

- Mispagel, M.E. 1981. Relation of oxygen consumption to size and temperature in desert arthropods. *Ecol. Entomol.* 6: 423-431.
- Sen-Sarma, P. K. 1964. The effect of temperature and relative humidity on the longevity of psuedoworkers of *Kaloterme flavicollis* (Fabr.) (Isoptera) understarvation condition. *Proc. Natl. Inst. Sci. India (B)* 30: 300-314.
- Smith, J. L. 1992. Effect of abiotic factors on subterranean termite behavior and control strategies. Ph.D.dissertation, University of California Riverside,
- Smith, J. L. & M. K. Rust 1990. Tunneling response and mortality of the western subterranean termite (Isoptera: Rhinotermitidae) to soil treated with termiticides. *J. Econ. Entomol.* 83: 1395-1401.
- Smythe, R. V. 1972. Feeding and survival at constant temperatures by normally and abnormally faunated *Reticulitermes virginicus* (Isoptera: Rhinotermitidae). *Ann. Entomol. Soc. Amer.* 65: 756-757.
- Smythe, R. V. & L. H. Williams 1972. Feeding and survival of two subterranean termite species at constant temperatures. *Ann. Entomol. Soc. Amer.* 65: 226-229.
- Steward, R. C. 1983. The effects of humidity, temperature and acclimation on the feeding, water balance and reproduction of dry-wood termites (*Cryptotermes*). *Entomol. experimen. appl.* 33: 135-144.
- Su, N.-Y., & R. H. Scheffrahn 1986. A method to access, trap, and monitor field populations of the Formosan subterranean termite (Isoptera: Rhinotermitidae) in the urban environment. *Sociobiology* 12: 299-304.
- Su, N.-Y. & R. H. Scheffrahn 1990. Comparison of eleven soil termiticides against the Formosan subterranean termite and eastern subterranean termite (Isoptera Rhinotermitidae). *J. Econ. Entomol.* 83: 1918-1924.
- Tamashiro, M., J. R. Yates & R. H. Ebesu 1987. The Formosan subterranean in Hawaii: problems and control, pp. 15-22. *In* M. Tamashiro & N.-Y. Su [eds.], *Biology and control of the Formosan subterranean termite. Proceedings of the International Symposium on the Formosan Subterranean Termite, 67th Meeting of the Pacific Branch Entomol. Soc. Amer., Res. Ext. Ser. 083, University of Hawaii, Honolulu.*



## Ethology of the Robber Stingless Bee, *Lestrimelitta limao* (Hymenoptera: Apidae)

by

Shôichi F. Sakagami<sup>1</sup>, David W. Roubik<sup>2</sup>, & Ronaldo Zucchi<sup>3</sup>

### ABSTRACT

Over 90 raids by colonies of robber stingless bees *Lestrimelitta limao* were observed in Brazil and Panama, in most cases involving a known robber colony or observation hive. Potential hosts included at least 39 native stingless bee species and Africanized *Apis mellifera*. In total, 17 species hosted raids. We report here studies made of the robbing process focusing on both host and parasite. Robbing colonies removed brood cell provisions, some honey, wax and resin, and pollen from meliponine nests, seldom exterminating colonies. Queens were not attacked, while provisions in brood cells were totally removed; honey was pillaged from honeybees and caused colony absconding. Preferred hosts had long, cerumen entrance tubes, moderate to large colony size, high quality brood cell provisions and were no larger than *L. limao*. *Scaptotrigona* and *Nannotrigona* resisted *L. limao* relatively strongly and were frequent hosts but always retreated if robbers successfully invaded. Adult bee mortality during raids usually was slight. Masses of immobile host adults remained near pupae and prepupae, left untouched by robbers. Host bees suddenly resumed activity and nest defense near the termination of raids, as robbers gradually withdrew.

Guarding *L. limao* were unaggressive at their own nest, but during raids formed a ring around the host nest entrance and defended it from all other insects. Guard-ring robbers occasionally elevated the abdomen and exposed a whitish intersegmental region between hind terga while fanning the wings. Except at the start or end of raids, when host and robber fought, there was no discernible release of citral odor from robber mandibular glands. No robber odor was noted inside nests through most of the raid. When raids were artificially interrupted by release of *L. limao* mandibular gland substances, robbers quickly departed and host bees arrived, killing robbers that remained. Repeating this experiment did not always produce the same result. Complex timing in the release of host and robber chemicals likely control the robbing

<sup>1</sup>Institute of Low Temperature Science, Hokkaido University, Sapporo 060, Japan; present address: Ainosato 1-6, 2-2-610; Kitaku, Sapporo 002, Japan

<sup>2</sup>Smithsonian Tropical Research Institute, APDO 2072, Balboa, Panama, or Unit 0948 APO AA 34002-0948, USA

<sup>3</sup>Faculty of Philosophy, Sciences and Letters, University of São Paulo 14040, Ribeirão Preto, Brazil

process.

Host nests were occupied by *Lestrimelitta* for up to 5 days but usually only one day for less than 4 hrs; multiple colonies were also raided simultaneously. Raids occurred more frequently in wet season when flower abundance and colony food stores were low; thus raid frequency was inversely related to potential harvest per raid. Mass raids incorporated up to 600 bees while mild raids for nest material were made by only a few bees. Large groups arrived at host nests following lone scouts and small groups. Throughout a raid, workers flew in an ellipse at the *L. limao* nest entrance. Raiding bees were guards, collectors of building material, or food collectors, appearing not to specialize according to age.

**KEY WORDS:** social bees, stingless bees, robbing behavior, *Lestrimelitta*, nest usurpation, tropical bee communities, Africanized honeybee

## INTRODUCTION

Evolution of flower visiting habits in bees led to storage of nectar and pollen in their nests as provisions for larvae. This microbially processed and enriched food attracts many types of predators and parasites, but few are so specialized and prevalent as parasitic bees (Bohart 1970; Michener 1974; Wcislo 1987; Roubik 1989). Obligate robbers, sometimes termed "cleptobiotic" bees, are tropical and eusocial, consisting of at least 7 neotropical *Lestrimelitta* and one African *Cleptotrigona* (Sakagami & Laroca 1963; Wille 1979; Camargo & Moure 1989; Ayala 1992). The robbing syndrome arose independently in these 2 lineages, indicated most by divergent genitalic characters (Michener 1990). All species but *L. limao* F. Smith apparently have restricted distributions, although up to 3 are sympatric (Camargo & Moure 1989). We have determined, by examining males, that we studied a single species which ranges at least between Panama and southern Brazil, corresponding to *L. limao* as interpreted by Schwarz (1948). Nonetheless, the ethology of robbing bees remains little studied due to rarity of the parasite and difficulties in observing colonies. Only 7 nests of *Lestrimelitta* were located in 3 community-wide surveys producing 507 nests of 54 meliponine species (Hubbell & Johnson 1977; Roubik 1979, 1983). Robber bees may usurp entire host nests but more commonly, as reported herein, selectively harvest from host colonies without exterminating them. In both honeybees and stingless bees, robbing seems fairly common, with the enigmatic exception of Asian stingless bees (Nogueira-Neto 1949; Michener 1975; Dyer & Seeley 1987, and personal observations). Certain ants rob food from other ant species as it is carried back to the nest (Wilson 1971). Such ants have been termed cleptobiotic but do not usurp the host nest or remove its food. Robber bees therefore appear unique among bees and the social insects.

Whereas occasional pillaging behavior has been shown for unspecialized

robbing stingless bees of the genera *Oxytrigona*, *Scaptotrigona* and *Melipona* (Roubik 1989), *Lestrimelitta* has never been seen at flowers and its anatomy, behavior and chemical ecology strongly confirm its specialized existence as an obligate robbing parasite of other stingless bee colonies. Raiding behavior in this genus has been repeatedly observed, but never analyzed in detail in natural forest communities or with the use of observation hives (Michener 1946; Nogueira-Neto 1950; Kerr 1951; Moure *et al.* 1958; Wille 1961; Sakagami & Laroca 1963; Nogueira-Neto 1970; Laroca & Orth 1984). Workers of *L. limao* use large cephalic gland reservoirs of citral and its chemical isomers, apparently to recruit nest mates for raids (Cruz 1962; Cruz-Landim 1967; Blum 1966; Blum *et al.* 1970). All pillaged food is carried internally in the crop, the bees possess a notably elongated gut, and they lack a corbicula on the hind leg, where pollen is ordinarily carried (Cruz-Landim & Rodrigues 1967).

Since a report by Kerr (1951) it has been suggested that the strong lemon odor emitted by *L. limao* repels the bees of victim colonies so that raids are performed with little resistance. Only slight damage supposedly results to either host or parasite. However, stingless bees such as *Trigona* (*Tetragonisca*) *angustula* Illiger respond to odors of *L. limao* with intense colony defensive behavior (Wittmann 1985; Wittmann *et al.* 1990). Stingless bee species that cannot effectively repel *Lestrimelitta* or otherwise diminish its raids are likely to have behavior for reducing net losses to the parasite. Nogueira-Neto (1970) proposed that odors emitted by the host bees might produce this result. These hypotheses are evaluated in light of new data presented here. Our principal objective is to outline the ethology of *Lestrimelitta limao* and a portion of its natural history as the most widely distributed and best known robber bee.

## METHODS

### 1) Studies in Brazil

Observations were made during winter at a subtropical locality, Ribeirão Preto, at 21°S. latitude. When temperatures drop to 5° bee activity starts well after bees open their nest entrance. Brazilian *L. limao* were studied in a colony domiciled in a glass-topped observation hive (Sakagami 1966). The nest and colony were collected at Aragarías in the state of Goiás and were observed in a meliponary on the Ribeirão Preto campus of the University of São Paulo, during 2 July to 21 October, 1971. A vinyl pipe corridor through the wall of the building in which the colony was kept provided a nest entrance tube. Only one colony of *Lestrimelitta* existed at this site. In this area there were approximately 75 colonies of 17 species of other stingless bees, and about 50 hives of Africanized *Apis mellifera* Latreille, the feral Neotropical honeybees still interbred occasionally with imported European apiary bees (Ruttner 1988;

Roubik 1989; Roubik *et al.* in press).

Inspections of the robber colony and its activity were made daily for 2 months at Ribeirão Preto. Bursts of activity signalled a raid. Meliponary colonies were checked to confirm which, if any, hosted a raid. On some occasions *Lestrimelitta* pillaged colonies outside the meliponary, indicated by the return of bees carrying cerumen nesting material on their hind tibiae (Fig. 1). In many instances, however, raided bee colonies were observed (Fig. 1), and the attacks had begun 5 to 10min prior to detection.

The host colony was observed intermittently until withdrawal of the raiding bees. In addition to observing robber behavior and hosts outside of the nest, the lids of hived host colonies were occasionally removed or replaced with a sheet of glass to record intranest behavior. Robbers sometimes invaded colonies in glass-topped observation hives, allowing continuous observation of the entire raiding process.

2) Studies in Panama

In Panama, near sea level and at 9°N. latitude, diurnal bee foraging activity begins at least by 06:20 throughout the year. A pronounced dry season lasts from January until April or May. The driest month is March and rainfall is heavy in October and May (Rand & Rand 1982). Four colonies of *Lestrimelitta* were maintained in sections of tree trunks in which they were found and 4 natural nesting colonies were observed in lowland forest. Seven of these colonies were found in black mangrove forest near the Pacific Panama Canal, or in the lowland wet forest 60km east of Panama City (Roubik 1983). Four natural colonies in trees, and 3 raids of *L. limao* on the nests of other species, were observed in Soberanía National Park, north of Gamboa within the Canal area, at the above field sites, and also on Barro Colorado Island (BCI), a nature preserve maintained by the Smithsonian Tropical Research Institute. Other raids, constituting most of those in the study, were noted during brief daily inspections of stingless bee nests during 1982 to 1992 at meliponaries kept in a residential area approximately 150m from mixed secondary and remnant primary lowland forest at Curundu, on the Pacific side of the Panama Canal area. Meliponaries contained from 40 to 60 colonies of 15 to 30 species, most in natural log nests found in lowland forests (Roubik 1983). The type and number of colonies changed from month to month as new colonies were brought in or removed. Some were maintained in wooden hives. Five different colonies of *L. limao* were kept for observation at different times. Field observations of colonies were made during all but 3 years between 1980 and 1992. Experiments performed at the meliponary during raids are described below in the Results section. Curundu raids, and those at field sites, resulted from the activity of natural colonies.

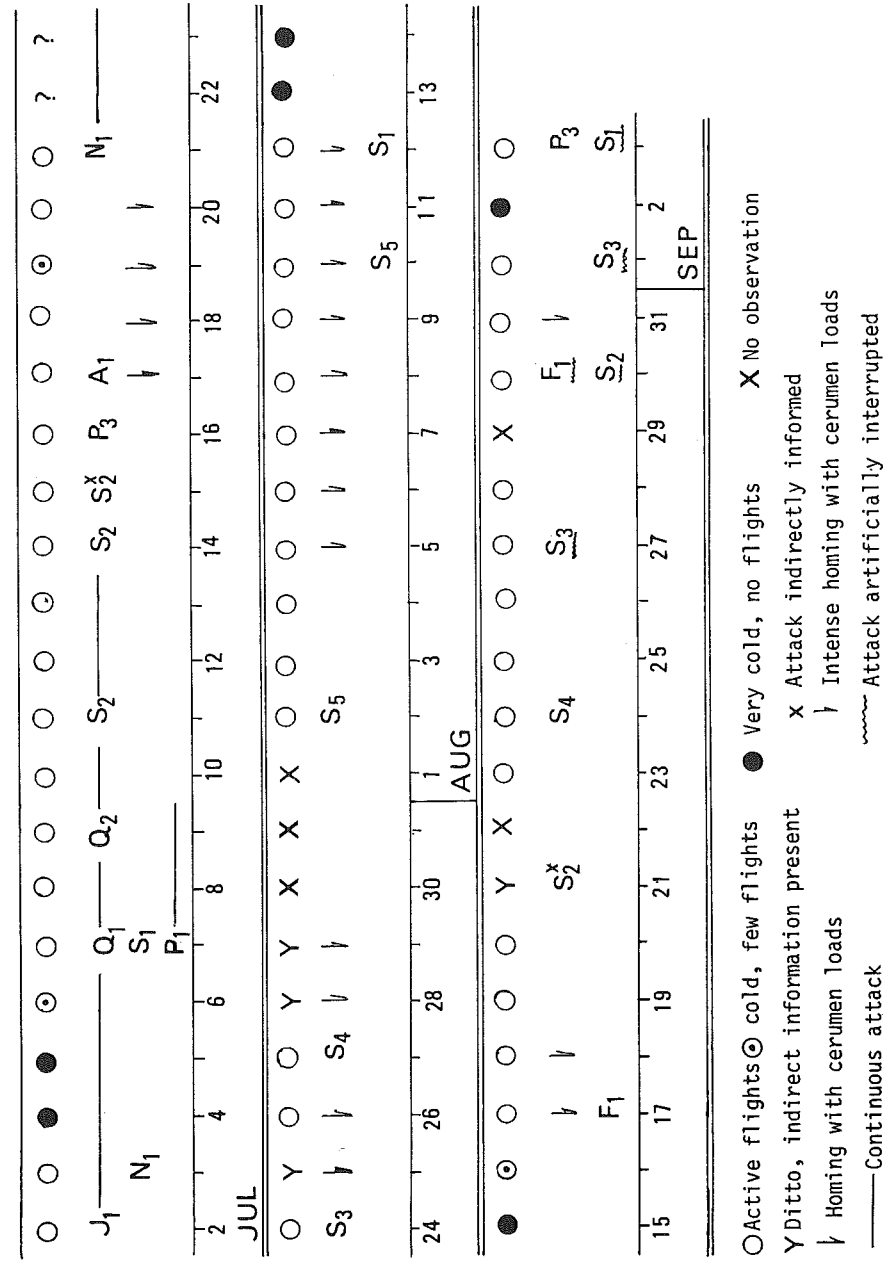


Fig. 1. Temporal distribution of raiding activity of *Lestrimelitta limao*, and of raids on host colonies under indicated weather conditions, during 2 July, 1971 to 3 September, 1971 at Ribeirão Preto, Brazil.

## RESULTS

## 1) Activity at Nest Entrances — Guarding

Despite their aggressive robbing behavior, workers of *L. limao* were unaggressive toward observers or insects approaching their nests. The Brazilian nest was constantly visited by honeybee workers and also by stingless bees from the meliponary, such as *Scaptotrigona postica* Latreille, *S. bipunctata* Lepeletier, *Melipona bicolor schencki* Gribodo, *M. quadrifasciata* Lepeletier, *M. rufiventris* Lepeletier and *Trigona spinipes* Fabricius (Table 1 & Fig. 1). Each collected cerumen from the soft protuberances of the nest entrance (Fig. 2B). Guarding *Lestrimelitta* exhibited no response besides extending the antennae toward these bees. Tubes were sometimes built by worker *Lestrimelitta* upon entrances of colonies being raided, but the natural nest entrance is usually larger, projecting up to 20 or 30cm from the nest of *Lestrimelitta* (Schwarz 1948: 79; Wille & Michener 1973; Fig. 2). Colony debris ejected from nests of *Lestrimelitta* accumulated under the entrance (Fig. 2A,C) and was often visited by workers of the stingless bee *Partamona* in Panama, which collected some of the pellets. These lone workers were not attacked by guard *Lestrimelitta*. Nest entrances always displayed several pliable, blind protuberances (Fig. 2A). Their apices were rounded but their shape and disposition changed. In the evening the bees closed the entrance by applying soft cerumen across the tip. When the entrance was open, it was guarded by up to a few dozen workers that positioned their heads along the inner rim (Fig. 2C). The Brazilian colony contained some individually marked workers that guarded the nest entrance for periods of 2-3 hours.

## 2) Flight Behavior

Departure from the nest was different from that of other stingless bees. They followed their predecessors for about 30-50cm from the entrance, forming a horizontal line of bees which then spread away in a funnel-like form. Groups leaving nests observed on BCI and Curundu repeatedly traced a horizontal ellipse of approximately 80cm by flying down and away from the nest entrance, largely facing the entrance during their retreat, looping up to return to the entrance, nearly touching it, then repeating the downward loop. Occasionally a circulating bee entered the nest. This activity continued as long as a raid was conducted. After mass departure, returning foragers waiting in flight quickly entered the nest. During sustained periods of high activity during pillaging, which we refer to below as a "mass raid" numbers of departing bees increased abruptly. For the Brazilian colony, 120 to 150 bees were recorded leaving the nest during 30 seconds. Departures and flight of bees flying in the ellipse near the nest entrance accelerated at these times.

Foraging by *Lestrimelitta* often began between 10:00 and 12:00 (24 of 36

Table 1. Relationship between number of colonies present and frequency of raids at meliponaries in Brazil and Panama.

Host species	No. colonies (A)	No. raids (B)	No. Colonies Raided	B/A
<i>Plebeia</i> sp.	2	2	1	1.00
<i>T. (Tetragonisca) angustula</i>	7	1**	1	0.14
<i>Plebeia droryana</i>	7	3	3	0.43
<i>Scaptotrigona postica</i>	9	14	3	1.55
<i>Melipona quadrifasciata</i>	12	2	2	0.17
<i>Nannotrigona testaceicornis</i>	13	2	2	0.15
<i>Apis mellifera</i>	68	1	1	0.02
<i>Scaptotrigona pectoralis</i>	3	6	3	2.33
<i>S. barrocoloradensis</i>	11	31	11	2.81
<i>S. luteipennis</i>	1	1	1	1.00
<i>Nannotrigona perilampoides</i>	3	3	2	1.00
<i>Paratrigona omaticeps</i>	3	4	3	1.33
<i>Lestrimelitta limao</i>	3	3*	3	1.00
<i>Plebeia (Scaura) latitarsis</i>	2	1	2	0.50
<i>Frieseomelitta nigra</i>	3	1*	1	0.34
<i>T. (Tetragonisca) angustula</i>	8	2*	2	0.25
<i>Melipona eburnea</i>	20	4	3	0.20

\*colonies exterminated by robbers. \*\*Species not raided and their colony numbers were: *Celetrigona longicornis* Friese 3), *Leurotrigona mulleri* Moure 2), *Frieseomelitta varia* 3), *Geotrigona* sp. 1), *Partamona cupira* 1), *Trigona hyalinata* Lepeletier 1), *Trigona spinipes* 1) *Melipona marginata* Lepeletier 1), *Melipona bicolor* 1), *Tetragona dorsalis* Smith 5), *Dolichotrigona longitarsus* Ducke 3), *Trigonisca atomaria* Cockerell 1), *Trigonisca* sp. 3), *Paratrigona lophocoryphe* Moure 1), *Plebeia frontalis* Friese 6), *Plebeia jatiformis* Cockerell 3), *Trigona (Cephalotrigona) zexmeniae* Cockerell 4), *Partamona* aff. *cupira* 1), *Geotrigona subgrisea* Cockerell 1), *Melipona micheneri* Schwarz 2), *Melipona fuliginosa* Lepeletier 2), *Melipona compressipes triplaris* Cockerell 3), *Trigona nigerrima* Cresson 6), *Trigona muzoensis* 1), *Trigona fulviventris* Guerin 1). Ribeirão Preto colonies were present for the duration of the study while those from Curundu were present for at least 6 months (see text).

days, range 09:00 to 15:00) during the cool season from July until August in Ribeirão Preto, which prevented earlier activity. In Panama the earliest raid started before 8:15.

## 3) Kinds of Raids — Mass Raids

Raids seldom did permanent damage a host colony, but mass raids could exterminate the host colony or, in the case of honeybees, make the colony abandon its nest (Roubik 1980, 1991). A mass raid is depicted in Fig. 3. When the raiding group arrived, returning host foragers avoided the nest entrance and hovered near it or retreated to nearby vegetation. Robber bees entered the nest and spread to the brood area containing provisioned cells with eggs or larvae, and also moved to the food storage pots to begin removal of their contents and building material. Host bees moved to the portion of the brood comb containing prepupae and pupae.

Mass raids were observed from their beginning at nests of *Melipona*

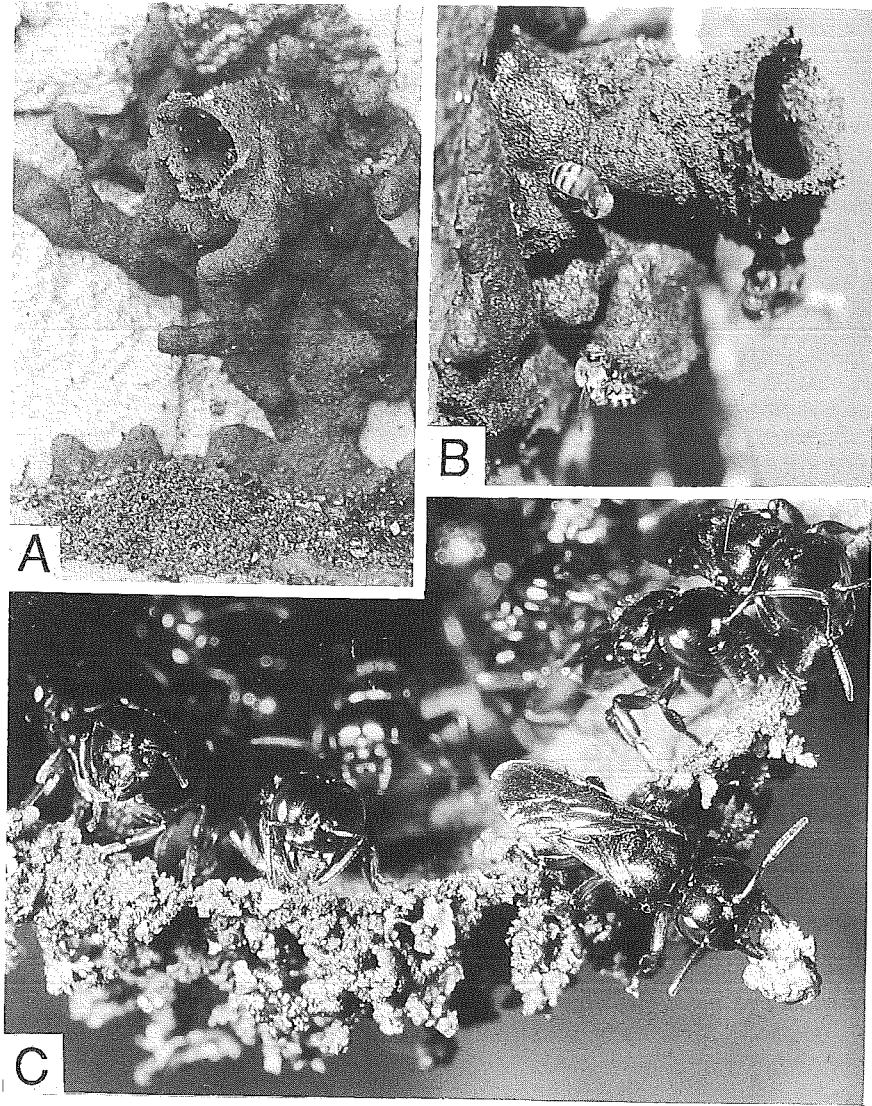


Fig. 2. Nest entrances of *Lestrimelitta limao*. A) entrance of observation colony in Brazil; note accumulated colony trash below. B) entrance showing honeybees collecting cerumen, guard bees and bee ejecting trash pellet from inside of natural nest in Panama.

*quadrifasciata* and *Scaptotrigona postica* in Ribeirão Preto. The former nest was just 10m from the nest of *L. limao*. A single *L. limao* was observed by chance at the hive entrance platform of *Melipona*. It was followed almost instantly by several dozen nest mates, which assumed a threatening posture by lowering the forebody, raising the abdomen, and fanning the wings

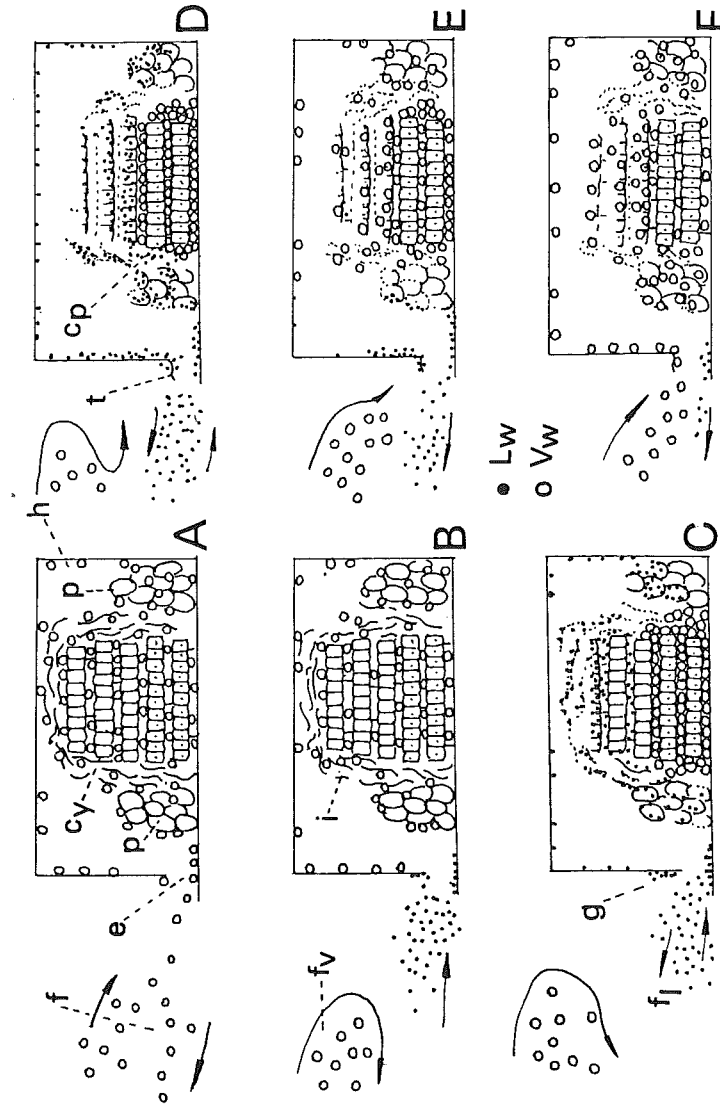


Fig. 3. Schematic sequence of a mass raid by *Lestrimelitta limao*. The sequence includes: A) host nest before mass raid; B) host nest at initiation of mass raid and occupancy of host nest in upper brood combs and food storage pots, D) peak of raid, in which provisioned brood cells and storage pots are pillaged and much of their cerumen removed, E) near termination of raid, when most robbers have departed and most host workers still are inactive, F) termination of the raid, when few *Lestrimelitta* remain at the nest entrance, host foragers are entering the nest, and host workers are resuming activity. LW = worker *Lestrimelitta*, Vw = host workers, h = hive entrance, e = storage pots of honey and pollen, cy = combs of young larvae or eggs, i = involucrum, cp = combs of post-feeding prepupae and pupae, f = returning foragers, fv = returning host foragers after raid initiation, fl = foragers of *Lestrimelitta*. Arrows indicate forager flight direction.

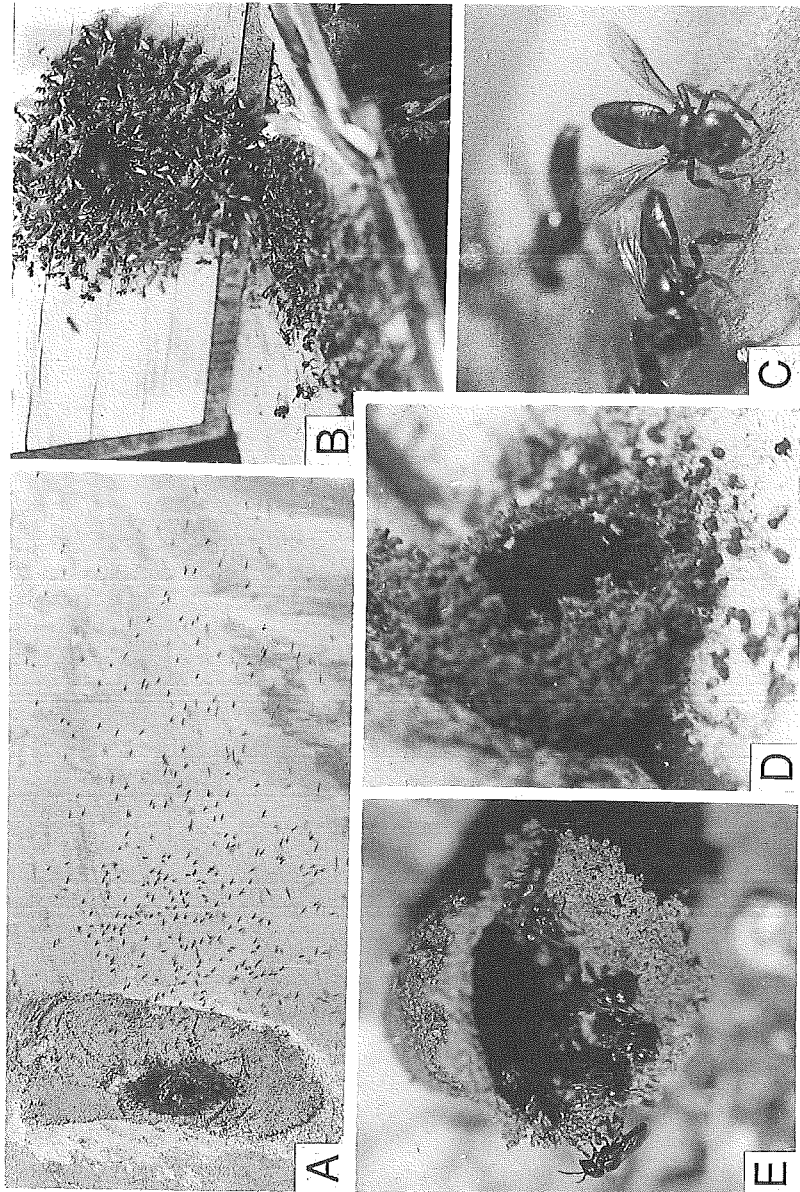


Fig. 4. Mass raid initiation and preparation of an entrance tube on the nest of *Scaptotrigona postica*. A) Mass raid group arriving at nest entrance, B) guard ring formed around host nest entrance and on platform below hive, C) magnified view of B), showing guards alighting and displaying threat posture, D) nest entrance of *Lestrimelitta* newly formed on the host nest entrance, E) guarding of the completed robber nest entrance at host nest entrance.

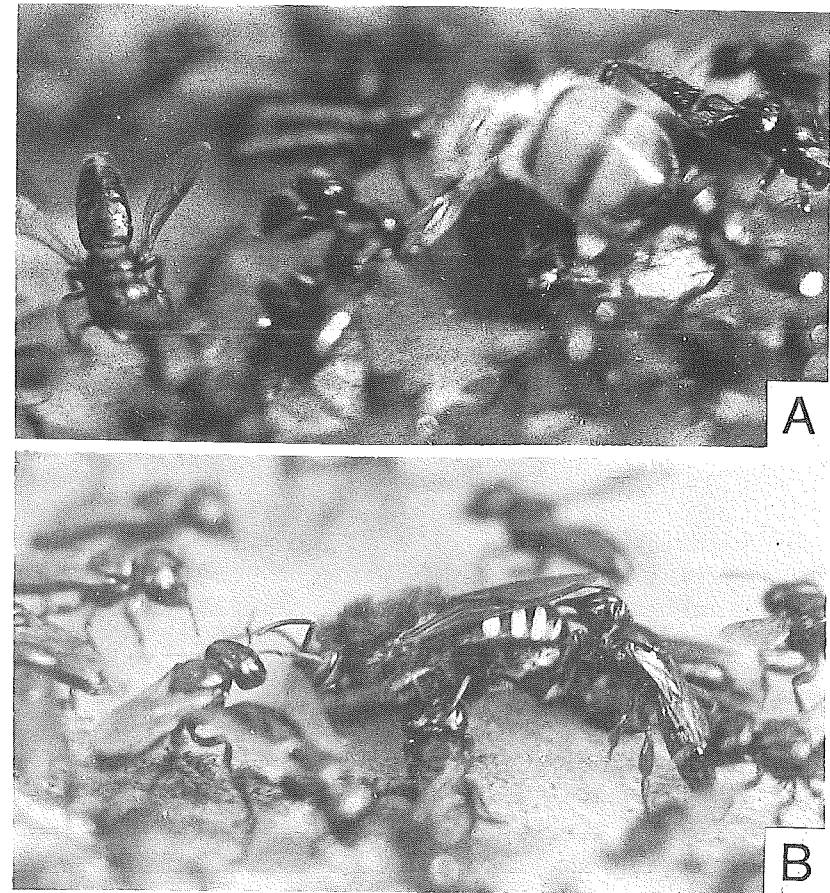


Fig. 5. Raiding *Lestrimelitta limao* attacking A) worker Africanized honeybee, and B) worker *Melipona quadrifasciata*.

(Fig. 4C). A strong citral odor was evident around the nest entrance. About 50 worker *Melipona* streamed from the entrance, each opening the mandibles and raising the abdomen, but they retreated when harassed by *Lestrimelitta*. The entrance platform became fully occupied by *L. limao*, as *Melipona* workers either retreated into the nest or were killed (Fig. 5B). During single combat *L. limao* primarily bit the mandibles of the larger *Melipona* (Fig. 6B,C). Joint attacks also occurred by 3 or 4 *Lestrimelitta*, some of which also locked their mandibles on the legs of *Melipona*. Three minutes after the raid began no *Melipona* was near the outside entrance; about 50 returning foragers flew around the nest but did not land. After another 2min the lid of the hive was opened, showing that no *Lestrimelitta* had passed through the internal nest entrance tube into the brood and food storage area. Invasion of the inner nest

started 25min after the raid began, at which time all returning *Melipona* foragers flew away and many more *Lestrimelitta* arrived. The brood cells containing small larvae or eggs on the top of the comb stack were opened and emptied by pillaging bees. In 10min this comb was removed; guarding *Lestrimelitta* attacked returning host foragers outside the nest.

The mass raid of *Scaptotrigona postica* in an observation hive began when many *Lestrimelitta* arrived in rapid succession and passed through the vinyl hive entrance tube. After 5min the entrance was surrounded by a dense aggregation of guarding *Lestrimelitta* (Fig. 4A,B). Shortly after the first few *Lestrimelitta* entered the nest, many spread throughout the inside of the hive, each displaying threat posture. The host workers rushed about within the nest, but without resisting the invading bees. Slight removal of the glass lid released a strong citral odor from within the observation hive. Ten minutes after the raid's start most of the remaining *Scaptotrigona* were beneath the combs of the brood nest, forming immobile masses (Fig. 3C-E). However, citral odor had been replaced by the strong mandibular gland odor of *Scaptotrigona*.

That a mass raid was initiated by a single worker, followed by arrival of a small group and later a larger group, was confirmed by observations in Panama. Two observations were made at wooden hives containing *Scaptotrigona pectoralis* Dallatorre and *Melipona eburnea panamica* Cockerell (known formerly as *M. fasciata panamica*, DWR, unpublished data). One raid of *Melipona eburnea* was initiated as a single worker of *L. limao* landed at the nest entrance, followed within seconds by the arrival of approximately 60 workers. At this point the hive was replaced by an empty hive of the same type, from which a colony recently had been removed. After another 4min a large group of *Lestrimelitta* entered the hive. The entrance was closed and 584 *Lestrimelitta* were later counted in the hive. The numbers of *L. limao* surrounding the nest entrance during the initial stages of other raids were approximately 500, 600, 100, 80 and 50 workers at 3 natural nests of *Scaptotrigona* and 3 of *Nannotrigona*, respectively. These bees assembled around the entrance when the smell of citral was strong in the air, and presumably were observed there shortly after the raid began. At the nests of *Scaptotrigona* having several hundred robbers at their entrances, a few dozen worker *Scaptotrigona* were seen crawling away from the nest on the ground, as if repelled and incapacitated by the raiding bees. An observation hive of *Scaptotrigona pectoralis* was opened 10min after a raid was initiated by a large group of *Lestrimelitta*. Worker *Lestrimelitta* moved throughout the brood area and food storage pots in the hive while a small cluster of *Scaptotrigona*, including the queen, had moved from the brood area and was relatively immobile in a corner of the nest. During this brief interval no fighting occurred between bees.

#### 4) Exterminating Raids and Competition Among Robbers

Raids resulted in host colony deaths on some occasions, after which the nest was either usurped or left unoccupied. *Lestrimelitta* killed a colony of *Plebeia (Scaura) latitarsis* Friese during a single raid. The colony was in its natural nesting niche, an active *Nasutitermes* termite nest. Small colonies of *Plebeia*, such as *P. franki* Friese, 1 colony of which was exterminated in a single raid of *Lestrimelitta* on BCI, show that the robber bee could not possibly occupy a host nest. Cavity nesting sites of these hosts are smaller in volume by at least an order of magnitude than those of *L. limao* (Roubik 1983).

Facultative robbing was seen in *Trigona (Tetragonisca) angustula*, which interacted aggressively with conspecific colonies and those of other species, including *Lestrimelitta*. Nests of *T. angustula* were removed from the Ribeirão Preto site after many *Lestrimelitta* were killed by this species the day the robber bee colony was installed. An exterminating raid took place only once during the 2mo study at Ribeirão Preto, when a colony of *T. angustula* succumbed after a single raid by *Lestrimelitta*, but its nest was not occupied thereafter. This species counter-attacked a colony of *L. limao* in Panama, and within 2wks abandoned its nest and occupied that of *Lestrimelitta*. The exterminated *Tetragonisca* colony had been attacked at the same time as the conspecific colony which later exterminated *Lestrimelitta*. At the end of the former raid, approximately 400 dead *T. angustula* and 50 *L. limao* were found outside the nest. Phorid flies then invaded, completely destroying the *T. angustula* colony. At the same site, another raid by *Lestrimelitta* on a natural nest of *T. angustula* in a log was successful; the *L. limao* abandoned the nest 2 days after the mass raid and no *Tetragonisca* appeared at the nest entrance until 9 days later. *T. angustula* also attacked a conspecific colony that was established in the former nest of *L. limao*. Natural colonies of *Melipona favosa* Fabricius, *M. eburnea*, *Nannotrigona perilampoides* Cresson and *Scaptotrigona barrocoloradensis* Schwarz were usurped by *T. angustula* in Panama. This was evident from host nest entrance tubes under those of *T. angustula* and by direct observation of usurpation. This suggests competition with *Lestrimelitta*. Intraspecific aggression seen among natural colonies of *L. limao* will be addressed in a companion paper.

Two of the 4 raids recorded at hives housing large colonies of Africanized honeybees in Panama resulted in colony absconding by *Apis*, rather than immediate colony death. Three of the attacks occurred in deep forest within Parque Nacional Soberanía and one in Curundu, during mid rainy season. After one raid, which did not result in colony absconding, about 60 dead *L. limao* and 10 dead, dismembered *Apis mellifera* were found in the hive (Roubik 1991).

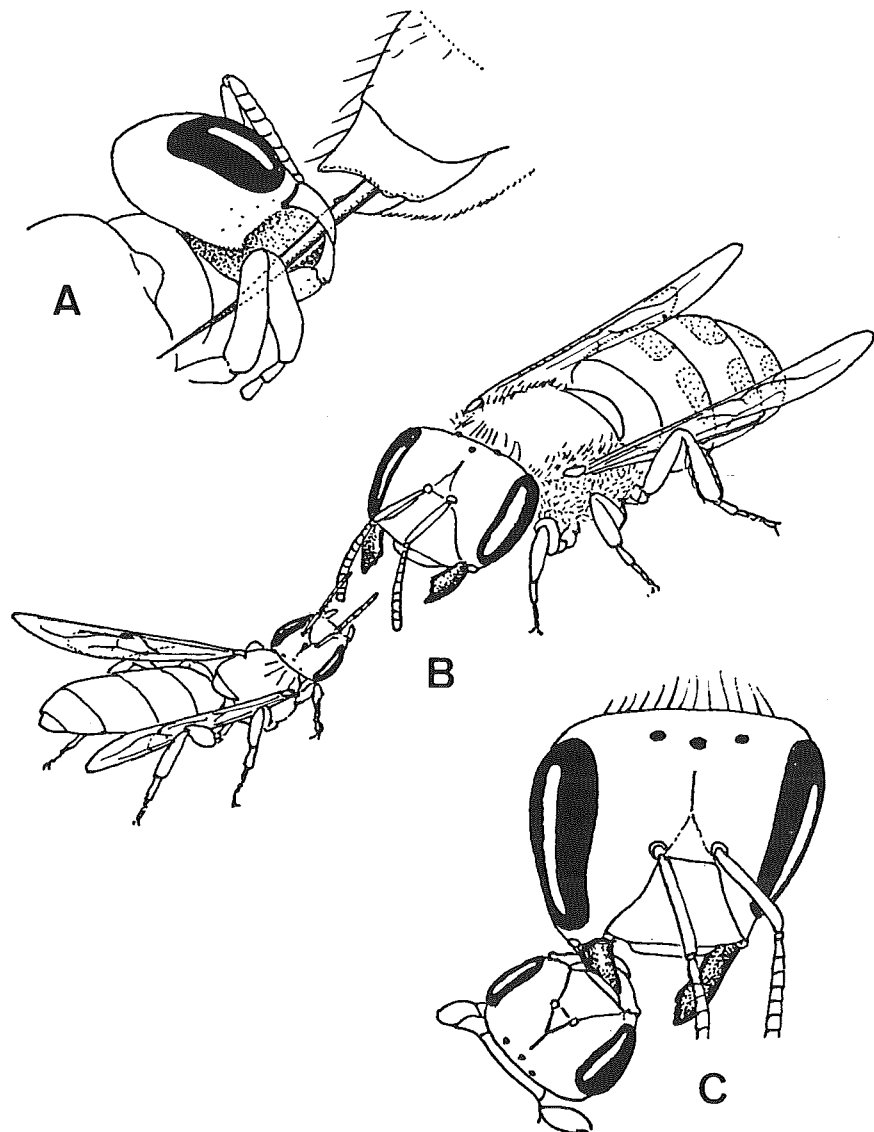


Fig. 6. Raiding *Lestrimelitta limao* attacking A) the sting apparatus of an Africanized honeybee, B) displaying threat posture confronting a worker *Melipona quadrifasciata*, and C) seizing the mandible of *Melipona quadrifasciata*.

### 5) Mild and Interrupted Raids

In addition to typical mass pillages during a few hours, mild raids or those

suddenly terminated for no apparent reason were seen in Brazil. Observations were also made at raids artificially interrupted in Panama. Fig. 1 shows that foragers returned with cerumen loads from outside the Brazilian meliponary. These were noted on 15 days. Cerumen was of light color, typical for locally abundant *Nannotrigona*, *Tetragonisca* & *Plebeia*. During 12 of 15 days returning foragers were few. They may have entered host nests, as described below, or cerumen was removed from nest entrances. Whether robbers worked in small groups or individually in this activity is unknown.

However, 3 raids took place at known nests, with neither mass flight activity nor conspicuous occupation of the host nest entrance by *Lestrimelitta*. These involved *Plebeia*, 2 different nests of *P. droryana* Friese and 1 other *Plebeia*. A few *L. limao* but no *Plebeia* occupied a nest entrance of *P. droryana* on July 5, and a few *L. limao* were found among honey storage pots. Later the same day there were 7 to 8 *L. limao* guarding the entrance, no *P. droryana*, and no bees in flight. *L. limao* closed the host nest entrance late in the day. On the following morning the nest entrance was guarded by 5 or fewer *L. limao*. Later the entrance was closed until just before midday when foraging *P. droryana* returned to the nest but did not enter. Three *L. limao* entered the nest and 5 departed, carrying cerumen loads on their hind legs. At 12:40 there were less than 8 guarding *Lestrimelitta* and no arriving bees. When the hive lid was completely removed an hour later, 6 *L. limao* left carrying cerumen, and 7 or 8 remained guarding the entrance. One more left with cerumen and 2 entered after the lid was replaced. No bees arrived after 16:00. On the following day *Plebeia* was the only bee at the nest entrance.

A second colony of *Plebeia droryana* gave evidence for a more complete temporary usurpation of the nest, but the foraging activity of *L. limao* was relatively slight. Host bee movement ceased and the adult population appeared paralyzed following invasion by 10-20 *L. limao* during 2hrs. The raid was apparently caused by artificial interruption of a raid on *Scaptotrigona*. The *Plebeia* nest contained 6 guarding *L. limao* and 10 inside the brood area; 2 immobile clusters of hosts were inside the hive lid and in a corner. At 14:25 2 or 3 *L. limao* guarded and 10 were inside the nest. At 14:55 2 guarded and 9 arrived at the nest, 2 returning *Plebeia* approached the nest but did not enter; simultaneously 5 *L. limao* left, only 1 carrying cerumen. At this time there were still 10 *L. limao* within the nest. Cells in the top comb of host brood had been opened. Small resin droplets had been deposited by the pillaging bees on the storage pots and outside of the nest entrance. One *L. limao* guarded, 7 arrived and 9 left, 1 carrying cerumen. Five minutes later 1 guard was present, 1 *L. limao* was in the nest, 5 left and 5 arrived. The same guard was present at 15:10, while 10 more *L. limao* arrived and 10 left, 1 with cerumen. After 10min a few *L. limao* were in the nest and 2 or 3 of the *Plebeia* started to move, some foragers arrived, and 1 took a guard position at the entrance tube. By 15:25 no



*L. limao* remained and the *Plebeia* moved freely.

The third case was more intense — a nest of *P. sp.* was occupied by 50 *L. limao*. There was no mass flight or occupation of the entrance. Two immobilized host masses were in the bottom of the hive; combs and storage pots were completely destroyed. No guarding *Lestrimelitta* was present.

Cessation of a raid after a short period was seen several times in Brazil and Panama. At a nest of *Nannotrigona testaceicornis* a few *Lestrimelitta* arrived and grappled with guard bees. *Nannotrigona* returned but did not enter the nest, while some *L. limao* entered and grasped *Nannotrigona* with their mandibles. After 30min *L. limao* began to retreat; all were gone an hour after the raid began. During a raid on Africanized *Apis mellifera* the *L. limao* arrived in 2 groups that remained outside the hive and killed 7 *Apis*, but none of the robbers was killed (Fig. 5A). Arriving honeybees or those leaving the nest were attacked and killed. Robbers outside the nest exhibited typical threat posture by approaching honeybees with their wings spread in a "V" position while raising the abdomen (Figs. 4C, 5A). Although returning honeybees were unable to enter the nest, *L. limao* also failed to advance the raid and withdrew after 30min. During this time 27 honeybees and 31 *L. limao* were killed, while several *Lestrimelitta* had attached to the legs, mandibles and sting of the honeybee (Fig. 6A). Short raids terminating in 10min (2), 16min, and 40min (2) were seen in Panama, all involving *Scaptotrigona barrocoloradensis*.

In Panama 2 raids of *Scaptotrigona pectoralis* were interrupted artificially in brief experiments. There was no odor at the nest entrance. Then several *Lestrimelitta* were crushed to release their odor. *Lestrimelitta* streamed from the nest and abandoned the raid, while 200 to 300 *Scaptotrigona* immediately approached. There had been no hovering host bees near the nest. These experiments were repeated 4 times using different colonies of *Scaptotrigona barrocoloradensis*, when no detectable citral odor in the air. Only 1 of the 4 trials triggered the emigration of *Lestrimelitta* and return of the hosts.

#### 6) Guarding Behavior During Mass Raids — The Guard Ring.

Except in the mild raids already described, victim nest entrances were encircled by guards, forming a ring up to 8cm diameter and consisting of about 100-600 workers (Fig. 4B). Outermost guards primarily faced outward and were relatively immobile, whereas workers within the ring, especially those nearest the entrance, faced various directions and were more active. When stimulated all guards assumed a threat posture by elevating the metasoma and opening the mandibles (Fig. 4C).

The number of guards in a raid was always far greater than that at the entrance to their own nest (Fig. 2A, C) and were aggressive, in marked contrast to robber nest guards. Occasionally a narrow outer ring of guards was formed, separated by about 2 to 3cm from the main ring. The Ribeirão Preto colony

contained some workers that were color-marked. Some stayed in the ring continuously, particularly those in the outer margin. These kept the same position for hours, during the 5 raids in which the marked bees were observed. At the same time, there were other marked workers that only carried cerumen loads. Task allocation among pillaging workers seemed to take place, at least within a single raid sequence. The area outside the nest covered with the guards was spotted with resin taken from the interior (Fig. 4D).

During observations of 1991 and 1992, video camera recordings were made of guard rings. A few bees exposed a whitish intersegmental membrane between the 5th and 6th terga, while raising the abdomen and fanning the wings. No odor was discernible, but this behavior raises the possibility of either additional chemical or visual signals during raids.

Marked *L. limao* engaged in guarding victim nests or foraging from 28 to 46 days of age. No shift between these 2 activities was age-related. The oldest marked worker of *L. limao* (46 days longevity) was the last remaining guard in an attack of *Nannotrigona*.

#### 7) Entrance Tube Preparation

During raids of more than 1 day the host nest entrance was modified or new entrances were built (Fig. 4E). On a nest of *Melipona quadrifasciata* having a single bee-sized hole of hardened mud and resin, *L. limao* prepared a short entrance tube of cerumen, 5cm in diameter. The tube was lengthened to 1cm in 2hr during a raid. Tubes of *Nannotrigona testaceicornis* were either replaced with entrances like those made on *Melipona* nests, or existing tubes were modified by robbers, giving them a coarser appearance. Nests of *Scaptotrigona postica* had entrance tubes similar in texture to those of *L. limao* and were extended without much modification (Figs. 4D,E). When tubes were elongated, guards were restricted primarily to the inner walls, as were guarding *Lestrimelitta* at their own nest; the number of guards thus decreased. When *L. limao* finished a raid, the nest entrance tube was always removed by the victim colony.

#### 8) Area Guarding

Figure 5B shows the extension of *Lestrimelitta* from the ring of guards on the landing platform of a *Scaptotrigona postica* hive. Guards at this position were very alert and maintained a threatening posture, readily darting at intruders. Most of the stingless bee hives at Ribeirão Preto were placed on stands so that entrances were well above ground. When the entrance was only 10 to 20cm high, a group of guarding *Lestrimelitta* often extended in an ellipse up to 15cm to the ground in front of the entrance. Area guards were aggressive, frequently assuming a threat posture, and scrambling, darting at, and chasing away approaching insects, workers of the victim nest, honeybees searching for pieces of cerumen, and parasitic phorid flies. Area guards even attacked an

inanimate object, the pencil of one observer that was thrust at it. These bees often grappled briefly with one another. One of the 4 upper corners of the hive containing a colony hosting a raid was often occupied by guarding *Lestrimelitta*. A front corner was occupied during the pillage of *Apis*, *Melipona*, *Plebeia* and *Scaptotrigona*, and a rear corner was occupied twice at a nest of *Scaptotrigona*. In most cases the corners were patched with resin by the resident bees to seal openings in the hive, and *Lestrimelitta* was possibly attracted to this deposit. In Panama, workers of *Plebeia (Scaura) latitarsis* and *T. (Trigonisca) angustula* covered raiding *Lestrimelitta* with resin extensively, although this did not always prove to be an effective defense. Guards at the corners readily attacked host worker bees or other animals of moderate size that approached or landed nearby.

Except during the initiation of the mass raid, no strong citral odor was evident near guards. Evidence of aerial guarding was noted once in Panama. This behavior was the constant circulation of workers in a horizontal ellipse in front of the host nest entrance, identical to the behavior of *L. limao* at its own nest during a raid. This occurred at a nest of *Scaptotrigona barrocoloradensis*.

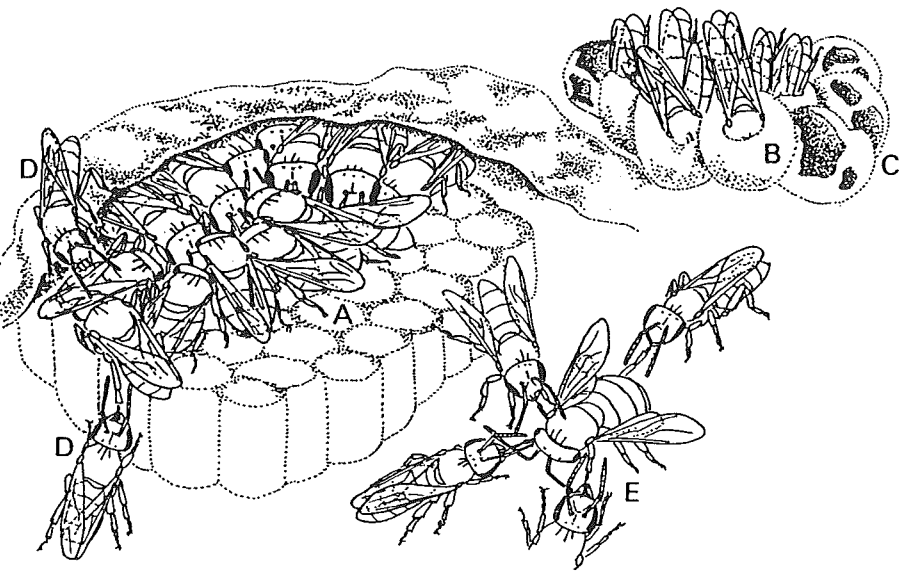


Fig. 7. Workers of *Lestrimelitta limao* and host *Scaptotrigona postica* within the nest during a mass raid. A) immobile mass of host bees on a comb containing pupae, B) *Lestrimelitta* removing honey from storage pots, C) empty storage pots showing partial removal of cerumen construction material, D) *L. limao* insepecting immobile masses of host workers and the queen, E) host worker attacked and killed by *Lestrimelitta*. The immobile mass of host bees is normally formed under or between pupal and prepupal brood cells and not on a top comb, shown here in a simplified view.

### 9) Ethology of The Pillage

All immature bees in the host nests except pupae in their cocoons and prepupae were killed or discarded from nests during mass raids. *Lestrimelitta* began pillage by opening the cells containing eggs or young larvae and imbibing larval provisions. Eggs and larvae were discarded randomly on the comb surface or dropped. Combs made up of pupae or prepupae never were damaged.

Stored honey and pollen were also taken during pillage of larval food, but removal of liquid brood cell provisions was performed separately and earlier, probably because each cell could be emptied by one or 2 bees. Pollen in storage pots became moistened by honey or brood provisions regurgitated by robbers to allow consumption.

Robbing bees appeared to specialize on either cerumen and resin, carried on the hind legs, or on food stores, carried in the crop. Building materials present in host nests — cerumen and resin deposits — were carried back to the robbers' nest. Cerumen is required by all stingless bees for building storage pots, brood cells and most of the involucre, and in some species the entrance tube. *Lestrimelitta* pillaged cerumen primarily from involucre but also from pillaged brood combs and storage pots (Figs. 7B,C). Resin was taken wherever encountered in the nest. In 2 nests of *S. postica*, roughly half the invaders robbed building materials. Trophallaxis between *Lestrimelitta* was observed 90 times among food foragers, 6 times from food foragers to materials collectors, and never from bees that collected cerumen or resin. During raids by *Lestrimelitta* in the nests of *T. (Trigonisca)* and *Scaptotrigona*, thieving of pillaged cerumen took place between *Lestrimelitta*.

Placing a glass plate on the hive caused pillaging workers to quickly fill gaps with resin. This was seen during raids upon 2 *Melipona quadrifasciata* and *Trigonisca*. The *Lestrimelitta* also covered pillaged combs of *Melipona* with incomplete, coarse cerumen envelopes.

### 10) Withdrawal of Robbers After Mass Raids

Increasing host arrival and robber withdrawal occurred over 5 to 58min until all robbers departed; in 8 of 10 cases within 14min. Robber bees often poured from the nest entrance, followed by the mass entrance of hosts. Larval food in brood cells was nearly always completely removed before withdrawal, but stored food remained, especially pollen. Resin and cerumen never were depleted. At withdrawal the following sequence of events took place: 1) the number of departures by *Lestrimelitta* gradually exceeded that of arrivals, even though the number of guarding robbers did not change; 2) host foragers previously unable to land near the entrance returned although blocked by guarding *Lestrimelitta*; 3) after some foragers entered, guarding *Lestrimelitta* seemed unable to prevent mass entrance, resulting in limited combat between

hosts and robbers; 4) host workers in the nest gradually resumed movement and fanned the wings, and 5) all guarding *Lestrimelitta* departed while host bees resumed normal entrance guarding activity. Withdrawal series differing in some details are described below.

Withdrawal of *Lestrimelitta* from a nest of *Plebeia droryana* (Fig. 7) shows this retreat was completed 10min after entry of *Plebeia* foragers. In the final phase the host bees returned despite the presence of guarding *Lestrimelitta* and often attacked them. Some of the returning foragers of *Plebeia* stayed at the nest entrance, fanned their wings, and grappled with the robber guards.

Variation occurred among conspecific host colonies. Withdrawal after a 2 day occupancy of the nest of *Nannotrigona testaceicornis* was completed in 8min. Robbers left en masse while their guards diminished in number and host foragers began to arrive. The last *Lestrimelitta* left with a cerumen load shortly after the first guarding *Nannotrigona* appeared at the nest entrance. At a 2nd nest, no foragers of *Nannotrigona* returned until all *Lestrimelitta* departed, although workers in the nest resumed activity in the presence of about 50 robbers.

During the 2nd day of a raid on one colony of *Melipona quadrifasciata* withdrawal was completed 19min after return of a host forager and 13min after the 1st entry of a host to the nest. Guarding robbers, however, displayed violent food solicitation toward departing conspecifics, yet were not fed. As in the case of *Nannotrigona*, host workers in the nest became active before *Lestrimelitta* began retreat. Guarding *Melipona* also prevented entry by nest mates near the end of withdrawal, a behavior occasionally seen in *Scaptotrigona postica*. Departure of guarding robbers while foraging *Lestrimelitta* remained occasionally produced relatively large mortality. Returning host foragers seemed responsible for attack of these robbers.

After many robbers departed a *Scaptotrigona postica* nest the entrance was dominated by returning foragers. Approximately 50 *Lestrimelitta* were within the nest and host workers there were immobilized. Five minutes later some hosts within the nest were active and several foragers entered. The *L. limao* were attempting to leave, running beneath the glass hive lid. Seven minutes later only one-third of the host adults were inactive and foragers entered continuously. Most of the remaining *Lestrimelitta* were running about on the underside of the glass lid, near the nest entrance. Inactivity was scarcely evident in nest bees; returning foragers ran excitedly through the nest, fanning the wings, and in 15min all the robbers had left. A second attack and withdraw sequence at the same host colony was witnessed on another day. As the withdrawal began, 2hrs after the raid started, the entrance was dominated by resident bees that prevented entry of their foragers. A half hour later most foragers had returned and many *Lestrimelitta* were still in the nest, along with many dead *Scaptotrigona*. Foragers of *S. postica* were active within the hive,

but younger adult bees that had remained in the hive were not. After younger *Scaptotrigona* in the nest had resumed activity about 100 *L. limao* were killed. The foregoing instance documents the only observation in Brazil of substantial losses by *Lestrimelitta*. A similar large loss of robbers occurred once in Panama during a raid of *S. barrocoloradensis*; approximately 300 dead *L. limao* and a few dozen *Scaptotrigona* were found on the ground outside of the nest entrance.

### 11) Intranidal Host Behavior During Mass Raids

The mass raid by *L. limao* illustrated schematically in Fig. 4 shows the congregation of nearly all host bees within space around brood combs of mature larvae and pupae. Detailed observations of intranidal behavior involved 8 raids on *Tetragonisca*, *Melipona*, *Plebeia*, *Nannotrigona*, and particularly *Scaptotrigona*. Host behavior was relatively uniform. A colony of *S. postica* observed at raid initiation reacted immediately as workers in the nest retreated to areas between brood combs or between sheets of involucre surrounding the brood area (Figs. 4, 7). This occurred 10min after *L. limao* dominated the entrance. Until that time, host workers excitedly rushed across the floor of the hive but did not move toward the entrance.

After their retreat from robbers, host workers formed dense, stationary masses in which they pressed closely upon each other. Host workers that failed to join were invariably killed by *L. limao*, often when 2 or more of the pillaging bees grasped appendages, pulling in opposing directions (Fig. 6E). Small masses of immobile workers at relatively exposed positions were also attacked. In a nest of *S. postica* 16 host workers had formed a disk 2cm in diameter. Several *L. limao* dragged the bees, one at a time, from the aggregation and gradually killed them all. A few individual host workers within larger masses also were dragged from them and killed, but this was rare. *Scaptotrigona* groups consisted of callows and older bees in all stages of adult pigmentation (adults become darker as they age), and thus could include foragers or guards that remained in the nest. In the nest of *P. sp.* the involucre is nearly absent and exposed masses of immobile bees were on the hive floor. Masses of other species were principally beneath the bottom brood combs.

Victim nests were occasionally checked by opening hive lids, the odor of *Lestrimelitta* was often not perceived in the 2 observation colonies of *Scaptotrigona* in Brazil, as well as in the wooden hive of *S. pectoralis* and at the nest entrances of numerous *S. barrocoloradensis* colonies in Panama. Instead, the strong odor of *Scaptotrigona* predominated.

### 12) Tolerant and Slightly Resistant Species

While intranidal behavior seemed to differ little among various species, extranidal behavior was distinctive, especially in the degree of resistance to robbers displayed by host foragers and guards. Foragers and guards of *Plebeia*

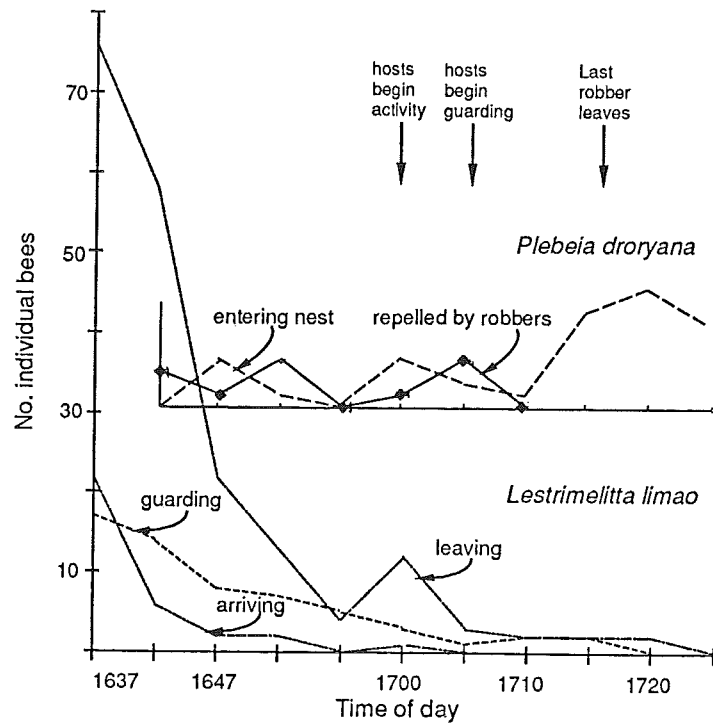


Fig. 8. The temporal sequence of robber and host activity during the process of withdrawal by robbers from the host colony. The host colony is of *Plebeia droryana*.

*droryana* and *P. sp.* displayed little resistance and seldom attempted to land at an entrance occupied by *L. limao*. This possibly was related to the occurrence of mild raids at nests of these species. *Melipona quadrifasciata* observed during 3 raids resisted only weakly. Guards attempted to prevent entry by the robbers primarily by blocking the entrance with their heads. Forager *Melipona* returned and approached the nest entrance, forming a dense mass of about 50 workers. They retreated without landing if threatened by guarding *Lestrimelitta*. Some returning foragers were bitten by guards and died (Fig. 4B); few others were killed.

### 13) Intermediate Response of *Nannotrigona*

Foragers of *N. testaceicornis* returned intermittently to the hive, forming a mass of 30 to 60 bees. They usually flew away but sometimes landed at the entrance and then fell to the ground. Combat between *Nannotrigona* and the area guards of *L. limao* was frequent but seldom resulted in deaths. Only 2 and 3 dead *Nannotrigona* were seen on the ground after 3 different raids, and none of *Lestrimelitta* were found. The host bee rarely bit *Lestrimelitta*. When it was

bitten by the robber it turned or raised the body during a series of slow, rocking motions and then, after remaining perfectly still, suddenly slipped away from the *Lestrimelitta*. This escape behavior was observed repeatedly. At night, groups of 10 to 15 guarding *Lestrimelitta* were seen at the entrances of pillaged colonies of *Nannotrigona*. Many foragers of the host bee returned to the nest carrying pollen loads in early morning on the next day, but they did not land and withdrew.

### 14) Strong Response of *Scaptotrigona*.

Attacks by *L. limao* on colonies of *Scaptotrigona* had hitherto been recorded only by Friese (1931), although *Scaptotrigona* was attacked frequently in both our study areas (Table 1). The workers at the nest entrance were fairly aggressive toward human observers, although far less than some of the *Trigona s. str.* that build large exposed nests. *Scaptotrigona* of diverse species emit strong mandibular gland odors that are chemically similar in the several species that have been studied (Blum 1981). The following observations apply to the *S. postica* studied at Ribeirão Preto. *Scaptotrigona postica* is as long as *L. limao* but more robust, having a mesosomal width of 2.2 to 2.4mm, compared to 1.8 to 1.9mm of *L. limao*.

Foragers driven away by guarding *Lestrimelitta* continued to fly in the area 10 to 30m from the nest. Beginning within 10min. of the initiation of a raid, this mass of bees repeatedly returned to the nest until the robbers withdrew. The interval between their approaches to the nest was on average 9min (range = 1 to 26min, N = 6 colonies, 55 intervals). Near the nest the group flew about 30cm above the entrance and emitted a strong odor. Some bees landed on the entrance but the remainder flew away. Workers landing on the entrance were chased away by guard *Lestrimelitta* and fell to the ground. Although some entrance guards of *L. limao* also fell to the ground grappling with *S. postica*, they normally did not take flight or leave the nest entrance. Fighting with *Scaptotrigona* was mainly executed by area guards (Fig. 4B). After each approach-and-retreat of the hovering group of *S. postica*, numerous dead bees were left near the nest entrance. Their numbers usually consisted of 6 to 7 times more host bees than robbers (range = 3 to 40, N = 10 colonies). In one instance the robber colony lost at least 148 workers in raids on *S. postica* during nearly 3mo, while various colonies of the host species lost a combined 1310 workers due to these raids. When the hovering bees began to land during robber withdrawal, fanning their wings, the remaining robbers withdrew within a few minutes.

### 15) Interspecific Conflicts Related to Robber Attacks

Colonies near those being raided occasionally responded by attacking host or robber bees. When *Lestrimelitta* attacked a colony of *Scaptotrigona luteipennis* Friese, a few dozen workers from a nest of *Melipona eburnea*

separated by 1 m from the *Scaptotrigona* attacked hovering host workers with their mandibles. Interspecific response to alarm odors, presumably all of mandibular gland origin, were seen several times among *T. angustula*, *S. barro-coloradensis* and *M. eburnea panamica*, all of which began to attack each other and *Lestrimelitta* after a raid of *Scaptotrigona* was initiated within a few meters of the nest of another species. Similar alarm behavior at this site was seen for *Paratrigona ornaticeps* Schwarz, *Trigona muzzyensis* Schwarz, and *Nannotrigona perilampoides*. When *Lestrimelitta* attacked the nest of the last species, the *Paratrigona* and *Trigona* also attacked and attempted to enter the nest of *Nannotrigona*. They also attacked each other and the *Lestrimelitta*. These skirmishes lasted for less than an hour, thus it is likely many similar incidents occurred that were unobserved. *Melipona bicolor* reacted to a raid on *Scaptotrigona* by gathering at the nest entrance of *Lestrimelitta* and grappling with guard bees. Possibly some workers of *Lestrimelitta* returning from the hive of *Scaptotrigona* inadvertently entered the nest of *Melipona*, separated by less than few meters. Workers of *M. bicolor* hovered in front of colonies of *M. quadrifasciata* and *Apis*, grappling in the air and falling to the ground with defenders of these colonies. Some of the 2 *Melipona* returned to their nests with dead workers of *L. limao* attached to legs. After such returns, colonies responded swiftly and many workers exited from the nest. Masses of hovering *M. eburnea* were formed artificially by presenting one or 2 crushed worker *Lestrimelitta* at nest entrances. Such hovering groups of bees were noticed several times on other occasions, and they persisted approximately 30min. Although no attack by *L. limao* was observed, it may have been a cause.

#### 16) Raid Duration

Raids lasted a few hours to 5 days, but the most common sustained raid lasted 4hrs during a single day. Raids during a single day ranged from 1 to 8 hrs (mode average 4hrs, 8 of 23 cases). The timing of raid initiation and termination in the Brazil study is given in Fig. 8. Raid initiation showed bimodality; the peak occurrence was at 10:00 to 11:00hrs and 12:00 to 13:00 hrs. Raids ended when all *Lestrimelitta* left the host nest, which occurred primarily between 16:00 and 17:00hrs. Temporal activity of *Lestrimelitta* in Panama confirmed trends recorded in Brazil. Foraging *Lestrimelitta* were seen at the nests of other bees during every hour of the day between 08:00 and 18:00, with a maximum noted between 11:00 and 12:00. A total of 42 raids was witnessed, 6 of which involved conspecific colonies of *Lestrimelitta*. Seven cases of simultaneous raids, probably originating from the same colony of *Lestrimelitta*, involved 2 host colonies of the same species or subgenus. Four were of 2 colonies of *Scaptotrigona barrocoloradensis*. One of the double raids affected a colony of *Scaptotrigona luteipennis* and 1 of *S. pectoralis*. Partly callow *Lestrimelitta*, having a fully black head and propodeum but white

metasomal segments found only in young bees, participated in the raids; suggesting their origin from the same colony. Other double raids were of 2 colonies of *T. (Tetragonisca) angustula* and 2 colonies of *Paratrigona ornaticeps*. None of the colony pairs were grouped close together. Several raids were observed at their inception or finish, but only in 1 instance were both the beginning and ending observed for a given host colony. In 11 cases, raids began from 08:15 to 13:00hrs and finished between 11:30 and 15:00hrs. A total raid duration of 2.5hrs, from 13:00 to 15:30, was recorded at 1 nest of *Scaptotrigona pectoralis*. Raids that lasted multiple days occurred for 2 days at one nest of *Nannotrigona testaceicornis* and for 2 days at a nest of *T. (Tetragonisca) angustula*. Nests appeared normal. Nest entrances were complete and occupied by host bees, on the next day for *N. testaceicornis* and 9 days later for *T. angustula*.

#### 17) Host Colony Selection

Summarizing the types of bees attacked by *Lestrimelitta* in Brazil and Panama, the group having the highest probability of hosting a raid was *Scaptotrigona* (Table 1). Intensity of attack was also relatively high on some *Plebeia*, *Nannotrigona*, *Melipona*, and *Paratrigona*. Numbers of raids observed on each species are related to their total colonies. The ratio of these 2 numbers is used as an index of host preference. Relative preference for hosts at Ribeirão Preto, calculated in this manner, is *S. postica* > *P. sp.* > *P. droryana* > *M. quadrifasciata* > *N. testaceicornis* > *T. angustula* >> *A. mellifera*. Nine species of stingless bees, of 14 colonies, never hosted raids (Table 1). The preference index does not apply when a colony was exterminated or its stores were completely depleted by a single attack, because it was then unavailable for the repeated attacks observed at many colonies. Attacks on *N. testaceicornis* were confirmed only twice but the actual frequency must have been higher. Many foragers of *L. limao* returned to their nest from host colonies that were not located (Fig. 1). These likely returned from natural colonies of *N. testaceicornis* and *P. droryana*, and *T. angustula*, since their cerumen is pale brown, as was cerumen carried by these *Lestrimelitta*. In Panama, the raiding intensity, in descending order, was *Scaptotrigona barrocoloradensis* > *S. pectoralis* > *Paratrigona ornaticeps* > *Nannotrigona perilampoides* = *S. luteipennis* = *Lestrimelitta limao* >> *P. (Scaura) latitarsis* > *T. (Frieseomelitta) nigra* > *T. (Tetragonisca) angustula* > *M. eburnea*. Almost all colonies attacked by *Lestrimelitta* in Curundu were in natural nesting cavities. Although the number of species varied at this site, the nests considered here were kept at least 6mo.

Raids in the forest of BCI, Parque Nacional Soberanía and elsewhere were seen on 2 nests of *Melipona eburnea* (Roubik & Wheeler 1982, and 1 nest on BCI), *Melipona favosa* (1 raid), *Apis mellifera scutellata* Latreille (4 raids),

*Scaptotrigona barrocoloradensis* (on BCI and Soberanía Park, 2 raids), *Partamona* sp. (1 raid), *Plebeia franki* (1 raid), and *Trigona muzoensis* (on BCI, 1 raid). Evidence of 2 raids of *Melipona eburnea* was obtained by examining nest entrances where those of *L. limao* had been built, which were unmistakably of the host species.

### 18) Species Not Attacked

Colonies never raided in Brazil included 4 species of *Melipona*, 3 species of *Plebeia*, and the following species groups or genera: *Dolichotrigona*, *Leurotrigona*, *Trigonisca*, *Celetrigona*, *Trigona*, *Tetragona*, *Partamona*, *Cephalotrigona*, and *Geotrigona*. Field observations in Panama demonstrated that successful raids affected some of the above species that have highly aggressive defending workers, *Trigona muzoensis* and *Partamona*. Data from Curundu indicate that 13 species of stingless bees were never attacked by *Lestrimelitta* but 4 of these were raided elsewhere in Panama: *Melipona favosa*, *Plebeia franki*, *Trigona muzoensis* and *Partamona* aff. *cupira* F. Smith. The sporadic nature of observations of the stingless bee colonies at Curundu may account for this discrepancy. Colonies of several more species, including *Nogueirapis mirandula* Cockerell, *Trigona amalthea silvestriana* Vachal, *T. (Tetragonisca) buchwaldi* Friese, and *Oxytrigona*, were kept for a few months at Curundu but were not attacked. Bee colonies both in outdoor hives and natural nests were raided by *Lestrimelitta*, as were hives kept in buildings at Ribeirão Preto, where most colonies were in hives. For all species, there is insufficient information on their attractiveness to *Lestrimelitta*. Nonetheless, all colonies present at Curundu were observed with approximately equal frequency, thus the colonies in place for several months establish the foraging choice of *L. limao*.

It was uncertain whether a species never successfully attacked was avoided, merely missed by scouting *Lestrimelitta*, or perceived as far less suitable as a host than other accessible species. Further notes were taken on the possible occurrence of avoidance. Considering the smallest species not attacked, the 'Hypotrigona' or *Trigonisca* group, nest entrances were too small to permit the entry of *Lestrimelitta*. One attack of *L. limao* on *Plebeia* sp. prompted removal of this colony, whereupon it was replaced with a nest of *Trigona (Frieseomelitta) varia* Lepeletier. Eight minutes after the raid began, workers of *Frieseomelitta* defended their nest from *Lestrimelitta*, which then abandoned the raid, leaving one dead worker. On another occasion, shortly after the nest entrance of *Scaptotrigona postica* was occupied by *Lestrimelitta*, the nest was removed and replaced with one of *Frieseomelitta varia*. About 100 workers of *Lestrimelitta* arrived near the nest entrance, which was promptly blocked by the heads of *Frieseomelitta*. *Lestrimelitta* landed near the nest entrance but did not approach it. When the hive was slightly opened, numerous

workers of *Frieseomelitta* left the nest and flew at the *Lestrimelitta*, taking them to the ground, locked in combat. Forty-five minutes after *Frieseomelitta* was introduced, all *Lestrimelitta* departed. Two were killed by *Frieseomelitta*, which sustained no mortality. Two later attacks on *Scaptotrigona* were intentionally interrupted by substitution of a colony of *Frieseomelitta varia*, with the same result.

## DISCUSSION

Our observations of 93 raids by *L. limao* in widely separated areas include the only detailed studies of these events. Consistency in general pattern and conformity with many observations by other researchers show that *Lestrimelitta* is a highly specialized parasitic insect that conducts raids not only in large groups, but also as a few individuals. In general, some workers are sacrificed at the beginning or end of a raid (Kerr 1951), and host species show unmistakable signs of minimizing losses. The possibility of effective mechanical, behavioral or chemical defense against *Lestrimelitta* by species that were never attacked can only be resolved through further study.

### 1) Host Selection

Preference of *Lestrimelitta limao* for *Scaptotrigona* was very clear in Brazil and Panama. Since both taxa share ranges throughout the lowland forests from northern Argentina to perhaps Mexico, they should interact frequently. Similar overlap in distribution exists between *L. limao* and its other 2 principal hosts, *Plebeia* and *Nannotrigona*. These 3 genera contain about 80 species, which suggests *L. limao* is potentially quite generalized, even considering that only 17 of 40 closely-observed species were raided in Panama and Brazil (Table 1). Michener (1946) also witnessed raids by *L. limao* in Panama, noting that all 10 raids were on *Nannotrigona* among 49 colonies, despite the presence of 92 colonies and 8 species of *Partamona*, *Melipona*, *Trigona*, *Tetragonisca*, *Frieseomelitta* and *Scaptotrigona*. Nogueira-Neto (1970) observed 23 attacks by *L. limao* within a meliponary. These were upon *Plebeia* and *Nannotrigona*, while less abundant colonies of *Melipona*, *Tetragonisca* and *Scaptotrigona* were untouched. Furthermore, Laroca & Orth (1984) reported a raid on *Plebeia* and listed 10 host species of stingless bees, also suggesting that *L. limao* changes its raiding habits, to some degree, probably according to regional availability of species. Other information summarized by Laroca & Orth (1984) confirms the repeated association of *Lestrimelitta* with host *Scaptotrigona*, *Nannotrigona*, *Plebeia*, and *Melipona*.

The preference that we noted for *Scaptotrigona* had not been suggested since Friese (1931), nor had interaction with *Tetragonisca* been noted since Nogueira-Neto (1970), and Wille (1961). In addition to our results, J. González (personal communication) has observed an attack of *Frieseomelitta* by a

Mexican *Lestrimelitta* of a currently undescribed species (Ayala 1992), where the robber also frequently raids nests of *Nannotrigona*. In Ribeirão Preto, a raiding colony of *L. limao* avoided entering a nest of *Frieseomelitta varia*, which may indicate a broad range of responses by the robbers to stingless bees of a genus. Eight observed species of *Melipona* included only 3 attacked by *L. limao*. Some variation in host selection within genera seems likely.

There are species and supraspecific groups that are attacked frequently, those attacked less frequently, species unsuccessfully attacked, and those not attacked. A number of factors may cause such a range of robber behavior. Relatively high colony density in the natural habitat, over a substantial period of time, could contribute to evolution of fixed preferences, perhaps reflected in the proportional attack frequencies quantified in our study (Table 1). Suitable victim species should be expected to muster relatively poor or at least occasionally ineffective defenses, consist of small colonies or relatively small bees, and possess a relatively conspicuous or wide nest entrance. These factors would permit frequent discovery and invasion.

## 2) Nest Entrances of Hosts

Characteristics of the host nest entrance seem particularly important to *Lestrimelitta*. It is significant that *Nannotrigona*, *Scaptotrigona*, some *Plebeia* and some *Trigonisca* make relatively long, soft, tube-shaped nest entrances. This is also the case for nests of *Tetragonisca*, *Scaura* and some *Paratrigona*. All of these groups were pillaged by *Lestrimelitta* in our studies. In contrast, those of most other stingless bees are short or are made of hard resin or mud. Therefore, the nest entrances of most preferred hosts of *Lestrimelitta* are not only conspicuous, they provide cerumen for nest construction and, perhaps more importantly, for building an entrance tube for raids. During some mass raids the nest entrance on the host colony was quickly extended, and droplets of resin were added to the perimeter (Fig. 3D,E). Long, narrow, and hard resin nest entrances are made by *Melipona*, *Cephalotrigona*, *Geotrigona* (inside of the nest) and *Tetragona* which do not host raids of *L. limao*. Such tubes may allow defense and deprive *L. limao* of cerumen needed for a raiding nest entrance.

## 3) Larval Provisions of Hosts

Larval provisions were usually depleted but considerable honey and pollen was left in host nests. We conclude that such provisions are important in host selection by *Lestrimelitta*. It would be surprising if *Lestrimelitta* did not carefully select the colonies it attacks. Host colonies were not necessarily those possessing the most stored food, or even the largest colony populations of immatures, hence fresh larval provisions suitable for pillage. For example, colonies of *Apis mellifera* were attacked infrequently, despite their large stores of honey and pollen, and inability to defend the nest. Their nests lack stored

provisions needed for the larvae of stingless bees, since the larvae of *Apis* are fed directly by workers. The complex mixture of glandular and other prepared food upon which bee larvae feed is therefore unavailable in honeybee nests. Large colonies of native stingless bees often are maintained by highly aggressive species of the genus *Trigona* s. str. Such colonies appear to be avoided by *L. limao*, although one raid on a colony of the aggressive *T. muzoensis* was observed in Panama. The robber bee attacked resident bee colonies within most of the observation area in Brazil and Panama. Searching and foraging activity were apparently continuous and proceeded almost on a daily basis during favorable weather conditions.

The quality of larval provisions (Hartfelder & Engels 1989; Roubik 1989), and also resin or cerumen (Sakagami & Camargo 1964) stored in nests of stingless bees vary much among species. Carbohydrates, crude protein, and symbiotic microbes vary widely between the supraspecific groups of stingless bees. The preference of *L. limao* for *Scaptotrigona*, *Nannotrigona* and *Plebeia* and some *Melipona* may be strongly influenced by such characteristics. These are complemented by the quantity of resource available and the defensive behavior of potential hosts. It is noteworthy that *Scaptotrigona* maintains colonies which are among the largest of those found in central Panama and may have several thousand brood cells at a given time (Roubik 1983). Colonies of *Scaptotrigona postica* are also among the largest found in Brazil (Michener 1974). These large colonies are defended by aggressive guards, but their defense is weakened and defeated by the specific communication pheromone and allomone of *Lestrimelitta*. *Nannotrigona*, while not among the largest colonies of stingless bees, has larval provisions with high concentrations of protein and both they and *Scaptotrigona* have high energy content (Hartfelder & Engels 1989; Buchmann & Roubik, quoted in Roubik 1989). The concentrations of these nutrients are from 2 to 10 times higher than those recorded for other stingless bees. The species to which *Nannotrigona* and *Scaptotrigona* have been compared, both in Panama and Brazil, are *Trigona*, *Melipona*, *Partamona*, *Tetragonisca*, *Tetragona*, *Trigonisca* and *Lestrimelitta*. This range of species, similar to that of the experimental meliponaries where raiding by *Lestrimelitta* was observed, indicate that robber preference is likely influenced by the quality and quantity of host larval provisions.

## 4) Competition Between Robber Colonies

Intense conflict ensued between conspecific *Lestrimelitta* in Panama and conflicts with *Tetragonisca* also were striking. Fatal combat between these colonies has many implications that require further study. A raid by *L. limao* of *Tetragonisca*, culminating in usurpation of the robber's nest by the raided colony was reported by Weyrauch (quoted in Schwarz 1948). The observation in Panama confirms this phenomenon, and also other reports that relatively

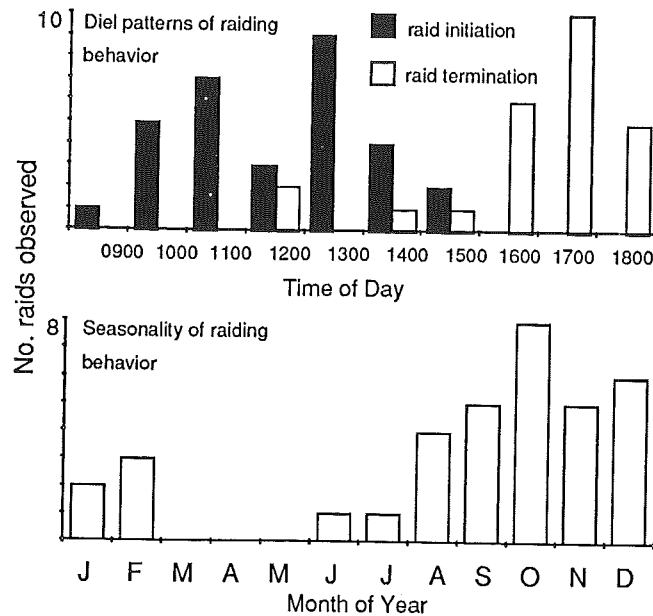


Fig. 9. Daily and seasonal raiding activity patterns of *Lestrimelitta* in Brazil and Panama. The daily raiding patterns are of an observation colony in Brazil, and the seasonal observations were recorded from attacks by natural colonies on observation colonies see text).

small *Tetragonisca* not only defends itself exceedingly well from *Lestrimelitta*, causing raiding parties heavy damage, but also usurps the nests of conspecifics and other meliponines (Mariano quoted by Schwarz 1948; Bertoni 1911; Wille 1961; Sakagami & Laroca 1963; Nogueira-Neto 1970; Wittmann 1985). Observations in Panama and Brazil on the extermination of *T. angustula*, without occupation of this colony's nesting site, stand out as examples of aggression between 2 species that have similar life styles, utilizing the nesting sites and materials of other social bee colonies.

The occasional usurpation of nests by *Tetragonisca* may be an artifact of crowding or weakened colonies sometimes found in meliponaries, but potential facultative parasitism of other stingless bee colonies by this bee is strongly implicated from field data. Furthermore, 3 separate citations exist of *Scaptotrigona* as facultative robbers of other stingless bees (Kerr 1951; Weaver 1975; Posey & Camargo 1985). Although this has not been confirmed in a natural setting, it is conceivable that *Lestrimelitta* seeks to diminish loss of its forage by attacking colonies of potential competitors, as it does conspecific colonies.

Effective defense by *Tetragonisca* against raids of *Lestrimelitta* appears to depend upon the use of the robber bee odor as a kairomone, which leads to

massive defensive response in *Tetragonisca* (Wittmann 1985, Wittmann *et al.* 1990). This small bee, which has moderate colony size (3000 - 5000 workers), is almost the only Neotropical stingless bee that perpetually maintains a group of hovering workers guarding the external nest entrance. These bees are successful in fights due to simultaneous attack by 2 or 3 bees on a rival. A consubgeneric species observed in Panama, *Tetragonisca buchwaldi*, displayed no hovering guard workers at the entrance to its subterranean nest. If a live *Lestrimelitta* approaches the nest entrance of *T. angustula*, it is attacked from behind by hovering guard bees. Dead bees, and live stingless bees of other species produce no such response (Wittmann 1985). Moreover, if the odor of the *Lestrimelitta* is released at the entrance or within the nest, hundreds of guarding workers issue from the nest and may hover there for hours (DWR unpublished data; Wittmann *et al.* 1990). Similar hovering defense in response to *L. limao* odor is known for *Melipona* (Nogueira-Neto 1970; Roubik 1989), but can be induced by a variety of colony disturbances. No hovering bees are normally at the colony entrance, and some *Melipona* also possess effective alarm pheromones (Smith & Roubik 1983).

##### 5) The Robbing Process — Raiding Frequency

Raids on particular stingless bee nests may last for a single day or continue intermittently for as many as 34 days (Sakagami & Laroca 1963, and the present study). Attacks continuing for 2 or 3 days seem relatively common (Nogueira-Neto 1970). Repeated attacks on the same victim nests were seen at one colony of *Nannotrigona* and 3 of *Scaptotrigona*, all of which were attacked twice in the site at Ribeirão Preto. Two other colonies of *S. postica* were attacked at least 3 and 4 times during the 2mo observation period. In Panama, although it could not be ascertained that a single natural colony of *L. limao* was responsible for all raids, colonies of *Scaptotrigona pectoralis* and *S. barrocoloradensis* were raided in succession at least 3 and 4 times, respectively, during 38 and 25 days. *Nannotrigona testaceicornis* was raided twice over a 2 day period and *Lestrimelitta* abandoned the nest 6hrs after beginning the raid on the first day. Lucas de Oliveira (quoted by Sakagami & Laroca 1963) saw repeated raids by *L. limao* at a nest of *Plebeia emerina* domiciled in a hive, over intervals of 2 to 2½ months for a few years.

Raiding visits to various colonies in our studies were irregularly spaced in time, and the intervals were longer than the adult longevity of stingless bees, particularly foragers that usually are the oldest bees of the colony (Sakagami 1982). If repeated visits to a nest occur during a month or less, it is likely that the same raiding bees participate, and they probably remember the location of the host colony. Over longer periods of time, repeated discovery of the host colony is necessary.

Seasonality of raiding behavior in Panama was precisely the inverse of



floral abundance (Roubik & Boreham 1990; Roubik *et al.*, 1986). Most raids occurred during the poorest time of the year for floral resources, the late wet season, and no raids were recorded at the meliponary during March, April and May. Since only the raids of hosts and not the raiding behavior of a colony of *L. limao* were observed at this site, raids probably occurred during the 3 months of the late dry and early wet season but were less frequent. This is when stored colony food is most plentiful. The robbing behavior of *L. limao* seemed conservatively adjusted to the richness of its resources. Less frequent raids were made when maximum resources were available per host colony, and no other explanation seems to fit the changing frequency of raids during the course of the year.

Our observations demonstrate that *L. limao* is capable of performing raids even when the colony is queenless and there are few foragers. This was seen after the Brazilian colony was divided and many workers were lost when the new nest was removed. Even young bees lacking complete adult pigmentation participated in raids in Panama. In addition, several colonies were raided in one day, or more than one colony simultaneously, and the workers of a raiding party remained overnight in the host nest. The last observation, given also by Nogueira-Neto (1970), may in part account for the fact that adult populations of colonies seem far too small for the number of brood present in their nests (Roubik 1979, 1983). Simultaneous attacks on 2 colonies are documented by Michener (1946) and Moure, Noeugira-Neto & Kerr (1958), involving *Nannotrigona* or *Plebeia*.

#### 6) Guarding the Host Nest

Occupation of the host nest entrance by raiding *L. limao* was recorded by Michener (1946), Nogueira-Neto (1950), Lucas de Oliveira (quoted in Sakagami & Laroca 1963) and Wille (1961) at nests of *Nannotrigona*, *Plebeia* and *Trigonisca*, respectively. Similar behavior in *Cleptotrigona* was noted by Portugal-Araújo (1958). Lucas de Oliveira and Michener [1946 and quoted by Sakagami & Laroca (*op. cit.*)] reported that guarding *Lestrimelitta* chewed at the victim nest entrance. Removal of the entire nest entrance tube of *Scaptotrigona* and *Nannotrigona* by *L. limao* was seen occasionally in Panama. Construction of a new entrance tube resembling that at nests of *Lestrimelitta* was seen by Nogueira-Neto (1950) and also mentioned in Schwarz (1948). Michener (1946) and Wille (1961) report that guards do not permit entry by returning foragers of the victim colonies, and Michener also noted that the guards display an elevated body posture and threaten by opening the mandibles.

No previous studies have described behavior of area guards and bees guarding other positions noted here. These guarding behaviors may not occur at some natural nest sites lacking structures like the corners of hives, or for

nests that are high above the ground and without a horizontal substrate near their bases. However, such behavior is evidently effective at keeping away potential secondary invaders such as ants and phorid flies. A secondary invasion of phorid flies completely destroyed 2 colonies, one of *Tetragonisca angustula* and one of an unknown species in Panama. We suggest that placement of resin droplets around the host nest entrance (Fig. 4D) may repel ants or phorid flies. Ants have long been known to avoid these resin barriers (Schwarz 1948; Khoo & Yong 1987). The biochemistry of the resin compounds and corresponding bioassays with live ants have not been carried out. The aggressive specializations among guarding bees would, under natural conditions, permit continued survival of a host colony after the raid, particularly in the interval of host colony recovery before the nest entrance is repaired and guarded normally. Defense of victim nests is also known in the giant hornet, *Vespa mandarinia*. This wasp occupies nests of *Apis* and also the nests of other *Vespa* and species. It defends the victim nests from any potential enemies (Matsuura & Sakagami 1973). Functions of host defense by *Lestrimelitta* seem to be slightly different. Guards obviously defend their temporary resource from natural enemies and competitors, but the guarding bees, substantially more aggressive than those found at nests of *Lestrimelitta*, have likely evolved modified behavior patterns to repel foragers of the host colony, which return intermittently to the nest. Preparation of an entrance tube at the victim nest is probably to reinforce nest defense, but we lack information on whether the presence of this structure alone is sufficient to deter returning foragers of the host colony or bees of other colonies.

Only Michener (1946) had adequately recorded the final withdrawal of *L. limao* from victim nests, and our information shows variations in the sequence, according to victim species. During the last 30min of the raid witnessed by Michener at a nest of *Nannotrigona*, the number of *L. limao* departing from the host nest exceeded arriving robbers; there was no sudden mass exodus. In our study, the final guarding robbers incessantly lunged with open mandibles at returning host foragers. After robber withdrawal, the host colony quickly resumed normal activity. We have shown the important role of guarding robbers in the gradual exit of pillaging bees, which otherwise were killed in the nest. Factors that trigger the withdrawal sequence are still unknown.

Exploitation of larval provisions and infrequent use of stored pollen and honey indicate that the gradual depletion of provisions may cause a decrease of arrivals relative to departing, pillaging *Lestrimelitta*. Robbers prefer the larval food, which is much more dilute and undoubtedly more easily imbibed than the pasty stored pollen that must be mixed with honey or liquid in order to be removed. Observations including periodic measurement of resources remaining in the victim nest and numbers of arrivals and departures of robbers might clarify the causal relationship. A fairly clear allocation of tasks existed

within a raiding party. Vigorous food solicitation by guarding *Lestrimelitta* from the departing foragers demonstrates a prolonged participation in this form of guarding. Trophallaxis between guards and pillaging bees may help to regulate the duration of the raid, and especially that of mass guarding. The reason that cerumen foragers cease to pillage, despite the presence of much more of the resource than they carry back to the raiding colony, is also unknown.

### 7) Host behavior and Low Mortality

Formation of immobile masses of workers in victim nests was undoubtedly the proximate cause of their low mortality during raids, both of host and robber. This applies even to *Scaptotrigona*, which were aggressive and sometimes lost many workers outside the nest when attacked by *Lestrimelitta*. Low mortality in nests was in part due to tight aggregation of victims at places of little interest to robbers, such as in corners, beneath combs of prepupae and pupae-away from cells containing larval provisions (Fig. 3B,C). Robbers attacked and killed host bees outside aggregations, or those in very small groups (Fig. 7), thus the number of hosts in a small area seemed to regulate the behavior of robbers.

Our observations did not show that workers of victim colonies fill their crops with liquid food before their retreat from robbers, as suggested by Nogueira-Neto (1970) as a possible defense from their depredations. However, the massive hovering of worker *Melipona eburnea* in front of a nest disturbed by *Lestrimelitta* or other agents can consist of bees engorged with nectar, probably for ripening to honey (DWR, unpublished data). This apparently does not involve imbibition of brood provisions, which is the preferred resource of *L. limao*.

### 8) Chemical Aspects of Raiding

That raiding is permitted through use of citral and other volatile gland compounds of *Lestrimelitta*, not only for intraspecific communication guiding nest mates to a host colony but to effectively subvert the odor recognition of nest mates and also repel the host bees, has been stated many times (Kerr 1951; Moure *et al.* 1958; Wille 1961; Sakagami & Laroca 1963; Blum *et al.* 1970; Wilson 1971). Despite the surprising lack of detailed investigation of this essential point (Nogueira-Neto 1970), it is a logical prediction that host species repeatedly damaged by *Lestrimelitta* learn to recognize the odor of the robber bees and implement defensive tactics following its detection. A more profound and immediate response, such as that displayed by *Tetragonisca angustula* when a live worker of *Lestrimelitta* is crushed at the nest entrance (Wittmann 1985) indicates possible innate response of stingless bees to the odor of *Lestrimelitta*. Recruitment of more guards is accomplished by host alarm pheromone containing not only some monoterpenes released by robbers but

also benzaldehyde, absent in robber bees (Wittmann *et al.* 1990). Controlled tests are needed to establish whether such a response exists in colonies composed solely of workers previously unexposed to the odor of *L. limao*. The merely presumed use of mandibular gland products to guide nest mates to a host seems plausible, as they fly through the air, as does recruitment via scouts (Blum *et al.* 1970). There is no alternative hypothesis for their chemical navigation by means of mandibular gland odors.

Several observations cannot be satisfactorily explained if the odor of *Lestrimelitta* is only a repellent or allomone. First, when a few workers of *Lestrimelitta* are placed at the nest entrance, they are approached or attacked by *Plebeia* and *Melipona* (Nogueira-Neto 1970). This behavior is similar to the mass exit of defending *Tetragonisca angustula*. Nogueira-Neto (1970) established its probable role as a kairomone when he saw *Tetragonisca* react by attacking *L. limao* after robber's odor was released at a nearby nest of *Nannotrigona*. Second, some stingless bees steal cerumen from the entrance tube of *L. limao*, and are thus apparently not repelled by its smell (although not physically harassed by the bees, and the chemical may not attain sufficient concentration there). Third, foragers of *Nannotrigona testaceicornis* driven away by guarding *L. limao* return to their nest, although the entrance is blocked by *L. limao* (Nogueira-Neto, 1970). Finally, a strong smell of citral is not apparent outside of nests being raided or within them. Throughout our observations it was replaced by the host odor in *Scaptotrigona*, the genus hosting more than half the pillages.

Two experiments at nests of *Scaptotrigona pectoralis* but only 1 in 4 with *S. barrocoloradensis* showed that release of the strong odor of *Lestrimelitta* after the raiding bees had dominated the host nest entrance triggered mass return of worker *Scaptotrigona* and the retreat of *L. limao*. This behavior pattern had not been previously recorded. The immediate arrival of hundreds of host workers and simultaneous departure by several hundred *L. limao* implies significance of the odor in retreat of the pillaging bees. Apparently, it signifies a "robber retreat message" (Nogueira-Neto 1970) originating with *Lestrimelitta* that occupy the nest entrance area. At high concentration outside of the nest after the entrance has been occupied by the robbing bees and no obvious smell of citral is in the air, it seems to function not as a repellent, but as an attractant for host workers. Considering *Scaptotrigona* observed in our study, their own strong mandibular gland odor, and not that of the raiding *L. limao*, was apparent within the nests. Further, when the odor of *Lestrimelitta* was released in the nest entrance by the first raiding bees, intranidal behavior of *Scaptotrigona* was chaotic. Ten min later, all host workers within the nest were immobile. This period seems too short if retreat appears after emission of a retreat message by some workers fighting with robbers, as originally proposed by Nogueira-Neto (1970). On the other hand, release of the alarm odor by the host

*Scaptotrigona* again occurs at the departure of *Lestrimelitta* as host workers become active in the nest and attack the remaining *Lestrimelitta*, so that the host pheromones alone probably do not induce the torpor of nest mates. The initial retreat and eventual immobility of hosts are separate and successive responses to mass raids. Again, controlled bioassay of worker response to controlled presentation of nest mate and robber odors would be useful in determining their roles in tranquilizing the host colony.

The above cases show that odors of *L. limao* are not always repellents and have different functions when at different concentrations (Nogueira-Neto 1970), or in different contexts. An uncontrolled variable in all observations made thus far is the rate of odor dissipation within and outside the nest, but it is reasonable to assume that the odor lingers at a higher concentration after release within a pillaged nest. At low concentrations, outside of the nest, the odor of *L. limao* may be a kairomone, attractant and pheromone, but within the nest it probably reaches a higher concentration after being released by just a few bees, and it is likely to be an allomone there. A question raised by Nogueira-Neto (1970) was whether the allomone alone caused the host bees to display immobility and cluster within the nest during a raid — the 'superseding odor hypothesis'. This idea was originally proposed by Kerr (1951) and extended by Moure *et al.* (1958). Nogueira-Neto (1970) observed that prior to the final withdrawal of *L. limao* from 2 pillaged nests of *Plebeia droryana*, some host workers resumed activity in the nest, although the odor of *L. limao* was still present. The length of time needed for host workers to recover from exposure to odor of *L. limao*, and their reactions to intermittent exposure to the odor clearly deserve further attention.

The odor of *L. limao* had differing effects on different species. Generally, intimidation of host workers resulting from the released mandibular gland contents of raiding *L. limao* was more evident among bees in a nest than among returning workers. In all victim species, foragers were never simply intimidated. Once driven away by aggressive guard *Lestrimelitta*, they invariably returned to locations near the nest, even in species like *Plebeia droryana* and *Melipona quadrifasciata* that are quite timid. Damage of adult workers was negligible in the timid *Plebeia* and *Melipona*, intermediate in *Nannotrigona testaceicornis* that exhibited mild resistance and counter attack, and highest in *Scaptotrigona* and *Tetragonisca*. *Scaptotrigona* showed strong resistance, evident in mass counter attack and skirmishing accompanied by release of the alarm and recruitment pheromone typical of this genus (Blum 1981). Blum *et al.* (1970) mentioned that *Scaptotrigona postica* and *Melipona rufiventris* were not at that time known to be attacked by *L. limao*, and they reacted mildly to citral, not being completely disrupted as were *Plebeia droryana*, *Nannotrigona testaceicornis* and *Melipona quadrifasciata*. However, citral from the mandibular glands of *L. limao* is not simply 1 compound but exists as isomers of

geraniol and nerol. Another major component is 6-methyl-5-hepten-2-one (Wittmann *et al.* 1990). Reagent grade citral elicited no response from colonies of *Tetragonisca angustula*, while live *Lestrimelitta* evoked strong reactions (Wittmann 1985). In the present study, *Scaptotrigona* was attacked by *L. limao* more frequently than several other species. The repeated attempts of *S. postica* to recover nests occupied by the robber bee, despite loss of many workers, suggest it is less repelled by the odor of *L. limao*. On the other hand, the returning workers are clearly affected by odor and the presence of guarding *Lestrimelitta*. Returning foragers usually do not land near the nest entrance, even when they are the first to arrive after a raid has been initiated.

We have established that *L. limao* extensively incorporates biting behavior during attacks of other bee colonies. This was thought to be even more important to successful raiding than the release of mandibular gland odors (Nogueira-Neto 1970). Citral may function as a venom if it is introduced through the cuticle of an insect, although pertinent assays are not known to us. Citral and its isomers may combine with biting as an effective weapon, as does formic acid in some ants, and in the stingless bees *Oxytrigona* (Blum 1981; Roubik *et al.* 1987). The effectiveness of biting behavior was particularly clear in combat between *L. limao* and *S. postica*, and also evident in the deaths of defending worker honeybees caused by fighting with *Lestrimelitta*. However, since the extranidal avoidance behavior of returning meliponine foragers takes place before bees have direct contact with *Lestrimelitta*, odor seems to play an important part in this reaction. Little landing at the entrance was seen in host species studied at Ribeirão Preto. A chemical basis for the behavior is very likely because *Lestrimelitta* is otherwise similar to many stingless bees in size, body form and coloration. We suggest that instinctive and probably species-specific behavior may be evoked by *Lestrimelitta*, but naive bee colonies are necessary to test the hypothesis.

Intranidal threat and aggression by robbers might in part be responsible for continued inactivity on the part of the hosts, even though direct attacks were relatively rare. It seems likely that both the sequence and combination of odors produced by mandibular glands of hosts and robbers are responsible for intranidal behaviors, as well as for the behavior of host foragers outside the nest. Biting is frequent only when robbers are first arriving or after they begin to depart, and it is at such times that odor concentrations appear highest. However, raiding parties from natural nests of *Lestrimelitta* seen in Panama consisted of hundreds of bees, while the gradual departure of pillaging bees recorded both in this study and by Michener (1946) at natural nests indicate that few *Lestrimelitta* are present at the end of a raid. We hypothesize that the ratio of host to robber odor concentration is low at the initiation of a raid but high at its conclusion, and that this is brought about by the normal course of fighting between resident and robber bees, and the steady dissipation of odors. When

the odor of the robber supersedes that of the host, the raid is initiated. While the host odor is present but that of the robber is weakening, the raid continues. And when the host odor is strong and there is a sudden addition of the robber odor, final retreat of the robber is initiated, host bees attack the intruders, and host bees outside of their nest return. Woven within the sequence is the exposure of intersegmental areas of abdominal terga, possibly releasing further chemicals that control host and robber behavior.

#### ACKNOWLEDGMENTS

Work in Panama was supported by Scholarly Studies grants to DWR from the Smithsonian Institution and work in Brazil was supported by grants from Fundação de Amparo a Pesquisas de estado de São Paulo, and Campanha Nacional de Aperfeiçoamento de Pessoal de Nível Superior (Rio de Janeiro) to SFS and RZ. They thank W. E. Kerr for use of the facilities in the Department of Genetics, Medical Faculty of Ribeirão Preto, and for the assistance of Mr. Z. A. Schiavoni, apiary manager. Help on manuscripts and field studies was given by Drs. G. C. Eickwort, C. D. Michener, and R. Jander.

#### REFERENCES

- Ayala, R. 1992. Revisión de las abejas sin aguijón de México. Thesis. National Autonomous University of Mexico, Mexico City.
- Blum, M. S. 1966. Chemical releasers of social behavior. VIII. Citral in mandibular gland secretion of *Lestrimelitta limao*. Ann. Entomol. Soc. Amer. 59: 962-964.
- Blum, M. S. 1981. Chemical defenses of arthropods. Academic Press, New York.
- Blum, M. S., R. M. Crewe, W. E. Kerr, L. E. Keith, A. W. Garrison, A. W. & M. M. Walker 1970. Citral in stingless bees: isolation and functions in trail-laying and robbing. J. Ins. Physiol. 16: 1637-1648.
- Bohart, E. 1970. The evolution of parasitism among bees. Utah State University 41st Faculty Honor Lecture, 33 pp. Utah State University, Logan, Utah.
- Camargo, J. M. F. & J. S. Moure 1989. Duas novas espécies de *Lestrimelitta* Friese (Meliponinae, Apidae, Hymenoptera) da região Amazônica. Bol. Mus. Para. Emilio Goeldi Nova. Ser. Zool. 5: 195-212.
- Cruz, C. C. 1962. Anatomia e histologia comparadas das glândulas mandibulares dos meliponíneos. Arq. Mus. Nac. [Rio de Janeiro] 52: 79-84.
- Cruz-Landim, C. C. 1967. Estudo comparativo de algumas glândulas das abelhas (Hymenoptera, Apoidea) e respectivas implicações evolutivas. Arq. Zool. [São Paulo]. 15: 177-190.
- Cruz-Landim, C. C. & S. Rodrigues 1967. Comparative anatomy and histology of the alimentary canal of adult Apinae. J. Apic. Res. 6: 17-28.
- Dyer, F. C. & T. D. Seeley 1987. Interspecific comparisons of endothermy in honeybees (*Apis*): deviations from the expected size-related patterns. J. Exp. Biol. 127: 1-26.
- Friese, H. 1931. Wie können Schmarotzerbienen aus Sammellienen entstehen? II. Zoologischer Jahrbüch., [Abteilung für Systematik, Ökologie und Geographie der Tiere] 67:235-318.
- Hartfelder, K. & W. Engels 1989. The composition of larval food in stingless bees:

- evaluating nutritional balance by chemosystematic methods. Ins. Soc. 36: 1-14.
- Hubbell, S. P. & L. K. Johnson 1977. Competition and nest spacing in a tropical stingless bee community. Ecology 59: 949-963.
- Kerr, W. E. 1951. Bases para o estudo da genética da populações dos Hymenoptera em geral e dos Apinae sociais em particular. Anales do Escola Superior Agricola "Luis de Queiroz", Universidade de São Paulo 8: 219-353.
- Khoo S.-G. & H-S Yong 1987. Nest structure and colony defence in the stingless bee *Trigona terminata* Smith. Nat. Malay. 12: 4-15.
- Laroca, S. & A. I. Orth 1984. Pilhagem de um ninho de *Plebeia catamarcensis meridionalis* por *Lestrimelitta limao* (Apidae, Meliponinae) em Itapiranga, Santa Catarina, sul do Brasil. Dusenica 14: 123-127.
- Matsuura, M. & S. F. Sakagami 1973. A bionomic sketch of the giant hornet, *Vespa mandarinia*, a serious pest for Japanese apiculture. J. Fac. Sci., Hokkaido Univ., VI. Zool. 19: 125-162.
- Michener, C. D. 1946. Notes on the habits of some Panamanian stingless bees (Hymenoptera, Apidae). J. N. Y. Entomol. Soc. 54: 179-197.
- Michener, C. D. 1974. The Social Behavior of the Bees: A Comparative Study. Harvard Univ. Press, Cambridge, Massachusetts xii + 404 pp.
- Michener, C. D. 1990. Classification of the Apidae (Hymenoptera). Univ. Kans. Sci. Bull. 54: 75-164.
- Moure, J. S., P. Nogueira-Neto & W. E. Kerr 1958. Evolutionary problems among Meliponinae (Hymenoptera, Apidae). Proc. 10th Inter. Cong. Entomol., Montreal 2: 481-493.
- Nogueira-Neto, P. 1949. Notas bionômicas sobre meliponíneos (Hymenoptera, Apoidea). II. Sobre a pilhagem. Pap. Avul. Dep. Zool., Sec. Agr., São Paulo, Brasil. 9: 13-32.
- Nogueira-Neto, P. 1950. Notas bionômicas sobre meliponíneos (Hymenoptera, Apoidea). IV. Colônias mistas e questões relacionadas. Rev. Entomol. 21: 305-367.
- Nogueira-Neto, P. 1970. Behavior problems related to the pillages made by some parasitic stingless bees (Meliponinae, Apidae). Development and Evolution of Behavior. Essays in Memory of T. C. Schneirla: 416-434. W. H. Freeman & Co., San Francisco.
- Portugal-Araújo, V. 1958. A contribution to the bionomics of *Lestrimelitta cubiceps* Hymenoptera, Apidae). J. Kans. Entomol. Soc. 31: 203-211.
- Posey, D. A. & J. M. F. Camargo 1985. Additional notes on the classification and knowledge of stingless bees by the Kayapó indians of Gorotire, Pará, Brazil. Ann. Carn. Mus. 54: 247-274.
- Rand, A. S. & W. W. Rand 1982. Variation in rainfall on Barro Colorado Island. In E. G. Leigh, Jr., A. S. Rand, and D. M. Windsor, editors. The ecology of a tropical forest: seasonal rhythms and long-term changes: 47-60. Smithsonian Institution Press, Washington, D. C.
- Roubik, D. W. 1979. Nest and colony characteristics of stingless bees from French Guiana (Hymenoptera: Apidae). J. Kans. Entomol. Soc. 52: 443-470.
- Roubik, D. W. 1980. New species of *Trigona* and cleptobiotic *Lestrimelitta* from French Guiana (Hymenoptera: Apidae). Rev. Biol. Trop. 28: 263-269.
- Roubik, D. W. 1983. Nest and colony characteristics of stingless bees from Panamá

- Hymenoptera: Apidae). J. Kans. Entomol. Soc. 546: 327-355.
- Roubik, D. W. 1989. Ecology and Natural History of Tropical Bees. Cambridge University Press, New York, x + 516 pp.
- Roubik, D. W. 1991. Aspects of Africanized honey bee ecology in tropical America. M. Spivak, M. D. Breed, and D. J. C. Fletcher, editors. The 'African' honey bee: 147-158. Westview Press, Boulder, Colorado.
- Roubik, D. W. & M. M. Boreham 1990. Learning to live with Africanized honey bees. Intercien. 15: 146-153.
- Roubik, D. W. & Q. D. Wheeler 1982. Flightless beetles and stingless bees: Phoresy of scotocryptine beetles (Leiodidae) on their meliponine hosts (Apidae). J. Kans. Entomol. Soc. 55: 125-135.
- Roubik, D. W., J. E. Moreno, C. Vergara & D. Wittmann 1986. Sporadic food competition with the African honeybee: projected impact on neotropical social bees. J. Trop. Ecol. 2: 97-111.
- Roubik, D. W., V. E. Moreno & O. Duran. Why are Panamanian honeybees African? Apidologie. (in press)
- Roubik D. W., B. H. Smith & R. G. Carlson 1987. Formic acid in caustic cephalic secretions of stingless bee, *Oxytrigona*. J. Chem. Ecol. 13: 1079-1086.
- Ruttner, F. 1988. Biogeography and Taxonomy of honeybees. Springer-Verlag, Berlin, xii + 282 pp.
- Sakagami, S. F. 1966. Techniques for the observation of behavior and social organization of stingless bees by using a special hive. Papeis Avulsivos do Departamento de Zoologia, Secretaria de Agricultura, São Paulo, Brasil 19: 151-162.
- Sakagami, S. F. 1982. Stingless bees. H. Hermann, editor. Social insects, Vol. 4: 361-423. Academic Press, New York.
- Sakagami, S. F. & J. M. F. Camargo 1964. Cerumen collection accompanied by thieving and attacking in a stingless bee, *Nannotrigona (Scaptotrigona) postica* Latreille, with a consideration on territoriality in social insects. Rev. Biol. Trop. 12: 197-207.
- Sakagami, S. F. & S. Laroca 1963. Additional observations on the habits of the cleptobiotic stingless bees, the genus *Lestrimelitta* Friese (Hymenoptera, Apoidea). J. Fac. Sci., Hokkaido Univ. VI, Zool. 15: 319-339.
- Schwarz, H. F. 1948. Stingless bees of the Western Hemisphere. Bull. Amer. Mus. Nat. Hist. 90: 1-546.
- Smith, B. H. & D. W. Roubik 1983. Mandibular glands of stingless bees (Hymenoptera: Apidae): chemical analysis of the contents and their biological function in 2 species of *Melipona*. J. Chem. Ecol. 9: 1465-1472.
- Wcislo, W. T. 1987. The roles of seasonality, host synchrony, and behaviour in the evolutions and distribution of nest parasites in Hymenoptera, Insecta, with special reference to bees (Apoidea). Biol. Rev. 62: 515-543.
- Weaver, N. 1978. Chemical control of behavior-interspecific. M. Rockstein, editor. Biochemistry of Insects: 392-419. Academic Press, New York.
- Wille, A. 1961. Las abejas jicótes de Costa Rica. Rev. Univ. Costa Rica 22: 1-30.
- Wille, A. 1979. Phylogeny and relationships among the genera and subgenera of the stingless bees (Meliponinae) of the world. Rev. Biol. Trop. 27: 241-277.
- Wille, A. & C. D. Michener 1973. The nest architecture of stingless bees with special

- reference to those of Costa Rica. Rev. Biol. Trop. Suppl. 21: 1-278.
- Wilson, E. O. 1971. The Insect Societies. Harvard University Press, Cambridge, MA xi + 548 pp.
- Wittmann, D. 1985. Aerial defense of the nest by workers of the stingless bee *Trigona (Tetragonisca) angustula* Latreille (Hymenoptera: Apidae). Behav. Ecol. Sociobiol. 16: 111-114.
- Wittmann, D., R. Radtke, J. Zeil, G. Lübke & W. Francke 1990. Robber bees *Lestrimelitta limao* and their host chemical and visual cues in nest defense by *Trigona (Tetragonisca) angustula* (Apidae: Meliponinae). J. Chem. Ecol. 16: 631-641.

