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LARVAL CASE-MAKING BEHAVIOR OF A PESTIFEROUS CHIRONOMID, *GLYPTOTENDIPES PARIPES* (DIPTERA: CHIRONOMIDAE), WITH SAND GRAINS OF DIFFERENT SIZES[1]

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ABSTRACT. Field-collected 4th-instar *Glyptotendipes paripes* larvae from a natural lake in central Florida preferred small-sized (<0.84–1.99-mm-diam) grains of sand to burrow in and make cases in the laboratory. The larvae buried or formed cases within 3 h of introduction into 120-ml paper cups containing 80 ml tap water and 20 g of the small-diameter sand. When placed in sand with grains of different size ranges, 98–100% of the larvae constructed cases in sand <0.84 mm in diameter, whereas 86% of these larvae built cases in 0.85–1.99-mm-diam sand. The larvae were unable to construct any recognizable cases in coarser sand (2–3-mm diam). The lengths of cases built with smaller sand grains were significantly longer than those constructed in the larger grains used in the experiment. For laboratory colonization and insecticidal bioassay purposes using *G. paripes* larvae, sand grains <0.84 mm in diameter should be utilized for better acclimation of the larvae.

Adult chironomid swarms frequently emanate from some aquatic habitats situated amid urban and suburban centers in many parts of the world (Ali 1995). At times, these swarms develop to the degree that in addition to being a severe nuisance they also cause economic problems to visitors, residents, workers, and businesses around the water resources (Ali 1991). Medical problems, such as human allergies, caused by Chironomidae have also been reported (Cranston 1995). Larvae of a large number of bottom-dwelling chironomid species either burrow or live in tubes (cases) constructed of substrate materials (McLachlan 1976, Brennan and McLachlan 1979). Thus, substrate particle size in natural aquatic habitats may affect the larval distribution (Cummins and Lauff 1969, Moore 1980) and dispersal (Davies 1976) of many species of chironomids.

In central Florida, *Glyptotendipes paripes* Edwards is one of the major pestiferous species of chironomids. This species has a high fecundity potential. Research on the bionomics and control of *G. paripes* and other chironomid species of central Florida were summarized recently (Ali 1996). As a part of this continuing research, case-building behavior of larval *G. paripes* when provided with different sizes of sand grains was studied and is reported here. Such information is necessary for understanding the distributional ecology of *G. paripes* in the field, as well as in the selection of appropriate sand size for laboratory use to provide substrates during insecticidal bioassays against larvae of this species.

To study case-making behavior, *G. paripes* larvae were collected from Lake Monroe, Sanford, FL, and 4th instars were separated in the laboratory. Different sizes of sand grains were obtained from 2 kg of construction sand sterilized at 100°C for 24 h. The sterilized sand was passed through a series of U.S. standard sieves to obtain size (diameter) ranges of <0.25, 0.35–0.58, 0.59–0.84, 0.85–1.99, and 2–3 mm. The study was conducted in 120-ml disposable papers cups (7-cm diam and 4 cm high). For each size group 3 replicates were utilized. Each cup contained 20 g of sand of each particle size range and 80 ml distilled water to which 20 4th-instar *G. paripes* larvae were introduced. The cups were maintained at 25–27°C in the laboratory. Burrowing behavior and construction of cases by the larvae in the cups were checked at 0.5, 1, 3, and 24 h after the larval introductions. The number of cases constructed and case lengths in each cup were counted and measured at 3 days postintroduction. The experiment was conducted on 3 different occasions and the resulting data were analyzed by using a χ^2 test and one-way analysis of variance (ANOVA) (Tukey's honestly significant difference [HSD] [at $P = 0.05$] was used for separation of means). Larval movement and behavior when buried in the substrates was observed by introducing 4th instars into a glass container (7 cm long, 3 cm wide, and 4 cm high) half filled with glass powder (0.425–0.6-mm beads; Sigma Chemical Company, St. Louis, MO) under 2 cm of distilled water.

Immediately after introduction of larvae to the test cups or glass container, the larvae attempted to enter sand or glass powder at the sediment–water

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Table 1. Mean percent (±SD) field-collected 4th-instar *Glyptotendipes paripes* larvae entering sand of different sizes and making cases when introduced into sand and water in disposable paper cups in the laboratory.

Sand size (mm)	No. larvae introduced	% larvae entered after introduction			% cases formed after 3 days
		0.5	1	3	
<0.25	60	100 ± 0	100 ± 0	92 ± 0.6	98 ± 0.6
0.35–0.58	60	98 ± 0.5	96 ± 0.5	92 ± 0.6	100 ± 0
0.59–0.84	60	98 ± 0.5	92 ± 0.6	67 ± 1.5	98 ± 0.6
0.85–1.99	60	96 ± 1.2	96 ± 1.2	46 ± 1.2	86 ± 0.6
2.00–3.00	60	91 ± 0.7	100 ± 0	8 ± 0.6	16 ± 0[1]

[1] Larvae with a fiberlike transparent secretion around their body. $\chi^2 = 113.335$, df = 12, $P < 0.001$ (for mean differences between all sizes and observation times [s×4]).

interface by probing with their anterior end (head). After 0.5 h, a majority of the larvae in all test cups and in the container had completely burrowed in bottom sediments. After 1 h, many larvae started moving towards the substrate surface and at 3 h postintroduction, some larvae completely exposed themselves and engaged in the maneuver of reentering the substrates. One day after introduction many larvae remained buried, whereas others built cases by binding bottom sediments with salivary secretions. These cases were of variable length and either projected vertically from the sediment–water interface or were placed horizontally on sediments in the cup bottom. A majority of the vertically oriented cases contained larvae with heads positioned towards the mouth of the cup. Some empty vertical and horizontal cases were also observed in each cup.

Data concerning *G. paripes* larvae entering and exiting sand of different sizes are summarized in Table 1. Three hours after introduction, 92% of the total larvae introduced to 2–3-mm-diam sand had exited, whereas in sand with diameters of 0.85–1.99, 0.59–0.84, and 0.35–0.58 mm, 54, 33, and 8% of the total larvae had exited, respectively. In the finest sand (<0.25 mm), 8% of the total larvae introduced had exited by 3 h after introduction. At day 3 after the larval introductions, 86% of the larvae introduced to 0.85–1.99-mm-diam sand had formed cases. Similarly, in sand <0.84 mm in diameter, 98–100% of the total larvae introduced had formed cases. Larvae were unable to form any recognizable cases in the largest size sand of 2–3 mm (Table 1). Sixteen percent of these larvae contained visible fiberlike transparent secretions around their body. This was probably their salivary secretions utilized for binding substrate materials.

The case lengths of larval *G. paripes* measured to the nearest mm ranged from ≤1 cm to ≥2.6 cm, with no cases recovered from 2–3-mm-diam sand (Table 2). In <0.25-mm-diam sand, 97% of larval tubes ranged from 1.1 to 2.5 cm in length, with mean case length being 2.1 cm. In 0.35–0.58-, 0.59–0.84-, and 0.85–1.99-mm sands, the mean case lengths were 1.8, 1.8, and 1.1 cm, respectively. Data analysis indicated that sand size significantly influenced case length ($F_{3,54} = 26.26$; $P < 0.001$). Multiple comparisons indicated a significant difference between case lengths in sand <0.25 mm and the 3 larger size sand groups. Case lengths in 0.85–1.99-mm-diam sand significantly differed from those in the smaller 3 sand sizes, but no significant difference was found between case lengths in 0.35–0.58-mm-diam and 0.59–0.84-mm-diam sand. Generally, a trend of longer cases in the smaller sized sand was observed.

The tube-building behavior of chironomid larvae may vary by species (Chaloner and Wotton 1996) and numerous other factors. Our study shows that sand particle size affected case-making behavior of larval *G. paripes*. The larvae preferred smaller sized sand to make longer tubes. For laboratory colonization and insecticidal bioassays, smaller sized (<0.84-mm-diam) sand should be used. Also, this species prefers field habitats with substrates of fine sand and mud (muck) (Cowell and Vodopich 1981) and its abundance and distribution are affected by

Table 2. Larval case lengths and mean length (percentage) of *Glyptotendipes paripes* cases in sand of different sizes in the laboratory.

Sand size (mm)	No. larvae introduced	No. (%) cases in various ranges of case lengths (cm)					Mean ± SD[1] (cm)
		≤1.0	1.1–1.5	1.6–2.0	2.1–2.5	≥2.6	
<0.25	60	0	13	53	31	2	2.1 ± 0.35
0.35–0.58	60	0	35	35	27	2	1.8 ± 0.33
0.59–0.84	60	4	47	38	11	0	1.8 ± 0.43
0.85–1.99	60[2]	78	18	0	0	0	1.1 ± 0.18
2.00–3.00	60	0	0	0	0	0	—

[1] $F_{3,54} = 26.26$; $P < 0.001$.
[2] Two larvae did not form cases.

substrate particle size (Cummins and Lauff 1969, McLachlan 1976, Moore 1980). In artificial aquatic habitats, the introduction of coarser substrate material may discourage the breeding and propagation of *G. paripes.*

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