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Parasitic Behavior of *Exaerete smaragdina* with Descriptions of Its Mature Oocyte and Larval Instars (Hymenoptera: Apidae: Euglossini)

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CONTENTS

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ABSTRACT

The behavior patterns of *Exaerete smaragdina* (Guérin-Méneville), a cleptoparasite of *Eulaema nigrita* (Lepeletier de Saint Fargeau), are described. Included are nest-searching behavior, behavior in the nest of the host, interactions between the host and parasite, and egg placement and number of eggs per host nest. Evidence suggests that the cleptoparasite's sting (or perhaps metasomal apex) usually kills the host egg (or possibly first instar) and that the second instar is capable of killing the host egg and the eggs (or young larvae) of other cleptoparasites in instances of multiple parasitism. Comparisons are made with *Exaerete dentata* (Linnaeus), the only other member of the genus whose behavior has been studied.

The mature oocyte of *Exaerete smaragdina* is described and found to be small relative to the intertegular distance of the female (egg index 0.55). Also described are the five larval instars of this species; the last larval instar is compared with last instars of other Euglossini whose larvae are known.

RESUMEN

Os padrões comportamentais de *Exaerete smaragdina* (Guérin-Méneville), um cleptoparasita de *Eulaema nigrita* (Lepeletier de Saint Fargeau), são descritos. Comportamento de localização do ninho, comportamento no ninho do hospedeiro, interações entre hospedeiro e parasita e colocação e número de ovos por ninho hospedeiro são apresentados. Evidências sugerem que o cleptoparasita adulto mata, geralmente, com seu ferrão (ou talvez com a extremidade metassomal) o ovo (ou possivelmente a larva de primeiro instar) do hospedeiro e que a larva de segundo instar é capaz também de matar o ovo do hospedeiro e, nos casos de parasitismo múltiplo, os ovos (ou larvas jovens) de outros cleptoparasitas. Comparações são feitas com *Exaerete dentata* (Linnaeus), o único outro membro do gênero cujo comportamento foi estudado.

O oócito maduro de *Exaerete smaragdina* é descrito e foi encontrado ser pequeno em relação à distância intertegular da fêmea (índice do ovo 0,55). Os cinco instares larvais dessa espécie são também descritos; o último instar larval é comparado com o último instar de outros Euglossini cujas larvas são conhecidas.

INTRODUCTION

The Euglossini contains three pollen-collecting genera, *Euglossa* (= *Eg.*), *Eulaema* (= *El.*), and *Eufriesea* (= *Ef.*), and two parasitic genera, *Aglae* (= *Ag.*) and *Exaerete* (= *Ex.*) (Michener, 2000). Of the parasitic genera, *Aglae caerulea* Lepeletier de Saint Fargeau and Serville, the sole member of the genus, has been reared from nests of *Eulaema nigrita* (Lepeletier de Saint Fargeau) (Myers, 1935), and *Exaerete*, with five species, parasitizes *Eulaema* and *Eufriesea* nests (Ducke, 1903, 1906; Friese, 1941; Moure, 1946; Dodson and Frymire, 1961; Zucchi et al., 1969b; Bennett, 1972; Ackerman and Montalvo, 1985; Roubik, 1990; Pereira-Martins, 1991). The parasitic behavior of both of these genera is poorly understood. Bennett (1972) provided the only available information, observing *Exaerete dentata* (Linnaeus) parasitizing *Eufriesea surinamensis* (Linnaeus). His information is summarized below in the section on Mode of Parasitism.

Exaerete smaragdina (Guérin-Méneville) has a broad geographic range from Mexico to northern Argentina (Moure, 1967). It has been reported to parasitize *El. nigrita* (Ducke, 1903; Moure, 1946; Pereira-Martins, 1991) and *Ef. surinamensis* (Dodson and Frymire, 1961). Studies on seasonal abundance and species richness of male euglossine bees in forest fragments in the state of São Paulo, Brazil, have demonstrated that *Ex. smaragdina* is active during the hot and wet season (September–April), although it is most abundant from January to March (Rebêlo and Garófalo, 1991, 1997).

In this paper we present information on the parasitic behavior of *Ex. smaragdina* in nests of *El. nigrita*. *Eulaema nigrita* nests communally, with nests found in preformed cavities. Cells, arranged in clusters, are more or

less vertical and have walls constructed from excrement, mud, and resin. Its nesting behavior has been thoroughly described by Zucchi et al. (1969b) and Santos and Garófalo (1994).

Recoveries of immatures of *Ex. smaragdina* from host cells permit us to record the anatomy of the larval instars of this cleptoparasitic genus for the first time.

The first author (CAG) was responsible for gathering and reporting the biological observations presented here. The second author (JGR) investigated the oocytes and ovaries of preserved females and interpreted and described the larval anatomy. Together we developed the conclusions regarding the mode of parasitism presented in the Discussion of Behavioral Observations.

MATERIALS AND METHODS

The observations were carried out on the campus of the University of São Paulo, Ribeirão Preto, State of São Paulo, Brazil, in October 1997, from October 1998 to January 1999, and from March to April 2000. Time is recorded on a 24-hour basis.

Of the six nests of *El. nigrita* attacked, three (N-1, N-2, and N-3) had been established inside hollow cement bricks (inside dimensions 9.0 × 10.5 × 14.0 cm) forming a wall (11.0 m long and 0.7 m high) built in front of a small sloping earth bank (1.8 m high). The bees entered circular holes about 2.0 cm in diameter, situated 10.0, 12.0, and 22.5 cm above the ground. These holes and 58 others had been made to attract *Eulaema* females to nest in those cavities. Activities of the bees within the nests were observed using an otoscope. Nests 1, 2, and 3 were collected 14, 2, and 1 day, respectively, after the parasite attacks had been observed. In the laboratory, cell clusters from N-1 and N-2 were placed in wooden boxes covered with glass plates, and they were observed daily until the adults emerged. All the sealed cells from N-3 and one cell from N-2 were opened to verify parasite egg placement.

The other three nests, N-4, N-5, and N-6, were maintained in observation boxes in the laboratory. Each consisted of a wooden box (inside dimensions 19.5 × 19.5 × 11.5 cm) with the bottom filled with soil, the top covered with a glass lid, and with a 2.0-cm circular hole on one side. The bees were allowed to leave the boxes freely through plastic tubes connecting the boxes to the outside through holes in the laboratory wall. Observations of bee activity in the nests were made through the glass. Daily recordings were made of the number of *Eulaema* females working in each nest, the number of sealed cells, and the number of cells being constructed or provisioned.

The most detailed observations of adult *Ex. smaragdina* behavior were made in March and April 2000 when parasites attacked N-6. This nest's development was interrupted when the *Eulaema* females were collected after they oviposited into 29 cells. Thirteen of those 29 cells were opened to gain more information on the parasite egg placement within each cell and to obtain material to describe larval anatomy of the different instars.

The female cleptoparasites were observed (N = 51 occasions) at the study area between 7:30 and 12:00 hr. Usually only a single parasite was seen at a time, but the presence of at least two was recorded on five occasions. While searching for host nests, the parasites flew quickly over the surface of the bank, inspecting the cracks and holes in the soil and also holes in the bricks and the laboratory wall. During these inspections, the parasite hovered in front of holes for several seconds. The parasites were never observed entering N-1, N-2, and N-3 while a host female was present (N = 3). On 10 occasions in N-6 (in an observation box in the laboratory), the parasites entered and left rapidly, seemingly because a host female was present. On nine other occasions, the parasites hovered in front of the entrance of N-6 and then flew away, even though no host female was in the nest on seven of those instances.

RESULTS OF BEHAVIORAL OBSERVATIONS

Nest-Searching Behavior

During the days while N-6 was being observed, the parasites were seen on three occasions sitting on the laboratory wall 15–20 cm from the nest entrance; they remained motionless, pointed toward the nest entrance.

TABLE 1
Nest Contents of *Eulaema nigrita* when Attacked by *Exaerete smaragdina*

Nest code	Date of attacks	Cells			*Eulaema* females in nest
		Sealed	Being provisioned	Under construction	
N-1	28 Oct 97	6	2	0	2
N-2	14 Oct 98	7	3	0	3
N-3	5 Nov 98	5	2	0	2
N-4	4 Dec 98	12	3	0	3
N-5	20 Jan 99	6	3	0	4
	21 Jan 99	7	2	0	4
N-6	31 Mar 00	1	2	0	4
	1 Apr 00	3	0	4	4
	11 Apr 00	18	2	3	5
	12 Apr 00	18	5	0	5
	13 Apr 00	21	2	3	5
	14 Apr 00	23	3	2	5

On the first occasion (April 13, 2000), a parasite sat on the laboratory wall at 9:55 hr; at 10:10 hr a host female returned from the field with larval food and hovered some seconds 10–15 cm from the laboratory wall, apparently perceiving the presence of the parasite. The host female entered the nest, discharged the larval food into her cell, and stayed in the nest. At 10:34 hr another host female returned to the nest bringing larval food, entered the nest, discharged the food into her cell, and left. At 10:39 hr another host female returned from the field with larval food, and, after she entered the nest, the parasite flew to the front of the nest entrance, hovered some seconds, then flew away. At 11:00 hr a female parasite entered N-6 and left rapidly, presumably because there were two host females in the nest. At 11:13 hr those two females left the nest, and at 11:20 hr a parasite entered the nest and attacked three cells. On two other occasions (April 14 and 15), female parasites remained poised on the laboratory wall less than 2 minutes; in one of these instances, the parasite flew away when a host female returned to her nest.

The attacked nests were in different stages of development. Nests 1 and 3 were being reused for the first time, and the other nests were being reused the second time. When the attacks occurred, the number of active females in nests ranged from two to five (table 1).

Parasite Behavior in the Nest

On 21 occasions the parasites entered the nests while the host females were foraging. In four of these instances the parasites entered N-5 (one instance) and N-6 (three instances) but did not attack any cell; the parasites inspected the cells for 158, 259, and 180 (two instances) seconds and flew away. On two other occasions the parasites left the nest after a host female entered; in these instances the parasites remained in the nest less than 120 seconds. On three occasions two parasites entered a nest. In one of these instances, 2 minutes after a parasite entered N-5, another parasite entered. Apparently, one parasite disturbed the other, and both quickly departed. On another occasion, the second parasite entered N-6 some seconds after the first one; one of them rapidly left, and the other continued inspecting cells, even though a host female was present; this parasite flew away 8 minutes later when a host female returned.

Aggressive interactions between parasites were observed only once (April 12, 2000).

At 10:56 hr, as a parasite was introducing the apes of her metasoma into a cell, another parasite entered the nest; when the two met, they grappled furiously, and both fell to the observation box floor. The parasites remained clinched in a head-to-tail position until 11: 15 hr. During this time the parasites rolled intermittently on the floor, each apparently trying to sting or to get away from the other. Between bouts, they remained relatively quiet. No damage to either was detectable after each struggle. After separating, one of the parasites quickly left the nest; the other went to the cell that was being parasitized, oviposited in it, attacked two other cells, and flew away when a host female returned from the field.

Parasite Behavior During Cell Opening, Oviposition, and Cell Closing

Immediately after entering a host nest, a parasite begins to search for a suitable cell to parasitize. She walks on the cells while exhibiting an excited behavior, vibrating her wings for 2–4 seconds, repeatedly, and touching the cells with her antennae. During the attacks, the cells chosen by the parasite were usually those in which an egg had been laid a day before (N = 13) by *Eulaema* females (table 2). But in the absence of such cells, or when they were few, the parasite attacked cells that had received host eggs 2 (N = 6), 3 (N = 2), 5 (N = 2), and 6 days (N = 2) before (table 2). Some of these cells had already been parasitized one or more times, as observed in N-3 (all cells; see section on Egg Placement and Number of Eggs per Host Cell) and N-6 (cells 19 and 22; table 2). The time spent by the parasite to find a suitable cell ranged from 83 to 296 seconds (N = 7) and was significantly correlated with the number of sealed cells in the nest ($r = 0.834$; $P < 0.05$).

Once the cell is chosen, the parasite makes an opening on the lateral wall above the surface of the provisions. To do so, she removes bits of cell wall with her mandibles and attaches them to the inside of a cell that is being provisioned. This behavior continues until the size of the hole permits the parasite to introduce the apex of her metasoma into the cell. The time spent by parasites to open cells from 1 and 2 days after oviposition by host females ranged from 94 to 335 seconds ($x = 156.9 \pm 59.6$ sec; N = 18), significantly shorter than the time spent to open the older cells (range: 185–214 seconds; $x = 193.7 \pm 17.3$ sec; N = 6) (Mann-Whitney test, $Z = 2.50$; $P < 0.05$) (table 2). While ovipositing, the parasite stays motionless with her head directed downward. The time from metasomal apex insertion to apex withdrawal ranged from 24 to 115 seconds ($x = 49.1 \pm 23.4$ sec; N = 24) (table 2). After ovipositing, the parasite removes her metasomal apex from the cell and immediately begins to close the hole. She goes to a cell being provisioned and, with her mandibles, collects bits of cell collar, made of excrement and resin. She returns to the attacked cell and, with her mandibles, fills the hole with the collected material (fig. 1). In some instances, the material was taken from the building materials stored on the nest floor. The time spent to close the hole ranged from 87 to 310 seconds ($x = 177.5 \pm 73.1$ sec; N = 19) (table 2).

On seven occasions the parasites left the nests after attacking one (N = 3), two (N = 2), or three cells (N = 2). On seven other occasions their activities were interrupted by the return of host females; in these instances, the parasites flew away after attacking one (N = 4), two (N = 1), and three cells (N = 2) (table 2). On two occasions (April 12 and 14) N-6 was attacked by a parasite, which flew away when a host female returned, and then was attacked again the same day. We do not know if the second attack was by the same individual. When the parasites flew away before a host female returned from the field, the parasites remained in the nests from 592 to 2160 seconds (N = 7), a time significantly correlated with the number of cells attacked ($r = 0.834$; $P < 0.05$).

How is it possible for the parasite to recognize and attack only cells that contain live or dead (in the instances of multiple parasitism) host eggs? One possible explanation may be that they detect the presumably softer and more easily punctured cell walls of fresher cells; certainly such cells take them less time to open, as indicated above.

We have no direct evidence that the adult parasite kills early larval instars of the host. However, the first instars of many nonpara-

TABLE 2

Observations of 14 Attacks by *Exerete smaragdina* on Nests of *Eulaema nigrita:* Dates of Oviposition by *El. nigrita* and of Attacks by *Ex. smaragdina;* Timed Activity (in seconds) Includes Time Parasite Occupied Nest and the Following Behaviors: Cell Opening, Oviposition, and Cell Closing (Attacks separated by horizontal lines. For analysis, see text.)

Nest code	Cell no.	Oviposition dates	Attack dates	Duration of parasite activity (sec)			
				Parasite in nest	Opening cell	Ovipositing	Closing cell
N-1	6	27 Oct 97	28 Oct 97	900	?	?	?
N-2	7	9 Oct 98	14 Oct 98	840	185	115	224
N-3	3	30 Oct 98	5 Nov 98		204	68	96
	4	2 Nov 98	5 Nov 98		189	54	118
	5	3 Nov 98	5 Nov 98	2160	157	75	109
N-4	10	3 Dec 98	4 Dec 98		179	48	111
	11	3 Dec 98	4 Dec 98		148	60	92
	12	3 Dec 98	4 Dec 98	1320	120	33	87
N-5	3	15 Jan 99	20 Jan 99	438[b]	166	75	80[a]
	7	20 Jan 99	21 Jan 99	490[b]	152	29	103[a]
N-6	1	29 Mar 00	31 Mar 00	592	161	86	225
	2	31 Mar 00	1 Apr 00		259	58	260
	3	31 Mar 00	1 Apr 00	1434	335	76	247
	18	10 Apr 00	11 Apr 00	480[b]	114	46	116[a]
	17	10 Apr 00	12 Apr 00	480[b]	117	44	104
	15	9 Apr 00	12 Apr 00		214	42	302
	11	6 Apr 00	12 Apr 00		204	37	161
	16	10 Apr 00	12 Apr 00	2100[b]	170	47	310
	22[c]	12 Apr 00	13 Apr 00		111	25	205
	21	12 Apr 00	13 Apr 00		140	25	127
	19[d]	12 Apr 00	13 Apr 00	1308[b]	182	28	92[a]
	22[c]	12 Apr 00	14 Apr 00		109	27	221
	20	13 Apr 00	14 Apr 00	804[b]	102	35	45[a]
	19[d]	12 Apr 00	14 Apr 00		175	24	211
	23	13 Apr 00	14 Apr 00	1616	94	29	163

[a] Activity interrupted by return of host female from field.
[b] Duration in nest interrupted by return of host female.
[c] Cells attacked twice.
[d] Cells attacked twice.

sitic Apidae (as well as of *Ex. smaragdina,* see below) remain more or less surrounded by the chorion for most of the stadium, if not the entire stadium, as observed in *Eulaema polychroma* (Mocsáry) (see Remarks under the description of the second instar of *Ex. smaragdina*). Pharate first instars are easily confused with eggs. Hence, we are uncertain whether the cells attacked 5 and 6 days after host oviposition actually contained host eggs or pharate host first instars.

Interactions Between Host and Parasite

On two occasions a host female returned to a nest occupied by a parasite. One of the

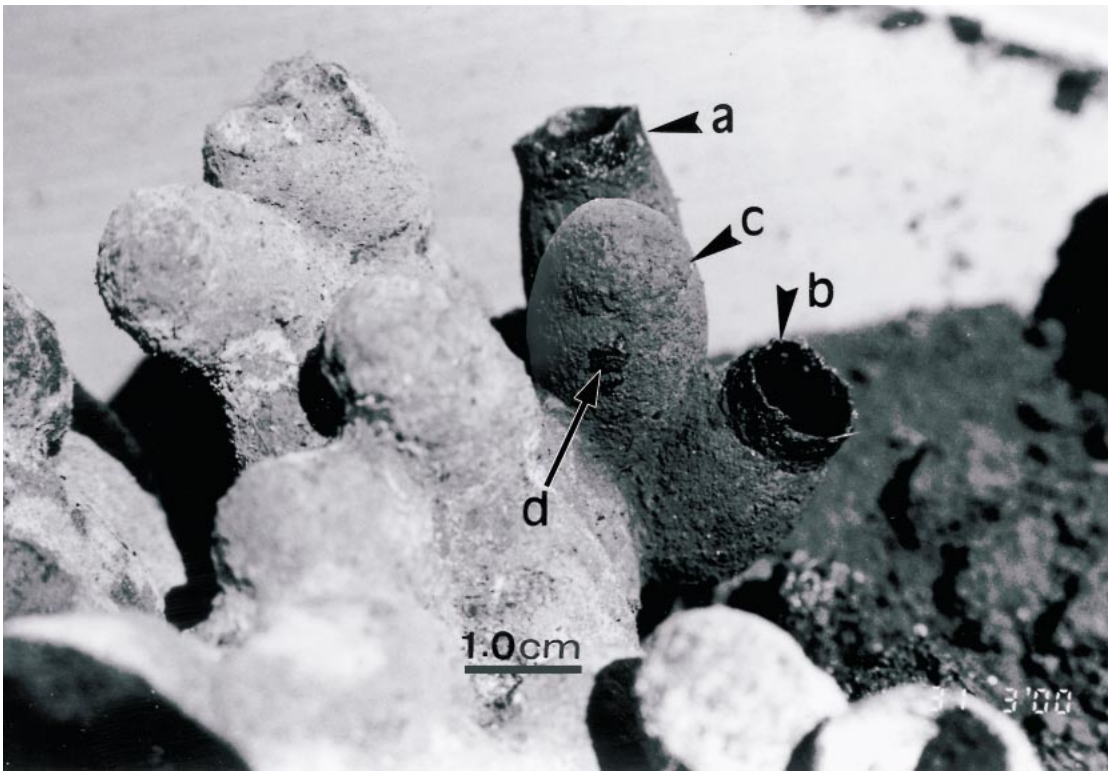


Fig. 1. Cells of *Eulaema nigrita* being provisioned (a and b) and parasitized (c) by *Exaerete smaragdina*; the arrow (d) indicates the plugged scar made by *Exaerete smaragdina.* The new cells (a, b, and c) were attached to one cell of the older cell cluster built during the first re-use process of N-6.

host females was bringing larval food, and the other was carrying construction material. Apparently, the host females did not detect the presence of parasites, one of which was ovipositing in a cell and the other was trying to open a cell. After depositing the larval food and construction material, the host females walked on the cell clusters where they met the parasites. In both instances, the parasites withdrew from the host females and quickly left the nest. Sometimes while searching for a cell, the parasite met the host female, withdrew from her, and left the nest. The parasites never showed any aggressive behavior toward the host females and vice versa.

Egg Placement and Number of Eggs per Host Cell

Parasite eggs are easily distinguished from host eggs because those of *Ex. smaragdina* are shorter and narrower than those of *Eulaema* (fig. 2). Both host and parasite eggs were laid on the top of the food mass, those of *Eulaema* at the center, while those of *Exaerete* were laid near the cell wall.

When the 13 cells from N-6 were opened, 5 contained eggs and the others contained larvae. Four of the five cells with eggs held a single *Exaerete* egg in addition to the host egg. The fifth cell, which had been observed being parasitized twice (cell 19; see table 2), contained the host egg and two eggs of *Exaerete,* although one was dead. Of the cells with eggs, the host eggs in four of them were dead, and the host egg in the fifth cell shriveled 4 days after the cell was opened. These data, as well as of those in the following paragraph, have a direct bearing on the mode of cleptoparasitism as explained below.

All five sealed cells from N-3 had been parasitized (fig. 2). The oldest cell had a mid-

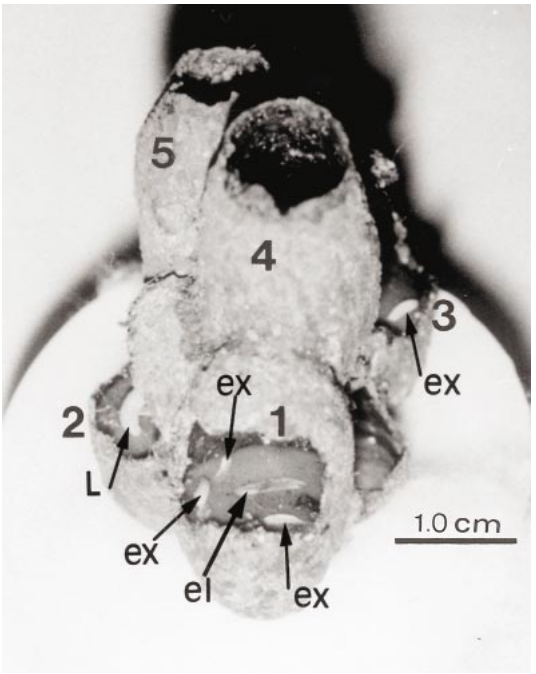


Fig. 2. Cell cluster of *Eulaema nigrita* from N-3. Cell 1 shows the position of eggs of *Exaerete smaragdina* (ex) and *Eulaema nigrita* (el) on the food mass. Cells 2 and 3 show one larva (L) and one egg of *Exaerete smaragdina,* respectively. Cells 4 and 5 were being provisioned.

sized parasite larva; three cells each had four *Exaerete* eggs and the host egg; and one cell had five *Exaerete* eggs and the host egg. In these cells all but one host egg was dead. In the cells with four *Exaerete* eggs, each of them had one dead parasite egg. In the cell with five *Exaerete* eggs, two were dead. In three cells one *Exaerete* egg had one end leaning on another egg. In the cell with five eggs, two of them had been placed on other eggs. Only one larva hatched in each cell; none completed development.

Parasitism Rate and Egg-to-Adult Periodicity

In N-4 no individual, host or parasite, emerged because larvae of an unidentified species of Lepidoptera consumed the contents of all the brood cells. In the other five nests the rates of parasitism by *Ex. smaragdina* ranged from 11.1% to 100.0% (table 3). Two additional parasites, the meloid beetle *Meloetyphlus fuscatus* Waterhouse and the bombyliid fly *Anthrax* sp., were reared from N-5 (table 3).

In N-3 and N-5, which were active during the hot, wet season (September to March), the time between observed attacks and emergence of *Exaerete* adults was 70, 71, and 73 days for males (N = 3) and 73 days for females (N = 2). On the other hand, the attack-to-emergence period for N-6, which was active in the early cool, dry season, was 118, 127, and 170 days for males and 172 and 181 days for females. Thus, the length of the egg-to-adult period of *Ex. smaragdina* is affected by climatic conditions, in that different values were obtained for different seasons, as was reported by Garófalo (1985) for *Euglossa cordata* (Linnaeus), Santos and Garófalo (1994) for *El. nigrita,* and Garófalo et al. (1998) for *Euglossa annectans* Dressler.

DISCUSSION OF BEHAVIORAL OBSERVATIONS

Nest-Searching Behavior

The flight activity exhibited by *Ex. smaragdina* at the nesting area of *El. nigrita* was similar to that reported by Bennett (1972) for one or more females of an unidentified species of *Exaerete* visiting a nest of *Eulaema meriana* (Olivier) (as *El. terminata* (Smith)). The morning visitation by the parasites to the host nests increases the possibility of successful attacks, because during that time the foraging activities of host females are more intense (Bennett, 1972; Garófalo, unpubl. data). Thus, the probability of a host female being in her nest in the morning is lower than that in the afternoon when she tends to spend more time within her nest ovipositing (Santos and Garófalo, 1994).

As related by Bennett (1972) and observed in this study, the parasites on several occasions approached but did not enter the host nest, and other times they entered only to leave quickly without ovipositing. In most instances these behaviors by *Ex. smaragdina* occurred when at least one host female was in the nest, indicating that the parasite avoids entering the nest when a host female is present. Thus, the behavior of perching on some substrate in the vicinity of nest, as reported by Bennett (1972) and observed in this study,

TABLE 3
Parasitism Rates by *Exaerete smaragdina* in Five Nests of *Eulaema nigrita*

	Nest code				
	N-1	N-2	N-3	N-5	N-6
Number of sealed cells	8	9	5	29	29
Cell contents destroyed by molds	5				
Dead immatures				6	3
Emerged hosts					
Females		5		9	3
Males	2	3		6	
Emerged parasites					
Exaerete					
Females				1	2
Males		1		3	3
Meloetyphlus fuscatus (Meloidae)				1	
Anthrax sp. (Bombyliidae)				1	
Dead parasites					
Exaerete					
Females	1			1	2
Males				1	4
Opened and parasitized cells[a]			5		12
Parasitism by *Exaerete*[b]	33.3%[c]	11.1%	100.0%	26.1%[d]	88.5%[d]

[a]Cells containing eggs or larva of *Exaerete*.
[b]Including emerged and dead individuals.
[c]Excluding cells with mold.
[d]Excluding dead immatures.

and apparently watching for host females entering and leaving the nest may ensure the host female's absence during the parasite's visit to the nest. This behavior has been observed in several other parasitic bees such as *Coelioxys sodalis* Cresson (as *ribis* Cockerell) (Megachilidae) (Graenicher, 1927), *Coelioxys octodentata* Say (Michener, 1953b), *Coelioxys flagrata* Baker (Bohart and Youssef, 1972), *Stelis lateralis* Cresson (Michener, 1955), *Leiopodus singularis* (Linsley and Michener) (Apinae: Protepeolini) (Rozen et al., 1978), and *Holcopasites ruthae* Cooper (Nomadinae) (Danforth and Visscher, 1993).

Mode of Cleptoparasitism

Mode of cleptoparasitism refers to the method by which a cleptoparasite introduces her egg into the host cell and the method by which the host offspring is killed, allowing the parasite offspring to develop on the provisions originally supplied for the host's larva (Rozen, 1991). Because cleptoparasitism has evolved de novo at least 27 times among bees (Rozen, 2000), it is not surprising that various mechanisms for egg introduction and host elimination have been detected. Also not surprising is the discovery of evolutionary parallelisms in these activities among various lineages. Always interesting are the investigations, such as this one, that encounter suspected new modes of parasitism. Some apparent novelties in the parasitism by *Ex. smaragdina* are discussed below.

As described above, the female of *Ex. smaragdina* makes a small hole in the cell wall above the level of the provisions, inserts the tip of her metasoma, deposits an egg through the hole, and then reseals the opening. Information recorded in the section on Egg Placement and Number of Eggs per Host Cell indicates that N-3 and N-6 together contained seven dead host eggs and only two live host eggs in cells that also contained eggs of *Ex. smaragdina*. In each of the five cells containing more than one parasite egg, one parasite egg was killed in four of the cells, and the other cell contained two para-

site eggs that had been killed. These statistics strongly suggest that the dead host and parasite eggs had been killed by an attacking cleptoparasite at the time she oviposited. Since only the apex of her metasoma penetrates the cell lumen, the implication is that the parasite uses her sting. A videotape of a female attacking a cell taken outside the cell is consistent with this inference, although there were no views of the cell interior. If we are correct, then this is the first known instance in which a cleptoparasitic female eliminates the host offspring with her sting. With all other cleptoparasites where the female kills the host offspring (*Sphecodes*: Bohart, 1970; Rozen, 2000; *Hoplostelis bilineolata* (Spinola): Bennett, 1966; Augusto and Garófalo, 1998; and *Ex. dentata*: Bennett, 1972) she is thought, or is known, to do so with her mandibles. *Exaerete smaragdina* is also the first known cleptoparasite to use two methods for eliminating hosts, in that we interpret the mandibular morphology of the second instar to be hospicidal (see description below and figs. 8–11).

Dr. Charles D. Michener, who kindly reviewed the manuscript, pointed out that the terminal part of the female metasoma in this genus is narrow and is inserted into the host cell during egg laying. Thus, it might damage the host egg without stinging. Before drafting this section, JGR had noted that the metasomal apex of the female of *Ex. smaragdina* is more attenuated than that of *Ex. frontalis* (Guérin-Méneville) or *Ex. dentata.* He attributed this to the fact that the female of *Ex. smaragdina* inserts her metasomal apex through a small hole in the cell wall, not through a large hole at the top of a cell, as does *Ex. dentata* (see below). Furthermore, the setae of sternum 6 were, if anything, slightly weaker than those the other species and therefore less effective for injuring host eggs. After receiving Michener's comments, CAG reviewed the videotapes and reevaluated the size of the hole. We both think that Michener's idea may be correct, but that our explanation is more probable.

Why two methods of killing the host offspring should have evolved is unclear. However, there is evidence that assassination by stinging is uncertain in that one host egg remained alive in N-6, as did one host egg and eight parasite eggs in N-3. If stinging were known to be the primary means of eliminating host offspring, then hospicidal second instars would be a logical backup system. Perhaps when other species of *Exaerete* are studied, they will shed light on this matter.

A consequence of the female parasite using her sting (or, for that matter, metasomal apex) to kill the host egg is that any parasite egg deposited in the cell before an attack is also at risk. This is evidenced by the fact that attacking *Exaerete* presumably had killed 5 of the 17 parasite eggs in N-3. With other parasitic bees having hospicidal early instars, one assumes that the first parasite egg to be deposited in a cell with more than one parasite egg will have the greatest chance of survival; the first larva to hatch can kill all competitors before they eclose. The situation for *Ex. smaragdina,* however, is different; statistics from N-3 indicate that in instances of multiple parasitism, the parasite eggs deposited earlier are in jeopardy.

Comparison with *Exaerete dentata*

The method by which *Ex. smaragdina* introduces her egg into the host cell and kills the host egg differs from the behavior of *Ex. dentata* regarding these matters, as described by Bennett (1972). He reported that the female of *Ex. dentata* made a hole in the cell cap large enough so that she could introduce her head and part of her mesosoma, enabling her to reach in, remove the host egg, and crush it with her mandibles. She then reversed her position, inserted the tip of her metasoma into the cell, and after several moments extended her metasoma farther to deposit an egg. Afterward she resealed the cell. This contrasts with the female *Ex. smaragdina* making only a small hole in the sidewall of the cell and inserting her metasomal apex presumably to sting the host egg at the same time she deposits her own egg.

These two species of *Exaerete* share two similarities. First, they attack only closed host cells (i.e., not cells still being provisioned and without host offspring). Second, the hole made for oviposition is resealed with bits of material removed from the cell cap in the case of *Ex. dentata,* and, in the case of *Ex. smaragdina,* removed from the cell col-

lar, from a cell in the process of being constructed, and/or from stored construction material.

With most cleptoparasitic bee taxa, one assumes that all species within a clade will have the same mode of cleptoparasitism because once a successful mode has been established in a cleptoparasitic lineage, there is no apparent reason for selection pressure to develop another mode. Certainly this seems to be the case in the Nomadinae, the largest taxon of cleptoparasitic bees (Rozen, 1991); in all known species, a female enters the host cell while it is being provisioned, hides her egg by imbedding it in the cell wall, and then departs. When the egg hatches, a small hospicidal first instar emerges to kill the host offspring. Hence, the detection of two modes of cleptoparasitism in *Exaerete* (killing with female's mandibles vs. stinging plus hospicidal second instar) is noteworthy.

Multiple Parasitism

As observed in this study, *Ex. smaragdina* females may attack more than one cell during a visit to the host nest, and only a single egg is laid in each of them. Thus, multiple parasitism of host cells, as observed in N-3 and N-6, reflects the frequency of attacks on those cells. It may suggest that *Ex. smaragdina* females are unable to recognize cells that have been parasitized either by other individuals or by themselves on previous visits. Alternatively, it may suggest that, in the case of this species, there is a selective advantage not to be the first parasite to oviposit in a cell, as discussed above. After locating a host nest, *Ex. smaragdina* females return to it several times to parasitize the new cells as they are completed, as observed in N-5 and N-6. In the absence of new cells or when they are few, the parasites attack cells that have been parasitized before, as occurred in the cells from N-3 and in two cells from N-6. These periodic revisits to the nest lead to high rates of parasitism, as observed in four of the five nests analyzed here and as also reported by the following authors: Moure (1946), 50.0% parasitism by *Ex. smaragdina* in one nest of *El. nigrita*; Zucchi et al. (1969b), 33.3% parasitism by *Ex. dentata* in one nest of *Eufriesea auriceps* (Friese); and Ackerman and Montalvo (1985) and Roubik (1990), 76.0% and 27.3% parasitism, respectively, in nests of *El. meriana*, all by *Exaerete frontalis*. Nest-location learning behavior has also been reported in *Epeolus minimus* (Robertson) (Graenicher, 1906), *Nomada opacella* Timberlake (Linsley and MacSwain, 1955), *Leiopodus singularis* (Rozen et al., 1978), *Melecta separata callura* (Cockerell) (Thorp, 1969), and *Holcopasites ruthae* (Danforth and Visscher, 1993), and according to Rozen et al. (1978), it may be widespread among parasitic bees.

Effect of Host Females on Parasitism Rate

As observed in this study, the mere presence of one host female in the nest prevented attacks by *Ex. smaragdina*. Therefore, it could be hypothesized that the more females sharing the nest, the higher the probability of one of them being present and, consequently, the lower the probability of the nest being attacked. This, however, did not occur, as evidenced by the fact that nests containing two and three host females (N-1, N-2, and N-3) had a combined parasitism rate of 41.2%, while nests of 4 and 5 females (N-5 and N-6) had a combined parasitism rate of 61.7%. Thus, the suggestion that one advantage of communal nesting, as occurs in *El. nigrita* (Santos and Garófalo, 1994), could be improved defense against parasites that must enter the nest to parasitize the immatures (Lin and Michener, 1972; Michener, 1974; Abrams and Eickwort, 1981; McCorquodale, 1989; Garófalo et al., 1992) is not corroborated by the present study. This may be due to the absence of guarding behavior in *El. nigrita* (Santos and Garófalo, 1994; Garófalo, personal obs.), because the females do not show aggressive behavior toward the parasites and also because the parasites visit the host nest frequently, seeking an opportunity to attack the cells.

FUTURE BEHAVIORAL STUDIES

The presence of two modes of parasitism within one genus should be confirmed. It is possible, but unlikely, that we have misinterpreted the hospicidal nature of the second instar of *Ex. smaragdina*. Observing a live lar-

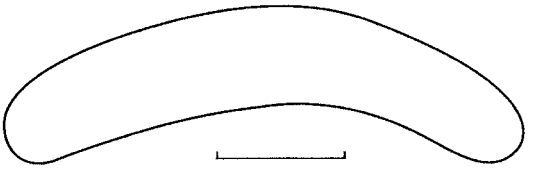


Fig. 3. *Exaerete smaragdina,* mature oocyte, anterior end to the left. Scale = 1.0 mm.

va in the act of killing a host or conspecific egg could easily settle this matter. Assuming our conclusions are correct, then an investigation into the biology of other species presents a fascinating opportunity to explore the evolution of the modes in the genus, particularly in light of Engel's (1999) cladistic study of euglossine phylogeny.

MATURE OOCYTES AND EGG INDEX OF *EXAERETE SMARAGDINA*

Two females of *Ex. smaragdina,* preserved in Kahle's solution after being taken from a *Eulaema* nest, were dissected to examine their ovaries and oocytes. Both had four ovarioles per ovary (i.e., ovarian formula 4: 4), the number typical for most Apidae and not a higher number as exhibited in the Nomadinae (Alexander, 1996) and a few other apid taxa. The maturity of oocytes in these specimens was difficult to evaluate because follicular tissue adhered closely to the delicate chorion. One specimen contained four large oocytes 3.70–3.85 mm long. Two of these appeared to a have partially developed chorions, and one, described below, was thought to have a fully developed chorion. The other specimen contained two large oocytes, each 4.0 mm long, one lacking a chorion, the other possibly developing a chorion.

DESCRIPTION OF MATURE OOCYTES (fig. 3): Size small (3.70 long, 0.8 mm maximum diameter) relative to distance between outer rims of tegulae (i.e., egg index 0.55). Shape (fig. 3) elongate, symmetrical around its curved axis, and with its anterior end slightly less pointed than the posterior end; maximum diameter near middle; posterior half tapering slightly; micropylar pore not evident. Color nearly white. Chorion smooth, lacking sculpturing and other ornamentation, presumably transparent.

REMARKS: The egg index is a measure of egg/oocyte size (length) relative to overall body size (distance between outer margins of tegulae) as defined by Iwata (1955) and Iwata and Sakagami (1966). More recently this index has been modified by Alexander and Rozen (1987) so as not to distinguish Iwata's category A and B oocytes. However, here we have judged the maturity of the oocyte by the development of the chorion; the only clearly mature oocyte was 3.70 mm long. If we had taken the largest of the four large oocytes (thus adhering to the Alexander/Rozen modification), the index would have been 0.58 for that female. For the other female, the index would have been 0.60, with the mean of the two being 0.59. The egg index is calculated by dividing the oocyte length by the intertegular distance of the female (both 6.7 mm) from which the oocyte comes.

Thus, *Ex. smaragdina* has small eggs, as is characteristic of most parasitic bees (e.g., Alexander, 1996). With this species, the value of small eggs may be twofold: it reduces the risk of the egg being killed by a subsequent visit of a female *Exaerete*; and several mature or nearly mature oocytes can be contained within the metasoma so that when a nest is found, the parasite can oviposit in more than one cell.

LARVAL INSTARS OF *EXAERETE SMARAGDINA*

Michener (1953a) pioneered modern-day comparative studies of bee larvae by describing preserved specimens available at the time and reviewing previously recorded accounts in the literature. Since most bees spend 10 months out of the year as mature larvae, his descriptions dealt almost exclusively with last larval instars. Accounts of earlier instars appeared sporadically in the literature (see for example Graenicher, 1905; Iwata, 1939) over a long period. However, comparative studies of early instars began in the mid-1900s. Many of these studies treated early instars of cleptoparasitic taxa whose early stage larvae exhibit striking adaptations that enable them to find and kill host larvae (e.g., Rozen, 1954; Michener, 1957; Lucas de Oliveira, 1966a; Torchio and Youssef, 1968). Other studies were of nonparasitic taxa, of other instars, and /or of all larval instars (e.g.,

Lucas de Oliveira, 1960, 1966b; Rozen, 1964, 1967; Baker, 1971). The following are taxonomic descriptions of the five larval instars of *Ex. smaragdina*, the first descriptions of any larvae of that genus.

First Instar

Figure 12

The following is based on exuviae of the first instar found partly still attached to the second instar, the specimen having been preserved while molting. Part of the chorion clung to the first instar, an indication that the first instar does not crawl from the egg but rather stays partly attached to the chorion until the second instar emerges, as has been observed in other taxa by Torchio (1989) and Alves dos Santos et al. (in prep.). The description is incomplete because the exuviae reveal only a limited number of larval features.

Diagnosis: The less apically attenuate mandibles (fig. 12) of the first instar and lack of dense ventral spicules on the body immediately distinguish the first instar from the second.

Head: Labrum faintly bilobed, perhaps faintly pigmented; labral sensilla not tuberculate like those of second instar. Mandible short, pointed apically, without attenuated apex like that of second instar, sclerotized and pigmented toward apex. Salivary opening a transverse slit, without lips.

Body: Integument without spicules but with narrow band of granules extending between most spicules along side of body, as described for *Tetrapedia diversipes Klug* (Alves dos Santos et al., in prep.).

Material Studied: One first instar partially cast exuviae with part of chorion attached, all attached to second instar, accompanied by rest of cast chorion, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, April 19, 2000 (C. A. Garófalo), N1/99, cell 23. *nigrita.*

Remarks: The function of the linear band of granules extending between some and perhaps all spiracles (also observed in first instar *Tetrapedia diversipes*) is unknown. That it occurs in two taxa not particularly closely related suggests that the phenomenon might be found elsewhere in the Apidae or even the Apoidea. These bands appear where the chorion first splits during eclosion, thus suggesting a linkage with hatching.

Second Instar

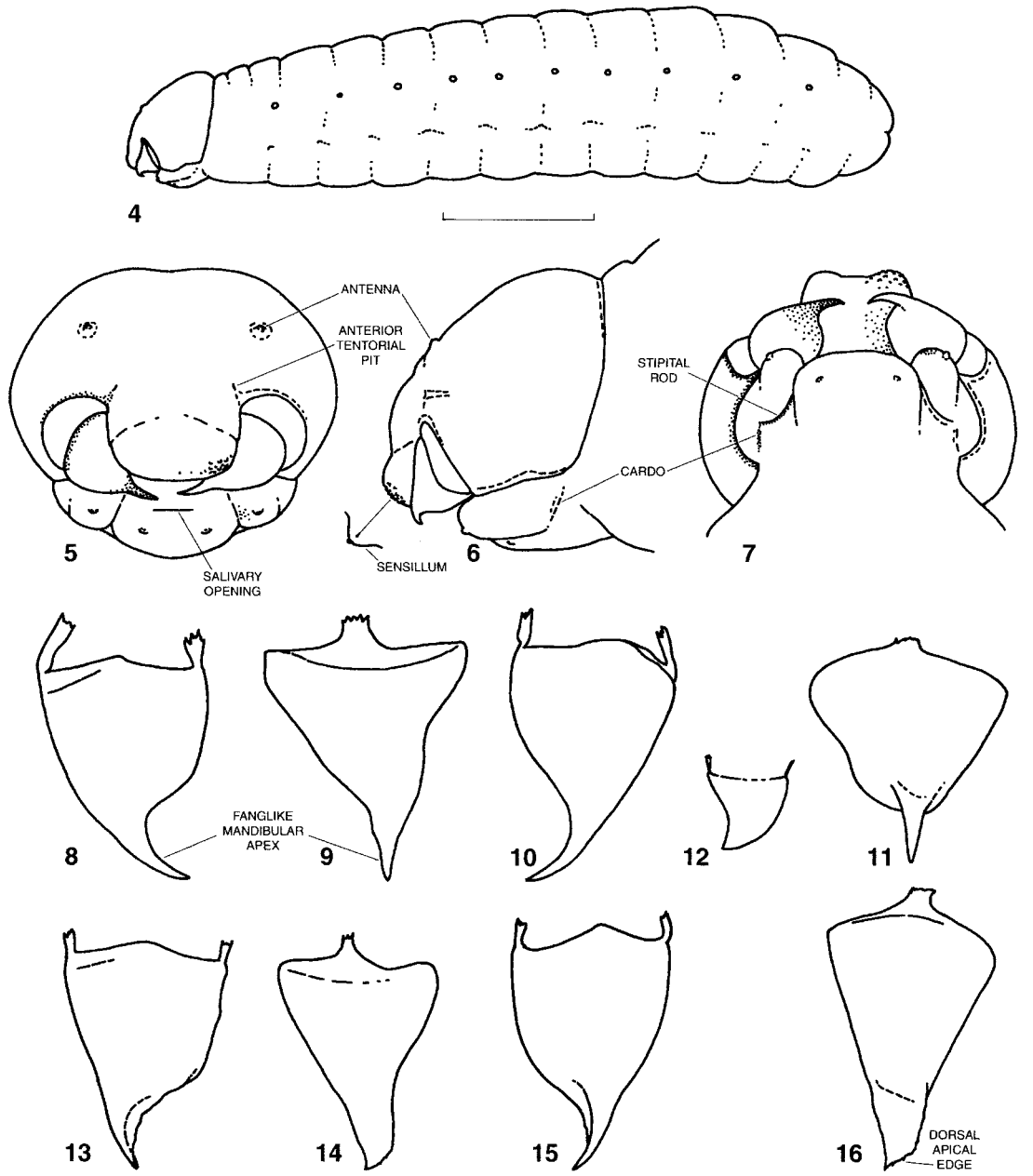
Figures 4–11

Diagnosis: The sharply pointed, apically sclerotized, fanglike mandibles of this instar will distinguish it from other instars of this species and presumably from all instars of the host. The much shorter, simple mandible of the first instar (fig. 12) contrasts with the more elongate mandible of the second instar (fig. 10). For a further discussion of the unusual shape of the second-instar mandible, see Remarks, below.

This is the only cleptoparasitic apid whose second instar is thought to be capable of killing the host egg or early instar, although the modes of parasitism of *Aglae* and the Osirini are unknown.

Length: About 4.0 mm; distance between antennal apices 0.375 mm (N = 1).

Head (figs. 5–7): Shape hypognathous; parietals neither swollen, elongate, or otherwise modified (figs. 5, 6), not usually constricted behind; sclerotization of parietals not extending below hypostomal ridge. Integument of head capsule moderately sclerotized, faintly pigmented except internal ridges tending to be darkly pigmented; labrum faintly pigmented; mandible moderately pigmented with apices more strongly sclerotized and pigmented; cardo faintly pigmented; stipital rod moderately pigmented. Head capsule with inconspicuous, nonsetiform sensilla, without spinulae (as defined by Melectini); labrum apicolaterally with numerous conspicuous, tuberculate, nonsetiform sensilla; dorsal surface of labium not spiculate; hypopharynx spiculate medially (not visible in fig. 5). Tentorium complete including dorsal arms, moderately developed; posterior pit in normal position; coronal ridge vague; other internal head ridges moderately developed except epistomal ridge between anterior tentorial pits absent. Parietal bands absent. Antennal prominence scarcely evident; antennal papilla poorly differentiated from disc, moderately projecting, bearing approximately 3–5 tuberculate sensilla (fig. 6). Labral sclerite absent; labrum moderately broad, broadly



Figs. 4–11. *Exaerete smaragdina*, second instar. **4.** Entire body, lateral view. **5.** Head, frontal view. **6.** Head, lateral view, showing tuberculate labrum sensillum enlarged. **7.** Head, ventral view. **8–10.** Right mandible, dorsal, outer, and ventral surfaces, respectively. **11.** Right mandible, apex in maximum profile. Fig. 12. Same, first instar, right mandible, ventral view, drawn to same scale as ventral view of second-instar mandible, fig. 10. Scale (= 1.0 mm) refers to fig. 4. Figs. 13–15. *Eulaema polychroma*, second instar, right mandible, dorsal, outer, andventral views, respectively. Fig. 16. Same, right mandible with apex in maximum profile.

rounded apically, bearing rounded mound on each side but lacking paired tubercles; labral apex with numerous tuberculate sensilla; epipharyngeal surface finely spiculate.

Mandible (figs. 8–11) short compared with mandibles of many hospicidal early instars, robust at base, tapering rapidly to narrow, strongly curved apical region which tapers to attenuated, sharply pointed, fanglike apex; distinct cusp not defined. Maxillae and labium not greatly fused, each represented by apical projection. Cardo represented by pigmented sclerite; stipes a sclerotized pigmented rod extending along mesal surface of maxilla; articulating arm of stipes not evident; maxillary palpus evident but shorter than basal diameter; galea not evident; dorsal inner surface of apex with fine spicules. Labium apparently not divided into prementum and postmentum; labial apex not projecting quite as far as maxillary apices, bearing apical salivary slit; labial palpus evident, similar but less pronounced than maxillary palpus. Hypopharyngeal groove weakly or not evident; hypopharynx slightly projecting. Salivary opening a broadly transverse slit, without lips.

BODY (fig. 4): Form linear; intersegmental lines moderately incised; abdominal segments not divided into cephalic and caudal annulets; abdominal dorsal tubercles and lateral body swellings absent; neither prothorax nor abdominal segment 9 protruding ventrally; abdominal segment 10 apically attached to 9, without any apparent modification for crawling. Integument without setae, that of venter of each body segment densely spiculate; integument elsewhere less densely spiculate with some dorsal areas on anterior part of body apparently mostly nonspiculate. All spiracles present, apparently flush with body surface, subequal in size except those of metathorax smaller, about one-half diameter of other spiracles. Anal area an apical transverse depression.

MATERIAL STUDIED: Three second instars, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, April 19, 2000 (C. A. Garófalo), N1/99, cells 21, 22, 23.

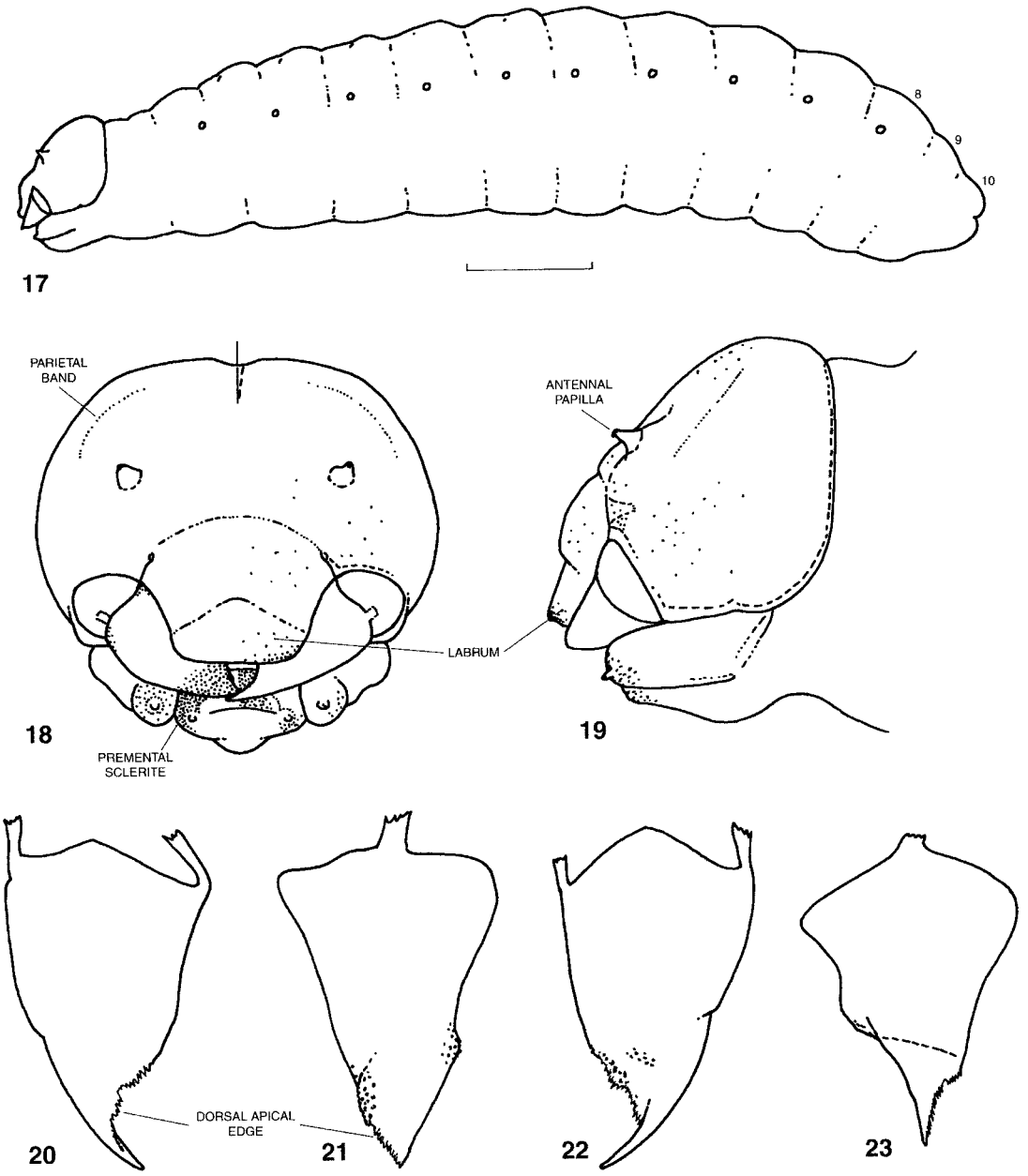
REMARKS: This is believed to be the only cleptoparasite belonging to the Apidae whose second instar is the hospicidal form; so far as we have been able to detect, the first instars of the following taxa are the primary hospicidal stage: Nomadinae, Isepeolini, Protepeolini, Melectini, Ericrocidini, Tetrapediini (*Coelioxoides*), and Rhathymini (first instars of *Aglae* and the Osirini unknown). Compared with the first instars of these cleptoparasites, the second-instar *Exaerete* is remarkable in that it exhibits so few modifications adapting it to hospicidal activity. It shows no modification of its cranium for projecting its mandibles forward or for increasing muscle attachment for stronger mandibles, no enhanced labral tubercles or elongate antennae for host detection, and no development of terminal pygopod-like structures for crawling in search of host immatures. If it were not for the modified mandibles, one would not suspect hospicidal activity of the second instar of this species.

To evaluate further the hospicidal anatomy of the second-instar mandible of *Ex. smaragdina,* JGR examined the mandible of the second instar of *Eulaema polychroma.* The specimen (Peru: Lima Dept., San Bartolomé, May 10, 1996 [J. G. Rozen, A. Ugarte, M. Laime]) had been collected from a communal nest with numerous immatures; its stage was determined by comparison with a first instar still partly surrounded by the chorion. The second-instar mandible (figs. 13–16) of this species is sharply pointed. However, in contrast with the fanglike mandibular apex (figs. 8–11) of *Ex. smaragdina,* its apex is asymmetrical (figs. 14, 16), basally broad in inner or outer view, minutely jagged along the dorsal edge as seen in maximum profile (fig. 16), not strongly curved in dorsal and ventral views (figs. 13, 15), and with inner surface somewhat scoop shaped. If it represents the plesiomorphic condition in the Euglossini, then the curved, slender, smooth mandibular apex of *Ex. smaragdina* is derived, presumably for piercing the chorion and/or integument of host or rival.

THIRD INSTAR

Figures 17–23

DIAGNOSIS: The third instar can most easily be differentiated from the first by the much more pronounced antennal papilla, which is clearly longer than its basal diameter. Further, the mandibles are now clearly though mi-



Figs. 17–23. *Exaerete smaragdina*, third instar. **17.** Entire body, lateral view. **18.** Head, frontal view. **19.** Head, lateral view. **20–22.** Right mandible, dorsal, inner, and ventral views, respectively. **23.** Right mandible with apex in maximum profile. Scale (= 1.0 mm) refers to fig. 17.

nutely denticulate along the dorsal apical edge, unlike the smooth, fanglike mandibular apices of the second instar.

LENGTH: About 6.0 mm; distance between antennal apices M = 0.613, range 0.5875–0.625 mm (N = 3).

HEAD (figs. 18, 19): As described for first instar except for following: Apical part of maxilla faintly pigmented; premental sclerite faintly but distinctly pigmented. Some head sensilla finely setiform. Dorsal surface of labium spiculate. Coronal ridge well developed

but short; epistomal ridge between anterior tentorial pits not evident but fine external suture evident. Parietal bands faint but evident. Antennal papilla much more conspicuous than that of second instar, distinctly longer than basal diameter, well differentiated from disc. Labrum as seen from front with apicolateral angles moderately defined.

Mandible (figs. 20–23): with apical part less fanglike than in second instar, shorter in relation to mandible proximal to cusp; dorsal apical edge with numerous sharp, small teeth; ventral apical edge with several irregularities, too indistinct to be termed teeth; cusp moderately defined, separating apical, vaguely scooplike mandibular apex from robust mandibular base; ventral edge of cusp bearing patch of small rounded denticles. Articulating arm of stipes now evident. Maxillary palpus somewhat longer than basal diameter; galea faintly evident as low, sensilla-bearing mound. Labial palpus about as long as basal diameter. Hypopharyngeal groove well defined; hypopharynx strongly projecting. Salivary opening a broad slit about as wide as distance between labial palpi.

BODY (fig. 17): As described for second instar except for following: Form slightly less linear than that of second instar; posterior mid-body becoming slightly enlarged; body segments vaguely divided into cephalic and caudal annulets dorsally; venter of abdominal segment 10 slightly more elongate than dorsum of 10 (fig. 17). All spiracles subequal in size.

MATERIAL STUDIED: Two third instars, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, April 19, 2000 (C. A. Garófalo), N1/99, cells 11, 15; one third instar, same data except April 23.

FOURTH INSTAR
Figures 24–28

DIAGNOSIS: Except for the larger size and more pigmented head capsule, this instar is similar to the third. It can, however, be distinguished from the previous one by the emarginate apex of the labrum when viewed from the front and the development of the small subapical dorsal mandibular tooth.

LENGTH: About 10.0 mm; distance between antennal apices 0.80 mm (N = 1).

HEAD (figs. 25, 26): As described for third instar except for following: Sclerotized areas more darkly pigmented than in previous instar. Most head sensilla setiform. Antennal papilla more conspicuous than that of third instar. Apical edge of labrum as seen from front (fig. 25) now somewhat emarginate.

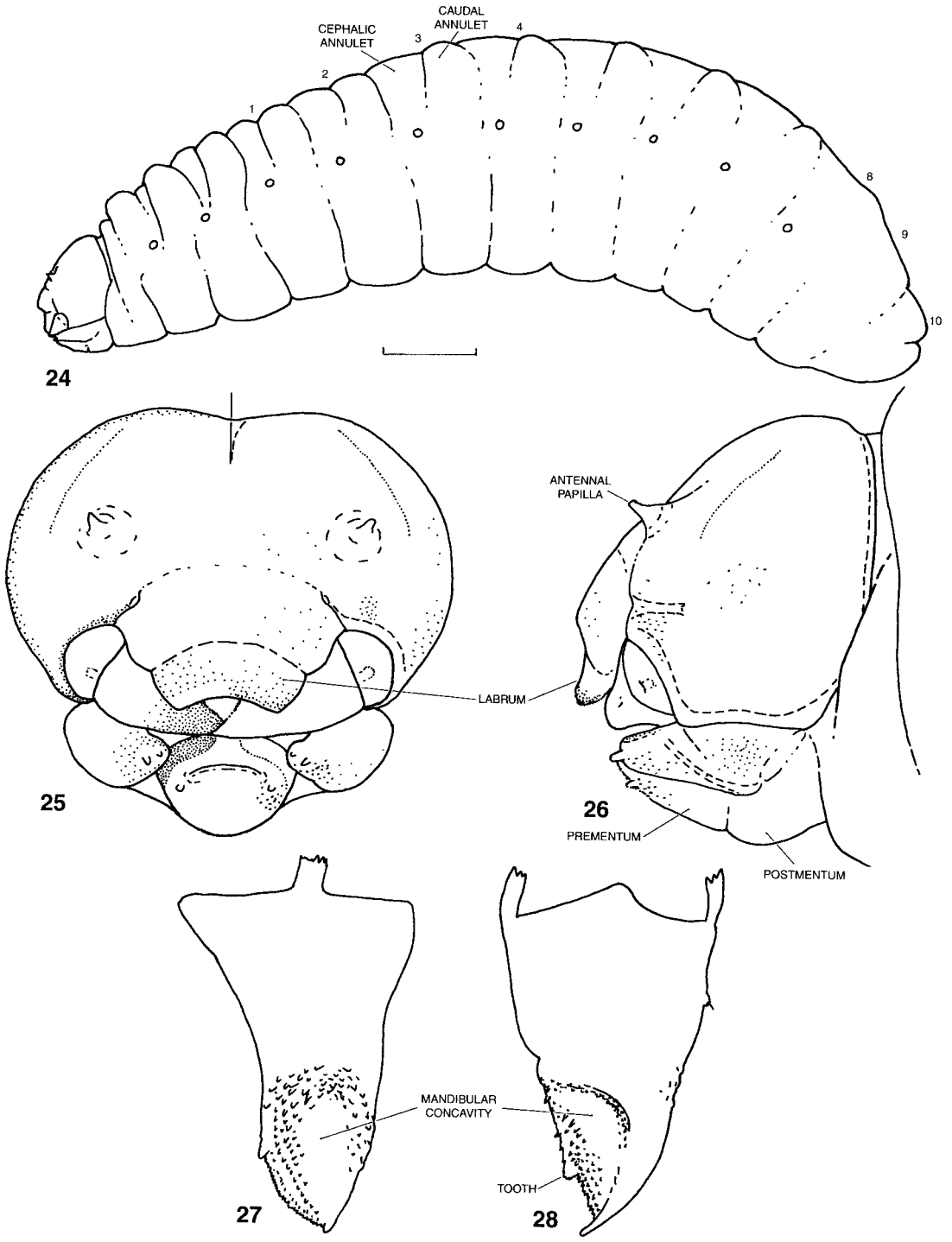
Mandible with extreme apex shorter relative to total length as seen in ventral view (fig. 28) than in third instar; dorsal apical edge now with small subapical tooth; scoop-shaped apical concavity more defined and denticles more extensive than in previous instar; outer surface with small tubercle near base. Labium (fig. 26) clearly divided into pre- and postmentum. Hypopharynx now bilobed. Salivary opening now with thin, slightly projecting lips, which are microscopically fringed apically.

BODY (fig. 24): As described for third instar except for following: Form somewhat curved; mid body thicker than at front or rear; most body segments dorsally clearly divided into cephalic and caudal annulets with intrasegmental lines well expressed dorsally; venter of abdominal segment 10 longer than dorsum so that anus appears somewhat dorsal relative to segment 9 and area below anus projects posteriorly more than area above anus (fig. 24). Most of integument spiculate but ventral surface more conspicuously so; abdominal segment 10 with scattered setiform sensilla.

MATERIAL STUDIED: One fourth instar, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, April 28, 2000 (C. A. Garófalo), N1/99, cell 20.

LAST LARVAL INSTAR

As seems the case with bee larvae in general, the last larval instar of *Ex. smaragdina* is the stage that consumes much, if not the most, of the provisions. Consequently there is a dramatic modification of its postcephalic region during this stage. Traditionally taxonomic descriptions of mature larvae are based on specimens that have defecated. Hence, an account of the postdefecating larva is presented below, given in sufficient detail to be compared with postdefecating larvae of other taxa. Following this description, predefecating larvae are treated.



Figs. 24–28. *Exaerete smaragdina,* fourth instar. **24.** Entire body, lateral view. 2**5.** Head, frontal view. **26.** Head, lateral view. **27, 28.** Right mandible, inner and ventral views, respectively. Scale (= 1.0 mm) refers to fig. 24.

Postdefecating Larva
Figures 29–31, 34, 35, 37–40

DIAGNOSIS: The fully developed salivary lips of this instar are characteristic of the last larval stage, and the relatively smooth, scooplike mandibular concavity (figs. 39, 40) contrasts with the heavily dentate apical concavity of the fourth instar (figs. 27, 28). Body shape readily distinguishes the postdefecating larva (fig. 29) from the predefecating forms (figs. 32, 33).

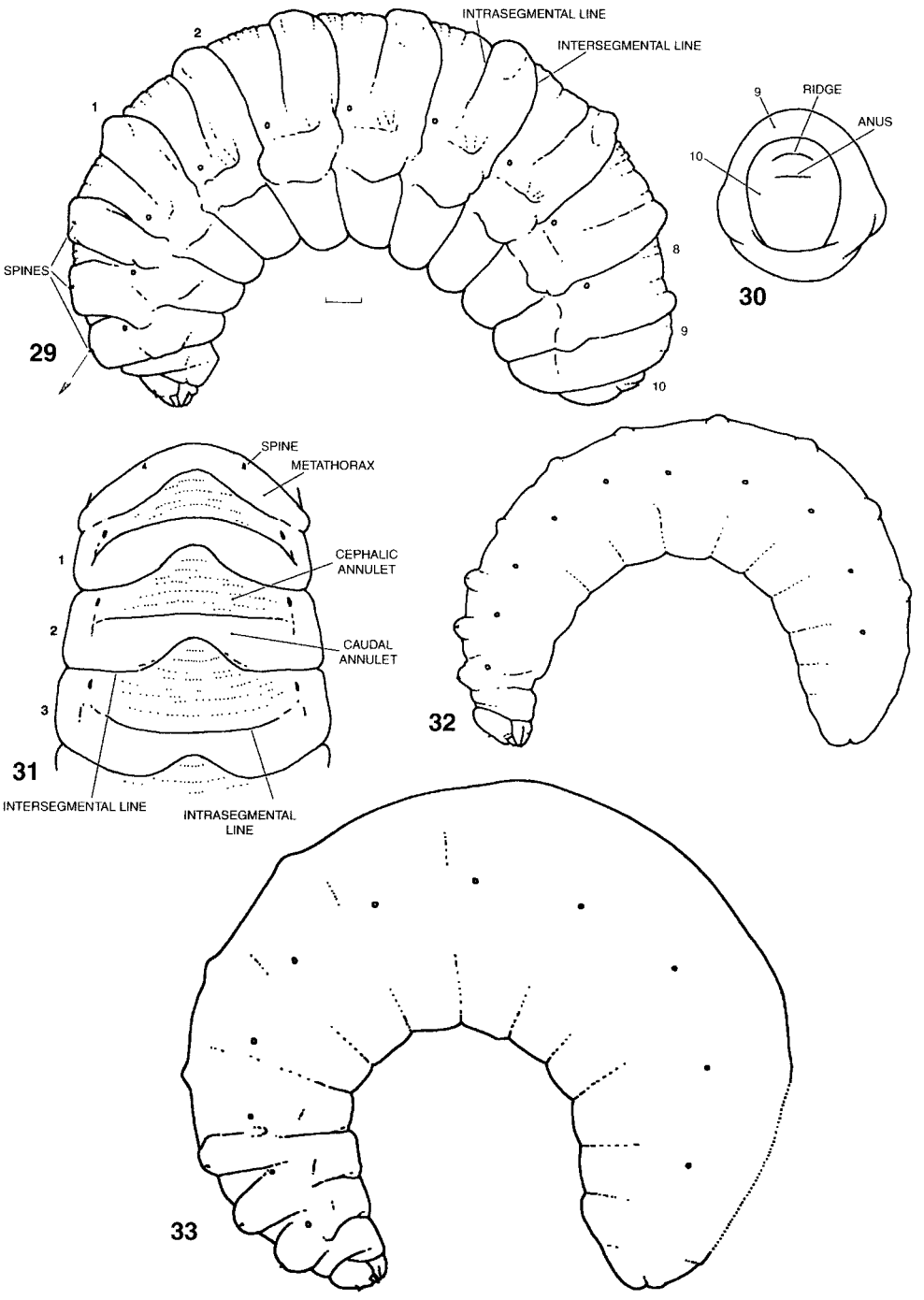
Larvae of other species of *Exaerete* are unknown, except for a cast skin of a mature larva of *Ex. dentata* from a cell of *Ef. surinamensis* (Trinidad: Caratal, Cumuto, February 26, 1964 [F. D. Bennett, D. Bharath]) in the collection of the American Museum of Natural History (AMNH). Features of the peritreme, maxillae, and mandibles appear indistinguishable from those described below. The larva also possessed paired, darkly pigmented dorsal spines on at least some of the thoracic segments. No features could be identified that distinguish this species from *Ex. smaragdina*.

LENGTH: About 18 mm (when body curved); distance between antennal apices 1.05 mm (N = 1).

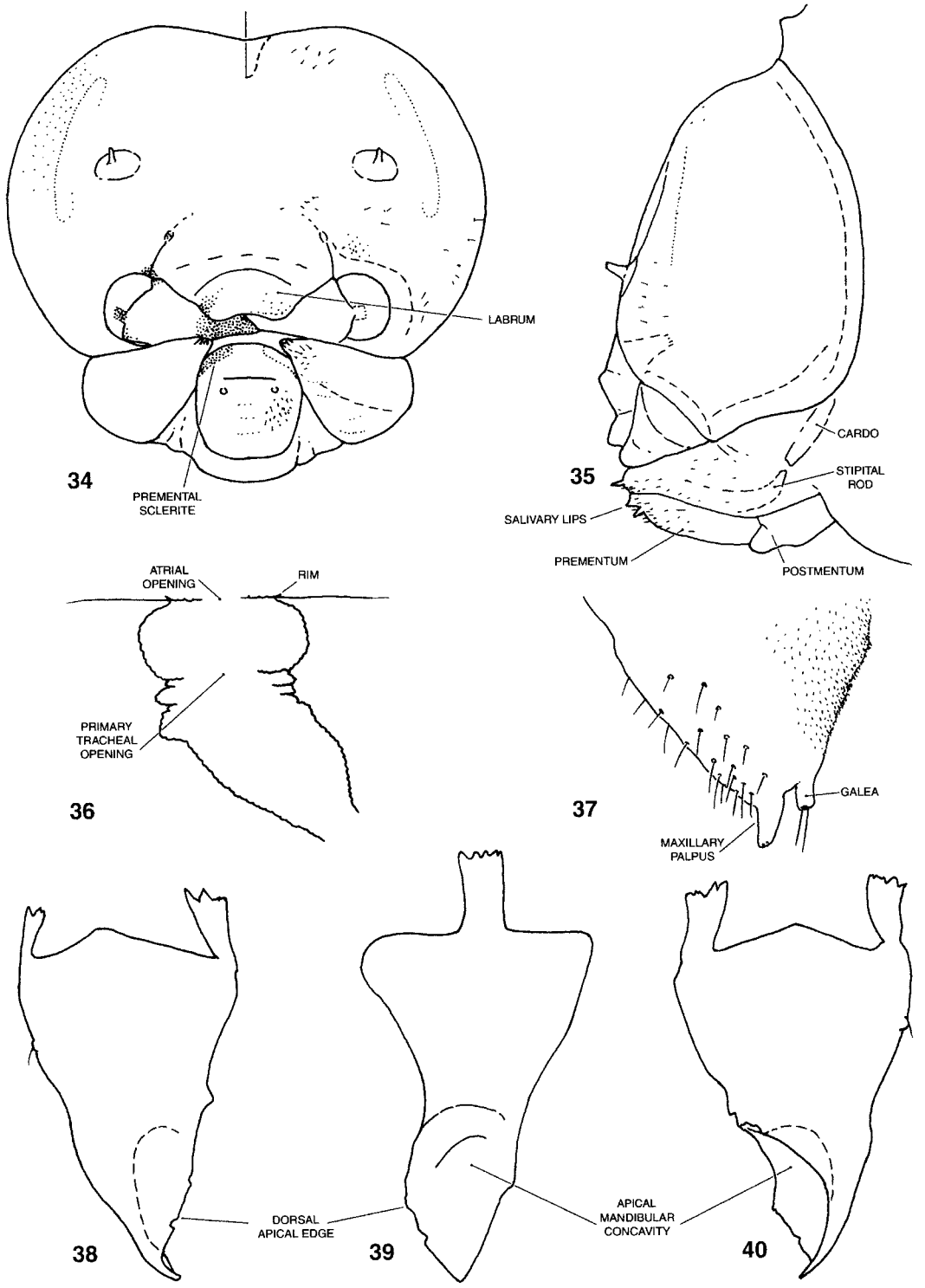
HEAD (figs. 34, 35): Shape hypognathous. Integument of head capsule strongly sclerotized, parietals faintly pigmented laterad of parietal bands; some internal ridges and apodemes with pigmentation when seen on cleared specimen; labrum darkly pigmented laterally; mandible moderately pigmented with apices more strongly sclerotized and pigmented; cardo pigmented; stipital rod faintly pigmented. Head capsule with moderately fine setiform and nonsetiform sensilla; labrum with numerous conspicuous sensilla and with apical row of tuberculate sensilla; dorsal surface of labium not spiculate; hypopharynx spiculate. Tentorium complete, robust, including dorsal arms; posterior tentorial pits deeply imbedded; coronal ridge pronounced but limited to vertex; other internal head ridges strongly developed except epistomal ridge between anterior tentorial pits scarcely evident. Parietal bands well expressed. Antennal prominence scarcely evident; antennal papilla (fig. 35) projecting, length about two times basal diameter, bearing approximately 5 sensilla. Labrum small relative to head size as seen in frontal view (fig. 34); basal labral sclerite apparently absent but dark pigmentation on sides of labrum may give impression of sclerite; labrum apically bearing rounded mound on each side but lacking paired tubercles; labral apex shallowly emarginate apically; epipharyngeal surface finely spiculate in some areas.

Mandible (figs. 36–40) robust, acutely pointed apically, with deep, scoop-shaped apical concavity; dorsal apical edge irregularly uneven; ventral apical edge less uneven; inner surface of concavity now without denticles but with irregularities at base. Maxillae and labium well separated apically. Cardo represented by pigmented sclerite; stipes a sclerotized but weakly pigmented rod extending along mesal surface of maxilla; articulating arm of stipes evident and darkly pigmented; maxillary apex tapering, with palpus and distinct galea close together on narrow apex; maxillary palpus moderately long but slightly smaller than antennal papilla. Labium divided into long prementum and postmentum; premental sclerite pigmented dorsolaterally; labial apex in repose projecting about as far as maxillary apices, bearing broadly transverse salivary lips apically; labial palpus evident, similar but less pronounced than maxillary palpus. Hypopharyngeal groove evident; hypopharynx projecting, bilobed.

BODY (fig. 29): Form linear, robust; intersegmental lines well incised; each thoracic segment dorsally with pair of small, darkly pigmented, acute spines; abdominal segments distinctly divided into cephalic and caudal annulets dorsally; cephalic annulets with fine transverse wrinkling; caudal annulets projecting farther in lateral view (fig. 29) than cephalic annulets; intersegmental lines of most abdominal segments curving forward medially as seen in dorsal view (fig. 31) so that caudal annulet invaded by cephalic annulet of following segment and caudal annulet reduced in height medially; thus caudal annulets of most abdominal segments somewhat taking on appearance of paired dorsal transverse tubercles; lateral swellings below level of spiracles pronounced; abdominal segment 9 not protruding ventrally; abdominal segment 10 apically attached to 9; anus



Figs. 29–33. *Exaerete smaragdina*, fifth instar. **29.** Postdefecating larva, lateral view. 3**0.** Same, posterior view of terminal abdominal segments. **31.** Same, dorsal view of posterior part of metathorax and first three abdominal segments. **32.** Predefecating, early fifth instar, lateral view. **33.** Predefecating, late fifth instar, lateral view. Scale (= 1.0 mm) refers to all figures.



Figs. 34–40. *Exaerete smaragdina*, fifth instar. **34.** Head, frontal view. **35.** Head, lateral view. **36.** Spiracle of predefecating larva, side view. **37.** Apex of right maxilla, dorsal view. **38–40.** Right mandible, dorsal, inner, and ventral views, respectively.

somewhat dorsal in position (fig. 30) with area immediately above projecting, this area appearing in posterior view (fig. 30) as curved transverse ridge dorsal of anus. Integument with many areas having inconspicuous spicules; setae widely scattered, fine, inconspicuous. Spiracles subequal in size; atrial wall finely, concentrically ringed; peritreme flush with body surface, its surface finely, concentrically ringed, with its rim projecting slightly farther than inner rings; atrial opening small, about one-half diameter of primary tracheal opening; primarily tracheal opening with collar that is also finely concentrically ringed; subatrium with integument ringed similar to integument of rest of spiracle; subatrium of postdefecating larva longitudinally collapsed, optically dense, thus difficult to interpret; subatrium (fig. 36) of predefecating larva consisting of two or three weakly defined expandable chambers broadly attached to wide chamber that attaches to trachea.

MATERIAL STUDIED: One postdefecating larva, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, May 22, 2000 (C. A. Garófalo), N1/99, cell 16.

Predefecating Larva
Figures 32, 33, 36

Figures 32 and 33 are based on larvae that developed from eggs that had been deposited 20 and 24 days earlier, respectively. Anatomical structures of the head are the same as reported for the postdefecating larva described above; for example the antennal apices on these two specimens were separated by 1.00 mm (N = 2), virtually the same as 1.05 mm for the postdefecating larva. However, the spiracular subatrium is not collapsed longitudinally, so that it can be interpreted differently, as in figure 36. Furthermore, the small acute spines on the dorsum of the thoracic segments are less pigmented, and on the two predefecating larvae examined these spines tended to be more transverse than those on the postdefecating larva, presumably a phenomenon of individual variation.

The main differences, other than size, between the postdefecating larva and the two predefecating larvae pertain to the degree of expression of the inter- and intrasegmental lines, the extent of expression of the dorsal annulations of the abdominal segments, and the appearance of the dorsal tubercles. On the postdefecating larva the inter- and intrasegmental lines are deeply incised; on the predefecating form, the intersegmental lines are faint and the intrasegmental lines are absent or nearly so. On the postdefecating form, the dorsal abdominal annulations are distinct for the most part, with the caudal annulations projecting more than the cephalic ones; on the predefecating forms the annulations are invisible except for the low, paired, somewhat transverse tubercles that represent the caudal annulations. These tubercles are scarcely evident on the postdefecating larva. Although these differences seem to be due to the postdefecating larva having voided the massive amount of fecal material, this is not entirely the case. Otherwise, why do some of the postdefecating larval features not appear in the younger (smaller) of the two predefecating forms, which had eaten much less?

MATERIAL STUDIED: Two predefecating larvae, Campus de Ribeirão Preto, Universidade de São Paulo, Brazil, May 2, 5, 2000 (C. A. Garófalo), N1/99, cells 18, 25.

DESCRIPTION OF THE EUGLOSSINI BASED ON THEIR MATURE LARVAE

Mature larvae of the following Euglossini have been described: *Eufriesea violacea* (Blanchard) (Michener, 1953a), *Euglossa (Glossura) imperialis* Cockerell (Roberts and Dodson, 1967), *Euglossa (Glossura) intersecta* Latreille (Zucchi et al., 1969a), and *Eulaema nigrita* (Zucchi et al., 1969b). From these accounts and the mature larvae of *Ex. smaragdina,* the following tentative, brief characterization of the mature larvae of the Euglossini emerges:

HEAD: Top of head with (*Euglossa, Eufriesea*) or without (*Eulaema, Exaerete*) pair of pigmented spines; antennal papilla elongate; labrum small relative to size of head, without distinct tubercles, shallowly emarginate apically; mandible robust with conspicuous scoop-shaped apical concavity, with apex acute, and often with toothlike projection along dorsal inner edge near base of

concavity; maxilla and labium well separated; galea present, presumably with a number of long setae; galea and maxillary palpus arising from narrow apex of maxilla, which is not bent mesad; labium divided into pre- and postmentum; salivary opening a pair of transverse, projecting salivary lips borne on labial apex.

BODY: Form robust with posterior segments thicker than anterior segments; most abdominal segments divided into cephalic and caudal annulets dorsally (these annulets apparently not evident on predefecating forms); thoracic segments each bearing pair of pigmented spines; sometimes these spines represented by transverse series of pigmented projections. First abdominal segment with (*Euglossa, Eufriesea*) or without (*Eulaema, Exaerete*) pair of similar spines.

Exaerete (fig. 29) and *Eulaema* agree in that known postdefecating larvae lack a pair of pigmented spines on the top of their heads in contrast with larvae of *Eufriesea* and *Euglossa*. *Exaerete* (figs. 29, 31) and *Eulaema* also agree in lacking a pair of pigmented spines on the first abdominal segment, a feature also shared by *Eg. intersecta,* but not by *Eg. imperialis.* The large primary tracheal opening of *Ex. smaragdina* (fig. 36), which is about twice the diameter of the atrial opening, will readily separate this species from *El. nigrita* (Zucchi et al., 1969b: fig. 9D) in which the diameter of the two openings are approximately equal.[3]

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[3] The primary tracheal opening of the mature larva of *Eulaema polychroma* is somewhat smaller than the atrial opening.

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