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## EXAMINATION OF METHODS FOR FORMOSAN SUBTERRANEAN TERMITE (ISOPTERA: RHINOTERMITIDAE) FECES RECOVERY

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Chemical assay of undigested food from termite fecal material has been reported for Formosan subterranean termites (*Coptotermes formosanus* Shiraki) and other species (Itakura et al. 1995; Hyodo et al. 1999; Mishra & Sen-Sarma 1979; Katsumata et al. 2007). Unlike drywood termites, which produce solid fecal pellets with a conspicuous appearance (Lewis et al. 2010), subterranean termites produce liquid feces (Gouge et al. 2001) that is potentially difficult to recognize and obtain. The current study describes the outcome of attempts at recovery of *C. formosanus* feces from laboratory arenas in which no food had been placed, and from arenas containing filter paper or wood.

Termites used for this study were collected at Audubon Park, New Orleans, LA. Arenas without food were maintained and processed 2 different ways. For the first set of arenas, 18 groups of approximately 50 workers that had been fed southern yellow pine (Pinus sp.) for at least 72 h were transferred to pre-weighed bowls shaped from aluminum foil. Bowls containing termites were placed in polystyrene boxes (32 cm × 25 cm × 10 cm) (Tri-State Plastics, Latonia, KY) lined with saturated paper towels. The boxes were covered and maintained at 23°C. At 3, 6, 20, 24, 40, and 48 h, workers were removed from three randomly selected bowls by allowing them to crawl up a filter paper disc. After removal of dead insects and body parts, bowls were dried 15 min at 70°C, and folded closed immediately upon removal from heat. Fecal dry weight was determined to 0.01 mg from the difference between the initial and final weight of the bowl. For the second set of arenas, 3 groups of approximately 500 workers fed southern yellow pine for at least 72 h were transferred to polystyrene boxes lined with aluminum foil over saturated paper towels. Containers were covered and maintained at 23°C. After 4, 8, 12, 28, and 38 h, termites were transferred to clean polystyrene boxes. Following each transfer, dead termites were removed, and air-dried feces transferred to pre-weighed aluminum foil bowls with a watercolor brush. Feces were further dried 15 min at 70°C before weighing. For arenas containing filter paper, 3 groups of 50 workers were placed in plastic screw-top vials (40 mm  $\times$  15 mm diameter) along with a piece of moistened Whatman 1 filter paper (Whatman Inc., Piscataway, NJ). Vi-



Fig. 1. Mean (+SEM) dry weight of *C. formosanus* feces recovered from laboratory arenas without food. Graph A: mean feces recoveries from 3 randomly selected foil bowls holding 50 workers. Graph B: repeated feces collection from containers that each held 500 workers and soldiers.

als were loosely capped and maintained sideways at 28°C in the dark. After 1 week insects were removed and collection of fecal material attempted by scraping vials and paper with a razor blade and spatula. For laboratory arenas containing wood blocks, 5 groups of approximately 200 to 500 workers and soldiers were placed in clear polystyrene containers lined with a piece of aluminum foil over saturated pa-



Fig. 2. Cemented structures constructed by *C. formosanus* termites in a laboratory arena containing southern yellow pine blocks.

per towels. Southern yellow pine blocks (either 50 mm  $\times$  20 mm  $\times$  5 mm, or 20 mm<sup>3</sup>) that had been soaked in distilled water for 1 h were added to arenas. Containers were covered and stored at 28°C. Accumulated debris on foil and wood was periodically collected from 4 arenas with a spatula and razor blade, while a fifth arena was left undisturbed for 6 weeks.

The weight of fecal material recovered from arenas that had not contained food averaged approximately 0.03 mg to 0.2 mg feces (dry weight) from foil bowls, and about 1 mg total feces (dry weight) after repeated collections from the same groups of insects (Fig. 1). Feces in these arenas were apparent either as liquid droplets that dried to dark solid specks, or collections of small, sticky black piles mixed with dead termites, body parts, and trapped live termites. No feces were recovered from arenas containing filter paper, although liquid fecal material was apparent from dark spots on the paper. For arenas with wood, solid debris was apparent on wood blocks and arena surfaces within 72 h. Debris was formed into branched structures in the single undisturbed arena (Fig. 2). The structures were soft and friable immediately upon removal from arenas, and when pulverized dried to a fine powder. Considering the minute quantity of feces recovered from arenas without food (Fig. 1), little fecal material appears to have been available for use in construction of the structures. Feces were not visually discernible from any arena containing wood blocks.

Observations from the current study underscore the need for caution in collection of *C. formosanus* fecal material for assay because it can easily be contaminated by foreign particles or unrecoverable if food is present. Furthermore, collection of feces for assay from arenas without food may be impractical because of low yield (Fig. 1). For instance, this species digests wood carbohydrate very efficiently (Yoshimura 1995), so large numbers of termites would be needed to produce fecal material with enough undigested sugars for detection by conventional chromatographic methods.

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## SUMMARY

The current study attempted different methods for collection of Formosan subterranean termite feces. When food was present, the liquid fecal material was not recoverable either due to absorption into food, or occlusion by food particles that were used instead for construction purposes. Uncontaminated fecal material was recoverable from arenas without food only in minute quantity.

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