

# Hydramethylnon Uptake by *Blattella germanica* (Orthoptera: Blattellidae) by Coprophagy

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**ABSTRACT** *Blattella germanica* (L.) that were fed hydramethylnon bait produced residues that were toxic to exposed conspecifics. Insecticidal activity was traced to the feces of treated insects by feeding radiolabeled material, where  $\approx 50\%$  of the recovered radioactivity was unmetabolized parent compound. Ingestion of toxicant-laden feces by all life stages was evident, but the effect of this behavior was greatest on early instar nymphs. Baits containing toxicants with delayed activity, such as hydramethylnon, probably affect cockroach field populations indirectly through coprophagy.

**KEY WORDS** Insecta, *Blattella germanica*, coprophagy, hydramethylnon

HYDRAMETHYLNON has been used in bait form (COMBAT Roach Control System, MAXFORCE, American Cyanamid, Clifton, N.J.) for several years against indoor cockroach populations (Patterson & Koehler 1989, Appel 1990). This chemical has delayed action, with no symptoms of intoxication generally occurring for 24 h after exposure (Silverman & Shapas 1986). Such delayed activity is of obvious importance for the control of ants (Banks et al. 1981, T. H. Su et al. 1980) and termites (N. Y. Su et al. 1982) because it permits toxicant transfer among colony members. The importance of delayed toxicant activity in the control of insects that are not eusocial, such as cockroaches, is not as obvious.

Many arthropods, particularly detritus feeders, use a coprophagous strategy to obtain nutrients. Most research on arthropod coprophagy, however, has focused on ruminant dung (Stevenson & Dindal 1987). A millipede, *Apheloria montana* (Bollman) (McBrayer 1973), and passalid beetle, *Popilius disjunctus* (Illiger) (Mason & Odum 1969), require ingestion of their own feces for survival. Because both species lack internal symbiotes to process food materials, specifically cellulose, extraintestinal microbial decomposition and coprophagy permit more efficient nutrient utilization. Coprophagy by cockroaches has received little attention. Schal & Bell (1982) observed *Xestoblatta hamata* (Giglio Tos) males feeding on bird droppings and demonstrated male-to-female-to-offspring transfer of uric acid obtained from high nitrogen diets. Schowalter & Crossley (1982) speculated that coprophagy had some effect on the non-feces food consumption rate by the cockroach detritivore *Gromphadorhina portentosa* (Schaum). Burnett et al. (1969) demon-

strated that *Cryptocercus punctulatus* Scudder does not ingest its own feces.

We noticed considerable mortality when individual German cockroaches, *Blattella germanica* (L.), were placed in containers harboring residues produced by insects that fed on hydramethylnon bait. Here we describe this phenomenon in some detail. We also discuss delayed toxicant activity and coprophagy as they relate to cockroach behavior and control.

## Materials and Methods

Analytical grade hydramethylnon (98.6%) and [<sup>14</sup>C]pyrimidinyl-labeled hydramethylnon were obtained from the Agricultural Research Division, American Cyanamid, Princeton, N.J. The specific activity of the labeled compound was  $8.7 \times 10^5$  Bq/mg (11.9 mCi/mmol). Radioactive baits were prepared using a 1:100 dilution of radiolabeled hydramethylnon in unlabeled hydramethylnon in the COMBAT bait base. The final specific activity of the baits was  $8.7 \times 10^3$  Bq/mg hydramethylnon.

All wild-type *B. germanica* used in these experiments were from colonies maintained at American Cyanamid. The colonies have been free from insecticide exposure for >30 yr. Orange body mutants were obtained from M. Ross, Virginia Polytechnic Institute and State University, Blacksburg, Va. Insects were reared and experiments were performed at  $27 \pm 1^\circ\text{C}$ ,  $50 \pm 2\%$  RH, and a photoperiod of 12:12 (L:D).

**Exposure to Residues.** Hydramethylnon formulated at a concentration of 2% in the COMBAT bait base was fed to 10 male *B. germanica* (six replicates). The insects, bait, water, and harborage (folded 4-cm-diameter filter paper disk) were held in 0.55-liter, wide-mouth jars with a light film of petroleum jelly-mineral oil along the inner rim to

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prevent cockroach escape. After the treated cockroaches died (3–4 d after treatment), all insects and bait remnants were removed. Fifty first and second instars were added to each of these jars. Three replicates received untreated dog food (Ralston Purina, St. Louis) in addition to the residues. Jars containing residues from males fed untreated dog food served as a control. Nymphal mortality was assessed at 3 and 14 d.

**Origin of the Cockroach-Derived Residues.** Residues were obvious in containers harboring *B. germanica* that were fed hydramethylnon bait. The source of these residues (oral secretions or feces) was therefore determined. Two strips of corrugated cardboard (2 cm wide) were held together along the long edge by a strip of modeling clay (3 mm wide). One hundred and ten male *B. germanica*, previously starved for 3 d, were fed 2% hydramethylnon for 3 h, anesthetized with CO<sub>2</sub>, and placed across the clay so that each insect's head (ventral side down), was resting within the trough of one cardboard strip and the terminal abdominal segments were on the adjacent strip. A strip of clay was placed against the insects' dorsa to hold them in place. After the death and removal of the males, each cardboard strip was placed in a separate 0.55-liter jar containing 50 first and second instars, dog food, and a water vial. Nymphal mortality was assessed for 18 d.

In addition, 48 male *B. germanica* were fed <sup>14</sup>C-hydramethylnon bait (specific activity,  $8.7 \times 10^9$  Bq/mg) and handled as above. Material collected on four cardboard sections from beneath the cockroach head or abdomen was combined (12 replicates). These samples were combusted in a Packard Model 306 biological oxidizer (Packard Instrument, Downers Grove, Ill.) and counted in a Beckman LS 9800 liquid scintillation spectrometer (Beckman Instruments, Irvine, Calif.).

**Stage-Specific Hydramethylnon Excretion.** Various developmental stages of *B. germanica* were housed separately in 0.55-liter jars, starved for 2 d, and fed 2% <sup>14</sup>C-hydramethylnon baits for up to 72 h. The average amount of bait ingested per insect was determined by weighing baits before and after they were fed upon, then dividing by the number of insects exposed to each bait. This value times the specific activity of the original bait gave the level of radioactivity ingested per insect. After death, insects from each stage treated with hydramethylnon were sorted and replicated as follows: first and second instars ( $n = 10$ , 10 replicates), third and fourth instars ( $n = 5$ , 7 replicates), fifth instar ( $n = 5$ , 7 replicates), non gravid females ( $n = 4$ , 10 replicates), gravid females ( $n = 4$ , 10 replicates), and males ( $n = 5$ , 6 replicates). Each replicate was combusted and counted separately. The level of carcass radioactivity was subtracted from the level within the ingested bait to give the amount of <sup>14</sup>C excreted. Parent hydramethylnon was assayed as described by Hollingshaus & Little (1984). Briefly, feces from males were collected and

weighed, then homogenized three times in 5 ml MeOH. The supernatant was collected after centrifugation and reduced under a stream of N<sub>2</sub>. Proteins were precipitated with acetone at 0°C. Extracts were spotted on precoated silica gel 60 F-254 plates of 0.25 mm thickness (MCB Reagents, Gibbstown, N.J.) and developed with toluene/dimethoxyethane/ammonium hydroxide (50:50:1, vol/vol) in the first dimension and acetonitrile/2-propanol/acetic acid (33:7:1, vol/vol) in the second dimension. Spots were visualized under UV light or by autoradiography using Kodak XOMAT AR film (Eastman-Kodak, Rochester, N.Y.). Baits, feces, and thin-layer chromatography spots containing radioactivity were combusted and counted.

**Toxicant Activity in Feces Following Topical Application.** Male *B. germanica* (starved 2 d,  $n = 20$ , 5 replicates) were treated topically between the metacoxae with 50 µg hydramethylnon in 1 µl acetone. They were subsequently fed bait without toxicant for 1 h. Dead males were removed and first and second instars were exposed to the feces produced by these males. Feces from males fed hydramethylnon bait served as a positive control. All insects were provided with dog food and water. Nymphal mortality was assessed at day 7.

**Effect on Developmental Stages.** We considered the possibility that certain *B. germanica* developmental stages might restrict foraging and instead exploit cockroach feces in the harborage as a food source. First instars ( $n = 42$ –91), second and third instars ( $n = 60$ –194), fourth and fifth instars ( $n = 73$ –86), gravid females ( $n = 7$ –16), and non gravid females ( $n = 8$ –14) were caged separately in plastic containers (38 by 26 by 15 cm). An inverted cardboard harborage box (11 by 6 by 6.5 cm) was placed within each container, and dog food and a water vial were placed within each harborage. Twenty replicates were used for each stage. One-half (10) of these replicates received an additional 50 male cockroaches. All insects were allowed to acclimate for 3 d, at which time a 2% hydramethylnon bait was placed about 25 cm from the harborage. Baits were removed after 2 d and mortality was assessed 5 d after bait removal.

Mixed stages of phenotypically wild-type *B. germanica* ( $n = 113$ –218) were allowed to acclimate for 3 d in the containers described previously. However, no insecticide bait was provided. Instead, males from a strain of orange body *B. germanica* ( $n = 25$ ), previously starved for 2 d, were fed 2% hydramethylnon bait for about 1 h and then released into the containers with the wild-type insects. This procedure was replicated 10 times. Mortality of both cockroach strains was assessed 7 d after the orange body insects were placed in the containers.

Percentage data were transformed to arcsine of the square root for data analysis. Nonpaired *t* tests were performed and analysis of variance (ANOVA) was conducted with Fisher's multiple comparison procedure (Ryan et al. 1985) for mean separations.

**Table 1.** Excretion of  $^{14}\text{C}$ -hydramethylnon and metabolites by various stages of *B. germanica* after ingestion of 2% baits

Stage	$^{14}\text{C}$ Excreted, $\mu\text{g}$	$^{14}\text{C}$ Excreted as % ingested
1st & 2nd Instar	1.3 $\pm$ 0.25a	29.5 $\pm$ 5.63a
3rd & 4th Instar	13.8 $\pm$ 2.21b	32.1 $\pm$ 10.90a
5th Instar	41.1 $\pm$ 4.06c	29.7 $\pm$ 6.98a
Nongravid ♀	53.8 $\pm$ 6.96d	44.7 $\pm$ 7.15a
Gravid ♀	12.2 $\pm$ 1.80b	38.5 $\pm$ 5.66a
♂	35.8 $\pm$ 2.45c	22.3 $\pm$ 2.85a

Results are the mean  $\pm$  SEM. Column means followed by the same letter are not significantly different ( $P = 0.05$ ; Fisher's multiple comparison procedure [Ryan et al. 1985]).

Nonparametric analyses were performed with the Kruskal-Wallis test (Ryan et al. 1985).

### Results

**Exposure to Residues.** All male *B. germanica* fed hydramethylnon bait died within 3 d. Ninety-nine percent of the nymphs exposed to the residues produced by the males fed toxicant died by day 3. These nymphs were observed feeding on the residues. When nymphs were provided with untreated dog food in the presence of hydramethylnon residues produced by males, mortality was 93% by day 3 and 100% by day 14. Mortality of control nymphs exposed only to the residues from untreated males was 0% at day 3 (Kruskal-Wallis test;  $H = 10.87$ ,  $df = 3$ ,  $P < 0.025$ ) and 88% by day 14. Because no control mortality was evident when untreated food was also provided, these insects probably died from starvation.

**Origin of Cockroach-Derived Residues.** More than 99% of the total radioactivity from  $^{14}\text{C}$ -hydramethylnon fed males was recovered in the feces. Whereas, none of the nymphs exposed to cardboard strips on which the treated male's heads were resting died, 90% mortality occurred from the cardboard contaminated by abdomens.

**Stage-Specific Hydramethylnon Excretion.** All stages of *B. germanica* excreted hydramethylnon (Table 1), with nongravid females excreting the most and first and second instars the least ( $F = 29.03$ ;  $df = 5, 44$ ;  $P < 0.0001$ ). When the amount excreted was considered as a percentage of that ingested, there were no significant differences be-

tween the stages ( $F = 1.26$ ;  $df = 5, 44$ ;  $P > 0.05$ ). Analysis by thin-layer chromatography revealed that 51% of the radioactivity recovered from the feces was parent hydramethylnon compared with the level of parent compound in the prepared bait.

**Toxic Activity in Feces after Topical Application.** When exposed to the feces of males that had been given a topical dose of hydramethylnon, 61  $\pm$  5.5% of the nymphs died. Nymphal mortality was 95  $\pm$  3.0% in the presence of feces from males fed hydramethylnon bait ( $t = 5.21$ ,  $df = 8$ ,  $P < 0.0001$ ).

**Effect on Developmental Stages.** Mortality of first through third instars increased significantly ( $P < 0.001$ ) in the presence of adult males when both were exposed to hydramethylnon baits (Table 2). The harborages from the replicates containing males were noticeably contaminated with feces having an amber appearance, characteristic of material from insects intoxicated by hydramethylnon bait. This material was not apparent in containers harboring only young nymphs. The addition of males to containers with older nymphs or females did not increase mortality of these stages. By the end of the study, male mortality was complete in all containers.

Ninety-eight percent of the orange body cockroaches fed hydramethylnon bait and released among wild-type insects died by day 7. In addition, 41% of all the wild-type insects died. The mean  $\pm$  SEM% mortalities for each of the following stages of wild-type insects were males, 81.5  $\pm$  2.1; nongravid females, 47.6  $\pm$  4.4; gravid females, 22.0  $\pm$  4.2; and nymphs, 35.0  $\pm$  2.0. The differences between these stages were significant ( $F = 62.56$ ;  $df = 3, 36$ ;  $P < 0.001$ ).

### Discussion

Our results demonstrate that hydramethylnon is excreted in the feces of all stages of *B. germanica*. These feces are toxic to other individuals when consumed (hydramethylnon from bait and excreted in feces is not effective by cuticular contact [J.S., unpublished data]). Furthermore, uptake of hydramethylnon by coprophagy increases the effect of the toxicant because more insects are killed in addition to those consuming the bait directly. Necrophagy may be of some importance in the transfer of hydramethylnon and other toxicants in the

**Table 2.** Effect of coprophagous uptake of hydramethylnon on various developmental stages of *B. germanica*

Stage	% Mortality $\bar{x} \pm$ SEM		<i>t</i>	<i>P</i>
	♂♂ present	♂♂ absent		
1st Instar	20.4 $\pm$ 4.59	1.8 $\pm$ 0.95	3.97	0.0009
2nd & 3rd Instar	77.2 $\pm$ 4.03	37.4 $\pm$ 3.80	7.21	0.0001
4th & 5th Instar	94.2 $\pm$ 1.96	90.3 $\pm$ 2.95	0.85	0.41
Nongravid ♀	93.0 $\pm$ 2.82	91.8 $\pm$ 2.74	0.39	0.70
Gravid ♀	70.8 $\pm$ 5.57	69.0 $\pm$ 4.79	0.47	0.64

$df = 18$  for each row comparison.

field. However, in studies where cockroaches had access to insects fed bait in the laboratory, no consumption of insect carcasses was evident. Cockroaches were fed hydramethylnon in a single bait base in our studies. Bait base composition may affect the level of secondary mortality by influencing hydramethylnon excretion, feces palatability, or both. Although metabolites were evident in *B. germanica* feces, parent hydramethylnon is most probably the actual toxicant because the identifiable metabolites have no insecticidal activity (Hollingshaus & Little 1984). Hydramethylnon also appears in the feces at lethal levels after topical application. Although poorly understood, transport of hydramethylnon from the cuticle to the gut via the hemocoel is apparent.

To our knowledge, this is the first report of an insect that is not eusocial acquiring a toxicant from conspecifics. Hydramethylnon, mirex, and some other materials have been used successfully in ant control because they are distributed to colony members before the death of the foraging workers (Lofgren et al. 1964, Manley 1982). The "bait block method" of subterranean termite control also relies upon intracolony toxicant transfer (Beal & Esenther 1980). An example of toxicant transfer between unrelated taxa is the inhibition of dung-breeding hematophagous dipteran development by compounds, such as ivermectin, that retain their activity when fed to livestock (Roncalli 1989).

The delayed action of hydramethylnon and its poor metabolism by insects are probably responsible for its activity against individuals that do not feed directly on the toxic bait. Before death, individuals that have fed on hydramethylnon bait can return to and defecate in harborage areas frequented by other cockroaches. Our demonstration that early instar nymphal mortality was significantly increased in the presence of males (Table 2) suggests that hydramethylnon may exert its greatest effect on early instars through coprophagy. In general, movement by early instar *B. germanica* nymphs is limited (Ross et al. 1984, Bret & Ross 1985). Consequently, newly emerged nymphs may feed within the harborage on the feces produced by other cockroaches after maternally-derived nutrients are depleted. The aggregation pheromone, secreted with the feces of *B. germanica* and other cockroaches (Ishii & Kuwahara 1967), may, in addition to signaling a suitable harborage, mark a potential food source. Access of mixed stages of *B. germanica* to hydramethylnon exclusively in fecal form resulted in high male mortality. Competition for the limited amount of feces could account for the observed stage specific mortality differences.

Coprophagous behavior in cockroaches has not received serious attention, although Cochran (1985) suggested that it may be of some importance in the few species that excrete uric acid. *B. germanica* does not excrete uric acid in the feces (Cochran 1973), although males do void stored urates during deposition of the spermatophore, which are sub-

sequently consumed by females (Mullins & Keil 1980). Hydramethylnon may be incorporated within the spermatophore but its appearance in the excreta of all cockroaches makes the feces the most probable source of toxicant. Nitrogen is excreted as ammonia by many cockroaches including *B. germanica* (Mullins & Cochran 1976); however, the volatility of ammonia may limit its availability to coprophages. In addition to ammonia, *Periplaneta americana* (L.) excretes amino nitrogen, tryptophan metabolites, and water-soluble and insoluble nitrogen products (Mullins & Cochran 1973). Unfortunately, a complete analysis of nitrogen-containing compounds is unavailable for *B. germanica* feces. A thorough biochemical analysis of *B. germanica* excreta would provide further insight into the purpose of a coprophagous strategy.

The indirect uptake of hydramethylnon and perhaps other bait toxicants might be exploited further in a somewhat novel manner. In locations where conventional cockroach control is inappropriate because baits or liquids cannot reach most of the target population, it may be possible to trap, bait, then release toxicant-laden individuals with the expectation that they will return to harborages and defecate before death. *P. americana* also is affected by hydramethylnon by fecal ingestion (J.S., unpublished data). Baiting of this species in sewers is generally difficult because of limited placement sites. Excretion in harborages by bait-fed and released individuals might make toxicant more accessible to most of the insects.

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#### References Cited

- Appel, A. G. 1990. Laboratory and field performance of consumer bait products for German cockroach (Dictyoptera: Blattellidae) control. *J. Econ. Entomol.* 83: 153-159.
- Banks, W. A., H. L. Collins, D. F. Williams, C. E. Stringer, C. S. Lofgren, D. P. Harlan & C. L. Mangum. 1981. Field trials with AC 217,300, a new amidinohydrazone bait toxicant for control of the red imported fire ant. *Southwest. Entomol.* 6: 158-164.
- Beal, R. H. & G. R. Esenther. 1980. A new approach to subterranean termite control—the bait block method. *Sociobiology* 5: 171-174.
- Bret, B. L. & M. H. Ross. 1985. A laboratory study of German cockroach dispersal (Dictyoptera: Blattellidae). *Proc. Entomol. Soc. Wash.* 87: 448-455.
- Burnett, A. M., W. H. Mason & S. T. Rhodes. 1969. Reingestion of feces and excretion rates of Zn<sup>65</sup> in *Popillius disjunctus* versus *Cryptocercus punctulatus*. *Ecology* 50: 1094-1096.
- Cochran, D. G. 1973. Comparative analysis of excreta from twenty cockroach species. *Comp. Biochem. Physiol. A Comp. Physiol.* 46: 409-419.

- 1985.** Nitrogen excretion in cockroaches. *Annu. Rev. Entomol.* 30: 29-49.
- Hollingshaus, J. G. & R. J. Little, Jr.** 1984. Toxicity, penetration, and metabolism of AC 217,300 (AMDRO) in the tobacco budworm (*Heliothis virescens*) by various methods of application. *Pest. Biochem. Physiol.* 22: 329-336.
- Ishii, S. & Y. Kuwahara.** 1967. An aggregation pheromone of the German cockroach *Blattella germanica* L. (Orthoptera: Blattellidae). I. Site of the pheromone production. *Appl. Entomol. Zool.* 2: 203-217.
- Lofgren, C. S., F. J. Bartlett, C. E. Stringer & W. A. Banks.** 1964. Imported fire ant toxic bait studies: further tests with granulated mirex-soybean oil bait. *J. Econ. Entomol.* 57: 695-698.
- Manley, D. G.** 1982. Efficacy of AMDRO bait on fire ants in South Carolina. *J. Ga. Entomol. Soc.* 17: 410-412.
- Mason, W. H. & E. P. Odum.** 1969. The effect of coprophagy on retention and bioelimination of radionuclides by detritus-feeding animals, pp. 721-724. In D. J. Nelson & F. C. Evans [eds.], Proceedings, 2nd national symposium of radioecology. U.S. Atomic Energy Commission, Technical Information Service, Washington, D.C.
- McBrayer, J. F.** 1973. Exploitation of deciduous leaf litter by *Apheloria montana* (Diplopoda: Eurydesmidae). *Pedobiologia* 13: 90-98.
- Mullins, D. E. & D. G. Cochran.** 1973. Nitrogenous excretory materials from the American cockroach. *J. Insect Physiol.* 19: 1007-1018.
- 1976.** A comparative study of nitrogen excretion in twenty-three cockroach species. *Comp. Biochem. Physiol. A Comp. Physiol.* 53: 393-399.
- Mullins, D. E. & C. B. Keil.** 1980. Paternal investment of urates in cockroaches. *Nature (London)* 283: 567-569.
- Patterson, R. S. & P. G. Koehler.** 1989. Peridomestic cockroach suppression with hydramethylnon bait. *J. Agric. Entomol.* 6: 37-42.
- Roncalli, R. A.** 1989. Environmental aspects of use of ivermectin and abamectin in livestock: effects on cattle dung fauna, pp. 173-182. In W. C. Campbell [ed.], Ivermectin and abamectin. Springer-Verlag, New York.
- Ross, M. H., B. L. Bret & C. B. Keil.** 1984. Population growth and behavior of *Blattella germanica* (L.) (Orthoptera: Blattellidae) in experimentally established shipboard infestations. *Ann. Entomol. Soc. Am.* 77: 740-752.
- Ryan, B. F., B. L. Joiner & T. A. Ryan, Jr.** 1985. Minitab handbook, 2nd ed. PWS, Boston.
- Schal, C. & W. J. Bell.** 1982. Ecological correlates of paternal investment of urates in a tropical cockroach. *Science* 218: 170-173.
- Schowalter, T. D. & D. A. Crossley, Jr.** 1982. Bioelimination of <sup>51</sup>Cr and <sup>85</sup>Sr by cockroaches, *Gromphadorhina portentosa* (Orthoptera: Blaberidae), as affected by mites, *Gromphadorholaelaps schaeferi* (Parasitiformes; Laelapidae). *Ann. Entomol. Soc. Am.* 75: 158-160.
- Silverman, J. & T. J. Shapas.** 1986. Cumulative toxicity and delayed temperature effects of hydramethylnon on German cockroaches. *J. Econ. Entomol.* 79: 1613-1616.
- Stevenson, B. G. & D. L. Dindal.** 1987. Functional ecology of coprophagous insects: a review. *Pedobiologia* 30: 285-298.
- Su, N. Y., M. Tamashiro, J. R. Yates & M. I. Haverty.** 1982. Effect of behavior on the evaluation of insecticides for prevention of or remedial control of the Formosan subterranean termite. *J. Econ. Entomol.* 75: 188-193.
- Su, T. H., J. W. Beardsley & F. L. McEwen.** 1980. AC 217,300, a promising new insecticide for use in baits for control of the bigheaded ant in pineapple. *J. Econ. Entomol.* 73: 755-756.

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