Comparative Biology of Six Species of Coccinellid Beetles (Coleoptera: Coccinellidae) Predaceous on the Mealybug, *Phenacoccus herreni* (Homoptera: Pseudococcidae), a Pest of Cassava in Colombia, South America

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ABSTRACT The mealybug, *Phenacoccus herreni* Cox & Williams, is a pest of cassava in Colombia, South America. Six species of coccinellid beetles that are predaceous on this mealybug were studied in the laboratory: *Cleothera notata, Cleothera onerata, Cleothera* sp., *Hyperaspis* sp. 1, *H.* sp. 2, and *H.* sp. 3. Comparisons were made of adult coloration, sites of oviposition and pupation, larval feeding behavior and wax production, developmental times, sex ratios, and potential for use in biological control programs in South America and Africa.

KEY WORDS Insecta, Coccinellidae, behavior, cassava mealybug

CASSAVA (yuca, manioca), *Manihot esculenta* Crantz, is an important tropical crop whose starchy roots are basic to the diet of hundreds of millions of people (Bellotti & van Schoonhoven 1978). It is attacked by several species of mealybugs in the genus *Phenacoccus*. Among these, *P. manihoti* Matile-Ferrero is a serious exotic pest in Africa that originated in South America. It has been the target of a classical biological control program at the International Institute of Tropical Agriculture (IITA) in Ibadan, Nigeria (the "Africa-wide Biological Control Programme" of the IITA is now located in Cotonou, République de Bénin). The solitary, encyrtid endoparasitoid, *Epidinocarsis lopezi* (De Santis), introduced from Paraguay into Nigeria in 1981, has been especially successful (Herren et al. 1987, Neuenschwander & Herren 1987, Neuenschwander & Hammond 1988, Gutierrez et al. 1988). Coccinellid beetles, however, have not had an important impact in Africa.

In South America, a related mealybug, *P. herreni* Cox & Williams, also attacks cassava in the plains of eastern Colombia, and in the Cauca Valley near Cali in southwestern Colombia (Varela & Bellotti 1981, Bellotti et al. 1983). A related encyrtid parasitoid, *E. diversicornis* (Howard), has been used as a biological control agent in Colombia (Van Driesche et al. 1987).

To broaden the complex of natural enemies in Colombia and also for possible use in Africa, studies were conducted on six species of coccinellid beetles in the Tribe Hyperaspini that are predaceous on *P. herreni*. The Tribe Hyperaspini is in the Subfamily Scymninae and includes some 500 species in 14 genera, with most of these species belonging to *Hyperaspis* (Pope 1979). The centers of distribution are the Americas, although species of *Hyperaspis* are found in Europe, Asia, and Africa. However, most of the species of this huge genus are neotropical (Gordon 1985). Species of *Hyperaspis* prey only on Homoptera, especially the Pseudococcidae (mealybugs) and Coccidae (scale insects).

Unfortunately, because of insufficient taxonomic information on the Hyperaspini in the tropics, and confusion about the genus *Cleothera*, only two identifications could be made to the species level. Comparisons were made of adult coloration, sites of oviposition and pupation, larval feeding behavior and wax production, developmental times, sex ratios, and potential for use in biological control programs in South America and Africa.

Materials and Methods

Rearing of *Phenacoccus herreni*. Using a modification of the method described by Van Driesche et al. (1986), cultures of this mealybug were maintained on potted cassava plants (variety CMC-40), in organdy-covered cages (80 by 90 by 200 cm) in the greenhouse at the Centro Internacional de Agricultura Tropical (CIAT) in Cali, Colombia. Each week, approximately 75 potted cassava plants (25–40 cm in height) were infested with mealybugs by placing 8–10 ovisacs on the underside of three different leaves. The colonies were maintained at 35.0 ± 4°C, 70% RH, and natural photoperiod (approximately 12:12).

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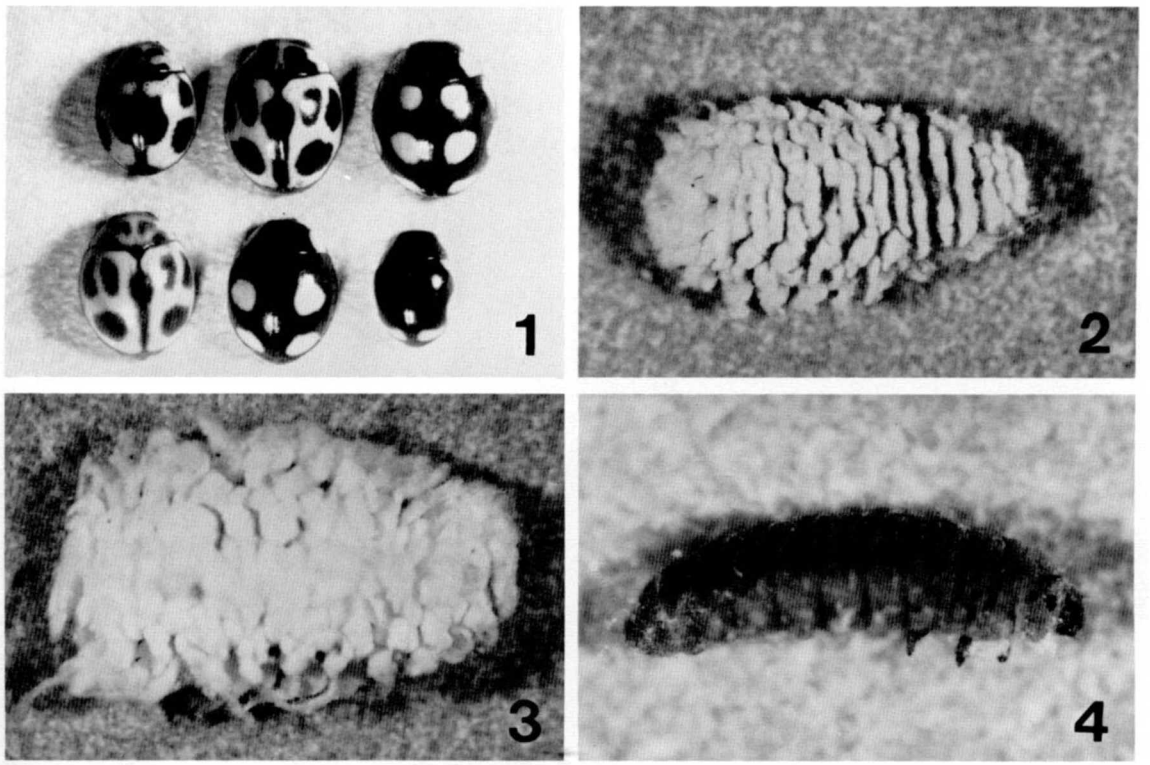

Fig. 1. Six species of adult coccinellids. Top row (left to right): *Cleothera onerata*, *C. notata*, *C.* sp. Bottom row (left to right): *Hyperaspis* sp. 3, *H.* sp. 1, *H.* sp. 2 (×10; figure reduced to 64%).
Fig. 2. Fourth-instar larva of *Cleothera notata* with wax secretions (×20; figure reduced to 64%).
Fig. 3. Fourth-instar larva of *Hyperaspis* sp. 1 with wax secretions (×20; figure reduced to 64%).
Fig. 4. Fourth-instar larva of *Hyperaspis* sp. 2—note absence of wax secretions (×25; figure reduced to 64%).

Coccinellid Species. Two genera and two of the six species were identified by R. D. Gordon (Systematic Entomology Laboratory, USDA, Beltsville, Md.): *Cleothera notata* Mulsant: Palmira, Departamento del Valle del Cauca, Colombia; and *Cleothera onerata* Mulsant: Palmira, Departamento del Valle del Cauca, Colombia; however, he had no time to describe the four remaining species: 1) *Hyperaspis* sp. 1 Redtenbacher: Carimagua, Departamento del Meta, Colombia; 2) *Hyperaspis* sp. 2 Redtenbacher: Carimagua, Departamento del Meta, Colombia (only these four species were feeding on *Phenacoccus herreni* on *Manihot esculenta*); 3) *Hyperaspis* sp. 3 Redtenbacher: Timaná, Departamento del Huila, Colombia—feeding on *Nipaecoccus nipae* on *Psidium quajaba*; 4) *Cleothera* sp. Mulsant: Pucalpa, Departamento de Ucayali, Peru—feeding on *Planococcus* sp. on *Iatrophobia gossypiense* (Pioroto).

Rearing of Coccinellids. Six potted cassava plants, well-infested with mealybugs, were placed in each rearing cage and then 800–1,200 adult males and females of the particular coccinellid species being studied were released, where they remained for 48 h of oviposition. After eclosion, the coccinellid larvae fed on the mealybug eggs, and completed their development to adults. This procedure was repeated for each of the six coccinellid species, which were kept in separate rearing cages.

Laboratory Experiments. A large Petri dish (15 by 3 cm) was used. A circular hole (9 cm diameter) was cut in the cover, and was covered below with organdy to permit aeration. Then 40–50 mealybug ovisacs and two adult females were removed from the greenhouse colony, and placed on a square piece of paper towel cut to fit inside the Petri dish. Two ends of the paper towel were bent at a 90° angle so that it could be turned upside down without the ovisacs contacting the bottom of the Petri dish, thus avoiding crushing of the ovisacs.

Five females and three males of each coccinellid species were placed in a Petri dish, which was then sealed around the circumference with a 4-cm-wide strip of Parafilm "M." The Petri dish was placed in a walk-in environmental room which was kept at a constant temperature of 28°C, 55 ± 4% RH, and 12:12 photoperiod. To obtain sufficient numbers and similar-aged cohorts of coccinellid larvae, the adults were permitted to oviposit over only 24 h, so that eclosion would normally be restricted to a similar period later. After the adult coccinellids were removed, the large Petri dish was resealed

Table 1. Comparative coloration and behavior of six species of coccinellid beetles

Species	Elytra	No. & color of spots	Oviposition site (within/ outside ovisac)	Larval wax	Pupation site
C. notata	Yellow	6 black	Within	Yes	Soil
C. onerata	Yellow	4 black	Outside	Yes	Leaf
C. sp.[a]	Black	10 yellow	Within	Yes	Soil
H. sp. 1	Black	4 yellow	Outside	Yes	Leaf
H. sp. 2	Black	2 yellow stripes	Outside	No	Leaf
H. sp. 3[a]	Yellow	6 brown	Within	Yes	Soil

[a] When collected in the field, these two species were not feeding on *Phenacoccus herreni*.

with parafilm and returned to the environmental room for 5–6 d. The eggs were then examined daily and each newly-hatched coccinellid larva was placed in an individual, aerated, but smaller Petri dish (100 by 15 cm) containing only 20–30 ovisacs plus one adult female mealybug on a piece of paper towel.

These coccinellids have four instars, which can be distinguished from each other. Depending on the species and individual variation, pupation occurs 16–25 d after oviposition. Hence, the ovisacs were replaced about 12–14 d after oviposition, in order to provide fresh eggs for the individual larva in each of the replicates. They were left undisturbed until adult emergence. The sex of the adults was then determined, and they were released into the coccinellid colony rearing cage in the greenhouse.

Results and Discussion

Adult Size. In comparison with many other coccinellid beetles, adults (Fig. 1) of the six species are quite small (2.0–4.0 mm in length). However, they are within the normal range for the Tribe Hyperaspini of 1.0–5.0 mm in length (Gordon 1985).

Adult Coloration. The adults of three species have yellow elytra with a varying number of spots or dots (Fig. 1 and Table 1), while the three remaining species have black elytra with either yellow spots or stripes. There seems to be no generic consistency in the color patterns of the elytra to distinguish *Cleothera* from *Hyperaspis*. In coccinellid taxonomy, color is not a major consideration because other diagnostic characters are usually more important, such as male genitalia, and (in the genus *Hyperaspis*) the number of antennal segments. Although R. D. Gordon (personal communication) used both generic names, he indicated that *Cleothera* and *Hyperaspis* may not be distinct. However, this can only be resolved by a complete study of the South American hyperaspine fauna. Hence, it would be premature to name these species at this time.

Oviposition and Pupation. *C. notata*, *C.* sp., and *H.* sp. 3 lay ovoid-shaped, colorless or cream-colored eggs inside the ovisacs of the mealybug (Table 1). Observations in the field and in the large rearing cages indicate that after the larvae feed on the eggs, they then pupate in the soil at the base of the cassava plant. On the other hand, *C. onerata*, *H.* sp. 1, and *H.* sp. 2 deposit flat, grey eggs outside the ovisacs (but near them on the leaf substrate). After moving to the eggs for feeding, these larvae subsequently pupate on the leaves of the cassava plant on which mealybugs have been feeding.

Larvae. Upon hatching, first instars of five of the six species are yellowish, resembling the color of the first instar mealybugs. In the case of *H.* sp. 2, however, the larvae are grey. When disturbed or prodded by a dissecting needle or probe, all species of larvae react vigorously with movements of the body, and may discharge one or more droplets of a sticky, yellow liquid from the dorsum. Our observations agree with those of Nsiama She et al. (1984) on *H. jucunda*—concerning feeding on mealybug eggs within the ovisacs, discharge of sticky droplets, and wax production—discussed below. The grey color of the larva of *Hyperaspis* sp. 2, however, was unusual.

Larval Wax Production. The second, third, and fourth instars of five species (Table 1) produce white wax secretions from the entire dorsal side of the body (Fig. 2 and 3). However, the larva of *H.* sp. 2 does not produce these secretions at all, and remains grey throughout the four instars (Fig. 4). The pupa also keeps the wax secretions of the fourth-instar larva. The function of coccinellid wax secretions was discussed by Pope (1979).

Duration from Oviposition to Adult Emergence. Comparative developmental time in days is given for the six species in three parts: from oviposition to pupation, from pupation to adult emergence, and from oviposition to adult emergence (Table 2).

A short developmental period from oviposition to adult emergence may be important in a potential biological control agent by permitting more generations per season or year. In comparison with the five other species, therefore, *H.* sp. 2 has the advantage. *H.* sp. 2 needs only 26.0 d to reach adult emergence (Table 2). The ANOVA indicates that this is a significant difference in comparison with the five other species. *H.* sp. 2 has a notably short life cycle because it has the shortest pupation period (6.8 d) in relation to the other species. At the

Table 2. Comparative developmental time in days from egg to pupa, pupa to adult, and egg to adult for six species of coccinellids

Species	*n*	Developmental time, $\bar{x}$ ± SD (range)		
		Egg–pupa	Pupa–adult	Egg–adult
H. sp. 1	40	20.9 ± 1.2a (19–24)	10.3 ± 0.9a (8–12)	31.2 ± 1.3a (28–34)
C. onerata	40	20.0 ± 2.0b (17–25)	8.8 ± 1.5b (6–12)	28.8 ± 1.6b (25–32)
C. sp.	41	18.3 ± 1.6c (16–21)	10.4 ± 1.9a (7–14)	28.7 ± 1.3bc (26–32)
C. notata	40	19.1 ± 1.1bc (17–22)	8.8 ± 0.9b (7–11)	27.9 ± 1.4c (26–31)
H. sp. 3	40	19.0 ± 1.2c (16–21)	8.9 ± 0.8b (7–11)	27.9 ± 1.2c (26–30)
H. sp. 2	39	19.2 ± 1.4bc (16–21)	6.8 ± 1.0c (5–9)	26.0 ± 1.8d (23–30)

Means in each column followed by different letters indicate a significant *T*-grouping difference at $P < 0.05$ (ANOVA multiple *T* test, Fisher's LSD [SAS Institute 1985]).

same time, however, it does not do this at the expense of its feeding period as a larval predator; its developmental time from oviposition to pupation is equal to the others, without any statistical difference in relation to four of the five other species. In addition, *H.* sp. 2 is a very aggressive coccinellid. In the laboratory, both larvae and adults tend to move very quickly and seem to search more actively for prey in comparison with the other species.

Sex Ratios. The sex ratios ($n = 240$) indicated that only *H.* sp. 2 had a highly significant deviation (2:1 [female : male]) from the five other species, where the sex ratio was 1:1.

Field Observations. *Cleothera notata* was originally collected at CIAT (Palmira, Departamento del Valle del Cauca) in Colombia, and it is the only one of the six coccinellid species that has been released and successfully established elsewhere—in Villavicencio, Departamento del Meta, Colombia. Field evaluations are still in progress, however; when completed, they will be given in a separate paper. Probably this same species had been introduced earlier into Africa under the name *Hyperaspis notata*, but it has had only limited success there.

Based on the short developmental time and aggressive searching behavior in the laboratory, *H.* sp. 2 may be the best candidate for use in a biological control program. However, field trials of *H.* sp. 2 have not been completed in Colombia or in Africa where it is being tested at the IITA. On the other hand, *C.* sp. and *H.* sp. 3 were not collected while feeding on *P. herreni*, nor even on cassava. Hence, although both of these species were easily reared on this factitious host in the greenhouse insectary and in our laboratory experiments at CIAT, we have not yet released or tested them in cassava fields. We gave them low priority in comparison with the four other species, and we would not be surprised if it were shown later that they were ineffective colonizers in the cassava ecosystem.

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