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Mortality of the cassava mealybug, *Phenacoccus manihoti* MAT.-FERR. (Hom., Pseudococcidae), associated with an attack by *Epidinocarsis lopezi* (Hym., Encyrtidae)[1]

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Cassava mealybugs (CM) of different stages, which had been stung by an *E. lopezi* female, were inspected after 6 and 20 days. The killing power of the parasitoid was several times higher than the production of parasitoid off-spring. Host-feeding and mutilation were responsible for 6–22% and 11–34% mortality respectively. Both were significantly more important on the younger hosts. 11–33% of the stung CM yielded live parasitoids. Reproduction was significantly more successful on older CM. 30–56% of all CM survived a single oviposition attempt by the wasp. By contrast, mortality of the unstung control was 4–8%. In choice experiments, 3rd instar CM were slightly but not significantly prefered.

The major pest of cassava, the mealybug *Phenacoccus manihoti* MAT.-FERR. (CM) was accidentally introduced to Africa from South America in the early 1970's. The history of this introduction and its impact on cassava production have been reported by HERREN (1981), FABRES & BOUSSIENGUE (1981), NWANZE (1982), and HERREN *et al.* (1985).

In 1981, the specific and solitary parasitoid *Epidinocarsis lopezi* (DE SANTIS) was introduced from Paraguay and successfully established in southwestern Nigeria (HERREN & LEMA, 1982). It has since been released at over 30 sites in Africa. By Sept. 1985 it was established in 12 African countries, covering 650000 km^2 (HERREN *et al.*, 1985 and unpubl. results). First results suggest that it is capable of maintaining CM populations at a low level (HERREN & LEMA, 1983; LEMA & HERREN, 1985; W.N.O. HAMMOND, unpubl. results; NEUENSCHWANDER *et al.*, 1986).

This level of control is much higher than expected, considering that parasitization rates in the field remain mostly below 30% (LEMA & HERREN, 1985; W.N.O. HAMMOND, unpubl. results). *E. lopezi* is sometimes difficult to maintain at low host densities in the laboratory and, even at high host densities, reproductive capacity is lower than in other encyrtids (CLAUSEN, 1972). It was therefore suspected that *E. lopezi* destroyed more insects than it utilised for reproduction. The present study of CM mortality associated with an *E. lopezi* attack was undertaken to improve, if possible, rearing conditions and the interpretation of field results.

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MATERIALS AND METHODS

In the laboratory, individual *E. lopezi* females from an insectary culture were offered CM on leaves of *Talinum triangulare* (JACQ.) in a Petri dish. This common weed is a good though not preferred host of the CM in the field. A preliminary experiment demonstrated that CM survived better on the isolated fleshy leaves of this host, than on those of cassava, which rapidly curl up and dry out. In a comparison, 30 Petri dishes with *T. triangulare* and 30 with cassava leaves, each with 10 CM per Petri dish, were maintained at 3 different temperatures in the upper temperature range (30–34°C) for 5 hrs and then stored at 28°C. Average mortalities after 2 days were 33% on cassava leaves compared to 1.3% on *T. triangulare*. All experiments were therefore done on this substitute host in Petri dishes, where no escape of the CM was possible and dead CM could be recovered easily.

In the first experiment, in Nov. 1983, a total of 160 Petri dishes each with 10 young 4th instar CM and one female *E. lopezi* were set up and stored overnight at 10–15°C in the dark. The next day, they were divided into lots of 40 each and placed under 3 light-temperature regimes. The 3 laboratory regimes were: 30.5°C/800 lux, 31.2°C/2700 lux, and 34.2°C/6500 lux with temperatures kept accurate to ± 0.5°C and lux to ± 20% between different Petri dishes. In the glasshouse temperatures ranged from 36.5°C to 38.6°C and lux up to 21500 during the day. All measurements were made inside the Petri dishes. Ten *E. lopezi* females were removed at the beginning of the experiment (0 hrs), 10 after 1.5 hrs, 10 after 6 hrs, and 10 after 24 hrs from each of the 4 regimes. During the period of exposure, the oviposition attempts and host-feeding of all females (except those exposed 0 hrs) were directly observed in each Petri dish during 1 hr. Oviposition attempts and host-feeding activity were noted. After 24 hrs all Petri dishes, now without adult wasps, were stored at 28 ± 2°C. On day 6, all CM were inspected, and their survival was recorded. Survivors and already formed mummies were kept on fresh leaves up to day 20. Since mean developmental time of *E. lopezi* from egg to adult at 27°C is 14.3 d (LEMA & HERREN, 1982), all surviving parasitoids had emerged at that time. The percentage parasitism was calculated on the basis of the emerged parasitoids and the living unparasitized CM of day 6. This parasitization rate was then compared with the non-reproductive mortality on day 6.

In the second experiment, in 1984, the development of individual mealybugs attacked by an *E. lopezi* female was followed. In the laboratory at about 28°C, wasps were observed stinging different stages of CM. As soon as the wasp had finished and lost interest, the CM was carefully removed and reared in a Petri dish on *T. triangulare*. Mortality of the CM on day 6 and production of live parasitoids were noted as in the first experiment, and the results compared with the survival of unattacked CM kept under the same condition (control C). In the experiment A_1, a leaf with numerous CM in the 2nd to 4th instar (preoviposition females) was offered to one female wasp of undetermined age. This experiment involved many females and lasted several weeks. In A_2 the wasp could again choose among different stages, but each female was offered exactly 150 CM, 50 of each stage, for 2 hrs only. The experiment was replicated 4 × with different females. Only CM that had been stung are reported. In the third experiment (B), the female wasps were offered only one CM stage.

RESULTS

In the first experiment, mortality of the CM was compared with the number of progeny of the parasitoid. Under all temperature and light conditions, parasitization rates increased with the time of exposure of the host to the parasitoid. As many as 8 ovipositions per hour were observed, but the mean was less than one per hour. Although numerous unparasitized CM were available, the wasps often made repeated oviposition attemps on the same host. (But no mummy was ever found in the field or the laboratory where more than one *E. lopezi* adult emerged). Also, after having stung, the females often turned around and fed on hemolymph and large amounts of host tissue from the wound made by the ovipositor, a behaviour called host-feeding.

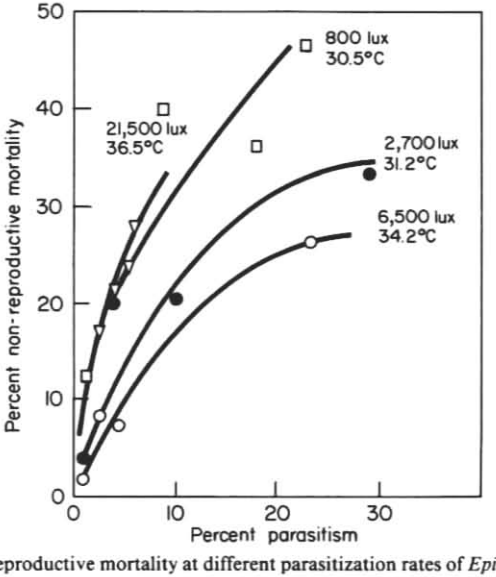

Fig 1. Increase in non-reproductive mortality at different parasitization rates of *Epidinocarsis lopezi* under various temperature – light conditions. Successive points represent about 100 cassava mealybugs exposed to 10 females for 0, 1.5, 6, and 24hrs.

The quantitative results in Figure 1 show that CM mortality not caused by parasitoid larvae was clearly correlated to the percentage of parasitism. This non-reproductive mortality differed for the various conditions, but even the lowest light intensities did not stop *E. lopezi* oviposition. In all treatments, the non-reproductive mortality surpassed the one from mummy formation. In 14 out of 16 treatments it was equal or greater than twice the mortality caused by successful reproduction of the wasp. Clearly, the killing power of the parasitoid was much greater than that indicated by the rate of parasitism. This non-reproductive mortality was partly attributed to host-feeding, but further investigation was required to determine all the mortality factors.

In the second experiment, we followed the development of individual mealybugs of different stages which had all been stung once by *E. lopezi*. Mortality

of stung and unattacked mealybugs (control C) and the production of parasitoid offspring were determined (Table 1).

When attacking the CM, *E. lopezi* slightly, but not significantly, preferred the 3rd instar (37.8% of all attacks in A_1, 44.3% in A_2) as compared to 2nd instars (A_1: 30.8%; A_2: 29.5%) or 4th instars (A_1: 31.4%; A_2: 26.2%). Where the female had a choice of host stages (experiments A_1, A_2), a significant host-feeding preference for the younger CM, which offer less resistance, was demonstrated (Table 1). (In other experiments, occasional host-feeding was observed even on first instars.) All CM host-fed upon died immediately. By contrast, the percentage of CM that yielded a live parasitoid increased significantly on the older stages but generally remained relatively low. Even under no-choice conditions (experiment B) and excluding the host-fed CM, successful parasitization of the 3rd instar was only 37.8%. A large percentage of the CM died after oviposition by the wasp without forming a mummy. About two thirds of these CM were killed right after being stung, especially when oviposition lasted more than 7 minutes. Death was delayed for the others for a few days. Mortality was significantly higher in the younger stages. Since it was much higher than in the unstung control (C), it is attributed to the oviposition behaviour of the wasp. Finally, a fairly high percentage of CM, 30–56% survived a wasp attack. All three experiments gave essentially the same results.

Tab 1. Fate of *Phenacoccus manihoti* attacked by *Epidinocarsis lopezi* in the laboratory, each CM stung once. Two choice experiments (A_1, A_2), one no-choice experiment (B), and an unstung control (C).

CM instar/ type of experiment	total No. CM	% CM host fed 1)	% CM prducing live parasitoids 2)	% CM dead on 6th day, but not host-fed 3)	% CM surviving sting more than 6 d
2nd instar:					
A_1	100	20.0	18.0	20.0	42.0
A_2	18	22.2	11.1	33.3	33.4
B	123	13.0	17.9	34.1	35.0
C	230	0	0	4.3	95.7
3rd instar:					
A_1	123	10.6	27.6	31.7	30.1
A_2	27	11.1	22.2	11.1	55.6
B	102	11.8	33.3	23.5	31.4
C	130	0	0	8.5	91.5
4th instar:					
A_1	102	8.8	28.4	10.8	52.0
A_2	16	6.3	25.0	12.5	56.2
B	100	9.0	29.0	14.0	48.0
C	130	0	0	6.2	93.8

A test of homogeneity between instars was performed for each mortality factor separately. A_1 and A_2 were pooled in order to fulfil the requirements of the test. $\chi^2_{0.05,\,2\,d.f.} = 5.99$, * significant at $p = 0.05$.

1) $\chi^2_{A1+A2} = 8.48^*$, $\chi^2_B = 1.77$

2) $\chi^2_{A1+A2} = 4.82$, $\chi^2_B = 7.49^*$

3) $\chi^2_{A1+A2} = 11.61^*$, $\chi^2_B = 12.12^*$

Parasitization rates of *E. lopezi* in the field rarely exceed 30% (maximum 60%) of all parasitizable host stages (2nd to 4th instars) (LEMA & HERREN, 1985; W.N.O. HAMMOND, unpubl. results). Higher rates of up to 79% were reported from Congo (GANGA, 1984) because 2nd instar CM were excluded from the calculations and empty mummies were counted as parasitoids. Despite the low degree of active parasitism, *E. lopezi* is an effective biological control agent. This tentative conclusion was reached from monitoring of CM densities in southwestern Nigeria following the establishment of *E. lopezi* (HERREN & LEMA, 1983; LEMA & HERREN, 1985; W.N.O. HAMMOND, unpubl. results) and from parasitoid exclusion experiments (NEUENSCHWANDER *et al.*, 1986).

The reproductive capacity of *E. lopezi* in the laboratory reaches about 40 per female (GANGA, 1984; and unpubl. results) which is smaller than the 100 to 150 reported for many other encyrtids (CLAUSEN, 1972). Also, parasitoid production on plants where thousands of CM were available averaged only 672 per plant (NEUENSCHWANDER *et al.*, 1984). The demonstrated effectiveness in spite of low reproductive capacity suggests that *E. lopezi* destroys more hosts than is indicated by the rate of production of mummies.

Killing of hosts by a parasitoid female in addition to those used for the immature stages is often important in biological control (DEBACH, 1943). Host-feeding has been observed mainly in the Ichneumonoidea and Chalcidoidea. In most families it is a normal part of oviposition in some species, and an act of predation separate from oviposition in other species (CLAUSEN, 1972). With *E. lopezi*, CM which yielded mummies were never observed to be host-fed upon. In our experiments, each female *E. lopezi* on the average killed another 1.5 to 2 CM for each CM which yielded an off-spring. This non-reproductive mortality was greater on 2nd (2.5–5.0) than on 4th instars (0.7–0.8). 35% of this non-reproductive mortality was due to host-feeding which provides proteins for the female. The rest was caused by mutilation through host probing or aborted parasitism, which does not directly benefit the parasitoid. Most non-reproductive mortality figures reported for other species in literature are lower than those for *E. lopezi*. Ratios of 1–2 additional hosts killed for each host yielding an off-spring were found among several *Aphytis* spp. Yet, these aphelinids effectively control their diaspine scale hosts at active parasitization rates not exceeding 30% (DEBACH, 1943; 1969; ALEXANDRAKIS & NEUENSCHWANDER, 1980). They prefer younger stages of their hosts as a prey (GULMAHAMAD & DEBACH, 1978). For the eulophid *Chrysocharis* sp., a parasitoid of leaf-miners, the ratio was 2 (SUGIMOTO & MASAKI, 1979); it was only about 0.3 for 2 pteromalid parasitoids of fly puparia (LEGNER, 1979) and 0.25 for *Encarsia*, an aphelinid parasitoid of white-flies (ARAKAWA, 1982). In most of these investigations, the different forms of non-reproductive mortality were not distinguished. The reason for the relatively high number of CM which survived the sting of *E. lopezi* is being further investigated by dissection of CM.

The high rate of non-reproductive mortality for *E. lopezi* makes it necessary to have high host numbers in rearing cages in order to avoid the killing of already parasitized hosts by host-feeding. It means that the rate of reproductive mortality is not sufficient for measuring and explaining the efficiency of this parasitoid.

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