Mayr, which is an open-land inhabitant, at the margin of woody patches (Wakashiba and Yamane, 1994). However, at least during the present survey, *Pheidole* noda is the only member of the genus occurring in the park. Its foraging activity is high on the shaded ground in the daytime and nighttime (Table 2). Temporal segregation in foraging between P. noda and Technomyrmex albipes in hedges (Transect C) may be due to their severe competition: P. noda (a primarily ground forager) frequently encounter T. alpipes (a primarily arboreal forager) in hedges.

Tetramorium bicarinatum, one of the major tramp species widely distributed in the warmer part of Asia and Pacific islands, is relatively dominant in woody patches in Nanatsujima Park. It forages on the ground and vegetation in the daytime and nighttime.

Technomyrmex albipes, one of the major tramp species, is very frequently encountered in Transect A, B and C, but not in D. This indicates that it depends on vegetation, probably due to its close association with aphid colonies (Yamane et al., 1994). It forages intensively on the vegetation and the ground near bush in the daytime and nighttime (Table 2). In the lowland of the Satsuma Peninsula, Kagoshima City may be the northern frontier of T. albipes range; this year (13/iv/2005) we found this species for the first time in the botanical garden of the Korimoto Campus of Kagoshima University (the core area of Kagoshima City).

Formica japonica: This species actively forage on the ground in Transect D, i.e., open habitats. It is a completely diurnal forager (Table 2).

Paratrechina amia is distributed in western and southern Japan, the Ryukyus and Taiwan. Its range has been at least partly expanded by humans (Terayama, 1999). In Kyushu it is a dominant ground forager in urban open habitats, on the other hand Paratrechina flavipes is dominant in woody and more humid habitats (Table 2; Ogata et al., 1998; Kawahara et al., 1999).

**Table 2.** Frequency of occurrence as measured by the number of honey baits attracting ants. A total of 540 baits were set in the present survey.

Transect	A			В			С		D		Gross		
Ground (G) / Tree (T)	(	G	٦	Γ	(	3	7	Γ	(	ĵ	(	3	
Daytime (D) / Nighttime (N)	D	N	D	N	D	N	D	N	D	N	D	N	
attracting ants	33	24	37	38	45	45	44	45	45	44	42	13	455
attracting one species	16	20	36	36	18	30	29	38	32	36	30	10	331
attracting two species	12	2	1	2	17	13	11	7	9	8	11	3	96
attracting three species	4	2	-	-	6	2	4	-	2	-	1	-	21
attracting four species	1	-	-	-	4	-	-	-	2	-	-	-	7
Crematogaster matsumurai	-	-	-	_	1	1	5	4	_	-	-	_	11
Crematogaster vagula	-	-	-	-	-	-	1	-	-	-	-	-	1
Monomorium chinense	10	-	-	-	13	-	-	-	1	-	2	1	27
Pheidole noda	18	13	-	-	22	15	5	3	5	29	2	7	119
Pristomyrmex punctatus	1	-	-	-	2	4	5	6	-	2	-	-	20
Pyramica membranifera	-	-	-	-	-	-	-	-	1	-	-	-	1
Solenopsis japonica	-	-	-	-	6	6	-	-	7	5	10	1	35
Tetramorium bicarinatum	2	4	-	6	8	2	14	10	2	_	3	4	55
Tetramorium tsushimae	-	-	-	-	1	1	-	-	1	1	5	-	9
Technomyrmex albipes	19	10	36	33	10	13	15	15	35	8	-	-	194
Camponotus japonicus	-	-	-	-	6	16	5	10	-	-	1	-	38
Camponotus sp. A	-	-	-	-	-	-	1	_	-	-	-	-	1
Formica japonica	1	_	1	-	2	-	2	-	5	-	25	_	36
Formica hayashi	-	-	-	_	3	-	1	-	_	-	-	-	4
Lasius japonicus	-	-	-	-	4	1	2	1	-	-	-	-	8
Paratrechina amia	-	1	-	1	-	-	2	-	1	-	7	3	15
Paratrechina flavipes	1	1	-	-	8	3	5	3	6	7	-	-	34
Paratrechina sakurae	4	1	1	_	_	-	_	-	-	-	_	-	6

# Protocol for monitoring ants in urban and industrial zones

Recently Yamaguchi (2004) extensively studied influences of urbanization on ant distribution in Tokyo and Chiba (Kanto District, Honshu) under a temperate climate region, and the progression of urbanization reduces ant species richness in urban parks, most likely because it isolates the parks from the surrounding area. On the other hand, urban and industrial zones may act as major gateways for tramp/introduced ant species and harbors of their initial populations. Thus, monitoring ants there is important in order to prevent hazards by introduced ants. However, such monitoring programs may be done with limited man-power, budget and time. Thus, we should take this limitation into consideration while designing protocols.

The methods employed in the present study almost

clear limitation in man-power and budget: most of sampling trials were done by one person; equipment (hand-sifter, white pan, a scoop, forceps, vials, cotton pieces, honey) costs ca. 50 US\$ in total. When results from Ouadra-Protocol and 30-min. Time-Unit Sampling are compared, species composition is a little different between the two protocols, while the numbers of species, genera and subfamilies are almost or completely the same between the two. Eight hours were spent for obtaining 16 samples by 30-min. Time-Unit Sampling, and about 20 hours were spent for surveying one transect by Quadra-Protocol (it means that at least 3 days are needed). Thus, in efficiency measured by species obtained per hour 30-min. Time-Unit Sampling (16 trials) is better than Quadra-Protocol. On the other hand, Quadra-Protocol can gathers quantitative data on life style of dominant species as mentioned above.

In man-made habitats, especially urban areas,

Table 3. Frequency of occurrence as measured by the number of soil samples containing ants. Number of individuals of each species are given in parentheses.

Transect	Α	В	C	D	Gross
containing ants	8	11	14	14	47
containing one species	7	3	1	5	16
containing two species	-	3	3	2	8
containing three species	1	3	2	3	9
containing four species	-	2	1	3	6
containing five species	-	-	4	1	5
containing six species	-	-	2	-	2
containing seven species	-	-	1	-	1
Hypoponera sauteri	1 (1)	1 (1)	8 (88)	-	10 (90)
Hypoponera nubatama	-	-	1 (1)	-	1 (1)
Pachycondyla pilosior	-	-	2 (4)	-	2 (4)
Monomorium chinense	2 (239)	6 (12)	7 (37)	8 (1778)	23 (2066)
Monomorium intrudens	-	1 (1)	-	-	1 (1)
Oligomyrmex sauteri	-	-	1 (1)	-	1 (1)
Pheidole noda	1 (2)	6 (7)	7 (36)	5 (9)	19 (54)
Pristomyrmex punctatus	-	2 (3)	2 (8)	-	4 (11)
Pyramica membranifera	-	-	4 (19)	2 (12)	6 (31)
Solenopsis japonica	3 (75)	8 (264)	14 (569)	13 (1087)	38 (1995)
Strumigenys sp. (cf. lewisi)	-	-	1 (56)	-	1 (56)
Tetramorium bicarinatum	-	-	-	1 (1)	1 (1)
Tetramorium tsushimae	-	-	1 (1)	4 (65)	5 (66)
Technomyrmex albipes	2 (2)	1 (1)	1 (23)	-	4 (26)
Camponotus sp. A	-	-	1 (3)	-	1 (3)
Camponotus nipponicus	-	-	1 (1)	-	1 (1)
Lasius japonicus	-	-	-	1 (1)	1 (1)
Paratrechina flavipes	-	1 (1)	4 (157)	-	5 (158)
Paratrechina sakurae	1 (1)	-	1 (2)	1 (1)	3 (4)

methods extracting soil/litter ants, such as sifting, are often less adoptable because of poor litter accumulation and hardened or paved ground surface. Thus, we here propose a new sampling protocol consisting of Time-Unit Sampling and baiting.

Sampling Protocol for Urban Ants (SPUA) is primarily designed for rapidly surveying ant faunas in urban areas and other man-made open habitats, such as annual-cropping fields. SPUA may be less effective in woody habitats where cryptic litter/soil ants largely contribute to the whole ant species richness. It is strongly desirable that SPUA users have been already trained for observing and collecting ants in the fields to some extent. SPUA consists of the following two methods.

10-min. Time-Unit Sampling: Twenty 10-min. time units of collecting are done by one person within 200+a

minutes (nearly 3.5 hours), or two persons within 100+α minutes (nearly two hours). Ants are visually searched on the ground, in/under shelters (stones, logs, litter, etc.) and on lower vegetation (up to ca. 1.5 m), and collected by forceps (and by fingers). The basic premise of this method is to collect as many ant species as possible within a single 10-minute time unit. Each of the time unit samples is independent, and thus the specimens from a single time unit are kept in a vial which is labeled with its own serial code for the time unit. Ogata (2001) mentioned that, if a researcher is to stay in a target site for 4 hours, a set of 24 x 10 min. samples is better than 8 x 30 min. samples, as long as the unit time is enough for searching under stones, in rotten wood etc. Thus, 10-min. Time-Unit Sampling is adopted in this protocol.

Bait Attracting (Cheese Eaiting): Thirty baits are placed on the ground with 4 to 5 m intervals, and inspected continuously during an hour by one person. The food item at each bait station is supplied before being exhausted by foraging activities of ants. Ant species visiting each station are collected and kept in a vial which is labelled with its own serial code for the station. As an option ant behaviors on/around baits are recorded by the second person (if present). Possible food items are divided into two categories, "sugar-rich" and "protein-rich". By using sugar-rich (honey+banana) and protein-rich (tuna) baits Hahn and Wheeler (2002) showed that a greater percentage of arboreal species preferred protein-rich baits and a greater percentage of terrestrial species preferred sugar-rich baits. This strongly advises us to use the same food item or food items of the same category in a series of surveys. Honey (20% honey solution) is one of the possible baits: honey bait is accepted by a major portion of ground and arboreal dwelling ants (Hashimoto et al., 1997), and the material is available in markets over the world. Processed cheese is a possible protein-rich item. Powdered cheese is useful when ones follow foragers carrying cheese particles and find out their nests (Eguchi et al., 2004), but in open habitats powdered cheese may be swept out by strong wind. Thus, solid cheese may be better than powdered cheese in such cases. Incidentally, it is worth noting that recruitment pattern of Pheidole indica Mayr is different between powdered and solid cheese (Magata and Yamane, 1989). SPUA adopting cheese baiting (powdered cheese) has been already done in Java (Hasin et al., in prep.) and in the northern Ryukyus (Takahashi, in prep), and is now conducted by T. V. Bui (Institute of Ecology and Biological Resources, Hanoi) and K. Eguchi in N. Vietnam.

In SPUA 4.5 hours may be required for one person, or three hours for two persons, for getting data set. If time and/or man power is sufficient, it is also possible to conduct optional surveys on ants in litter/soil in hedges by "Tirne-Unit Sifting", and/or on ant activities/behaviors, such as foraging distance and nesting site (see Eguchi et al. 2004). On the other hand, baiting can be omitted if time is insufficient, because Time-Unit Sampling may cover almost all species which are collected by baiting.

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# Appendix: Species Recorded in the Present Study

Species recorded are listed below. Abbreviations are: HGD, honey baiting on the ground in the daytime; HGN, honey baiting on the ground in the nighttime; HTD, honey baiting on the tree trunk in the daytime; HTN, honey baiting on the tree trunk in the nighttime; S, soil sifting; L, litter sifting; FS, free searching; TUSmi, 30-min. Time-Unit Sampling by K. Iwata; TUSsky, 30-min. Time-Unit Sampling by Sk. Yamane; Trans. A, Transect A; Trans. B, Transect B; Trans. C, Transect C; Trans. D, Transect D. Serial code for each bait or sampling unit is given in parentheses following the abbreviation of the sampling method.

Samplings were done in the following schedule in 1999: HGD and HTD in Trans. A on 22<sup>nd</sup> May; HGN and HTN in Trans. A on 29<sup>th</sup> May; S in Trans. A on 4<sup>th</sup>, 5<sup>th</sup> and 9<sup>th</sup> June; L in Trans. A on 25<sup>th</sup> May; FS in Trans. A on 28<sup>th</sup> May; HGD and HTD in Trans. B on 6<sup>th</sup> June: HGN and HTN in Trans. B on 30<sup>th</sup>

June; S in Trans. B on 10<sup>th</sup>, 12<sup>th</sup>, 15<sup>th</sup> and 16<sup>th</sup> July; L in Trans. B on 15<sup>th</sup> and 16<sup>th</sup> July; FS in Trans. B on 22<sup>nd</sup> July; HGD in Trans. C on 27<sup>th</sup> Aug; HGN in Trans. C on 19<sup>th</sup> July; S in Trans. C on 4<sup>th</sup>, 9<sup>th</sup>, 10<sup>th</sup> and 14<sup>th</sup> Aug; L in Trans. C on 22<sup>nd</sup> Aug; FS in Trans. C on 28<sup>th</sup> Aug; HGD and FS in Trans. D on 6<sup>th</sup> Oct; HGN in Trans. D on 26<sup>th</sup> Sept; S in Trans. D on 17<sup>th</sup>, 19<sup>th</sup>, 25<sup>th</sup>, 26<sup>th</sup>, 27<sup>th</sup> and 29<sup>th</sup> Sept, and 4<sup>th</sup> and 9<sup>th</sup> Oct.

#### PROCERATIINAE (1 species/1 genus)

Proceratium itoi (Forel) Trans. A: L-(b). TUSsky-(6, 7).

## PONERINAE (5 species/3 genera)

- Hypoponera nubatama Terayama et Hashimoto Trans. C: S-(b5).
- Hypoponera sauteri Onoyama Trans. A: S-(b5). Trans. B: S-(c1); L-(b). Trans. C: S-(a1, a2, a3, a4, a5, b1, b2, c5). TUSmi-(3, 4, 6, 7); TUSsky-(2, 4, 5, 6).
- Odontomachus monticola Emery Trans. B: FS-(b). TUSmi-(2, 4, 7); TUSsky-(4).
- Pachycondyla chinensis (Emery) Trans. A: FS-(c). Trans. B: L-(a, b); FS-(a, b). TUSmi-(1, 2, 3, 5, 6, 7); TUSsky-(1, 2, 3, 5, 6, 7, 8).
- Pachycondyla pilosior (Wheeler) Trans. A: FS-(a). Trans. C: S-(a4, b4); L-(b); FS-(b). TUSmi-(4, 7).

#### MYRMICINAE (20 species/13 genera)

- Aphaenogaster famelica (F. Smith) Trans. B: FS-(c). TUSmi-(1).
- Crematogaster matsumurai Forel Trans. B: HGD-(27); HGN-(28); HTD-(26, 27, 28, 29, 43); HTN-(26, 27, 28, 43); FS-(b, c). TUSmi-(2, 8); TUSsky-(1, 2, 3, 8).
- Crematogaster vagula Wheeler Trans. B: HTD-(45). Messor aciculatus (F. Smith) Trans. A: FS-(a). Trans. B: FS-(c). TUSsky-(1).
- Monomorium chinense Santschi Trans. A: HGD-(2, 5, 8, 10, 15, 19, 26, 30, 34, 41); S-(a5, c3); L-(a, b); FS-(a, b, c). Trans. B: HGD-(2, 7, 26, 28, 29, 30, 31, 33, 34, 36, 41, 42, 44); S-(a1, a2, b1, b2, b3, b5); L-(a, b, c); FS-(b, c). Trans. C: HGD-(44); S-(a1, a4, a5, b1, b2, b4, c5); L-(b, c); FS-(a, b, c). Trans. D: HGD-(21, 33); HGN-(19); S-(a1, a4, a5, b3, b4, b5, c1, c2); FS-(b, c). TUSmi-(1, 2, 3, 4, 5, 6, 7); TUSsky-(1, 2, 3, 4, 5, 7).
- Monomorium intrudens F. Smith Trans. B: S-(b3); L-(b). TUSmi-(3).
- Oligomyrmex sauteri Forel Trans. C: S-(a5); L-(b). TUSsky-(4).
- Pheidole noda F. Smith Trans. A: HGD-(6, 7, 8, 18, 19, 21, 22, 25, 26, 29, 30, 31, 33, 34, 35, 37, 42, 43); HGN-(3, 10, 12, 18, 19, 22, 23, 25, 26, 27, 29, 37, 42); S-(b5); L-(a, b, c); FS-(a, b, c). Trans. B: HGD-(2, 4, 5, 6, 7, 8, 18, 19, 21, 25, 27, 28, 31, 32, 33, 34, 35, 40, 41, 42, 43, 44); HGN-(5, 8, 9, 19, 21, 26, 27, 28, 29, 30, 33, 34, 43, 44, 45); HTD-(18, 23, 32, 41, 45); HTN-(32, 34. 45); S-(a3, a5, b2, b4, b5, c4); L-(a, b, c); FS-(a, b, c) Trans. C: HGD-(3, 5, 6, 44, 45); HGN-(1, 2, 4, 5, 6, 7, 8, 9, 11, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23 25, 26, 27, 31, 40, 41, 43, 44, 45); S-(a1,a2, a3, a4, b1

b2, c5); L-(a, b, c); FS-(a, b, c). Trans. D: HGD-(17, 18); HGN-(11, 12, 13, 14, 15, 30, 33); S-(a3, b1, b4, b5, c2); FS-(b, c). TUSmi-(1, 2, 3, 4, 5, 6, 7, 8); TUSsky-(1, 2, 3, 4, 5, 6, 7).

Pristomyrmex punctatus (F. Smith) Trans. A: HGD-(22); L-(a, b, c); FS-(a, b, c). Trans. B: HGD-(16, 42); HGN-(16, 35, 40, 42); HTD-(16, 21, 35, 40, 42); HTN-(16, 21, 35, 37, 40, 42); S-(b3, b4); L-(b, c); FS-(b, c). Trans. C: HGN-(24, 41); S-(b4, c1); L-(c); FS-(b, c). Trans. D: FS-(c). TUSmi-(1, 5, 7, 8); TUSsky-(2, 3, 6, 8).

Pyramica hexamera (Brown) TUSmi-(3).

Pyramica membranifera (Einery) Trans. C: HGD-(14); S-(a3, a4, b5, c4); L-(a); FS-(a). Trans. D: S-(b3, b4). TUSmi-(4)

Solenopsis japonica Wheeler Trans. A: S-(b4, b5, c4); L-(b, c); FS-(c). Trans. B: HGD-(4, 21, 22, 23, 25, 43); HGN-(21, 22, 23, 24, 25, 26); S-(a1, a2, a5, b1, b2, b3, b4, b5); L-(b, c); FS-(b, c). Trans. C: HGD-(33, 34, 35, 36, 37, 38, 39); HGN-(32, 34, 35, 36, 37); S-(a1, a2, a3, a4, a5, b1, b2, b4, b5, c1, c2, c3, c4, c5); L-(a, b, c); FS-(a, c). Trans. D: HGD-(8, 9, 13, 15, 35, 38, 39, 40, 41, 42); HGN-(21); S-(a1, a4, a5, b1, b2, b3, b4, b5, c1, c2, c3, c4, c5); FS-(a, c). TUSmi-(1, 2, 3, 4, 5, 6, 7, 8); TUSsky-(1, 3, 4, 5, 6, 7, §).

Strumigenys sp. (cf. lewisi Cameron) Trans. B: L-(a); FS-(a). Trans. C: S-(b2); L-(b); FS-(b). TUSsky-(3, 4, 6). Temnothorax congruus (F. Smith) Trans. C: L-(c).

Tetramorium bicarinatum (Nylander) Trans. A: HGD-(41, 45); HGN-(29, 33, 43, 45); HTN-(8, 38, 39, 42, 44, 45); FS-(a, b, c). Trans. B: HGD-(17, 19, 20, 29, 34, 38, 39, 40); HGN-(17, 2)); HTD-(17, 19, 20, 21, 22, 23, 24, 25, 32, 33, 35, 39, 40, 44); HTN-(17, 18, 19, 20, 22, 23, 24, 25, 38, 44); L-(b, c); FS-(a, b, c). Trans. C: HGD-(3, 5); L-(b). Trans. D: HGD-(19, 26, 45); HGN-(12, 13, 24, 26); S-(b5); FS-(a, b, c). TUSmi-(1, 2, 3, 4, 5, 7, 8); TUSsky-(1, 2, 3, 5, 6, 8).

Tetramorium kraepelini Forel TUSsky-(8).

Tetramorium nipponense Wheeler Trans. A: FS-(a).

Tetramorium tsushimae Emery Trans. A: L-(b, c); FS-(a, b, c). Trans. B: HGD-(45); HGN-(37); L-(c); FS-(c). Trans. C: HGD-(43); HGN-(10); S-(c5); FS-(c). Trans. D: HGD-(3, 29, 30, 44, 45); S-(a1, b1, c1, c2); FS-(b, c). TUSmi-(1, 8); TUSsky-(1, 2, 5, 6, 7).

Vollenhovia benzai Terayama et Kinomura Trans. B: L-(c). TUSsky-(1, 4).

Vollenhovia emeryi Wheeler TUSsky-(2).

## DOLICHODERINAE (4 species/3 genera)

Ochetellus glaber (Mayr) TUSmi-(8).

Tapinoma melanocephalum (Fabricius) Only from complementary sampling.

Tapinoma sp. 3 TUSsky-(2).

Technomyrmex albipes (F. Smith) Trans. A: HGD-(1, 2, 4, 6, 8, 9, 10, 12, 15, 16, 17, 23, 24, 29, 33, 36, 41, 42, 43); HGN-(4, 16, 17, 20, 22, 29, 32, 34, 36, 41); HTD-(2, 3, 4, 5, 6, 7, 8, 9, 10, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 28, 30, 31, 32, 33, 34, 35, 36, 37, 41, 42, 43, 45); HTN-(1, 2, 3, 4, 5, 6, 9,

10, 13, 14, 15, 16, 17, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 30, 31, 32, 33, 34, 35, 36, 37, 42, 43); S-(b1, b2); L-(a, b, c); FS-(a, b, c). Trans. B: HGD-(1, 3, 6, 9, 10, 11, 12, 13, 14, 15); HGN-(1, 2, 3, 4, 6, 7, 9, 10, 11, 12, 13, 14, 15); HTD-(1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15); HTN-(1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15); S-(a2); L-(a); FS-(a). Trans. C: HGD-(1, 4, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 35, 36, 37, 40, 41, 42); HGN-(3, 27, 28, 29, 30, 38, 39, 42); S-(b1); L-(a, b, c); FS-(a, b, c). TUSmi-(1, 3, 4, 5, 6, 7, 8); TUSsky-(1, 3, 4, 5, 6, 7, 8).

#### FORMICINAE (9 species/4 genera)

Camponotus (Camponotus) japonicus Mayr Trans. A: L-(b); FS-(c). Trans. B: HGD-(27, 28, 29, 30, 38, 42); HGN-(18, 19, 24, 27, 28, 29, 30, 31, 32, 33, 34, 35, 38, 39, 41, 43); HTD-(29, 30, 31, 38, 39); HTN-(21, 27, 29, 30, 31, 33, 34, 38, 39, 41); FS-(b, c). Trans. D: HGD-(16); FS-(b, c). TUSmi-(1, 3, 5, 6, 8); TUSsky-(2, 3).

Camponotus (Colobopsis) nipponicus Wheeler Trans. A: FS-(b). Trans. C: S-(c5). TUSmi-(6); TUSsky-(2, 4, 5, 6).

Camponotus sp. A Trans. A: L-(a); FS-(a, b, c). Trans. B: HTD-(26); L-(b); FS-(b, c). Trans. C: S-(c2); FS-(a, c). TUSmi-(2, 3, 5, 8); TUSsky-(2, 3, 6). Remarks: Possibly two species of the subgenus Myrmamblys, C. (M.) bishamon and C. (M.) vitiosus, are included here.

Formica hayashi Terayama et Hashimoto Trans. B: HGD-(9, 10, 34); HTD-(28); L-(c). TUSmi-(1, 2); TUSsky-(1, 2, 4).

Formica japonica Motschoulsky Trans. A: HGD-(45); HTD-(45); L-(a, c); FS-(a, c). Trans. B: HGD-(17, 25); HTD-(10, 27); FS-(b, c). Trans. C: HGD-(3, 5, 33, 34, 37); FS-(a, c). Trans. D: HGD-(1, 2, 3, 4, 5, 6, 7, 11, 12, 13, 14, 15, 17, 19, 20, 21, 23, 24, 25, 27, 29, 34, 36, 43, 44); FS-(a, b, c). TUSmi-(1, 3, 4, 5, 6, 7, 8); TUSsky-(1, 3, 5, 6, 7, 8).

Lasius japonicus Santschi Trans. B: HGD-(24, 35, 36, 37); HGN-(36); HTD-(34, 37); HTN-(36); FS-(b, c). Trans.
D: S-(a1); FS-(a). TUSmi-(1, 2, 3, 8); TUSsky-(1, 3, 8).

Paratrechina amia (Forel) Trans. A: HGN-(20); HTN-(19); FS-(b, c). Trans. B: HTD-(26, 33); L-(e); FS-(a, b, c). Trans. C: HGD-(45); L-(e); FS-(b, c). Trans. D: HGD-(18, 24, 28, 29, 30, 31, 32); HGN-(21, 22, 42); FS-(a, b, c). TUSmi-(1, 2, 3, 4, 5, 6, 7, 8); TUSsky-(1, 2, 5, 7, 8).

Paratrechina flavipes (F. Smith) Trans. A: HGD-(22); HGN-(22); FS-(b). Trans. B: HGD-(2, 4, 8, 11, 19, 25, 28, 40); HGN-(27, 41, 45); HTD-(19, 20, 27, 28, 35); HTN-(9, 18, 28); S-(b2); L-(a, b); FS-(a, b, c). Trans. C: HGD-(2, 3, 5, 6, 12, 24); HGN-(2, 3, 7, 9, 10, 12, 19); S-(a1, a3, a5, b2); L-(a, b); FS-(a, b). TUSmi-(1, 3, 4); TUSsky-(1, 2, 3, 4, 6).

Paratrechina sakurae (Ito) Trans. A: HGD-(8, 19, 30, 37); HGN-(19); HTD-(29); S-(b3); L-(b, c); FS-(a, b, c). Trans. C: S-(c5); L-(b, c). Trans. D: S-(b5). TUSmi-(1, 3, 5, 6, 7, 8); TUSsky-(2, 3, 5, 6, 7).