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Evolution

Circumvention of prey defense by a predator: Ant lion vs. ant*

(predation/chemical defense/formic acid/*Myrmeleon*/*Camponotus*)

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ABSTRACT **The pit-dwelling ant lion *Myrmeleon carolinus*, although topically sensitive to formic acid, is able to prey on formic acid-spraying ants (*Camponotus floridanus*). It kills the ants without inducing them to spray, and it sucks out the ant's body contents without puncturing the acid sac. Ordinarily, when *Camponotus* is attacked it retaliates by simultaneously biting and spraying, but it usually refrains from spraying until it has secured a grip with the mandibles. When *Myrmeleon* pulls *Camponotus* into the sand at the bottom of the pit, the ant is seemingly unable to grasp the ant lion and it is killed without being induced to spray. When feeding on the ant, the ant lion sucks up the contents of the nutrient-laden crop. How the ant lion differentiates between crop and acid sac, managing to spare the latter while rupturing the former, remains unknown.**

We here ask a simple question: how do ant lions cope with the defensive spray of the ants they capture? Ant lions (larvae of the neuropteran family Myrmeleontidae) typically construct funnel-shaped pits in sandy soil (Fig. 1*A*) and lie in wait at the bottom with only the mandibles protruding (Fig. 1*B*), ready to catch any ambulatory arthropod that oversteps the pit margin and slides down the pit's slope (2–5). The principal prey of ant lions are ants, including species of the subfamily Formicinae, well-known for the formic acid-containing defensive spray they discharge (6–8).

Two personal observations suggested that ant lions might circumvent the defense of formicine prey. First, when feeding formicine ants to ant lions in the laboratory, we noted that we could detect no formic acid odor immediately above the pits when the ants were struggling in the ant lion's hold. And second, when dissecting formicine ant carcasses discarded by ant lions after their meal, we found these remnants to contain seemingly intact acid sacs.

We here provide evidence that ant lions can indeed subdue formicine prey with little risk of being sprayed and that they consume such prey without rupturing the acid sac. Our study was undertaken in the laboratory, at the Archbold Biological Station, Lake Placid, Highlands County, FL, with a species of ant lion (*Myrmeleon carolinus*) (9) (Fig. 1*C*) and a formicine ant (*Camponotus floridanus*) (10) (Fig. 1*D*), both native to the area.

MATERIALS AND METHODS

***M. carolinus*.** This ant lion is abundant in the sandy terrain of Highlands County, where its pits may occur with those of other species. Individuals (last instar only) were collected from pits and transferred singly to cages filled to a height of ≈5 cm with sand, in which they constructed normal pits. Following experimentation, those not dissected were returned to their habitat.

***C. floridanus*.** This ant typically nests in dead wood, and in Highlands County it shares the habitat of *M. carolinus*. Groups of hundreds of ants, scooped with brood and nesting material from individual colonies, were transferred to large laboratory enclosures, in which they quickly became established. They were maintained on water, aqueous honey solution, and freshly cut-up insects, and they were not used in experiments until several days after capture.

Handling of Ants. To remove ants from their laboratory colonies, they were seized by the alitrunk with broad-tipped (steel-spring) forceps. When prevented from biting the forceps when thus grasped, they almost never sprayed. This permitted transferring ants to individual vials for experimental purposes. Ants that sprayed during manipulation (discharges noticeable by odor) were excluded from the experiments.

Feeding of Ants. Ants were individually fed by being given a drop of aqueous (≈10%) honey solution in their vial and allowed to gorge to repletion. For some tests the solution was stained intense red with commercial food coloring. Ants that were fed were used experimentally within 1–2 hr after feeding. We had shown by dissection of ants fed dyed fluid that the ingested liquid is retained in the capacious crop for hours after feeding without being passed in substantial quantity to the midgut.

Killing, Dissecting, Weighing. Ants and ant lions were killed by refrigeration (to the point of near freezing) in vials. The procedure induced neither regurgitation nor (in case of the ants) spraying. Dissections (under chilled saline solution) were done within minutes after death. Ant carcasses discarded by ant lions after their meal were dissected before they became desiccated.

Weighings were to 0.1 mg. Ants and ant lions were weighed live in vials. The three anatomical chambers that were dissected for weighing purposes—the ant crop, the ant acid sac, and the ant lion's crop—are all of ectodermal origin and therefore lined with a thin cuticular membrane. This membrane is relatively tough and prevented the sacs from rupturing during dissection. Sacs were weighed full (after blotting off excess saline solution with filter paper), then empty after rupturing and draining of contents on filter paper, and the difference was recorded as the content mass.

Indicator Paper. Pieces of filter paper freshly soaked in deep-red alkaline solution of phenolphthalein were used to check for presence of acid, in either vapor or liquid form. The contents of the formic acid sac of *C. floridanus* turn such paper white, both on contact and near contact.

Ant Lion/Ant Encounters. Ants were fed to ant lions by being tossed from vials into the pits. Vials were always checked beforehand for formic acid odor, to ensure that the ants had not just discharged. The ant lions usually grasped the

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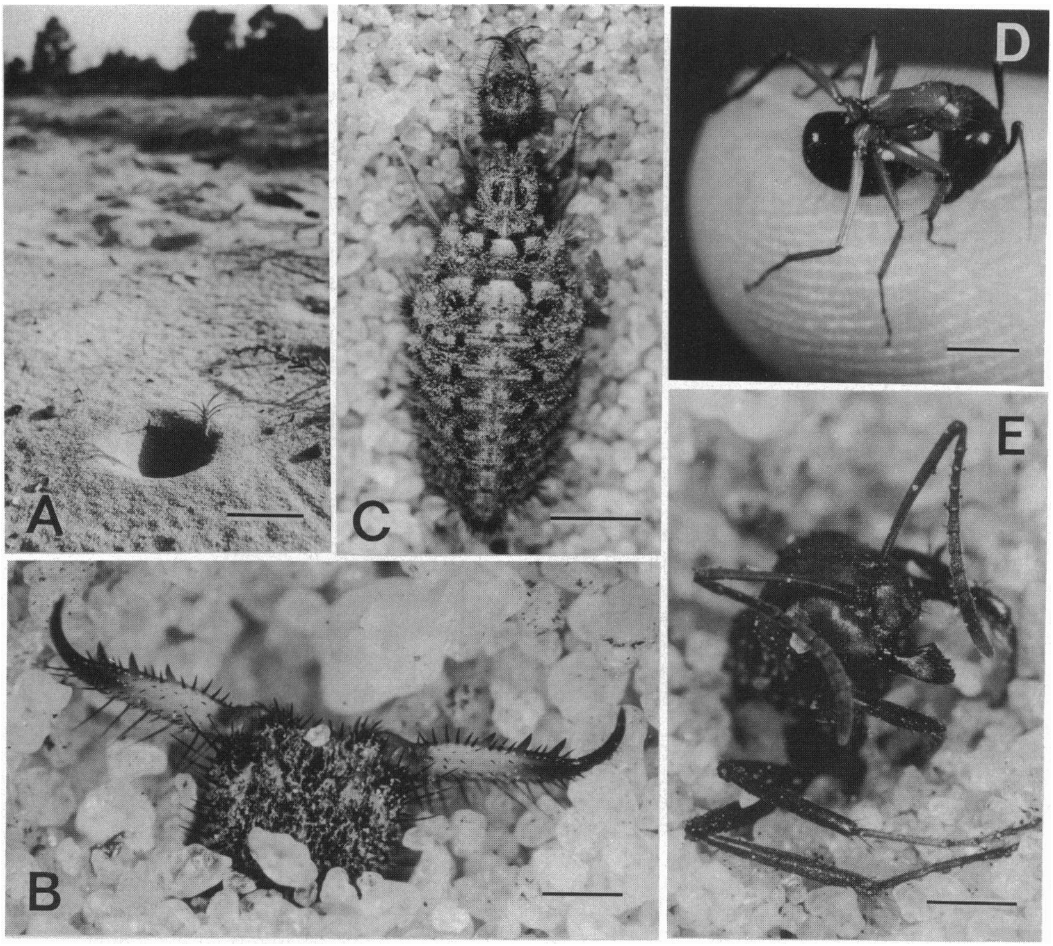

FIG. 1. (*A*) Ant lion pit (unidentified species) in sandy terrain, Lake Placid, FL. (*B*) Head of ant lion (unidentified species) lying in wait, at bottom of pit. (*C*) *Myrmeleon carolinus*. (*D*) *Camponotus floridanus* worker in typical defensive posture, biting with its mandibles while flexing the gaster to direct its acid spray toward the bitten site. (*E*) *C. floridanus* worker, pulled partly into the sand by an *M. carolinus* that has just seized it. [Bars = 5 cm (*A*), 0.5 mm (*B*), 2 mm (*C* and *D*), 1 mm (*E*).]

ants promptly by clamping onto their bodies with their hollow needle-sharp mandibles. Ants struggled for 2 to 20 min (Fig. 1*E*) before suddenly becoming immobile. Death presumably came to the ants when the ant lions pierced their integument and injected enteric fluid through their hollow mandibles. The ant lions then commenced the slow process of sucking up the enzymatically liquefied contents of the ants. During this process, which lasted on average over 2 hr, the ant lions thoroughly probed the insides of the ants, repeatedly shifting the angles of insertion of their mandibles, and even occasionally withdrawing the mandibles and reinserting them at new sites. When they finished their meal, they typically flipped the ant carcasses from the pits.

PROCEDURES AND RESULTS

Ant Lion/Ant Encounters: Body Mass Changes. Twenty-five preweighed ants were individually fed to repletion on honey solution and reweighed. Twenty-four of these were then offered to preweighed ant lions, which were reweighed after they fed on the ants. The discarded ant carcasses were weighed.

The results (Fig. 2) show that the ants gained substantially in mass by drinking (the acquired fluid amounted to 28.5% of the mass of the gorged ant), and lost 44.2% of body mass through predation. The ant lions gained 39.5% in mass as a consequence of feeding. In net equivalents, the ant lost 19.3 ± 7.1 (mean ± SD) mg, of which the ant lion gained about half (8.7 ± 5.0 mg); the difference could represent fluid lost by the ant through evaporation or through leakage during feeding.

Ant Lion/Ant Encounters: Acid Emission by Ants. In seven of the preceding tests, strips of indicator paper were held within millimeters from the ants as they struggled in the ant lion's hold, and then again at intervals while the ants were being sucked out by the ant lions. The papers failed to turn white, indicating that no acid vapor was emanating from the

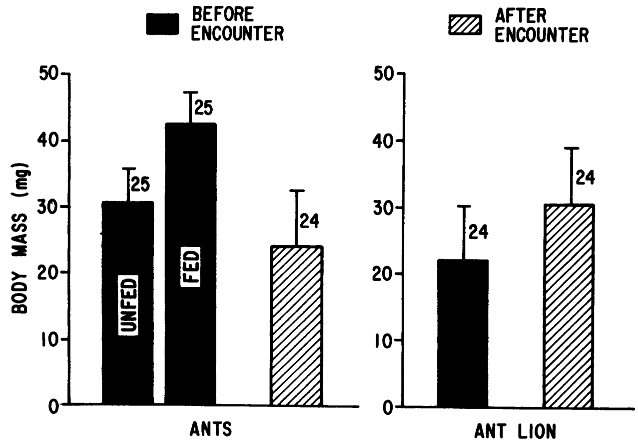

FIG. 2. Mean (±SD) body mass of ants and ant lions before and after encounter. Prior to being offered to the ant lions, the ants were fed to repletion. (Numbers above columns = sample size.)

ants. Control strips of paper held above areas of sand that were wetted with the contents of excised ant acid sacs became discolored instantly.

Ant Lion/Ant Encounters: Fate of Ant Crop and Acid Sac and of Ant Lion Crop. Preliminary dissection of ant carcasses had shown that while the acid sac seemed to be spared by the ant lion, the ant crop was drained. To obtain data on these points, as well as on the gain in content of the ant lion crop, the following organs were excised by dissection and weighed: (*i*) crops (n = 22) from ants gorged on dyed honey solution; (*ii*) crop remains (n = 23) from ant carcasses; (*iii*) acid sacs (n = 19) from ants freshly taken from their colonies; (*iv*) acid sacs (n = 25) from ant carcasses; (*v*) crops (n = 19) from unfed ant lions; and (*vi*) crops (n = 23) from ant lions that were fed ants replete with dyed honey solution.

The results (Fig. 3) confirmed the preliminary findings. The crop in the ant carcasses was essentially reduced to a shrivelled mass, while the acid sac underwent no significant loss in content (Fig. 4 *A* and *C*). The gain in mass of the ant lion crop was equivalent to the gain of the ant lion as a whole (Figs. 2 and 3). Part or all of the ant crop contents had been imbibed by the ant lions: the crop contents of the fed ant lions were distinctly red (Fig. 4*B*). Ingestion of the soft components of the ants had been thorough. While there were remnants of cuticular components in the carcasses (linings of the foregut, hindgut, trachea, and air sacs), little remained of such bulky body parts as the musculature.

Elicitation of Ant Spray Ejection. An observation, familiar to all who have collected formicine ants in the field, provided a clue to why the ants appeared not to spray when attacked by ant lions. When we allowed frenzied ants from a freshly distributed *C. floridanus* nest to swarm over our hands, we noted that the ants would not spray at random but only when biting. Biting seemed to be their first priority. All those that scurried over the hand would pause in an attempt to clamp down with their powerful mandibles, but it was only those that succeeded in biting that bent the gaster forward beneath the body and sprayed (Fig. 1*D*). The behavior seemed intended to combine spray delivery with abrasion or perforation of target site, and it may be common to many formicine ants (11, 12). It seemed possible that in the encounters with ant lions, *C. floridanus* had refrained from spraying because they had not succeeded in biting the predator.

A test was designed to clarify the factors that induce *C. floridanus* to spray. Individual ants from laboratory colonies were grasped by the alitrunk with steel-spring forceps and immediately held over indicator paper (Fig. 4*D*). After an interval, while still held over the paper, they were given a "bit" (an ≈3-mm segment of rubber band delivered to their mandibles with forceps), which they promptly grasped. A record was kept as to whether the ants sprayed when first seized and/or when given the bit. Thirty ants were thus tested (the interval between seizing with forceps and presentation of bit was 15–30 sec).

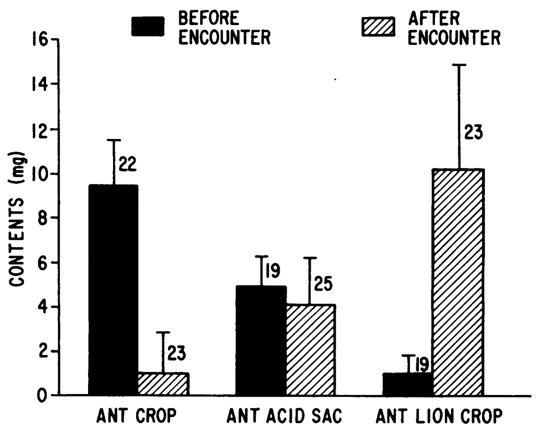

FIG. 3. Mean (±SD) content mass of ant crop, ant acid sac, and ant lion crop, before and after encounter. (Numbers above columns = sample size.)

Most ants (20 of 30) sprayed only when they were given the opportunity to bite. They bent the gaster forward, usually promptly upon seizing the bit, and discharged. The pattern of the spray became clearly demarked on the paper (Fig. 4*E*). Only 3 ants sprayed when seized, and 5 sprayed both when seized and when given the bit. There were 2 nonrespondents.

Volume of Acid Spray Ejected by Ants. Individual ants that were induced to discharge once on indicator paper (by being held in forceps and given a rubber bit as in the preceding test) were killed by refrigeration and their acid sacs were excised and weighed. Comparison of the mass of these sacs (mean ± SD = 2.2 ± 0.8 mg; n = 10) with that of the full sacs (mean ± SD = 4.9 ± 1.4 mg; n = 19; Fig. 2) leads to an estimate of ≈3 μl for volume of single spray ejections.

Sensitivity of Ant Lions to Formic Acid. Individual ant lions were stimulated with 40% aqueous formic acid solution [the spray of formicine ants may contain as much as 60% formic acid (7, 8)] while in the process of feeding on *C. floridanus*. The ant lions were given the ants, were allowed to kill these, and were then stimulated in one of two ways:

(i) *Direct application of acid (*n *= 18).* The ant lions were drawn from the sand by pulling on one of the ant's legs until only the ant lion's head projected free, and were then treated by application of a droplet of formic acid solution (3 μl, the equivalent of a single ejection of the ant) to the head with a micropipette. The time in seconds that the ant lions remained attached to the ants was scored. Failure to detach within 1 min was scored as no response. In 9 of 10 preliminary tests, carried out without application of acid, it was noted that the ant lions did not detach from the prey if partly pulled from the sand and held undisturbed for 1 min.

(ii) *Indirect application (*n *= 20).* The ant lions were allowed to remain concealed beneath the sand with their partly buried prey and were treated by application of a droplet of formic acid solution (6 μl) to the sand directly above their body. Time in seconds to detachment (which was visibly detectable by the sand perturbations induced by the retreating ant lion) was again scored.

Control tests to (*i*) and (*ii*) involved application of distilled water to exposed (3 μl; n = 20) and concealed (6 μl; n = 20) ant lions, respectively.

Direct application of the acid solution was 100% effective: all 18 treated ant lions released their hold within 3 sec of stimulation, unlike the controls, which remained for the most part attached (17 of 20). Indirect application, although it involved administration of twice the volume of acid solution, was no more effective than the control stimulus (17 of 20 and 19 of 20, respectively, held their prey).

DISCUSSION

It seems established that *M. carolinus* subdues *C. floridanus* without inducing the ant to spray and that it consumes the ant's contents without rupturing the acid sac. Given the ant lion's topical sensitivity to formic acid, the strategy is obviously adaptive to the predator.

The data suggest that if the ants had succeeded in biting the ant lions, they might have been prompted to spray. Why they failed to use their mandibles remains uncertain, although it is possible that with their bodies partly buried beneath sand and the ant lion totally buried, they were unable to sense the precise positioning of the predator and to direct their bites. Had the ants sprayed, they would not necessarily have secured their release, since under cover of sand the ant lions would not have been fully vulnerable to the spray. The

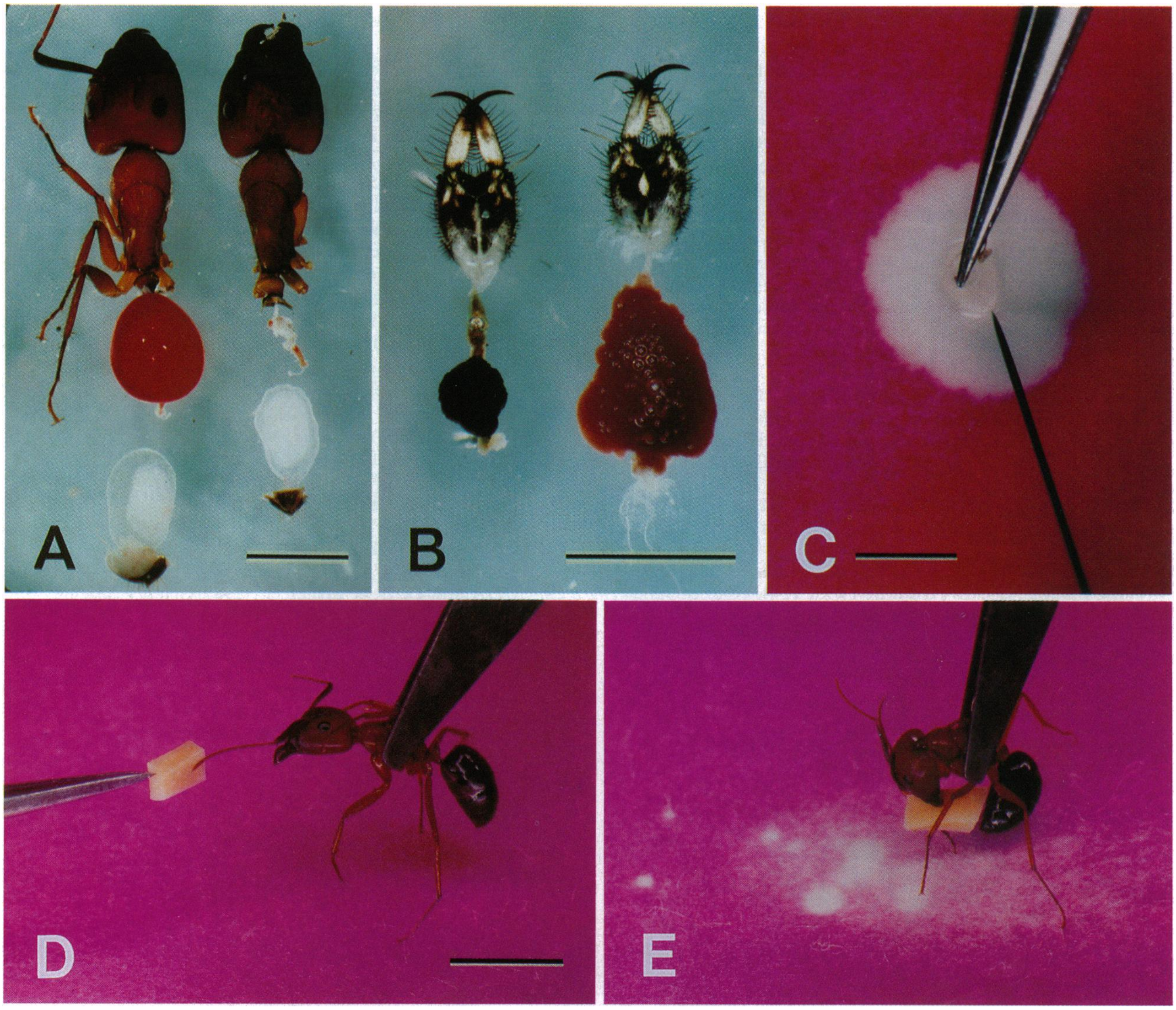

FIG. 4. (*A*) *C. floridanus* workers, partly dissected to reveal the crop and acid sac. The individual on left has been fed on stained (red) honey solution, and its crop is replete (midgut and hindgut have been removed). The individual on right has been killed and sucked out by an *M. carolinus*, which has imbibed the crop contents, leaving only the shrivelled remains of the crop wall. The acid sac (translucent chamber) remains replete in the individual eaten by the ant lion. (*B*) Heads, with crop attached, of *M. carolinus*, before (left) and after feeding on a *C. floridanus* worker. The ant had fed to repletion on stained honey solution. The red color of the crop on right attests to the ant lion's consumption of the ant's crop contents. (*C*) Acid sac recovered from a *C. floridanus* carcass, being drained on indicator paper. The sac, held in forceps, is being punctured with a pin. The leaking fluid is turning the paper white. (*D*) *C. floridanus* worker, held in forceps above indicator paper, being offered a rubber "bit." The bit was wiggled as it was presented to the ant's mandibles, to simulate liveliness. (*E*) Sequel to preceding: the ant has seized the bit and has flexed its gaster forward to spray upon it; the spray has discolored the indicator paper. [Bars = 3 mm (*A*, *B*, and *C*), 5 mm (*D*).]

protection conferred by sand has been shown to be decisive in enabling ant lions to capture bombardier beetles. These insects discharge an aimed jet of hot (100°C) quinones when disturbed (13). Ant lions are able to retain captured bombardier beetles only if they withdraw into the sand before the beetles discharge their first ejection (14).

Rupturing the acid sac could potentially render the ant's entire body contents unacceptable to the ant lion. How the ant lions manage to spare the acid sac in the course of their mandibular probings of the ant's innards, and how they differentiate between this sac and the crop, which is similarly thin-walled but is drained of its contents, remain a mystery. Conceivably the ant lions discriminate between the two sacs on the basis of taste (the cellular investiture of the acid sac could contain discernable amounts of formic acid), by use of chemoreceptors on the mouthparts.

The crop in worker formicine ants is capacious (15) and can take up much of the volume (together with the acid sac) left unfilled in the ant's gaster by the absence of developed gonads. In fully fed *C. floridanus* workers, the crop holds upward of 8 mg of fluid, a near equivalent of what the ant lion gains by feeding on the ant. The ant lion's ability to recognize and puncture this sac, and to imbibe its contents, must thus be viewed as an important concomitant of its predation strategy.

Ants other than Formicinae may be more successful in using their mandibles in defense. Workers of the fire ant, *Solenopsis invicta*, sometimes succeed in biting ant lions when captured and may thereby secure their release. They occasionally even commit "altruistic suicide," clamping onto an ant lion's mandible and dying without releasing their hold, with the result that the ant lion itself succumbs to starvation (16).

Many predators have evolved strategies that enable them to circumvent the defenses of their prey. Orb-weaving spiders, for instance, may shield themselves from the defensive chemical discharges of insect prey by encasing the latter in silk (17), while grasshopper mice subdue chemically protected beetles by first causing these to discharge their spray into the soil (18). Herbivores also may possess strategies for bypassing the defenses of plants (19–22). Ant lions are among the most successful of ant predators, and Formicinae are a dominant group of ants. The details of interaction described here for *M. carolinus* and *C. floridanus* may thus be pertinent to a type of encounter of widespread occurrence in nature.

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1. Attygalle, A. B., Xu, S.-C., McCormick, K. D., Meinwald, J., Blankespoor, C. L. & Eisner, T. (1993) *Tetrahedron*, in press.
2. Rösel, A. J. (1755) *Insekten Belustigung*, part III, pp. 101–132.
3. von Frisch, K. (1974) *Animal Architecture* (Harcourt Brace Jovanovich, New York).
4. Lucas, J. R. (1982) *Anim. Behav.* **30**, 651–664.
5. Youthed, G. J. & Moran, V. C. (1969) *J. Insect Physiol.* **15**, 867–875.
6. Wray, J. (1670) *Philos. Trans. R. Soc. London* **68**, 2063–2083.
7. Stumper, R. (1951) *C.R. Acad. Sci.* **233**, 1144–1146.
8. Osman, M. F. H. & Brander, J. (1961) *Z. Naturforsch.* **16**, 749–753.
9. Lucas, J. R. & Stouge, L. A. (1981) *Fl. Entomol.* **64**, 207–216.
10. Hölldobler, B. & Wilson, E. O. (1990) *The Ants* (Harvard Univ. Press, Cambridge, MA), p. 732.
11. Buschinger, A. & Maschwitz, U. (1984) in *Defensive Mechanisms in Social Insects*, ed. Hermann, H. R. (Praeger, New York), pp. 95–150.
12. Schmidt, J. O. (1986) in *Venoms of the Hymenoptera*, ed. Piek, T. (Academic, New York), pp. 425–508.
13. Aneshansley, D. J., Eisner, T., Widom, J. M. & Widom, B. (1969) *Science* **165**, 61–63.
14. Conner, J. & Eisner, T. (1983) *Psyche* **90**, 175–178.
15. Eisner, T. (1957) *Bull. Mus. Comp. Zool.* **116**, 439–490.
16. Lucas, J. R. & Brockman, H. J. (1981) *J. Kans. Entomol. Soc.* **54**, 228–232.
17. Eisner, T. & Dean, J. (1976) *Proc. Natl. Acad. Sci. USA* **73**, 1365–1367.
18. Eisner, T. (1966) *Nat. Hist.* **75**, 42–47.
19. Eisner, T., Johnessee, J. S., Carrel, J., Hendry, L. H. & Meinwald, J. (1974) *Science* **184**, 996–999.
20. Morrow, P. A., Bellas, T. E. & Eisner, T. (1976) *Oecologia* **24**, 193–206.
21. Common, I. F. B. & Bellas, T. E. (1977) *J. Austr. Entomol. Soc.* **16**, 141–147.
22. Dussourd, D. E. & Eisner, T. (1987) *Science* **237**, 898–901.