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# Toxicity, repellency, and laboratory performance of consumer bait products for German cockroach (Blattodea: Ectobiidae) management

Arthur G. Appel<sup>1,\*</sup>, Beatrice N. Dingha<sup>2</sup>, Marla J. Eva<sup>1</sup>, and Louis E. N. Jackai<sup>2</sup>

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## Abstract

Toxicity, repellency, and laboratory performance of consumer bait formulations were evaluated for control of 7 insecticide-resistant, field-collected strains of the German cockroach, *Blattella germanica* (L.) (Blattodea: Ectobiidae) in continuous exposure and Ebeling choice box assays. Solid and gel baits contained avermectin, dinotefuran, fipronil, or hydramethylnon as active ingredients, and were active for control of all German cockroach strains. However, the laboratory susceptible strain was generally more sensitive (lower median lethal times or  $LT_{50}$  values) than the field-collected strains. Resistance ratios in continuous exposure tests ranged from 0.37 for 0.01% fipronil (strain H) to 14.23 for 0.05% dinotefuran (strain DCC2). The  $LT_{50}$  values for most baits and strains generally were greater when tested in Ebeling choice boxes compared with continuous exposure tests. Resistance ratios ranged as high as 98.43 for 0.05% dinotefuran for control of strain DCC2. All strains had resistance to 0.05% dinotefuran bait, and the resistance ratios were greatest when tested in Ebeling choice boxes. All bait formulations had some repellency to most strains; however, repellency was never greater than 70% for any treatment–strain combination. Combining Ebeling choice box mortality and repellency data, maximum estimated performance index values ( $PI_{Max}$ ) reached 100, i.e., no repellency and complete mortality, for most treatments. There was not complete mortality of the majority of strains exposed to 0.05% dinotefuran bait and therefore the  $PI_{Max}$  value did not reach 100. The rate of increase in performance index value over time or  $t_{PI_{Max}/2}$  ranged 0.26 and 7.85 d. Our results indicate that although all baits were toxic to multi-resistant strains of German cockroaches, there was significant resistance or tolerance to many formulations that would likely negatively impact field control.

Key Words: *Blattella germanica*; insecticidal baits; continuous exposure; Ebeling choice box

## Resumen

Se evaluó la toxicidad, repelencia y desempeño en el laboratorio de formulaciones de cebo de consumo para controlar 7 cepas resistentes a insecticidas de la cucaracha alemana, *Blattella germanica* (L.) (Blattodea: Ectobiidae) y recolectadas en el campo, en exposición continua y ensayos de cuadro de selección de Ebeling. Los cebos sólidos y en gel contenían avermectina, dinotefurano, fipronil o hidrametilnon como ingredientes activos y fueron activos para el control de todas las cepas de cucarachas alemanas. Sin embargo, la cepa susceptible de laboratorio fue generalmente más sensible (tiempos letales medianos o valores  $LT_{50}$  más bajos) que las cepas recolectadas en el campo. Los índices de resistencia en las pruebas de exposición continua oscilaron entre 0,37 para fipronil al 0,01 % (cepa H) y 14,23 para dinotefurano al 0,05 % (cepa DCC2). Los valores  $LT_{50}$  para la mayoría de los cebos y cepas generalmente fueron mayores cuando se probaron en cajas de elección de Ebeling en comparación con las pruebas de exposición continua. Las proporciones de resistencia oscilaron hasta 98,43 para dinotefurano al 0,05 % para el control de la cepa DCC2. Todas las cepas tuvieron resistencia al cebo con dinotefurano al 0,05 %, y las proporciones de resistencia fueron mayores cuando se probaron en las cajas de elección de Ebeling. Todas las formulaciones de cebo tenían cierta repelencia a la mayoría de las cepas; sin embargo, la repelencia nunca superó el 70 % para ninguna combinación de tratamiento y cepa. Al combinar los datos de mortalidad y repelencia del cuadro de elección de Ebeling, los valores máximos del índice de rendimiento ( $IR_{Max}$ ) estimado alcanzaron 100, es decir, sin repelencia y mortalidad completa, para la mayoría de los tratamientos. No hubo mortalidad completa de la mayoría de las cepas expuestas al cebo con dinotefurano al 0,05 % y, por lo tanto, el valor de  $IR_{Max}$  no alcanzó 100. La tasa de aumento del valor del índice de rendimiento a lo largo del tiempo ( $IRT_{Max/2}$ ) varió entre 0,26 y 7,85 días. Nuestros resultados indican que, aunque todos los cebos fueron tóxicos para las cepas multiresistentes de cucarachas alemanas, hubo una resistencia o tolerancia significativa a muchas formulaciones que probablemente tendrían un impacto negativo en el control de campo.

Palabras Claves: *Blattella germanica*; cebos insecticidas; exposición continua; caja de elección de Ebeling

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The German cockroach, *Blattella germanica* (L.) (Blattodea: Ectobiidae), remains an important and common domiciliary pest. This species typically infests apartments, homes, and food preparation facilities where it feeds on all types of human and other animal food, trash, and waste ma-

terials (Appel 2021). They also can contaminate food and food preparation surfaces with their feces, oral secretions, and dead bodies, making them potential vectors of human pathogens (Alcamo & Frishman 1980; Brenner et al. 1987) and potent allergens (Gore & Schal 2007; Wang et al. 2008).

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Despite the successful adoption of integrated pest management to control many agricultural pests (Pedigo & Rice 2009), the use of insecticides is the primary means to manage German cockroach populations (Koehler et al. 1995; Dingha et al. 2013). Even though noninsecticidal tactics such as education, sanitation, sealing harborages and points of entry, trapping, and vacuuming are available (Kardatzke et al. 1981; Frishman 1995; Robinson & Zungoli 1985; Kaakeh et al. 1997), insecticides are the most common management tactic (Gondhalekar et al. 2021). Integrated pest management for German cockroach management is more effective than using only bait or liquid insecticides (Kramer et al. 2000; Miller & Meek 2004; Wang et al. 2006a; Williams et al. 2006). When integrated pest management methods, including insecticidal baits, are used to reduce cockroach populations, there is a strong correlation with reductions in cockroach allergens (Arbes et al. 2003, 2004; Sever et al. 2007).

Dingha et al. (2013) surveyed insecticide use practices and integrated pest management awareness and knowledge in rural North Carolina. Most residents were unfamiliar with integrated pest management and relied on spray formulations of insecticide, which they reported were ineffective. During the survey (Dingha et al. 2013), German cockroaches were collected from single-family residences and sent to Auburn University for rearing and to determine insecticide susceptibility. The insecticide treatment history of each strain was unknown. The toxicity and resistance levels of permethrin, chlorpyrifos, propoxur, imidacloprid, and fipronil to these field-collected strains were compared with a susceptible laboratory strain using topical application methods (Wu & Appel 2017). These insecticides were selected because they were or now are commonly used in cockroach control. The field-collected strains were significantly resistant to permethrin with resistance ratios ranging from 5.5 to 51.9 fold (Wu & Appel 2017); several strains also showed resistance to some essential oil components (Oladipupo et al. 2020). All strains were resistant to at least 1 of the tested insecticides. In addition, Wu and Appel (2018) determined these insecticides' repellency and laboratory performance and found that permethrin-treated surfaces repelled most strains. Only fipronil resulted in performance index values of 100 (no repellency and 100% mortality) in Ebeling choice-boxes indicating good potential field effectiveness of this insecticide (Rust & Reiersen 1978; Appel 1992, 2004).

Combining an educational program and application of insecticidal bait, Dingha et al. (2016) found that German cockroach infestations could be reduced by 86% in manufactured homes in North Carolina. They used a 0.03% fipronil-based dry bait product (small roach station, Combat® Source Kill Max<sup>®</sup>, Combat Insect Control Systems, Scottsdale, Arizona, USA) in child-resistant stations. The purpose of this study was to compare the activity and potential for effectiveness of several bait formulations for control of field-collected and insecticide susceptible strains of German cockroaches. We used continuous exposure and Ebeling choice-box methods to compare median lethal times (LT<sub>50</sub>

values), repellency, and performance index values for 5 bait products in resistance to 7 field-collected and a susceptible strain of German cockroach.

Materials and Methods

COCKROACH STRAINS

Seven field-collected strains of German cockroaches were used in this study along with a susceptible strain (S). These field-collected strains: B, D, DDC2, E, G, H, and I were collected from manufactured homes and daycare centers at least 32.2 km (20 miles) apart from each other in Franklin County, North Carolina, USA, in 2011 to 2012 (Dingha et al. 2013) and sent to Auburn University, Auburn, Alabama, USA. The insecticide treatment histories of these field-collected strains were unknown. However, all the strains exhibited significant resistance to permethrin in topical bioassays (Wu & Appel 2017). The field collections had between 10 and 25 individuals (mixed sexes) of each strain and were reared for between 10 and 15 generations (about 2.5 yr) to obtain sufficient numbers of cockroaches for testing. The susceptible strain was derived from the Orlando normal strain in the 1970s and reared in the laboratory at the University of California-Riverside, Riverside, California, USA, and Auburn University without exposure to insecticides for > 50 yr.

The susceptible strain (S) was maintained in 166 L plastic trash cans (Rubbermaid, High Point, North Carolina, USA), and the field-collected strains were reared in 3.8 L glass jars (Arkansas Glass Container Corp., Jonesboro, Arkansas, USA). All colonies were held at 28 ± 2 °C, with 40 to 55% RH, and a photoperiod of 12:12 h (L:D). All cockroach strains were provided rolls of corrugated cardboard as harborage, dry dog food (Purina Dog Chow, Ralston Purina, St. Louis, Missouri, USA), and water. Colonies were fed, watered, and cleaned each wk.

Only 1 to 2 wk old adult males were used in this study because of their relatively homogenous physiology compared with adult females and other stages (Abd-Elghafar et al. 1990). Cockroaches were anesthetized briefly with carbon dioxide (CO<sub>2</sub>) to facilitate handling.

INSECTICIDAL BAITS

Gel and solid bait formulations were used in the continuous exposure and Ebeling choice box tests. Gels (0.5 g) were applied in a 4.5 cm × 4.5 cm polystyrene weighing boat (VWR, West Chester, Pennsylvania, USA) and positioned in a jar or choice box. Solid baits were removed from their bait stations and about 0.5 g placed into a weighing boat as above. Active ingredients included avermectin B1, dinotefuran, fipronil, and hydramethylnon at various concentrations as sold by the manufacturer (Table 1).

Table 1. Consumer cockroach bait products and their active ingredients used in continuous exposure and Ebeling choice box studies against adult male German cockroaches. Both field-collected and laboratory strains were tested with these baits.

Brand	Manufacturer	Type	Active ingredient	% Active ingredient	EPA Reg. No.
Raid	SC Johnson & Son	Solid	Avermectin B1	0.05	4822-472
Hot Shot	Spectrum Group	Gel	Dinotefuran	0.05	9688-271-8845
Combat	SK* Combat	Solid	Hydramethylnon	2.00	64240-2
Combat	SK* Max Combat	Gel	Fipronil	0.01	64240-45
Combat	QK** Combat	Solid	Fipronil	0.03	64240-33

\*Source Kill  
\*\*Quick Kill

## CONTINUOUS EXPOSURE AND EBELING CHOICE-BOX TESTS

The toxicity of the bait formulations to the field-collected and susceptible strains of the German cockroach was evaluated in continuous exposure tests (Appel 1990, 1992). Groups of 10 cockroaches were confined in 0.95 L wide-mouth glass jars (Ball Corp., Broomfield, Colorado, USA) with a small water jar, a roll of cardboard harborage, a piece of dog chow (as an alternative food source), and a weighing boat with bait. There were 6 replicate jars for each cockroach strain and bait formulation combination. Untreated control jars were set up exactly as the baited jars, except the weigh boats were empty. Jars were exposed to  $28 \pm 2^\circ\text{C}$ , 40 to 55% RH, a photoperiod of 12:12 h (L:D), and examined daily (or bihourly for the susceptible strain exposed to avermectin and fipronil baits) for mortality for 14 d. Cockroaches were considered dead if they could not right themselves or move more than half a body length when probed.

Toxicity, repellency, and potential performance of the bait formulations were determined in Ebeling choice-boxes as described by Ebeling et al. (1966), Appel (1990, 1992), and Wu and Appel (2018). The choice-box assay has been used to evaluate the toxicity and repellency of insecticidal baits, dusts, and sprays, as well as wood paneling and other objects (Ebeling 1995). It is recommended by the US Environmental Protection Agency (EPA 2019) for laboratory testing of cockroach bait products. The choice-box is a square wooden box divided into 2 equal-size compartments by a wood partition. The base of the box is a solid sheet of polyvinyl chloride attached with brass screws. A 1.3 cm diam hole below the top of the partition allows cockroaches to move freely between compartments. Food and water were provided in 1 compartment and the insecticide treatment in the other. Insecticide treatments consisted of 0.5 g of bait in a weigh boat and untreated control boxes had empty weigh boats. Both sides were covered with clear Plexiglas®. The compartment with food and water (light side) simulated areas in a home normally illuminated containing food and water, such as a kitchen counter. The other compartment (dark side) was treated with insecticides and is meant to simulate dark voids where German cockroaches harbor during the d (photophase) and where insecticides are typically applied.

The hole in the partition was plugged using a cork, and 20 adult male German cockroaches were placed into the untreated light compartment. Cockroaches were allowed to acclimate for 2 h. Weighing-boats containing bait were positioned in 1 of the outside corners of the treated side of the box and, after the cork was removed from the partition, was covered with transparent Plexiglas® and an opaque sheet. Choice-boxes were exposed to a photoperiod of 12:12 h (L:D) at  $28 \pm 1^\circ\text{C}$ . Therefore, the light side with food and water was exposed to a daily photoperiod, whereas the dark side with treatment was in constant darkness. The number of live and dead cockroaches in both compartments was recorded 4 to 5 h into the photophase daily for 14 d. The total number of dead cockroaches found in each choice-box (both sides) was used to calculate mortality. The percentage of live cockroaches found in the untreated, light compartment of each choice-box during the photophase was defined as the percentage of repellency. Each treatment (bait and control) was replicated 6 times for the 8 German cockroach strains.

## DATA ANALYSIS

Toxicity ( $LT_{50}$ ) of the insecticides in both the continuous exposure and Ebeling choice-box tests were analyzed using probit analysis (SAS 2012). Differences among  $LT_{50}$  values were considered significant if the 95% confidence intervals did not overlap. Resistance ratios for continu-

ous exposure and Ebeling choice-box data were calculated for each insecticide and German cockroach strain as the quotient of the  $LT_{50}$  value of the field strain divided by the  $LT_{50}$  value of the susceptible strain.

Repellency measured in Ebeling choice-box tests was analyzed by repeated-measures ANOVA, and the least square means difference test was used to separate differences in repellency among treatments for each strain (SAS 2012).

The performance index (PI) is calculated as an estimate of potential field performance combining the effects of mortality and repellency:

$$PI = 1 - \left[ \frac{\text{Number alive} + \text{Number alive in light side}}{\text{Number dead} + \text{Initial total number}} \right] \times 100$$

Performance index values range from -100, complete repellency, and no mortality to +100, complete mortality, and no repellency. Untreated control boxes with no repellency and mortality have a performance index value of about 0 (Rust & Reiersen 1978; Appel 1992, 2004). Change in performance index over time was analyzed using nonlinear regression and SigmaPlot Version 13 software (Systat 2014). A rectangular hyperbolic model (Appel 1990, 1992; Appel & Abd-Elghafar 1990) was used to model the change in performance index over time. The following function was fit to the daily performance index values:

$$y = \frac{ax}{b+x}$$

where  $y$  = performance index,  $a = PI_{\text{Max}}$  or maximum asymptotic performance index value,  $b = t_{PI_{\text{Max}}/2}$  or the period required for half the  $PI_{\text{Max}}$  value to be reached, and  $x = d$ . The model was constrained so that the  $PI_{\text{Max}}$  was  $\leq 100$ . Correlations between resistance ratios determined in continuous exposure and choice-box assays, between resistance ratios and repellency, and between resistance ratios and  $t_{PI_{\text{Max}}/2}$  values were analyzed with Pearson correlation (Systat 2014).

## Results

The  $LT_{50}$  values of insecticidal bait formulations to control field-collected and a laboratory susceptible strain German cockroach are reported in Table 2 for continuous exposure tests and Table 3 for Ebeling choice-box tests. In the continuous exposure tests, mean untreated control mortality never exceeded 18% for any strain at 14 d. Avermectin bait (0.05%) was most toxic to strain B ( $LT_{50}$  of 3.0 d) and least toxic to strain DCC2 ( $LT_{50}$  of 5.8 d). Dinotefuran bait was most active for control of strain S ( $LT_{50}$  of 0.3 d) and least active for control of strain DCC2 ( $LT_{50}$  of 3.7 d). Fipronil bait at 0.01% ranged in activity from 0.6 to 3.4 d for strains H and I, respectively. However, for fipronil bait at 0.03%,  $LT_{50}$  values ranged from 0.8 d for strain H to 3.9 d for strain DCC2. Hydramethylnon was most active for control of strain S ( $LT_{50}$  of 2.2 d) and least active for control of strain D ( $LT_{50}$  of 5.3 d). Resistance ratios for all baits and strains ranged from 0.37 for fipronil bait at 0.01% for control of strain H to 14.23 for dinotefuran bait for control of strain DCC2. However, even the highest resistance ratio determined with any bait or strain combination in this study (14.23) represents an  $LT_{50}$  value of 3.7 d.

In Ebeling choice-box tests, untreated control mortality was < 20% at 14 d for all strains. In general, the  $LT_{50}$  values for the various baits and German cockroach strains were similar (no more than about 1 d different) when tested in Ebeling choice-boxes or continuous exposure (Table 2) with the exception of strain H. The  $LT_{50}$  values for dinotefuran, fipronil 0.01, and fipronil 0.03 were significantly greater (based on non-overlap of the 95% confidence intervals) for strain H

**Table 2.** Continuous exposure toxicity of solid and gel bait formulations to adult males of several strains of the German cockroach, *Blattella germanica* (L.)<sup>a</sup>.

Strain	Treatment	Slope ± SE	Median lethal time LT <sub>50</sub> (95% CI) d	χ <sup>2</sup>	RR <sup>b</sup>
B	Avermectin 0.05%	2.62 ± 0.52	3.00 (2.28–3.85)	25.87	0.84
	Dinotefuran 0.05%	0.89 ± 0.17	0.55 (0.15–1.04)	27.91	2.12
	Fipronil 0.01%	1.45 ± 0.20	1.30 (0.77–1.80)	51.04	0.79
	Fipronil 0.03%	2.37 ± 0.36	1.79 (1.30–2.24)	42.12	1.94
	Hydramethylnon 2.0%	3.63 ± 0.26	4.33 (3.98–4.68)	191.94	1.97
D	Avermectin 0.05%	5.85 ± 0.68	3.65 (3.34–3.96)	73.93	1.02
	Dinotefuran 0.05%	1.07 ± 0.17	0.88 (0.41–1.36)	39.79	3.39
	Fipronil 0.01%	1.63 ± 0.18	1.46 (1.04–1.86)	82.95	0.89
	Fipronil 0.03%	2.89 ± 0.35	1.47 (1.20–1.71)	70.23	1.60
	Hydramethylnon 2.0%	5.30 ± 0.54	2.39 (2.18–2.59)	97.27	1.09
DCC2	Avermectin 0.05%	6.35 ± 0.48	5.80 (5.50–6.09)	175.78	1.62
	Dinotefuran 0.05%	1.54 ± 0.15	3.70 (3.08–4.29)	108.34	14.23
	Fipronil 0.01%	1.82 ± 0.26	3.28 (2.41–4.08)	49.95	2.00
	Fipronil 0.03%	1.85 ± 0.19	3.87 (3.22–4.49)	95.44	4.21
	Hydramethylnon 2.0%	3.80 ± 0.30	6.95 (6.45–7.46)	159.44	3.16
E	Avermectin 0.05%	3.25 ± 0.33	4.76 (4.21–5.33)	99.02	1.33
	Dinotefuran 0.05%	1.43 ± 0.19	1.34 (0.87–1.76)	55.79	5.15
	Fipronil 0.01%	1.19 ± 0.16	1.49 (0.91–2.03)	51.72	0.91
	Fipronil 0.03%	1.53 ± 0.19	1.19 (0.78–1.59)	62.83	1.29
	Hydramethylnon 2.0%	3.18 ± 0.31	2.33 (1.98–2.66)	105.76	1.06
G	Avermectin 0.05%	5.65 ± 1.24	4.84 (3.15–4.56)	20.85	1.35
	Dinotefuran 0.05%	0.51 ± 0.18	0.30 (0.0003–1.01)	7.90	1.15
	Fipronil 0.01%	2.73 ± 0.38	1.20 (0.92–1.44)	51.03	0.73
	Fipronil 0.03%	2.20 ± 0.33	1.02 (0.69–1.29)	44.00	1.11
	Hydramethylnon 2.0%	4.08 ± 0.30	4.18 (3.85–4.49)	190.81	1.90
H	Avermectin 0.05%	4.07 ± 0.50	3.32 (2.89–3.75)	65.94	0.93
	Dinotefuran 0.05%	0.75 ± 0.21	0.64 (0.05–1.39)	12.29	2.46
	Fipronil 0.01%	0.93 ± 0.24	0.60 (0.08–1.20)	14.95	0.37
	Fipronil 0.03%	1.28 ± 0.19	0.81 (0.40–1.21)	43.72	0.88
	Hydramethylnon 2.0%	3.38 ± 0.25	5.47 (5.05–5.89)	183.96	1.54
I	Avermectin 0.05%	4.52 ± 0.34	4.52 (4.20–4.83)	179.61	1.26
	Dinotefuran 0.05%	1.04 ± 0.31	1.91 (0.47–3.23)	11.33	7.35
	Fipronil 0.01%	2.59 ± 0.26	3.43 (2.91–3.93)	100.11	2.09
	Fipronil 0.03%	2.56 ± 0.35	2.62 (2.06–3.19)	52.60	2.85
	Hydramethylnon 2.0%	5.32 ± 0.64	2.75 (2.45–3.04)	69.66	1.25
S	Avermectin 0.05%	4.13 ± 0.47	3.59 (3.19–4.01)	77.78	—
	Dinotefuran 0.05%	2.30 ± 0.16	0.26 (0.03–0.28)	13.56	—
	Fipronil 0.01%	4.73 ± 0.40	1.64 (1.54–1.77)	3.26	—
	Fipronil 0.03%	6.00 ± 0.35	0.92 (0.88–0.97)	12.38	—
	Hydramethylnon 2.0%	6.14 ± 0.72	2.20 (1.99–2.40)	73.16	—

<sup>a</sup>Probit analyses significant at < 0.0008, *n* = 60 for each strain and treatment combination.<sup>b</sup>Resistance ratio; LT<sub>50</sub> of field strain per LT<sub>50</sub> of susceptible strain (S).

in Ebeling choice-boxes and continuous exposure tests. However, the LT<sub>50</sub> value for hydramethylnon was significantly less in the Ebeling choice-boxes (Table 3). Avermectin bait was most active to strain I (LT<sub>50</sub> of 3.2 d) and least active to strain S (LT<sub>50</sub> of 4.3 d). Dinotefuran bait was most active to strain S (LT<sub>50</sub> of 0.1 d) and least active to strain DCC2 (LT<sub>50</sub> of 6.9 d). Fipronil bait at 0.01% ranged in LT<sub>50</sub> values from 0.8 to 2.9 d for strains S and E, respectively. However, for fipronil bait at 0.03%, activity ranged from 0.5 d for strain S to 2.2 d for strain H. Hydramethylnon was most active to strain S (LT<sub>50</sub> of 2.4 d) and least active to strain DCC2 (LT<sub>50</sub> of 4.2 d). Resistance ratios for all baits and strains ranged from 0.74 for avermectin bait against strain I to 98.4 for dinotefuran bait to strain DCC2. Even the highest resistance ratio determined with any bait or strain combination represented LT<sub>50</sub> values of < 7 d. Resistance ratios determined in continuous exposure

tests were significantly correlated with those determined in Ebeling choice boxes (*r* = 0.857; *n* = 35; *P* < 0.00001).

Mean repeated measures of repellency of cockroach baits tested in Ebeling choice boxes is reported in Table 4. Repellency in the untreated control boxes ranged from 15.6% for strain B to 46.1% for strain G. With the exceptions of strains G and H, untreated boxes had significantly lower repellency than most bait treatments (Table 4). The highest repellency was about 70% for strain E exposed to the hydramethylnon bait. Repellency was not correlated (*P* > 0.05) with either choice box or continuous exposure resistance ratios.

The PI<sub>max</sub> values for the untreated controls did not reach about 35 for any of the strains after 14 d (Fig. 1). There was, therefore, minimal mortality and repellency. Nonlinear regression, however, did estimate that the untreated controls would ultimately result in



**Table 3.** Toxicity of solid and gel bait formulations to adult males of several strains of the German cockroach, *Blattella germanica* (L.), in Ebeling choice boxes<sup>a</sup>.

Strain	Treatment	Slope $\pm$ SE	Median lethal time LT <sub>50</sub> (95% CI) d	$\chi^2$	RR <sup>b</sup>
B	Avermectin 0.05%	3.48 $\pm$ 0.16	3.61 (3.38–3.84)	454.61	0.84
	Dinotefuran 0.05%	1.12 $\pm$ 0.15	0.58 (0.25–0.95)	52.78	8.29
	Fipronil 0.01%	2.48 $\pm$ 0.27	1.27 (0.99–1.53)	82.11	1.59
	Fipronil 0.03%	1.76 $\pm$ 0.16	0.85 (0.62–1.07)	128.43	1.74
	Hydramethylnon 2.0%	3.23 $\pm$ 0.27	2.97 (2.62–3.32)	149.01	1.26
D	Avermectin 0.05%	2.88 $\pm$ 0.14	3.30 (3.05–3.54)	419.34	0.77
	Dinotefuran 0.05%	0.62 $\pm$ 0.10	0.69 (0.26–1.18)	40.49	9.86
	Fipronil 0.01%	1.70 $\pm$ 0.11	2.65 (2.29–2.99)	247.58	3.31
	Fipronil 0.03%	1.34 $\pm$ 0.30	1.14 (0.41–1.80)	20.02	2.33
	Hydramethylnon 2.0%	3.15 $\pm$ 0.31	3.28 (2.83–3.72)	106.93	1.40
DCC2	Avermectin 0.05%	2.48 $\pm$ 0.17	4.20 (3.77–4.61)	207.56	0.98
	Dinotefuran 0.05%	1.18 $\pm$ 0.20	6.89 (5.39–9.10)	33.43	98.43
	Fipronil 0.01%	1.77 $\pm$ 0.17	2.05 (1.60–2.47)	112.67	2.56
	Fipronil 0.03%	2.04 $\pm$ 0.22	1.93 (1.46–2.36)	86.31	3.94
	Hydramethylnon 2.0%	4.16 $\pm$ 0.25	4.22 (3.93–4.50)	282.44	1.80
E	Avermectin 0.05%	2.44 $\pm$ 0.20	4.86 (4.33–5.40)	146.29	1.13
	Dinotefuran 0.05%	1.03 $\pm$ 0.14	1.60 (0.94–2.21)	50.40	22.86
	Fipronil 0.01%	1.68 $\pm$ 0.13	2.91 (2.45–3.35)	163.22	3.64
	Fipronil 0.03%	2.28 $\pm$ 0.16	1.98 (1.68–2.26)	199.11	4.04
	Hydramethylnon 2.0%	3.22 $\pm$ 0.28	2.62 (2.30–2.93)	134.05	0.96
G	Avermectin 0.05%	2.84 $\pm$ 0.32	3.94 (3.32–4.54)	79.27	0.92
	Dinotefuran 0.05%	0.64 $\pm$ 0.21	1.10 (0.04–2.36)	9.15	15.71
	Fipronil 0.01%	1.56 $\pm$ 0.23	1.81 (1.16–2.41)	47.72	2.26
	Fipronil 0.03%	2.02 $\pm$ 0.20	1.82 (1.40–2.21)	100.30	3.71
	Hydramethylnon 2.0%	2.71 $\pm$ 0.13	3.81 (3.54–4.08)	435.79	1.62
H	Avermectin 0.05%	2.82 $\pm$ 0.16	3.36 (3.07–3.64)	326.85	0.78
	Dinotefuran 0.05%	0.92 $\pm$ 0.17	2.68 (1.55–3.67)	30.75	38.29
	Fipronil 0.01%	1.34 $\pm$ 0.12	1.95 (1.50–2.37)	124.94	2.44
	Fipronil 0.03%	2.41 $\pm$ 0.13	2.17 (1.93–2.40)	328.01	4.43
	Hydramethylnon 2.0%	3.04 $\pm$ 0.22	3.81 (3.42–4.18)	196.73	1.62
I	Avermectin 0.05%	2.99 $\pm$ 0.26	3.17 (2.75–3.58)	130.65	0.74
	Dinotefuran 0.05%	1.10 $\pm$ 0.14	0.90 (0.49–1.32)	66.09	12.86
	Fipronil 0.01%	2.56 $\pm$ 0.17	2.31 (2.03–2.58)	221.31	2.89
	Fipronil 0.03%	2.22 $\pm$ 0.24	1.85 (1.45–2.23)	89.22	3.78
	Hydramethylnon 2.0%	5.13 $\pm$ 0.32	2.67 (2.51–2.82)	254.79	1.14
S	Avermectin 0.05%	2.71 $\pm$ 0.20	4.30 (3.87–4.71)	190.25	—
	Dinotefuran 0.05%	0.56 $\pm$ 0.22	0.07 (0.0001–0.46)	6.19	—
	Fipronil 0.01%	1.93 $\pm$ 0.24	0.80 (0.53–1.05)	67.51	—
	Fipronil 0.03%	1.28 $\pm$ 0.23	0.49 (0.17–0.85)	30.81	—
	Hydramethylnon 2.0%	3.13 $\pm$ 0.34	2.35 (1.99–2.70)	84.12	—

<sup>a</sup>Probit analyses significant at  $< 0.05$ ,  $n = 60$  for each strain and treatment combination.<sup>b</sup>Resistance ratio; LT<sub>50</sub> of field strain per LT<sub>50</sub> of susceptible strain (S).

PI<sub>Max</sub> values of 100, but over extended periods. The rate of effect or  $t_{PI_{Max}/2}$  values were either slightly negative (controls for DCC2 and S strains) or  $> 18$  d indicating no or a very slow change in performance index over time (Fig. 1). The PI<sub>Max</sub> values for most bait treatments and German cockroach strains reached 100 by 14 d, indicating little repellency and complete mortality by the end of the experiment (Table 4; Fig. 1). The most notable exception was for the dinotefuran bait with a range of PI<sub>Max</sub> values from 78.1 for strain G to 100 for strain DCC2 (Table 4). The  $t_{PI_{Max}/2}$  values ranged between 0.26 d for dinotefuran bait against the susceptible strain S and 7.85 d for dinotefuran bait against strain DCC2 (Table 4); most  $t_{PI_{Max}/2}$  values were  $< 3$  d. Values of  $t_{PI_{Max}/2}$  were positively correlated with both continuous exposure ( $r = 0.438$ ;  $n = 35$ ;  $P = 0.0084$ ) and choice box ( $r = 0.521$ ;  $n = 35$ ;  $P = 0.0013$ ) resistance ratios.

## Discussion

Baits have become popular consumer insecticide products and contain several components, including the active ingredient, food base, attractants and feeding stimulants, and, in some cases, a pre-baited station (Reiersen 1995; Appel & Rust 2021). Bait formulations include solid and dissolvable granules, dry deposits, gels, dusts, and pastes, the most popular of which are dry deposits contained in stations and gels. In contrast to traditional fast-acting contact insecticides such as pyrethroids, the active ingredients for cockroach baits should be toxic at low concentrations, relatively slow-acting, and nonrepellent. The active ingredients tend to be metabolic inhibitors (hydramethylnon), acetylcholine agonists (dinotefuran), or affect GABA-gated chloride channels (abamectin,

**Table 4.** Percent repellency of solid and gel bait formulations to adult males of several strains of the German cockroach, *Blattella germanica* (L.), in Ebeling choice boxes<sup>a</sup>.

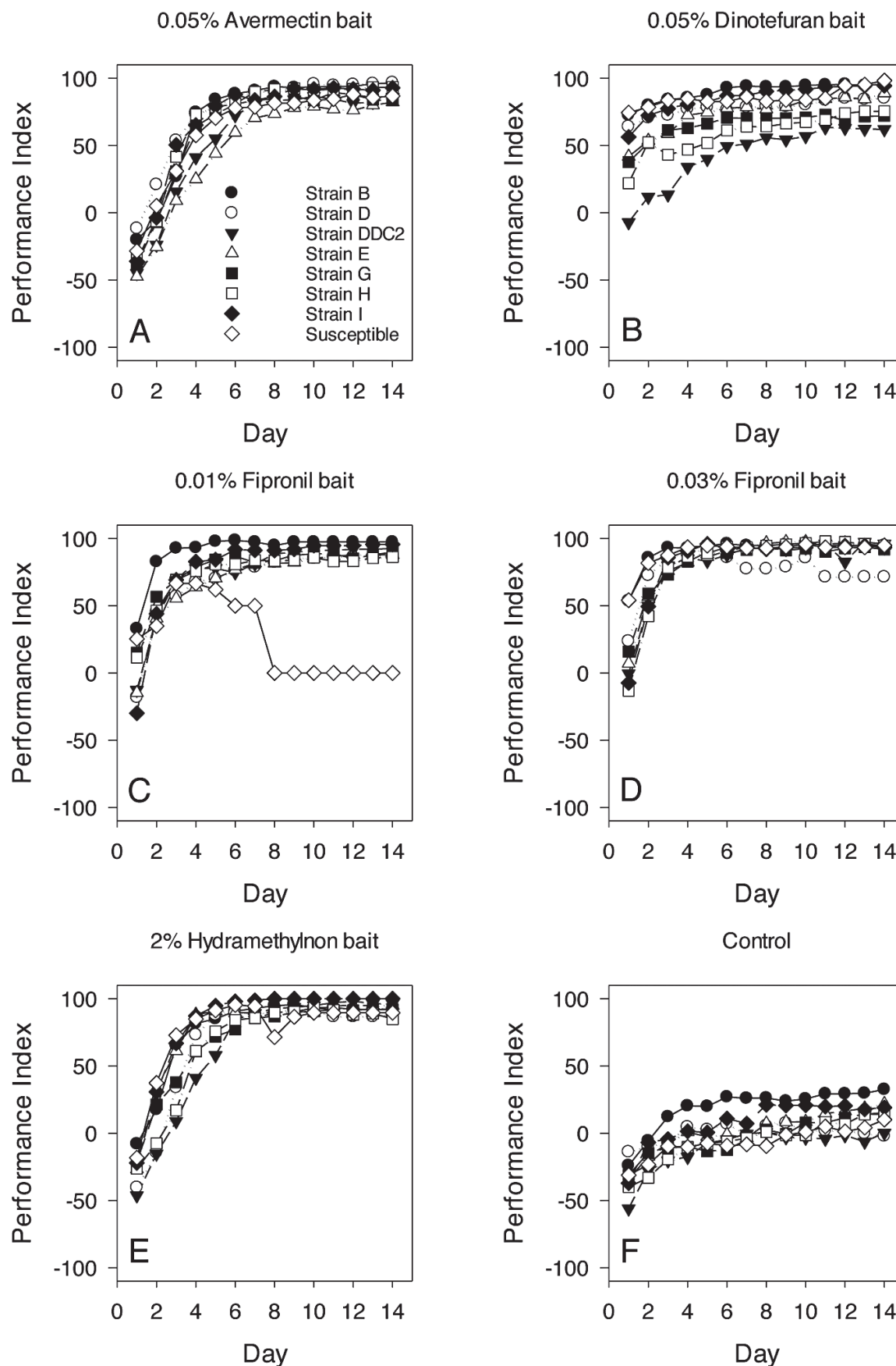
Strain	Treatment	Percent repellency LS Mean <sup>b</sup>	SE	t value
B	Avermectin 0.05%	28.1548 bc	4.6596	6.04
	Control	15.6153 d	4.4907	3.48
	Dinotefuran 0.05%	19.1021 cd	4.5185	4.23
	Fipronil 0.01%	12.2972 d	6.6316	1.85
	Fipronil 0.03%	60.0318 a	5.3254	11.27
	Hydramethylnon 2.0%	31.3003 b	5.1309	6.10
D	Avermectin 0.05%	23.8012 c	5.4215	4.39
	Control	22.9892 c	5.3123	4.33
	Dinotefuran 0.05%	29.5484 c	5.3123	5.56
	Fipronil 0.01%	54.8509 b	5.3123	10.33
	Fipronil 0.03%	47.778 b	6.7241	7.11
	Hydramethylnon 2.0%	66.7146 a	5.8562	11.39
DCC2	Avermectin 0.05%	49.8769 a	6.6179	7.54
	Control	33.9872 b	6.5153	5.22
	Dinotefuran 0.05%	38.2249 b	6.5952	5.80
	Fipronil 0.01%	51.8407 a	6.8608	7.56
	Fipronil 0.03%	37.8126 b	6.9829	5.42
	Hydramethylnon 2.0%	48.2527 a	6.8283	7.07
E	Avermectin 0.05%	60.2684 a	6.7199	8.97
	Control	29.2803 bc	6.6432	4.41
	Dinotefuran 0.05%	34.2005 b	6.7039	5.10
	Fipronil 0.01%	32.4765 b	6.6432	4.89
	Fipronil 0.03%	20.4367 c	6.8675	2.98
	Hydramethylnon 2.0%	69.5574 a	7.7281	9.00
G	Avermectin 0.05%	49.7124 a	9.5188	5.22
	Control	40.0633 a	9.2726	4.32
	Dinotefuran 0.05%	44.2306 a	9.2933	4.76
	Fipronil 0.01%	49.1331 a	9.4999	5.17
	Fipronil 0.03%	29.7612 b	9.5261	3.12
	Hydramethylnon 2.0%	33.5485 ab	9.2813	3.61
H	Avermectin 0.05%	38.2524 b	7.5258	5.08
	Control	46.0375 a	7.3848	6.23
	Dinotefuran 0.05%	53.6823 a	7.3848	7.27
	Fipronil 0.01%	43.7118 a	7.4267	5.89
	Fipronil 0.03%	13.7157 c	7.5919	1.81
	Hydramethylnon 2.0%	37.5451 b	7.5697	4.96
I	Avermectin 0.05%	51.1020 a	6.2255	8.21
	Control	25.9041 b	5.8803	4.41
	Dinotefuran 0.05%	27.0590 b	5.8803	4.60
	Fipronil 0.01%	45.0533 a	6.3021	7.15
	Fipronil 0.03%	23.5774 b	6.7056	3.52
	Hydramethylnon 2.0%	46.4824 a	12.773	3.64
S	Avermectin 0.05%	22.6354 b	6.9813	3.24
	Control	19.7521 b	6.8641	2.88
	Dinotefuran 0.05%	36.2516 a	7.0092	5.17
	Fipronil 0.01%	37.0412 a	8.3937	4.41
	Fipronil 0.03%	40.4984 a	7.7269	5.24
	Hydramethylnon 2.0%	47.3915 a	8.1788	5.79

<sup>a</sup>Repeated measures ANOVA significant at  $\alpha < 0.05$ ,  $df = 25$  for each strain and treatment combination.<sup>b</sup>Least squares means (LSmeans) within a strain and followed by different letters are significantly ( $P < 0.05$ ) different based on pairwise differences of least squares means.

fipronil). Pro-insecticides (indoxacarb) and insect growth regulators such as juvenile hormone analogs (fenoxycarb, hydroprene, methoprene, and pyriproxyfen) and chitin synthesis inhibitors (lufenuron and noviflumuron) as well as inorganics including boric acid, arsenic, and sodium fluoride also have been used in cockroach baits. In

this study, we evaluated baits containing abamectin, dinotefuran, fipronil, and hydramethylnon.

Insecticide resistance is the most important factor contributing to control failures and may be caused by the repeated use or the continuous presence of insecticide residuals in the environment.



**Fig. 1.** Performance index (PI) relationships for German cockroach bait products and an untreated control determined in Ebeling choice boxes against 7 field-collected and 1 susceptible strain. Points represent means of 6 replicate boxes, each containing 20 adult male German cockroaches.



German cockroaches have developed resistance to practically all insecticides used for their control and cross-resistance to other insecticides (Cochran 1995; Lee et al. 1996; Wen & Scott 1997; Wei et al. 2001; Chai & Lee 2010; Wu & Appel 2017; Fardisi et al. 2019). In addition, German cockroaches have developed behavioral resistance or aversion to glucose (Silverman & Bieman 1993) and other sugars, including fructose, sucrose, and maltose (Wang et al. 2004), that are used as feeding stimulants in many bait formulations. Like physiological insecticide resistance, sugar aversion is an inherited trait (Silverman & Bieman 1993; Wang et al. 2006b) and relatively widespread (Silverman & Ross 1994; Lee & Soo 2002). Wada-Katsumata et al. (2013) described the physiological mechanism involved in glucose aversion; glucose-averse cockroaches perceive glucose (normally a phagostimulant) as bitter resulting in aversion. Manufacturers have altered their bait formulations to overcome sugar aversion. None of the 7 field-collected strains (or the susceptible strain) exhibited bait aversion in preliminary studies (data not shown). Therefore, reduced activity of the baits in this study is likely due to physiological tolerance or resistance to the active ingredient.

Even though the detailed history of insecticide use against the field-collected cockroaches used in this study is unknown, we can make several reasonable assumptions. First, these cockroach populations were exposed to permethrin or other pyrethroids because all the strains exhibited significant resistance to permethrin in topical bioassays (Wu & Appel 2017) and continuous exposure experiments (Wu & Appel 2018) in previous studies. Second, 61% of residents where these cockroaches were collected indicated that they used aerosol spray insecticides (Dingha et al. 2013). According to the US EPA (2017) market estimates, permethrin and other pyrethroids are the most common active ingredients in consumer aerosol insecticides. Therefore, it is likely that these field populations of German cockroaches were exposed to and selected by pyrethroid insecticides. The exposure of these strains to any of the active ingredients in the bait products tested also is unknown.

All bait products were active against all German cockroach strains in the continuous exposure tests, although there were differences in  $LT_{50}$  values among products and strains. The  $LT_{50}$  values did not exceed approximately 7 d for any product-strain combination (Table 2). However, resistance ratios of approximately 1.5 and above (Fardisi et al. 2019) indicated tolerance or true resistance to several active ingredients. Continuous exposure and choice box resistance ratios were correlated highly indicating relatively consistent responses using the 2 assays. Since its introduction in the 1980s, bait formulations containing hydramethylnon (Combat™ and Maxforce™) revolutionized the control of German cockroaches (Appel & Rust 2021). Because of the popularity of Combat™ bait with consumers, it is likely that if the field strains had been exposed to baits, they were most likely to have been exposed to hydramethylnon. Based on  $LT_{50}$  values, the field-collected strains can be categorized into 2 groups: strains with similar susceptibility as an insecticide susceptible strain (D, E, and I) and strains with tolerance or resistance (B, DCC2, G, and H). The  $LT_{50}$  values of the susceptible strains averaged 2.4 d, whereas the tolerant or resistant strains averaged  $LT_{50}$  values of 5.2 d, or more than double that of the susceptible strain. Therefore, under field conditions in apartments and homes, residents could be expected to see live cockroaches a minimum of twice as long if the strains treated were insecticide-resistant. Though it is unlikely that an additional 2 to 3 d would affect the population dynamics and reproduction of a German cockroach population significantly, their presence alone could cause residents to assume the baits were not effective and lead to the purchase and use of aerosol pyrethroid products.

Repellency of insecticides also affects their performance, especially for German cockroaches (Ebeling et al. 1966; Rust & Reiersen 1978; Ebeling 1995; Appel 2004). Long-term repellency or avoidance is a result of an associated learning process of individuals exposed to even minute sublethal concentrations of an insecticide (repellent) (Ebeling et al. 1966; Metzger 1995; Ross & Mullins 1995). Immediate repellency also is possible because of instantaneous insecticide-receptor interactions. The bait formulations used in this study were not repellent, as evidenced by the relatively low (< 70%) repeated measures mean percentage repellency values (Table 4) and the positive performance index values (Table 5). The period required for half the  $PI_{Max}$  value to be reached, or  $t_{PI_{Max}/2}$ , was positively correlated with continuous exposure and choice box resistance ratios. That is, higher resistance or tolerance resulted in a longer period to reach maximum performance. In addition, repellency was not correlated with resistance ratios indicating the independence of these measures, at least for these baits. Other studies, however, have shown a positive correlation between resistance and repellency. These studies used soluble powder, wettable powder, and emulsifiable concentrate formulations, not baits (Rust et al. 1993). The repeated measures percentage repellency means are likely somewhat inflated because of the reduced numbers of individuals over time and the reduced movement of intoxicated cockroaches before their death. Intoxicated individuals were unable to return to the dark side of the choice box and were scored as repelled. The insecticide later overcame those individuals (Appel and Eva, personal observation) after 1 or even several days of being scored as repelled.

Some insecticidal baits may be contaminated and lose their effectiveness if they are inadvertently treated with repellent insecticides (Appel 2004). Similarly, areas near bait deposits may become contaminated when sprayed with insecticides. Pyrethroid insecticides, such as those used in most aerosol sprays, are toxic and usually quite repellent (Rust & Reiersen 1988). Avoidance of insecticide deposits, either directly or by avoidance of an area with deposits, results in decreased performance and lack of control (Ebeling et al. 1966). If repellent enough, the application of insecticides would spread an infestation to different parts of homes or buildings rather than reducing the pest population (Ebeling et al. 1966).

Integrated pest management programs for German cockroach management have included trapping to assess population size and composition, sanitation to remove harborage and competitive food, education of residents, and the judicious use of insecticidal baits. Wang et al. (2006a) and others have demonstrated the effectiveness of an integrated pest management approach for German cockroaches using baits. More recently, Miller and Smith (2020) found that gel bait applied onto squares of waxed paper and folded diagonally then distributed throughout infested kitchens was as or more effective than a combination of integrated pest management tactics. Their approach did not require residents to remove dishes or other items from cabinets before treatment. The cockroaches remained relatively undisturbed, while the addition of novel objects (bait-treated waxed paper) probably increased exploratory behavior and bait consumption. Elimination of competitive food through sanitation only could increase the effectiveness of this approach.

Our results demonstrate that field-collected German cockroaches have resistance or tolerance to several insecticidal bait formulations. In addition, the level of resistance is correlated with lower performance index values ( $PI_{Max}$ ) and slower activity ( $t_{PI_{Max}/2}$ ). These performance index parameters are directly related to field performance (Rust & Reiersen 1978; Appel 1990, 1992). Based on these laboratory results we would predict poor field results with many of these baits to most field strains. However, all of the baits eventu-

**Table 5.** Relationship between d and performance index (PI) of solid and gel bait formulations to several strains of the German cockroach, *Blattella germanica* (L.), in Ebeling choice boxes<sup>a</sup>.

Strain	Treatment	tPI <sub>max</sub> /2 b ± SE <sup>b</sup>	PI <sub>Max</sub> a ± SE <sup>b</sup>	r <sup>2</sup>	F
B	Avermectin 0.05%	2.52 ± 2.29	100.00 ± 24.65	0.615	19.19
	Control	17.99 ± 26.18	76.66 ± 72.56	0.653	22.62
	Dinotefuran 0.05%	0.40 ± 0.04	97.81 ± 0.84	0.937	177.84
	Fipronil 0.01%	0.64 ± 0.26	100.00 ± 5.09	0.679	25.41
	Fipronil 0.03%	0.50 ± 0.11	100.00 ± 2.41	0.806	49.75
	Hydramethylnon 2.0%	1.85 ± 1.20	100.00 ± 14.91	0.676	24.98
D	Avermectin 0.05%	2.08 ± 1.30	100.00 ± 15.33	0.714	29.94
	Control	32.99 ± 860.41	6.13 ± 122.25	0.037	0.467
	Dinotefuran 0.05%	0.37 ± 0.05	84.50 ± 1.02	0.866	77.27
	Fipronil 0.01%	2.47 ± 1.24	100.00 ± 13.48	0.763	38.54
	Fipronil 0.03%	0.71 ± 0.42	84.48 ± 6.85	0.409	8.30
	Hydramethylnon 2.0%	2.90 ± 3.05	100.00 ± 30.74	0.575	16.24
DCC2	Avermectin 0.05%	4.45 ± 5.15	100.00 ± 42.13	0.611	18.89
	Control	-0.88 ± 0.04	-6.87 ± 1.82	0.774	41.08
	Dinotefuran 0.05%	7.85 ± 3.37	100.00 ± 20.40	0.889	96.03
	Fipronil 0.01%	1.98 ± 1.04	100.00 ± 12.49	0.740	34.17
	Fipronil 0.03%	1.43 ± 0.72	100.00 ± 10.05	0.711	29.58
	Hydramethylnon 2.0%	3.51 ± 4.14	100.00 ± 38.13	0.624	21.53
E	Avermectin 0.05%	5.83 ± 7.34	100.00 ± 52.27	0.589	19.63
	Control	97.95 ± 1845	100.00 ± 1700	0.281	4.69
	Dinotefuran 0.05%	1.47 ± 0.14	95.17 ± 1.83	0.966	340.14
	Fipronil 0.01%	2.51 ± 1.19	100.00 ± 12.82	0.788	44.56
	Fipronil 0.03%	1.26 ± 0.57	100.00 ± 8.37	0.744	34.79
	Hydramethylnon 2.0%	1.87 ± 1.37	100.00 ± 17.00	0.637	21.08
G	Avermectin 0.05%	3.33 ± 3.11	100.00 ± 29.36	0.605	20.87
	Control	268 ± 3330	100.00 ± 1190	0.009	0.11
	Dinotefuran 0.05%	0.96 ± 0.09	78.13 ± 1.13	0.963	308.61
	Fipronil 0.01%	1.57 ± 0.44	100.00 ± 5.83	0.850	68.02
	Fipronil 0.03%	1.26 ± 0.43	100.00 ± 6.20	0.806	49.87
	Hydramethylnon 2.0%	2.69 ± 1.78	100.00 ± 18.61	0.726	31.73
H	Avermectin 0.05%	2.70 ± 2.52	100.00 ± 26.30	0.618	19.41
	Control	454 ± 20137	100.00 ± 4313	0.001	1.26
	Dinotefuran 0.05%	2.55 ± 0.53	86.59 ± 4.96	0.891	98.10
	Fipronil 0.01%	1.88 ± 0.12	100.00 ± 6.62	0.853	69.34
	Fipronil 0.03%	1.46 ± 0.95	100.00 ± 13.18	0.643	22.12
	Hydramethylnon 2.0%	3.16 ± 2.96	100.00 ± 28.63	0.631	20.47
I	Avermectin 0.05%	2.70 ± 2.45	100.00 ± 25.57	0.626	20.04
	Control	55.01 ± 377.68	100.00 ± 577	0.436	9.27
	Dinotefuran 0.05%	0.80 ± 0.04	99.10 ± 0.74	0.985	798.67
	Fipronil 0.01%	1.75 ± 1.30	100.00 ± 16.50	0.629	20.37
	Fipronil 0.03%	1.31 ± 0.81	100.00 ± 11.68	0.638	21.12
	Hydramethylnon 2.0%	1.52 ± 1.23	100.00 ± 16.76	0.595	17.60
S	Avermectin 0.05%	3.40 ± 2.56	100.00 ± 23.96	0.705	28.66
	Control	-0.86 ± 0.07	-4.36 ± 1.96	0.498	11.89
	Dinotefuran 0.05%	0.26 ± 0.09	90.34 ± 1.94	0.963	334.98
	Fipronil 0.01%	— <sup>c</sup>	—	—	—
	Fipronil 0.03%	0.57 ± 0.10	100.00 ± 2.00	0.869	79.46
	Hydramethylnon 2.0%	1.83 ± 1.22	100.00 ± 15.20	0.635	20.90

<sup>a</sup>Nonlinear regressions of bait treatments significant at < 0.01, n = 60 for each strain and treatment combination.<sup>b</sup>a = PI<sub>Max</sub> or maximum asymptotic performance index value, b = t<sub>PI<sub>max</sub>/2</sub> or the period required for half the PI<sub>Max</sub> value to be reached<sup>c</sup>Data did not meet the assumptions of the rectangular hyperbolic model.

ally did kill the majority of all strains. Therefore, any methodology that results in greater bait consumption likely would increase efficacy, even for strains that were tolerant or resistant to bait active ingredients.

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