Article

How to design an inventory method for ground-level ants in tropical forests.

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Abstract

To better clarify the optimal method of ground level ant inventory in tropical forests, sampling was conducted in a rain forest and lower montane forest in Sabah, Borneo using four methods: hand collecting, leaf litter sifting, soil core sampling, and honey bait trapping. Among the four methods, soil core sampling yielded the highest number of both total and unique species at both sites. Moreover, this method showed a high accumulation of species number with sampling efforts, as measured by either the number of samples or length of transect. However, to provide an adequate representation of the total ant fauna in a site, a combination of methods was preferable. Based on the results here, the combination of soil core sampling with leaf litter sifting and hand collecting is recommended for ground-level ant inventory in tropical forests. Their combined use was an efficient way to collect many species in a short time.

Key words: ants, inventory, sampling methods, tropical forest, Borneo

Introduction

Species inventory of ants has been viewed as an important task in tropical biodiversity and conservation studies (Agosti et al., 2000). Ants are one of the most abundant and diverse animal groups in tropical ecosystems (Stork, 1987, 1991), and they function at many levels in these ecosystems-as predators and prey, as detritivores, mutualists, and herbivores (Hölldobler and Wilson, 1990). Because of their ecological importance, ants have the potential to yield more meaningful biodiversity data than many other organisms, such as plants, birds, and butterflies. Moreover, ants have a potential role as indicators of environmental change since most species have stationary, perennial nests with fairly restricted foraging ranges. Thus, information from the ant inventory is indispensable to understand and monitor the ecosystems in tropical forests.

The choice of sampling methods is of critical importance in order to obtain useful information from an inventory. The primary objectives of an inventory are to record as many of the species present at a site as possible and to characterize community patterns in a site. Thus, one should choose a sampling method that can collect as many species as possible and provide unbiased representation of the local community. For an ant inventory, such an optimal method has not been universally agreed upon, and very few studies have addressed the question of how to seek out the optimal inventory method. For example, Agosti et al. (2000) presented the mini-Winkler extractor as a standard sampling method for ant inventory. However, because this method collects only ground-dwelling ants, it provides an inadequate representation of the total ant fauna. In this study, to better clarify the optimal method of ground and lower vegetation level ant inventory in tropical forests, inventories using each of four sampling methods-hand collecting, leaf litter sifting, soil core sampling, and honey bait trapping-were conducted in two different forest types in Sabah, Borneo. Based on the data from these inventories, each of the four methods was evaluated with respect to the number of species collected, sampling efficiency, and community characterization. Our final goal is to determine an optimal method for ground-level ant inventory in tropical forests, using few methods and minimal sampling effort, that would provide adequate results for estimating the species number and characterizing the total ant fauna at a site.

Materials and Methods

Study sites

The inventories using the four methods were tested in two different types of tropical forest in Sabah, Borneo. The first was a lowland rainforest (30-100 m alt.) in Tawau Hills Park (TRF), and the other is a lower mountain forest (around 1,000 m alt.) in Sayap, Kinabalu Park (SMF).

Sampling methods

In each forest, one transect was set, 180 m long and subdivided into three 60 m sampling sections. Each sampling section was sampled by the four different methods, hand collecting (HC), leaf litter sifting (LS), soil core sampling (SC), and honey bait trap (BT). The methods involved the following procedures:

HC: This method accessed the lower vegetation, logs, rocks, and ground surface of the forest. Ants were picked up using forceps or an aspirator. This procedure was carried out for 30 minutes per sampling section.

LS: The leaf litter was gathered up, sifted, and sorted on a white pan and ants collected from it. Such sampling was repeated for 30 minutes per sampling section.

SC: Five soil cores, each 20 x 20 x 15 cm deep, were taken at equal intervals along the transect in each sampling section. These soil cores were sifted using a hand sieve and white pan, and the ants collected.

BT: Fifteen baits with 20-30% honey solution were set on the forest floor at four-meter intervals along the transect in each sampling section. The same number of baits was set in both daytime (9:00 to 11:00) and nighttime (18:00 to 20:00). The ants attracted to the baits were collected with forceps.

Results

Species number

The four sampling methods yielded a total of 123 ant species at TRF, and 77 ant species at SMF. The number of species collected by each method is shown in Table 1. In both study sites, SC provided the largest total number of species (63 at TRF, 36 at SMF), followed by LS (47, 33), BT (35, 20), and HC (26, 18). However, the mean number of species collected per section was slightly greater in LS (16.0 ± 1.0) than in SC (13.7 ± 11.7) in SMF. In terms of the number of species sampled uniquely by each method, SC also recorded the largest numbers at both study sites (37 at TRF, 25 at SMF), followed by LS (22, 14), HC (19, 10) and BT (12, 4).

Species composition

The species collected at TRF belonged to five subfamilies and 40 genera, and at SMF five subfamilies and 38 genera. Of the five subfamilies, Myrmicinae, Ponerinae, and Formicinae were dominant in terms of species number at both sites. Although these subfamilies were sampled by all the methods, the proportions varied significantly among samples (Fig. 1). Species of Ponerinae were sampled most successfully by SC at both sites compared with the other

Table 1. Number of ant species collected from each of three sections (S1, S2 and S3) of the transects at the Tawau rain forest and Sayap lower montane forest, using the four methods.
HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

Tawau	HC	LS	SC	BT	Total
S1	10	17	26	18	58
S2	12	28	33	22	78
S3	16	23	28	19	67
Total (unique)	26 (19)	47 (22)	63 (37)	35 (12)	123
Mean±SD	12.7 ± 3.1	22.7 ± 5.5	29.0 ± 3.6	19.7 ± 2.1	

Sayap	HC	LS	SC	BT	Total
S1	5	15	27	16	48
S2	8	17	9	12	40
S3	12	16	5	9	35
Total (unique)	18 (10)	33 (14)	36 (25)	20 (4)	77
Mean±SD	8.3 ± 3.5	16.0 ± 1.0	13.7 ± 11.7	12.3 ± 3.5	

methods ($X^2 = 5.66$, $P < 0.05$ at TRF; $X^2 = 5.16$, $P < 0.05$ at SMF). Species of Formicinae were sampled most by HC at both sites ($X^2 = 12.38$, $P < 0.01$ at TRF; $X^2 = 9.33$, $P < 0.01$ at SMF). Species of Myrmicinae were sampled most by BT at TRF ($X^2 = 8.73$, $P < 0.01$).

The proportion of genera sampled by each method is shown in Fig. 2. The tests of equality revealed that the four methods showed significant heterogeneity in the composition of genera sampled at both study sites ($X^2 = 4.81$, $P = 0.18$ at TRF; $X^2 = 2.37$, $P = 0.49$ at SMF). *Hypoponera, Pheidole, Polyrhachis*, and *Paratrechina* were the dominant genera at both sites. The proportion of species belonging to these genera varied among sampling methods. Species of *Polyrhachis* were collected only by HC at both sites. Those of *Hypoponera* were collected only by LS and SC in both sites. *Pheidole* was recorded by all methods, but species of the genus were sampled most effectively by BT at both sites ($X^2 = 3.94$ $P < 0.05$ at TRF; $X^2 = 3.83$, $P < 0.05$ at SMF). Species of *Paratrechina* were sampled most effectively by BT at TRF ($X^2 = 5.42$, $P < 0.05$).

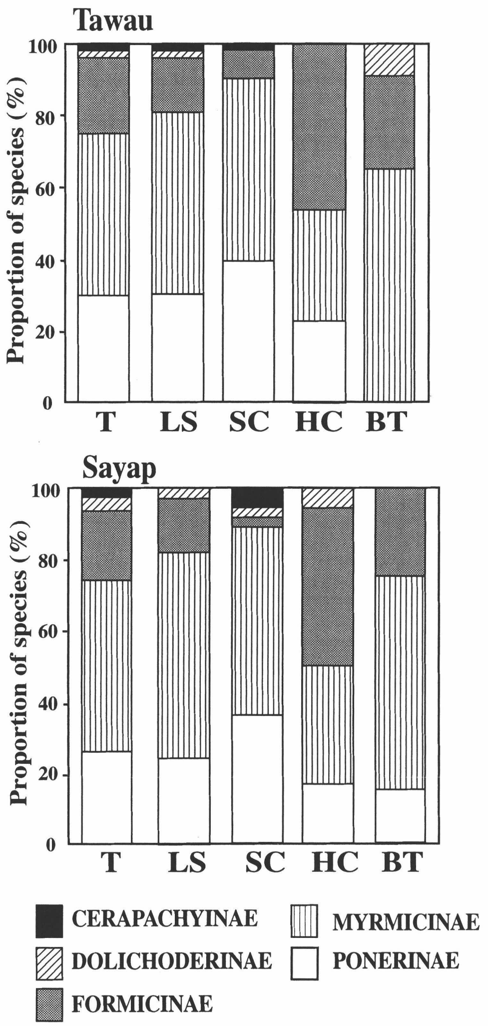

Fig. 1. Proportion of ant species belonging to different subfamilies in the four sampling methods. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap, T: all methods combined.

Complementarity of methods

A matrix of complementarity for the species lists sampled by the four methods is shown in Table 2. Complementarity (dissimilarity) between methods was derived using the measure "*C*" proposed by Colwell and Coddington (1994). *C* is the number of mismatches between two species lists divided by the total number of species in both lists. Thus, *C* varies from 0 (when the samples are identical) to 1 (when the samples are completely distinct). The highest values of *C* were generally those in comparisons between HC and other methods, especially between HC and SC (0.97 at TRF, 0.98 at SMF). On the other hand, the lowest values were between BT and the other methods, especially between BT and LS (0.78 at TRF, 0.74 at SMF).

For the BT method, complementarity between daytime and nighttime samples was calculated. The

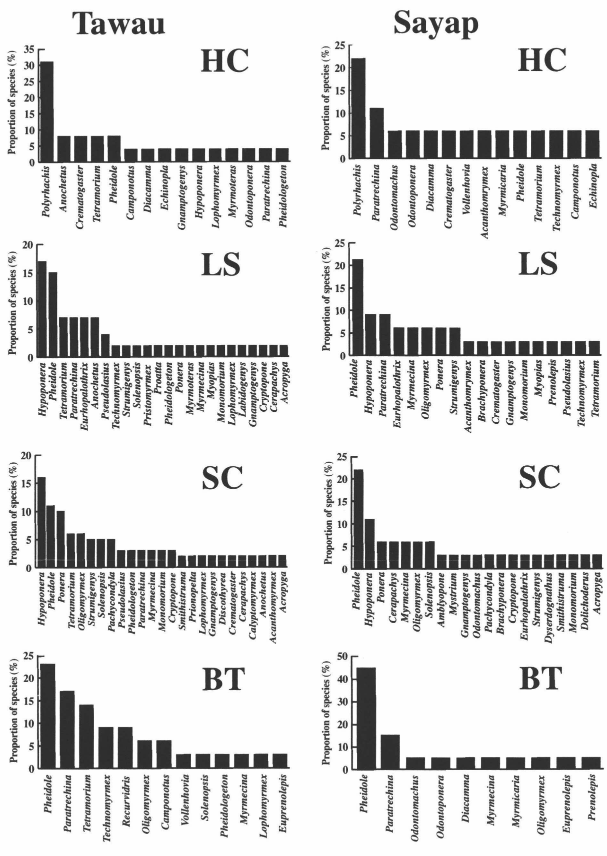

Fig. 2. Proportion of ant species belonging to different genera in the four sampling methods. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

Table 2. Complementarity between the four methods. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

Tawau

	BT	SC	LS
HC	0.87	0.97	0.96
LS	0.78	0.76	
SC	0.80		

Sayap

	BT	SC	LS
HC	0.90	0.98	0.87
LS	0.74	0.89	
SC	0.81		

value of *C* was 0.60 at TRF and 0.45 at SMF, which indicated quite high species overlap between the daytime and nighttime collections at both sites.

Sampling effectiveness of methods

The cumulative number of species was plotted against the number of sampling sections (length of transects) in each method (Fig. 3). In both sites, SC showed the greatest increase of species with successive sampling sections, followed by LS, BT and HC.

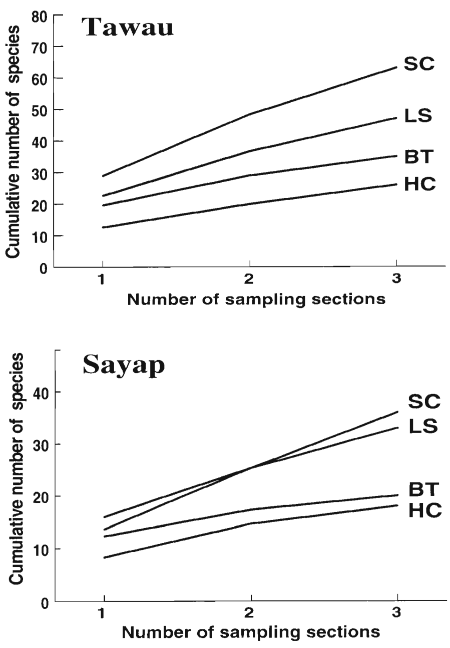

Fig. 3. Cumulative number of species plotted against number of sampling sections for the four methods pooled. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

For SC and BT, the large number of replicates meant that the species accumulation curve against the number of samples could be plotted, and their effectiveness compared by the steepness of the resulting curve. Fig. 4 shows that the species accumulation curves were much steeper in SC than BT, at both sites. This indicates that SC would be expected to collect more species by adding new samples, whereas BT is expected to collect few or no species through additional sampling.

BT samples can be partitioned into those from daytime and nighttime collecting. To estimate sampling effectiveness at each time of collection, the species accumulation curves were plotted for daytime and nighttime samples (Fig. 4). The species accumulation curves showed that one method is not markedly more productive in species number to sample number than the other. The curves were quite similar to the curve of daytime and nighttime combined.

Combination of methods

Table 3 lists the combinations of two and three methods that produced the five highest numbers of species at both sites. SC was an element in all such combinations, and SC combined with LS and HC produced the maximum number of species (111 species

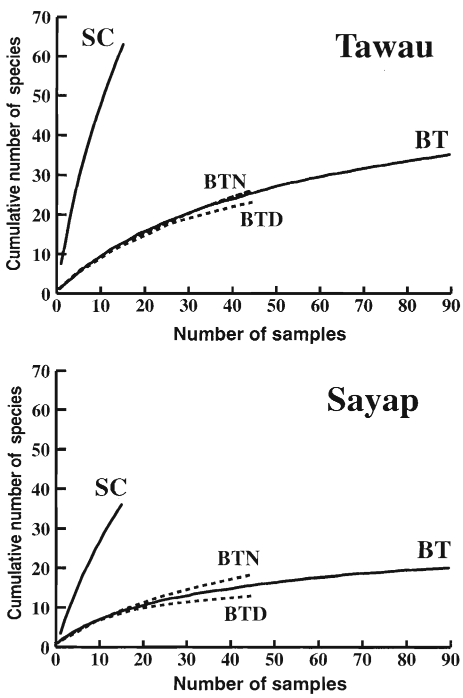

Fig. 4. Cumulative number of species plotted against number of samples pooled for the SC and BT. SC: soil core sampling, BT: honey bait trap, BTD: daytime collecting of BT, BTN: nighttime collecting of BT.

at TRF and 73 species at SMF) among two- and three-method combinations. At each site, these values represented more than 90% of the number of species collected by all methods combined. SC+LS+HC also collected the highest number of genera among two- and three-method combinations (38 genera at TRF and 37 genera at SMF).

Table 4 lists overall complementarities for species lists sampled by each combination of three methods. The overall complementarity (*OC*) was derived using modification of IBD (Koch, 1957), which is calculated as

$$OC = 1-(\sum_{j=1}^{3} Sj-St)/(3-1)\ St$$

where St is total number of species sampled and Sj is number of species in each sample. Thus, when the three samples are identical, *OC* equals zero, and when the samples are completely distinct, with no overlap, *OC* equals 1. At both sites, the highest values of *OC* were for SC+LS+HC and SC+HC+BT, and lowest values were from LS+BT+SC and LS+BT+HC.

Table 4. Overall complementarities (*OC*) of three method combinations. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

Tawau

Combination of Methods	OC
SC+HC+BT	0.41
SC+HC+LS	0.40
SC+LS+BT	0.34
LS+HC+BT	0.32

Sayap

Combination of Methods	OC
SC+HC+LS	0.39
SC+HC+BT	0.39
LS+HC+BT	0.37
SC+LS+BT	0.30

Discussion

The results presented here indicate that SC was the most efficient method used in this study for sampling ant species. SC yielded the highest number of both total and unique species at both sites. Furthermore, SC showed a high increase in species number as the number of samples increased. Although the sampling cost of the different methods is not explicitly compared, the time requirement was approximately equal by each of the four methods used in this study. Therefore, SC was the best way to collect the largest possible number of species by a single method in a short period of time.

Table 3. Combinations of two and three methods that produced the five highest number of ant species. HC: hand collecting, LS: leaf litter sifting, SC: soil core sampling, BT: honey bait trap.

Tawau

Combination of Methods	Number of Species	Number of Genera
SC+LS+HC	111	38
SC+HC+BT	104	36
SC+LS+BT	101	36
SC+LS	89	34
SC+HC	86	35

Sayap

Combination of Methods	Number of Species	Number of Genera
SC+LS+HC	73	37
SC+HC+BT	67	36
SC+LS+BT	63	35
SC+LS	61	34
SC+HC	53	34

However, to provide an adequate representation of the total ant fauna in a site, the use of more than one method is recommended. The present study showed that samples of the four methods had very low species overlap (i.e., high complementarity), and high heterogeneity in genus and subfamily composition. This suggests that each of the four methods collected different components of the ant fauna community. Thus, an assessment of the ant fauna using a single method would overestimate or underestimate the relative richness of a particular group of ants. For example, compared with the other methods BT tends to collect a large proportion of species of Myrmicinae, such as *Pheidole*. An inventory using this method alone would result in overestimating the proportion of Myrmicinae in a site. Therefore, for a more unbiased record of ground-level ants in a site, a combination of methods is required.

Based on the results here, the combination of SC with LS and HC is recommended for ground-level ant inventory in tropical forests. The number of species collected by this three-method combination represented more than 90% of the number of species collected by SC+LS+HC+BT (i.e., all methods combined) at both sites. BT revealed the lowest production of species number against sampling efforts. This, and the low complementarity between BT and the other methods, indicated that the use of BT was not beneficial in terms of the species number. Thus, additional effort with SC+LS+HC, rather than adding BT to the three methods, is the most efficient way to collect as many of the species as possible. Furthermore, SC+LS+HC was the set of methods used to collect the most genera, with high overall complementarity of the sample, at both sites. This reflects the fact that HC, LS, and SC were originally designed to collect ants from different strata in forests, such as from lower vegetation, litter, and the soil. In tropical forests, strong partitioning of the ant fauna among the different strata has been reported (Rosciszewski, 1995; Brühl et al., 1998). Therefore, the use of these methods in combination was found to be the best way to record the total fauna of ground and lower-vegetation level ants in tropical forests.

As shown in this study, the best approach to designing an inventory protocol is to identify the method that sampled the most number of species, and then to combine one and two complementary sampling procedures with this method. Such a minimal combination of methods will ensure a relatively complete representation of the fauna with less sampling effort. Thus, the above combination of methods can be used as a standardized protocol for comparisons between different habitats as well as for longer-term monitoring, to provide useful data in biodiversity and conservation studies.

Acknowledgements

We wish to acknowledge the staff of Sabah Parks for their support during our research in Tawau. We also deeply thank Katsuyuki Eguchi, Nordin Hj Wahid, and Zainal Awang, Arlina Abdullah for their assistance in enabling us to work in Sayap. This work was partly supported by a Grant-in-aid for Creative Basic Research No. 09NP1501 from the Ministry of Education, Science, Sports and Culture, Japan.

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Received: January 17, 2001
Accepted: March 21, 2001