

Effects of Two Temperature Storage Regimes on the Efficacy of 3 Commercial Gel Baits against the German Cockroach, *Blattella germanica* L. (Dictyoptera: Blattellidae)

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Abstract

Background: To compare the effectiveness of 3 commercial gel bait formulations containing fipronil (Goliath[®] Cockroach Gel 0.05% AI), chlorpyrifos-A (Clean Bait[®] Gel, 2% AI), and chlorpyrifos-B (Serpa[®] Gel 2% AI) against German cockroaches (*Blattella germanica*) when stored at 23°C and 30° C after treatment.

Methods: Laboratory bioassays consisted of placing groups of fifteen cockroaches (a random combination of adult, mixed sex and large nymphs-stage 6) into a 5 L cylindrical plastic container with one drop of product (avg 0.10 g ± 0.01 g) applied to a 76 mm x 26 mm glass microscope slide affixed to the bottom of each container (one product tested per container). Cumulative mortality was assessed at 6 h, 1, 2, 3, 4, and 5 d after application. To determine the further effects of storage temperature after treatment on residual activity of the gels, a drop of each product was applied to separate glass microscope slides and stored at either 23° C (warm) or 30° C (hot) under dark conditions for 0, 1, 7, 14, 30, 45, 60 and 90 d after application.

Results: Freshly applied baits (day 0) containing fipronil provided complete cockroach mortality (100%) within 5 d whereas chlorpyrifos-A and chlorpyrifos-B provided ≈72% and 88% mortality, respectively. Generally, cockroach mortality was greater when gels were stored at 30° C compared with 23° C.

Conclusion: The fipronil gel formulation proved to be as efficacious as the chlorpyrifos gels and in some instances surpassed the latter formulations depending on storage time and temperature by providing ≈90% mortality at 90 d post treatment.

Key words: *Blattella germanica*, *Chlorpyrifos*, *cockroach control*, *Fipronil*, *temperature*

Introduction

The German cockroach, *Blattella germanica* L. (Dictyoptera: Blattellidae) is one of the most prominent urban pests in the world. This species is known to transmit a vast array of pathogenic organisms e.g. bacteria, fungus, worms, and viruses (1, 2). Moreover, Tsai and Chen (3) reported that 40% of bronchial asthma patients in China were allergic to this cockroach species. German cockroaches may also contaminate human food and leave a persistent, objectionable odor in infested areas. Often times these pests are found in kitchens and bathrooms because they favor warm (27-28° C), humid areas especially when they are in close proximity to food and water.

Sanitation is key to reducing infestations as it limits the pest's access to food resources. Primarily, insecticide spray applications are often used for control of *B. germanica* but sometimes the presence of grease or caustic floor cleaners, especially in commercial food establishments, can adversely affect their effectiveness. Contamination of food handling areas is also a major concern with these types of insecticide formulations. Moreover, continual application of residual insecticides has resulted in cockroaches becoming resistant to several of the commonly used insecticide classes such as organochlorines, carbamates and synthetic pyrethroids (4-7).

The development of bait formulations has allowed more precise site-specific placement of toxicants into cockroach harborages where spray applications have had limited usefulness. A recent option for cockroach control is gel bait formulations and has been reported to perform well against field populations of the German cockroach (8). Gel baits generally contain a variety of attractive ingredients as carriers with high moisture content. These bait formulations have the added advantage over dry baits in that they can be applied in small amounts in a variety of locations that can improve bait efficiency (8, 9). Moreover, they are easily applied, safer, and exhibit longer residual activity (10-13). Moreover, baits can be removed when control is accomplished. However, if bait application occurs in areas that routinely have high temperatures, many gel baits may run and/or rapidly lose efficacy after a period of time. For example, the temperature in kitchens or food preparation areas (such as restaurants) is often greater (30° C) than the ambient temperature of the dining room (23° C). Therefore, after initial application, we wanted to determine the residual efficacy of several different commercial gel bait formulations against *B. germanica* populations with prolonged exposure (i.e. storage) to these two temperature regimes in laboratory bioassays.

Materials and Methods

Insects

An insecticide susceptible strain of *B. germanica*, originally obtained from Dr. Oner Kocak (Hacettepe University, Pesticide Test Laboratory, Ankara, Turkey) and maintained in the laboratory for 20 yr, was used in these experiments. The colony was maintained at 26±2° C, 60±10% relative humidity and a photoperiod of 12L12D.

Gel formulations

Three commercial gel baits were used: Goliath[®] Cockroach Gel (0.05% AI, fipronil, BASF Turk Kimya San. Tic. Ltd. Sti. Istanbul, Turkey), Clean

Bait[®] Gel (2% AI, chlorpyrifos-A; Kukbo Science Co. Ltd., South Korea), and Serpa[®] Gel (2% AI, chlorpyrifos-B; Mylva S. A. C./Verdi 24 08012 Barcelona, Spain).

Experimental set-up

Before testing, 15 cockroaches (a random combination of adult, mixed sex and large nymphs, stage 6) were placed into a 5 L cylindrical clear plastic container 48 h before testing for acclimation purposes. (Durier and Rivault (14) previously reported that cockroach age generally does not affect efficacy performance in gel bait assays.) The vertical walls of each container had been previously coated with talc (Utalk Pudra[®], Kurtsan İlacları A.S., Merter/Istanbul, Turkey) to prevent cockroach escape. During the acclimation period (and during testing) half of a sugar cookie, a cotton ball saturated with water and a cardboard tube (diameter 4.5 cm x height 10 cm) was placed in each container that provided food and harborage. After acclimation, one drop (or bead) of product (avg 0.10 g±0.01 g) was applied to a 76 mm x 26 mm glass microscope slide and placed on the bottom of containers at 0900h. Only one gel formulation was tested per container. Tests were conducted at 26±2° C, 60±10% relative humidity, and a photoperiod of 12L12D in a laboratory. Because fipronil is often slow acting and may possess secondary kill properties due to coprophagy, necrophagy, or cannibalism (15, 16), mortality was assessed at 6 h, 1, 2, 3, 4, and 5 d after application. Cockroaches were considered 'dead' when they were unable to right themselves. To evaluate efficacy under operational conditions, dead cockroaches were left in containers until the end of a test (5 d). In order to determine the effects of storage temperature on residual activity of the gels after placement, one drop of each product was applied to separate glass microscope slides and stored at either 23° C (warm) or 30° C (hot) under dark conditions for 1, 7, 14, 30, 45, 60 and 90 d after application. A sufficient number of slides were treated so that none were tested more than once. Tests were repeated on four different days.

Statistical analyses

Mean percent mortality data were subjected to one-way analysis of variance (ANOVA) and Student-Newman-Keuls (SNK) to determine differences within and between products, storage time and temperature (17). Differences were considered significant at $P < 0.05$. Cumulative 5 d mean data are presented in tables.

Results

Comparison of storage time and temperature on individual product efficacy

Generally, cockroach average mortality to chlorpyrifos-A baits stored at 30°C for 30, 45, and 90 d was significantly greater than at 23°C (Table 1). Conversely, at 23°C mortality was significantly greater to chlorpyrifos-A baits stored for 14 d and least to baits aged 90 d compared with the other storage time periods. At 30°C mortality was greatest to this product when stored for 1 and 30 d. At 14 d mortality significantly declined to 66%, the reason for this sudden decrease is unknown.

Cockroach average mortality to fipronil bait aged 1 d was not significantly greater than bait aged through 60 d at either storage temperature (Table 1). During this period average percent mortality ranged from 95 to 98.3%. However, when the bait had been stored for 90 d at 23°C, mortality had significantly decreased to <60%. Whereas, fipronil bait stored for the same amount of time at 30°C continued to produce 90% mortality.

Average mortality to chlorpyrifos-B bait stored at various time periods at either temperature was highly variable (Table 1). Mortality levels generally followed similar patterns of decline seen in chlorpyrifos-A as storage time increased. However, cockroach mortality to chlorpyrifos-B bait stored at 23°C for 60 d suddenly increased significantly to ≈87% compared with 56% at 30°C. In this instance, evaporation of gel moisture may have resulted in concentration of the active ingredient. Cockroach mortality to chlorpyrifos-B bait stored for 90 d continued to decline at both temperatures to >47% and were not significantly different from one another.

Chlorpyrifos-B baits stored at 23°C for 1 and 14 d exhibited the greatest cockroach mortality, but were not significantly different from one another, when compared with the other storage periods at this temperature (Table 1). However at 90 d, efficacy was significantly reduced compared with the other periods. At 30°C, chlorpyrifos-B baits exhibited the greatest mortality at day 14 compared with the other storage periods. At 60 and 90 d post application, mortality was significantly less than the rest of the storage periods but not significantly different from one another.

Product efficacy comparison

Cockroach mortality to fipronil or chlorpyrifos-B baits stored at 23°C for 1 day was significantly greater than chlorpyrifos-A (Fig. 1). However, no significant difference in toxicity between all 3 products was observed when baits were stored at this temperature for 14 d. Fipronil gel stored for 30 and 45 d at 23°C continued to provide significantly greater mortality (≥ 95%) than the chlorpyrifos gels. At 60 d cockroach mortality to chlorpyrifos-B and fipronil was not different from each other but were greater than chlorpyrifos-A. At 90 d mortality to the fipronil gel had declined to ≈60% but was still significantly greater than the chlorpyrifos gels.

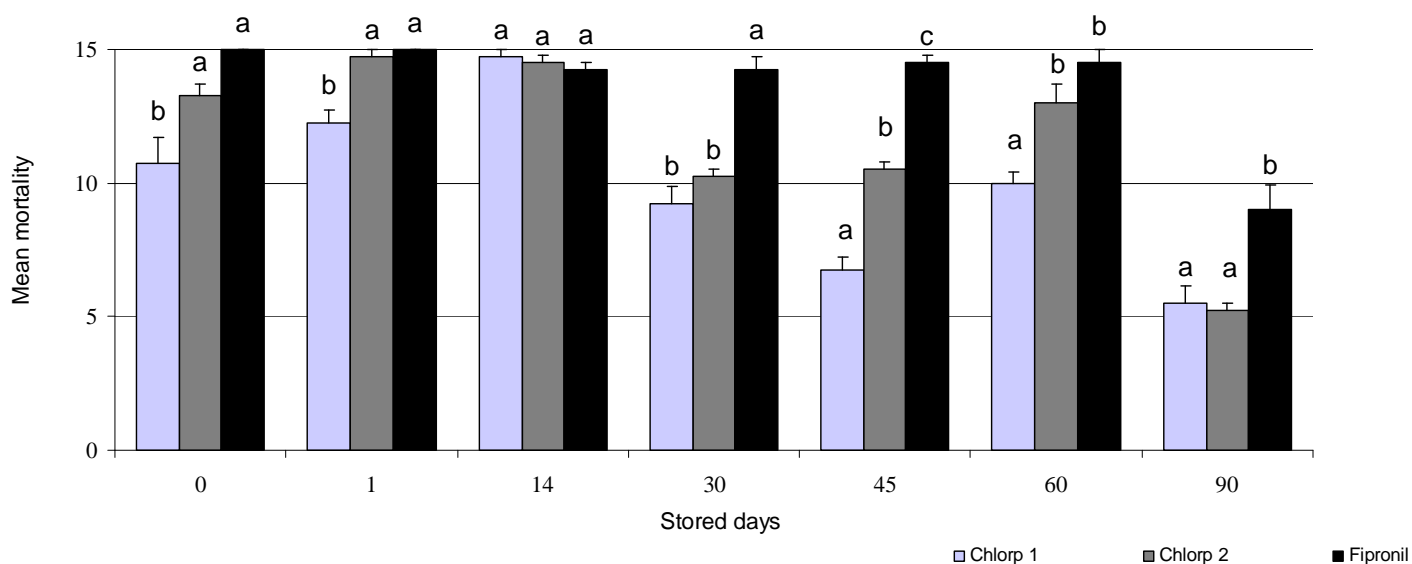
At 30°C, fipronil mortality was fairly consistent and ranged from 100 to 90% through 90 d (Fig. 2). Cockroach mortality from chlorpyrifos-A gels stored at this temperature were not significantly different than fipronil at days 1 and 30 while chlorpyrifos-B gels only performed as well as fipronil on day 14. At days 45 and 60, fipronil had performed significantly better than the chlorpyrifos formulations but at 90 d there was no difference between chlorpyrifos-A and fipronil when stored at 30°C.

Table 1: Evaluation of 3 commercial gel bait formulations against *Blattella germanica* stored at 23°C and 30°C under laboratory conditions (5 day cumulative mean percent mortality±SE).

Stored days	Clean Bait Gel (Chlorpyrifos-A)		Goliath Gel (Fipronil)		Serpa Gel (Chlorpyrifos-B)	
	23°C	30°C	23°C	30°C	23°C	30°C
0	71.5 ± 6.3 a ^x B ^y	71.5 ± 6.3 aA	100.0 ± 0.0 aB	100.0 ± 0.0 aB	88.3 ± 3.0 aC	88.3 ± 3.0 aB
1	81.8 ± 3.3 aC	98.3 ± 1.8 bC	98.3 ± 1.8 aB	100.0 ± 0.0 aB	98.3 ± 1.8 bD	81.5 ± 5.0 aB
14	98.3 ± 1.8 bD	66.3 ± 2.7 aA	94.8 ± 1.8 aB	100.0 ± 0.0 bB	96.5 ± 2.0 aD	100.0 ± 0.0 aC
30	61.5 ± 4.1 aB	100.0 ± 0.0 bC	95.0 ± 3.1 aB	96.0 ± 2.0 aB	67.8 ± 1.8 aB	71.5 ± 6.4 aB
45	45.0 ± 3.1 aA	83.3 ± 4.3 bB	96.5 ± 2.0 aB	100.0 ± 0.0 aB	69.5 ± 2.2 aB	74.8 ± 4.4 aB
60	66.3 ± 2.7 aB	81.8 ± 3.4 bB	96.8 ± 3.3 aB	96.5 ± 2.0 aB	86.5 ± 4.7 aC	56.5 ± 4.1 bA
90	36.8 ± 4.3 aA	81.5 ± 4.2 bB	59.8 ± 5.9 aA	90.0 ± 1.7 bA	34.8 ± 1.8 aA	46.8 ± 4.7 aA

^xMeans followed by the same lower case letter on the same store day between storage temperatures for each product (row) are not significantly different ($P > 0.05$).

^yMeans followed by the same capital letter within an each product and temperature between storage day (columns) are not significantly different ($P > 0.05$).



^aMeans followed by the same letter on the same day are not significantly different ($P > 0.05$).

Fig. 1: Laboratory evaluation of gel bait formulations against *Blattella germanica* stored at 23° C (5-day cumulative mean number dead±SE)

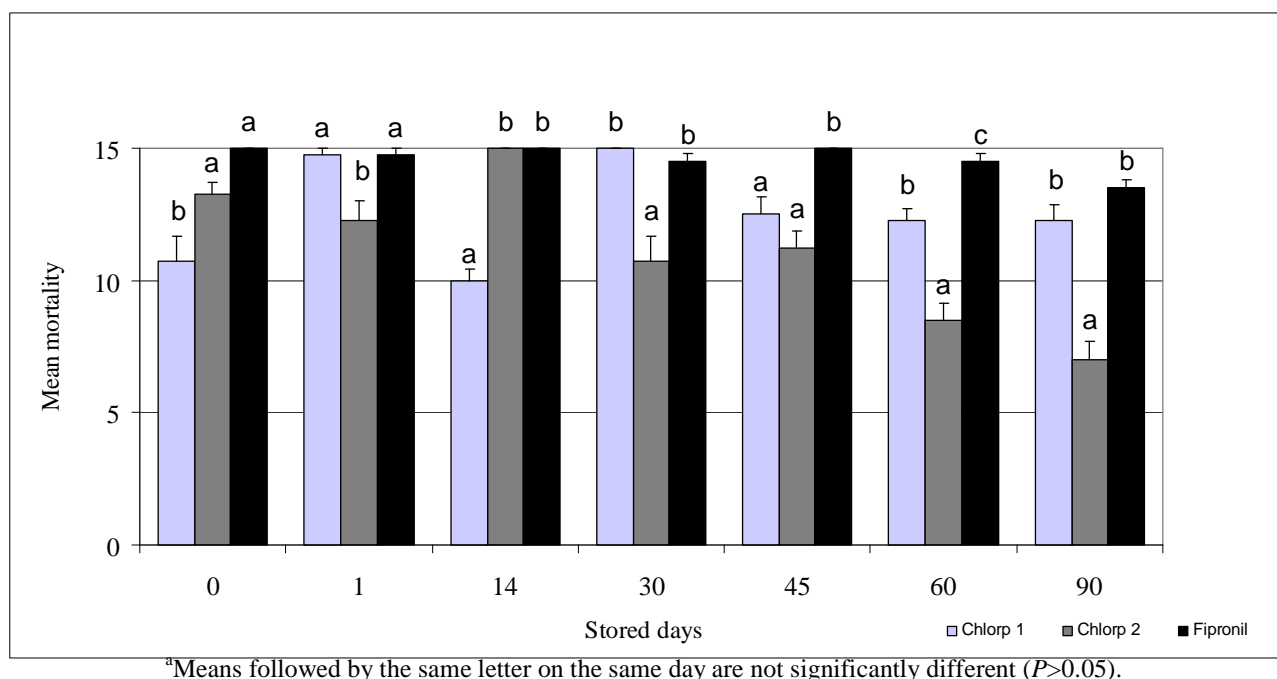


Fig. 2: Laboratory evaluation of gel bait formulations against *Blattella germanica* stored at 30° C (5-day cumulative mean mortality \pm SE)

Discussion

Our results showed that although all gel baits were highly effective they were not equally efficacious against *B. germanica*. Freshly applied baits containing fipronil provided complete cockroach mortality (100%) within 5 d followed by chlorpyrifos-B and chlorpyrifos A (Table 1). These data are in agreement with that reported by Kakeh et al. (18) who reported that fipronil was more toxic than chlorpyrifos. Indeed, complete mortality to fipronil was achieved at 2 d whereas the chlorpyrifos baits never achieved this level even at 5 d post application evaluation. Gahlhoff et al. (15) reported the LT_{50} of fipronil gel baits was 1.8 d and for chlorpyrifos (MaxForce FC) 8.5 d. It appeared that mortality in our study was greater than that reported by those authors. However, their study also found that secondary kill of male German cockroaches via cannibalism of nymphs that previously fed on fipronil or chlorpyrifos baits caused significantly greater mortality than when they fed upon untreated nymphs. Recent work by Buczkowski and Schal (16) on horizontal transfer of fipronil has confirmed this observation. Generally, we found that cockroach mortality was greater when the gels were stored at 30° C

compared with 23° C. It is possible that the active ingredients in these 3 formulations may have been concentrated as a result of increased evaporation of the gel moisture at the higher temperature. In any event, fipronil gel proved to be as efficacious as the chlorpyrifos gels and in some instances surpassed the latter formulations depending on storage time and temperature. Indeed, field trials by Ree et al. (19) found that fipronil gel baits applied in food handling establishments where temperatures can reach or exceed 30° C reduced *B. germanica* populations by 90.9% in Korean restaurants, 96.4% in Chinese restaurants, and 89.4% in beer-hall kitchens for 4 wk. Moreover, Miller and Peters (8) reported that at 12 wk Goliath cockroach gel applied at the rate of 1 drop (0.03 g) per m^2 achieved 94.8% reduction of field populations of Australian and German cockroaches. While Srinivasan et al. (20) found that this same fipronil formulation (0.03% AI gel) applied at rates from 0.025 to 0.125 g/m^2 on wood, cement, mud and thatch surfaces gave between 51-100% percent mortality of adult German cockroaches after 1 h exposure in laboratory bioassays.

Ethical considerations

Ethical issues (Including plagiarism, Informed Consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc) have been completely observed by the authors.

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