Journal of Economic Entomology, 118(5), 2025, 2536–2544
https://doi.org/10.1093/jee/toaf199
Advance Access Publication Date: 19 August 2025
Research

ENTOMOLOGICAL SOCIETY OF AMERICA
SHARING INSECT SCIENCE GLOBALLY
OXFORD

Household and Structural Insects

Toxicant translocation and colony impact in the Pharaoh ant (Hymenoptera: Formicidae) after consumption of gel bait-killed German cockroach (Blattodea: Ectobiidae) cadavers

Xingwei Feng, Dong-Hwan Choe, Michael K. Rust, Chow-Yang Lee*

Department of Entomology, University of California, Riverside, CA, USA

*Corresponding author. Department of Entomology, University of California, 900 University Avenue, Riverside, CA 92521, USA (Email: chowyang.lee@ucr.edu).

Subject Editor: Chin-Cheng Scotty Yang

This study investigated the translocation of cockroach bait toxicants to the Pharaoh ant, *Monomorium pharaonis* (L.), through feeding on cadavers of German cockroaches, *Blattella germanica* (L.), which were killed by commercial gel baits. The cockroaches were first treated with 1 of 4 gel baits: Advion Trio (0.6% indoxacarb, 0.1% pyriproxyfen, 0.1% novaluron), Maxforce FC Magnum (0.05% fipronil), Alpine Rotation 1 (0.5% dinotefuran), and Vendetta Nitro (0.5% clothianidin, 0.5% pyriproxyfen) before the Pharaoh ants were allowed to forage on the cadavers. The effects of the toxicants on the Pharaoh ant colonies were examined. Ants that fed on cadavers killed by Maxforce FC Magnum showed significantly higher ($P < 0.05$) mortality rates among queens and brood. Advion Trio initially led to the rapid reduction of queens, workers, and brood; however, the Pharaoh ant colony later recovered as the workers abandoned treated cockroach cadavers in favor of the regular diet. Examination of the queen ovaries revealed that, in the Vendetta Nitro-treated group, the size of mature oocytes was significantly reduced ($P < 0.05$). However, no structural damage was observed on any oocyte, as all exhibited a visible nucleus, multiple nurse cells, and well-developed follicular epithelium. The horizontal transfer efficacy of different cockroach gel bait toxicants is discussed.

Keywords: horizontal transfer, fipronil, clothianidin, dinotefuran, pyriproxyfen

Introduction

The Pharaoh ant, *Monomorium pharaonis* (L.), is one of the most significant cosmopolitan indoor pest species. Reports have shown that infestations of Pharaoh ants frequently occur in health care, homes, and food preparation facilities (Edwards and Baker 1981, Yap and Lee 1994, Man and Lee 2014, Tay et al. 2014). To effectively manage Pharaoh ant infestations, it is essential to select appropriate control strategies, as Pharaoh ant colonies may split and bud when disturbed, worsening the infestation (Buczkowski et al. 2005, Tay and Lee 2015). Studies have demonstrated that non-repellent residual treatments and baiting can control Pharaoh ants while preventing budding (Buczkowski et al. 2005). Current Pharaoh ant control strategies emphasize the use of bait, as residual sprays only kill a small portion of foragers, allowing the colony to recover from the resulting decline in worker population, while bait toxicants can be distributed throughout the colony (Vail 1996). Bait targeting Pharaoh ants should incorporate their dietary preferences to maximize toxicant uptake (Edwards and Abraham 1990).

In addition to the Pharaoh ant, the German cockroach, *Blattella germanica* (L.), widely recognized for its resilience and public health risks (Page et al. 2006, Tang et al. 2019, Schal and DeVries 2021), also presents a significant challenge in pest management (Fardisi et al. 2019). German cockroach infestations typically arise in residential homes, hospitals, restaurants, and supermarkets, where food, water, and shelter are plentiful (Alcamo and Frishman 1980, Tang et al. 2019). They serve as excellent carriers for the transmission of diseases and pathogens (Fotedar et al. 1991, Menasria et al. 2014). Given their synanthropic nature and potential to spread diseases within human environments, effective control methods are crucial for managing German cockroach infestations. A survey conducted in North Carolina revealed that most residents typically use aerosol sprays, baits, sprayers with concentrated sprays, and dust when addressing German cockroach infestations (Dingha et al. 2013).

Baiting offers several advantages, including ease of use and reduced chemical exposure to the environment (Schal 2011, Appel and Rust 2021). Additionally, the potential for

Received: 17 May 2025. **Revised:** 25 June 2025. **Accepted:** 17 July 2025
© The Author(s) 2025. Published by Oxford University Press on behalf of Entomological Society of America. All rights reserved. For commercial re-use, please contact reprints@oup.com for reprints and translation rights for reprints. All other permissions can be obtained through our RightsLink service via the Permissions link on the article page on our site—for further information please contact journals.permissions@oup.com.

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

horizontal transfer of bait toxicants may further enhance bait performance (Hamilton et al. 2023). In German cockroaches, mortality can occur in cockroaches not only through direct interaction with the bait, but also through necrophagy, coprophagy, emetophagy, and physical distribution of bait in the area (Kopanic and Schal 1999, Buczkowski and Schal 2001, Buczkowski et al. 2008, Hamilton et al. 2023). Similar toxicant distribution mechanisms also occur in ants, where workers translocate toxicants mainly through trophallaxis and necrophoresis (Choe and Rust 2008).

The global distributions of both Pharaoh ants and German cockroaches overlap, with infestation hot spots reported in the eastern United States, Europe, and the tropics (Leong and Grace 2008, Wetterer 2010, Lee and Wang 2021). Co-infestations of both species within the same building have been reported in sensitive environments where insecticide sprays and dust application are limited, such as hospitals and insectaries (Burden and Smittle 1975, Gliniewicz et al. 2003, Gliniewicz et al. 2006). Indoor aeroallergens from both Pharaoh ants and German cockroaches were also detected in homes in South Korea (Kim et al. 2007). Due to the coexistence of both species in the same building, an optimized bait formulation containing sulfluramid has also been developed to control both Pharaoh ants and German cockroaches simultaneously (Boase and Rupes 1996).

Given that Pharaoh ants often feed on live insects and insect cadavers in homes (Berenbaum 1994, Eow and Lee 2007, Klotz et al. 2008, Wetterer 2010), the potential for the horizontal transfer of bait toxicants may occur when Pharaoh ants consume the cadavers of German cockroaches killed by bait. Cross-species toxicant exposure has been demonstrated in the horizontal transfer of anticoagulant rodenticides, where nontarget wildlife feeding on contaminated rodent cadavers suffer accidental mortality or bioaccumulate toxicants through the food chain (Mendenhall and Pank 1980, Eason et al. 1999, Hoare and Hare 2006, van den Brink et al. 2018). However, little is known about this cross-species toxicant transfer pathway in insects. In this study, we investigated how Pharaoh ant colonies could be impacted by the bait toxicants of several cockroach gel baits after the colonies fed on the cadavers of German cockroaches killed by the baits, aiming to determine whether the ants would feed on the cockroach cadavers and whether these toxicants could be horizontally transferred through this pathway to a degree sufficient to affect colony survival.

Materials and Methods

Insects

The study was conducted using *M. pharaonis* obtained from USDA-ARS, Center for Medical, Agricultural, and Veterinary Entomology in Gainesville, Florida, in 2020 and reared in centrifuge tubes (12 cm in length, 1.5 cm in diameter) filled with unbleached paper towel strips under laboratory conditions of 27 ± 1 °C, 70 ± 2% RH, and a 12:12 L:D (light:dark) cycle in arenas (32 cm in length, 19 cm in width, 11 cm in height) coated with fluon at the University of California, Riverside over 4 yr. Water and a 10% sucrose solution were provided ad libitum, and Tetra Goldfish Flakes (Tetra GmbH, Melle, Germany) and fresh tuna (Kirkland Signature, Seattle, Washington) obtained from Costco as protein sources were provisioned once a week.

The German cockroaches used were from the UCR susceptible strain. The UCR strain was originally from the Orlando normal strain that was reared in UCR for over 40 yr and reared in 121-liter garbage bins equipped with an electrical barrier at 24 ± 2 °C, 40 ± 10 RH, and a 12:12 L:D cycle without any insecticide exposure. Dog food (Purina Dog Chow, Nestlé Purina Petcare, St Louis, Missouri), water, and corrugated cardboard harborage were provided ad libitum.

Experimental Design

The experimental setup consists of 3 round polyethylene containers (8.7 cm in diameter and 7.8 cm in height), with 2 containers, A and C, connected to a central container, B, each via a 5 cm Tygon tube on either side (Fig. 1). Ten third and fourth instar German cockroaches from the UCR susceptible strain were placed in container A and starved for 24 h. Following the starvation period, 0.3 g of bait was provided to the German cockroaches, allowing them to feed on it for 7 d. For the control groups, we used German cockroaches that were killed in the freezer at −20 °C for 24 h. Cockroach cadavers were only provided once at the beginning of the experiment for both treated and control groups. A Pharaoh ant subcolony consisting of 2 fertile queens, 100 workers, and 50 mg of mixed brood stage was introduced into container B 4 d after baiting the cockroaches, along with provisions of water, a 10% sucrose solution, and a piece of corrugated paper (5 × 5 cm) for harborage, allowing 3 days of acclimatization. Container C served as a clean arena where a regular diet of hard-boiled egg and canned tuna (Kirkland Signature, Issaquah, Washington, United States) was provided and replaced weekly. Tygon tubes were blocked

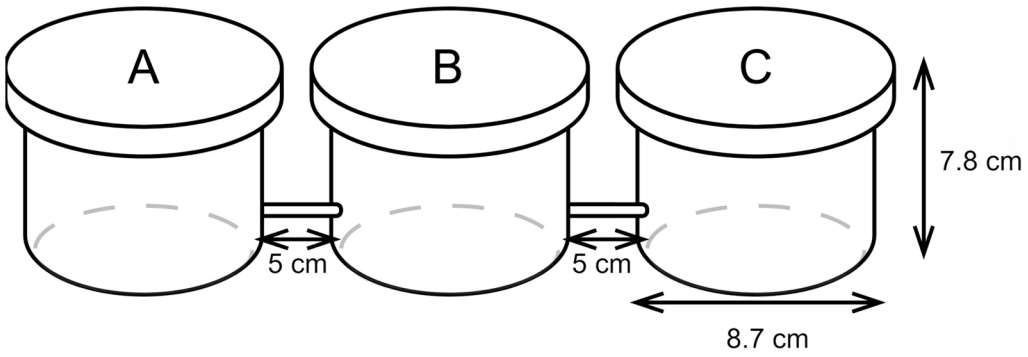

Fig. 1. The experimental setup used in this study. The polyethylene containers are labeled as: A) treated cockroach site; B) Pharaoh ant nest site; and C) clean site with regular diet.

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

with pieces of cotton until the experiment started to prevent Pharaoh ants from interacting with German cockroaches and the clean diet during the 3 d acclimatization period. The cottons were removed from the Tygon tube after acclimatization. All German cockroaches were killed after 7 d of baiting, and any leftover baits were removed after the baiting period.

Four common commercial cockroach gel bait products were evaluated (Table 1). The number of workers present in containers A and C was manually counted each week to determine whether the Pharaoh ant workers avoid contaminated cockroach cadavers. Worker mortality was assessed by manual counting the number of live ants every week, and the workers in container B, where the majority of colony resided, was determined by manually counting from photographs taken. Brood mortality was evaluated based on the reduction in total brood weight each week. Brood weight was measured using a digital electronic balance with a capacity of 220 g and a precision of 0.0001 g by placing the brood in a weighing boat. Queen mortality was assessed by manual counting the number of live queens in each replicate weekly. The presence of workers in containers A and C, as well as the mortality of workers, brood, and queens, was recorded in the morning weekly for 12 wk. The entire experiment was conducted twice, with 10 replicates in the first experiment and 8 replicates in the second one.

At the end of the study, selected remaining live queens were dissected under a stereo microscope (35×), and the ovary morphology along with the size of oocytes was examined using the Leica DMS1000 digital microscope with Leica LAS X software (version 5.1.1). To observe the presence of oocyte nuclei, nurse cells, and follicular epithelium, the ovaries were immediately fixed in Bouin's solution after dissection, stained with 4′,6-diamidino-2-phenylindole (DAPI), which marks DNA, and mounted on slides. Observations were made using the ZEISS Apotome 3 fluorescence microscope (400×) and ZEN Microscopy software (version 3.11).

Data Analysis

The weekly mean percentage reduction in worker population [(1 – number of live ants/initial number of workers in each subcolony)/total number of observations at each data collection point] and brood weight across different treatments [(1 – observed brood weight/initial brood weight in each subcolony)/total number of observations at each data collection point] were transformed ($\log_{10}$ + 2) and subjected to one-way analysis of variance (ANOVA). One-way ANOVA was also used to compare (i) the means of the number of workers present in containers A and C from each treatment, (ii) the number, and (iii) the size of mature oocytes. Means were separated using Tukey's Honestly Significant Difference (HSD) test. The impact of the baits on the survivorship of Pharaoh ant queens was analyzed with Kaplan–Meier survival analysis, and the survival curves between different treatments were compared using log-rank tests. All analyses were performed using IBM SPSS Statistics Version 30.0 (IBM Corp., Armonk, New York) at $\alpha = 0.05$.

Table 1. Commercial cockroach gel baits used in the study and their active ingredients

Product trade name	Active ingredient	Manufacturer
Advion Trio	0.6% indoxacarb 0.1% pyriproxyfen 0.1% novaluron	Syngenta Crop Protection, Greensboro, NC
Maxforce FC Magnum	0.05% fipronil	Bayer Environmental Science, Research Triangle Park, NC
Alpine Rotation 1	0.5% dinotefuran	BASF Corporation, Research Triangle Park, NC
Vendetta Nitro	0.5% clothianidin 0.5% pyriproxyfen	MGK, Minneapolis, MN

Results

Presence of Workers in Container A (Contaminated) and C (Clean)

In the control groups, the number of workers visiting both container A and C during the experimental period is not significantly different from each other (Fig. 2). In comparison, all treatments exhibited significantly higher activity ($P < 0.05$) in container C than in container A within each treatment, except for the Maxforce FC Magnum-treated group, where the number of ants present in container C was not significantly different from that in container A (Fig. 2). All treatments, except for the Maxforce FC Magnum-treated group, which had significantly fewer ($P < 0.05$) ants, showed no significant difference in the number of workers present in container C compared to that of the controls (Fig. 2). In container A, all treatments showed significantly fewer ants compared to the controls ($P < 0.05$), except for the Vendetta Nitro-treated group, where the number of ants present was not significantly different from that in the controls (Fig. 2).

Worker Mortality

In the first week posttreatment, the percentage reduction in the worker population was significantly greater in all treated groups than in the controls until the eighth week (Table 2, Supplementary Table S1). By the eighth week, Maxforce FC Magnum, Alpine Rotation 1, and Vendetta Nitro showed significantly greater percentage reductions in ant counts, whereas the Advion Trio-treated groups had recovered from the treatment, with their percentage reductions in ant counts no longer significantly different from those of the controls. This effect was sustained further up to the 12th week. At the 12th week, the percentage reduction in ant counts was not significantly different from that of the controls in the Maxforce FC Magnum- and Alpine Rotation 1-treated groups, whereas a significantly greater percentage reduction was observed in the Vendetta Nitro-treated groups (Table 2).

Brood Mortality

All tested baits showed a significantly greater reduction in brood weight during the first week posttreatment (Table 3, Supplementary Table S2). By the fourth week, Maxforce FC Magnum, Alpine Rotation 1, and Vendetta Nitro resulted in a significantly greater percentage reduction in brood weight (Table 3). In contrast, the percentage reduction in brood weight for Advion Trio-treated groups was not significantly different from that of the controls during weeks 3, 4, and 7 to 12 (Supplementary Table S2). At week 12, only Maxforce FC Magnum showed a significant reduction in brood weight (Table 3).

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

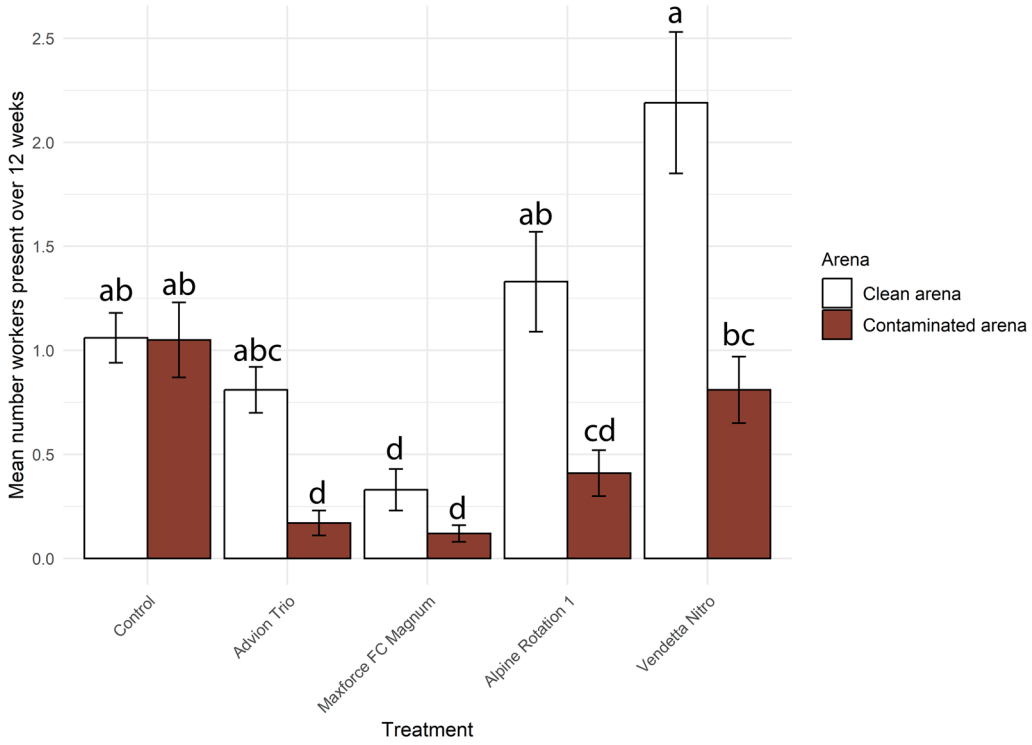

Fig. 2. Impacts of various treatments on the number of workers of the Pharaoh ant present in clean and contaminated arena. Means followed by the same letter indicate no significant difference ($P > 0.05$; Tukey's HSD test).

Table 2. Mean % reduction ± SEM of worker numbers of the Pharaoh ant at 1st, 4th, 8th, and 12th week after feeding on bait-killed German cockroach cadavers

	Mean % reduction ± SEM of worker numbers at *n*th week[a]			
Treatment	**1**	**4**	**8**	**12**
Control	13.4 ± 5.9 a	−62.6 ± 12.9 a	−65.3 ± 19.2 a	21.3 ± 22.4 a
Advion Trio	58.6 ± 7.9 b	−1.7 ± 10.4 b	−20.9 ± 7.3 ab	8.3 ± 17.0 a
Maxforce FC Magnum	65.6 ± 9.0 b	28.7 ± 18.3 b	43.1 ± 19.9 b	65.9 ± 15.7 ab
Alpine Rotation 1	41.7 ± 4.9 b	16.9 ± 11.5 b	46.4 ± 17.9 b	75.6 ± 15.3 ab
Vendetta Nitro	25.9 ± 9.8 b	−9.0 ± 15.8 b	20.4 ± 8.3 b	86.3 ± 3.0 b

[a]Means followed by the same letters within the same column are not significantly different ($P > 0.05$; Tukey's HSD test).

Table 3. Mean % reduction ± SEM in total brood weight of the Pharaoh ant at 1st, 4th, 8th, and 12th week after feeding on bait-killed German cockroach cadavers

	Mean % reduction ± SEM in brood weight (g) at *n*th week[a]			
Treatment	**1**	**4**	**8**	**12**
Control	−12.5 ± 10.3 a	15.7 ± 13.6 a	32.6 ± 10.0 a	58.0 ± 7.8 a
Advion Trio	32.3 ± 5.0 b	50.4 ± 6.1 ab	52.7 ± 9.5 ab	63.8 ± 10.8 ab
Maxforce FC Magnum	40.0 ± 9.0 b	73.9 ± 7.3 b	82.2 ± 8.1 b	90.1 ± 3.9 b
Alpine Rotation 1	40.1 ± 10.6 b	53.2 ± 8.5 b	70.2 ± 9.4 b	85.7 ± 8.8 ab
Vendetta Nitro	47.4 ± 11.1 b	66.0 ± 8.3 b	78.4 ± 4.5 b	82.8 ± 3.2 ab

[a]Means followed by the same letters within the same column are not significantly different ($P > 0.05$; Tukey's HSD test).

Queen Mortality

All treatments caused a significant decrease ($P < 0.05$) in survivorship compared to the controls (Fig. 3). Maxforce FC Magnum outperformed the other baits against Pharaoh ant queens, showing a significantly lower ($P < 0.05$) survival probability with a mean survival time of 6.8 wk, compared to 8.7 to 11.9 wk for the other treatments (Fig. 3).

Ovary Morphology

The reproductive system of queens consists of a visible spermatheca, a pair of oviducts, and a pair of ovaries in which the ovarioles with a mature oocyte are positioned toward the oviduct (Fig. 4A). The same morphology was also observed in Advion Trio- and Alpine Rotation 1-treated groups (Fig. 4B and D). In contrast, significantly fewer and smaller oocytes were

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

found in the Vendetta Nitro-treated group (Fig. 4E, Table 4). Additionally, although the reduction was not significant, fewer and smaller oocytes were also observed in the Maxforce FC Magnum-treated group (Fig. 4C, Table 4).

Oocyte Development

All treatments did not affect oocyte development (Fig. 5A–E). In the control group, healthy and well-developed oocytes were characterized by surrounding follicular epithelium, adjacent nurse cells, and a visible nucleus, which were also observed in all treatment groups. Although oocytes from the Vendetta Nitro-treated group were significantly smaller ($P < 0.05$) than those in the controls (Table 4), no structural damage was observed (Fig. 5E).

Discussion

In this study, foragers acquire bait toxicants by physically contacting and feeding on contaminated cockroaches, subsequently distributing the toxicants throughout the colony. The primary horizontal transfer pathway of toxicants in Pharaoh ants, either through physical contact or trophallaxis, was not determined in this study. Our results showed that Advion Trio and Maxforce FC Magnum initially demonstrated a significant reduction in the worker population, whereas no significant reduction was observed in the Alpine Rotation 1- and Vendetta Nitro-treated groups (Table 2). However, this effect did not persist throughout the observation period in the Advion Trio-treated group. In fact, the Advion Trio-treated group had completely recovered from the treatment, showing a larger average worker population compared to the controls by the end of the study, while a significant reduction in the worker population was observed only in the Vendetta Nitro-treated group, which initially did not show any significant reduction in worker population (Table 2). This phenomenon could be attributed to the difference in the number of ants visiting containers A and C. In the Advion Trio-treated group, significantly fewer ants visited container A than those in container C and the controls (Fig. 2), indicating that workers completely abandoned the contaminated arena after feeding on the contaminated cockroach cadavers and subsequently fed on a clean diet (Fig. 2). Maxforce FC Magnum and Alpine Rotation 1 also exhibited significantly fewer workers visiting container A compared to the controls (Fig. 2). On the other hand, the number of workers visiting container A in the Vendetta Nitro-treated group was not significantly different from that of the controls (Fig. 2), indicating that workers did not avoid the contaminated arena (Fig. 2). Additionally, it is noteworthy that only ant colonies that were exposed to Maxforce FC Magnum-treated cockroach cadavers exhibited significantly fewer average number of workers in container C compared to that of controls and other treated groups (Fig. 2), which could be a result of initial rapid worker population reduction, leading to fewer workers visiting container C (Table 2).

Table 4. Effect on queen ovary development of the Pharaoh ant after feeding on bait-killed German cockroach cadavers

Treatment (number of queens examined)	Mean number oocytes ± SEM[a]	Mean length of the largest oocyte ± SEM (μm)[a]
Control ($n = 22$)	9.3 ± 1.7 a	418.4 ± 17.7 a
Advion Trio ($n = 21$)	8.6 ± 1.4 ab	384.2 ± 19.8 ab
Maxforce FC Magnum ($n = 9$)	2.8 ± 0.8 ab	332.0 ± 37.6 ab
Alpine Rotation 1 ($n = 19$)	7.0 ± 1.9 ab	379.7 ± 16.4 ab
Vendetta Nitro ($n = 16$)	2.9 ± 0.6 b	327.8 ± 25.8 b

[a]Means followed by the same letters within the same column are not significantly different ($P > 0.05$; Tukey's HSD test).

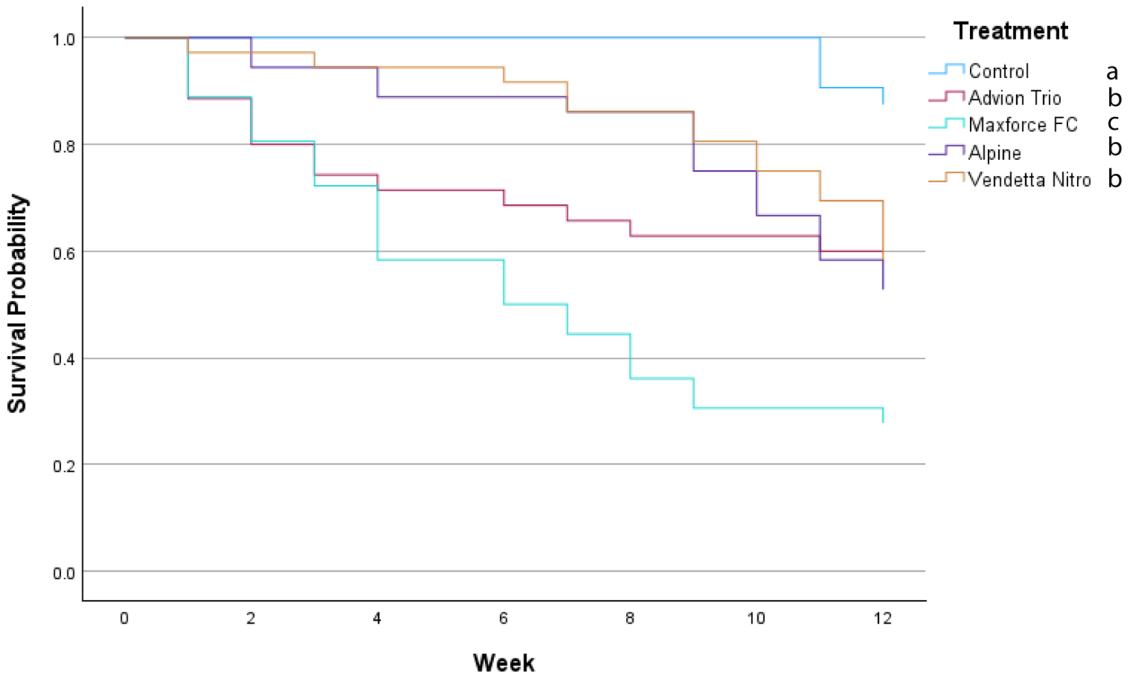

Fig. 3. Survivorship of Pharaoh ant queens exposed to bait-killed German cockroach cadavers. Treatment legends followed by the same letter indicate no significant difference ($P > 0.05$; Log-rank test).

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

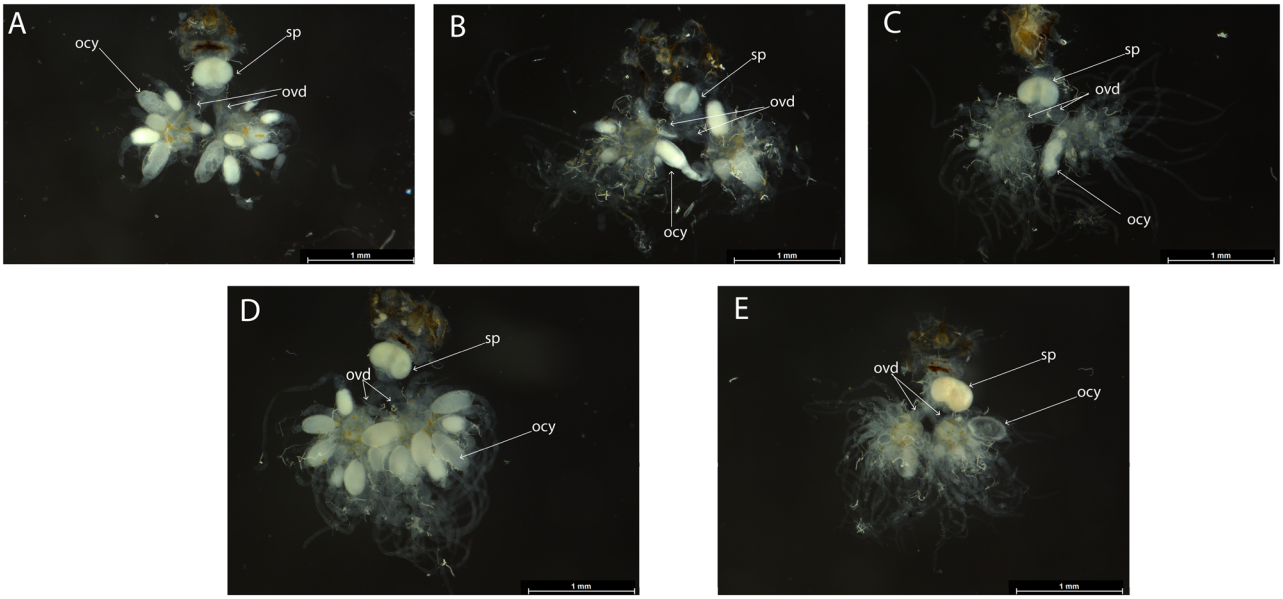

Fig. 4. Ovaries of queens of the Pharaoh ant after feeding on bait-killed German cockroach cadavers: A) Control, B) Advion Trio, C) Maxforce FC Magnum, D) Alpine Rotation 1, E) Vendetta Nitro. Abbreviations: ocy, oocyte; ovd, oviduct; sp, spermatheca.

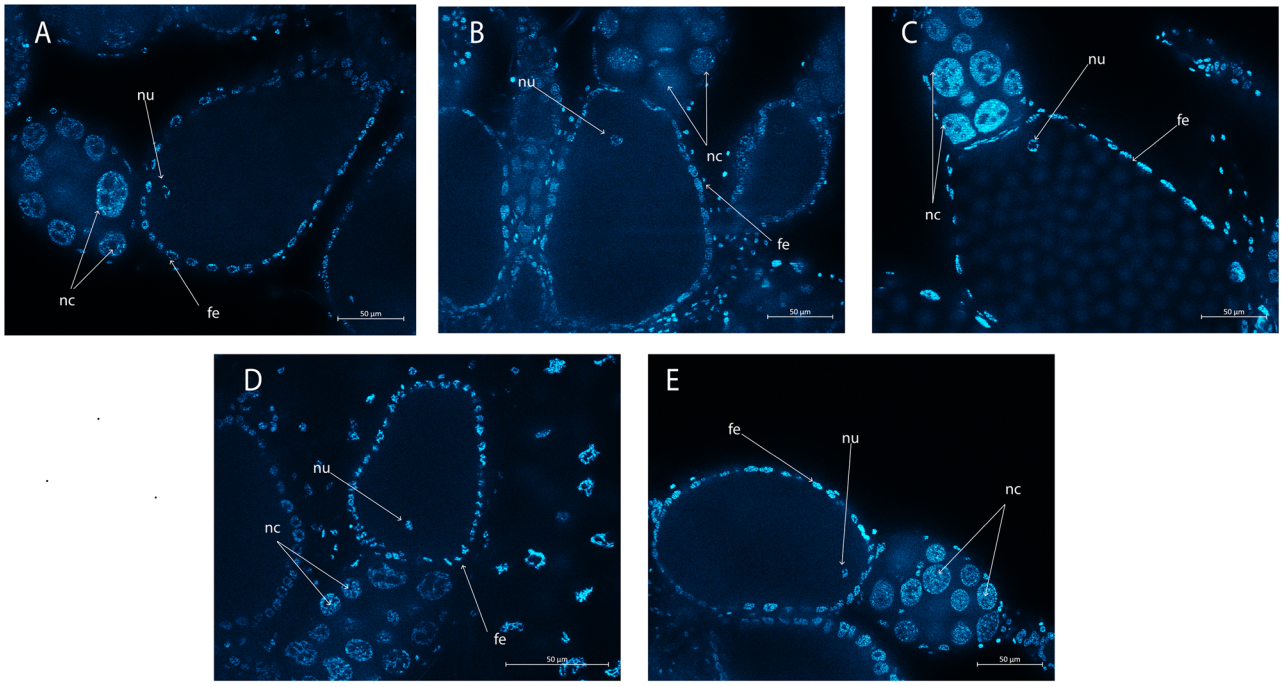

Fig. 5. Queen ovaries stained with DAPI after feeding on bait-killed German cockroach cadavers: A) Control, B) Advion Trio, C) Maxforce FC Magnum, D) Alpine Rotation 1, E) Vendetta Nitro. Abbreviations: fe, follicular epithelium; nc, nurse cell; nu, oocyte nucleus.

Hamilton et al. (2023) reported that fipronil and indoxacarb exhibit better horizontal transfer efficiency than dinotefuran and clothianidin. In addition, fipronil and indoxacarb demonstrate stable residual effects (Buczkowski et al. 2008, Jones and Bryant 2012), which align with the initial rapid elimination of workers, queens, and brood observed in the Advion Trio- and Maxforce FC Magnum-treated groups (Fig. 3, Table 2, Table 3). By the end of the study, only Vendetta Nitro significantly reduced the worker population (Table 2), likely because one of its active ingredients, pyriproxyfen, caused brood mortality, disrupted larval development, and contributed to worker attrition, along with direct worker mortality caused by clothianidin (Stanley 2004). A decline in the worker population was also observed in the control groups (Table 2), likely due to the lifespan of the workers. In Pharaoh ants, the longevity of workers ranges from 8 to 10 wk (Peacock et al. 1955, Edwards 1975, 1986). In this study, the initial cohort of 100 workers, along with those that emerged prior to week 4, likely reached the end of their lifespan between weeks 8 and 12, resulting in the observed decline (Table 2). An increase in the worker population may be expected if the experimental period is extended.

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

In addition to toxicant ingestion, physical contact with bait toxicants further enhances the efficacy of horizontal transfer, as undigested toxicants can be excreted and bait remnants may be physically distributed by German cockroaches within the arena (Kopanic and Schal 1999, Durier and Rivault 2000, Buczkowski et al. 2008). Among the active ingredients in the tested baits, fipronil exhibits strong contact toxicity (Lee 2000, Stanley 2004, Buczkowski et al. 2005). In this study, Maxforce FC Magnum resulted in significantly lower survivorship of queens compared to all other treatments and was the only treatment that caused a significantly higher brood reduction (Fig. 3, Table 3). In addition to being distributed through trophallaxis, fipronil may also be physically transferred on contact as workers groom the queens and brood.

Pyriproxyfen and novaluron are not lethal to adult insects and have been shown to have stable residual effects (Ameen et al 2005, Pitts-Singer and Barbour 2017, Joseph 2021); therefore, these active ingredients did not directly contribute to the mortality of workers and queens in this study. However, in addition to disrupting larval development, pyriproxyfen can still significantly impact colony dynamics by reducing the fecundity of Pharaoh ant queens (Tay and Lee 2014). Tay and Lee (2014) reported that pyriproxyfen decreases egg production, as well as the number and size of oocytes in treated queens. Consistent with these findings, a significant reduction in oocyte size was observed in the Vendetta Nitro-treated group in this study (Table 4). A similar reduction in oocyte size was also reported in Pharaoh ant queens treated with s-methoprene (Edwards 1975, Edwards and Clarke 1978). In contrast, Advion Trio did not cause any abnormalities in queen ovaries, likely because foragers abandoned the arena with contaminated cockroaches, preventing sufficient pyriproxyfen uptake to disrupt oocyte development (Fig. 2). Maxforce FC Magnum-treated group also did not show a significant difference in oocyte size compared to the controls (Fig. 4C). However, overall, a trend toward fewer and smaller oocytes was noted in treated queens compared to that of the control (Table 4). This change in ovarian morphology may be attributed to the significant reduction in brood. Børgesen and Jensen (1995) revealed that the fecundity of fertile queens depends on the presence of larvae, as the absence of larval secretions inhibits vitellogenin uptake by the ovarioles, thereby disrupting oocyte development (Børgesen 2000).

In this study, oocytes were stained with DAPI to observe oocyte development. Although a significant reduction in oocyte size was observed in the Vendetta Nitro-treated group, our results showed that queens from these colonies had oocytes with visible nuclei and nurse cells present throughout the ovarioles (Fig. 5E). In contrast, Tay and Lee (2014) demonstrated that directly feeding Pharaoh ant colonies with pyriproxyfen mixed with peanut oil led to severe oocyte developmental abnormalities within eight weeks, including the absence of oocyte nuclei, a lack of nurse cells, and underdevelopment of the follicular epithelium. This discrepancy suggests that the amount of pyriproxyfen delivered to the queens in this study was insufficient to induce any developmental deformities. This is most likely because the tested baits that contain pyriproxyfen also include indoxacarb and clothianidin. Given the faster-acting toxicity of indoxacarb and clothianidin compared to pyriproxyfen, queen mortality could likely have occurred before a sufficient amount of pyriproxyfen could induce oocyte developmental abnormalities (Vail and Williams 1995, Lim and Lee 2005, Tay and Lee 2014, Hamilton et al. 2023, Ma et al. 2024).

This study evaluated the horizontal transfer efficacy of commonly used cockroach gel baits by examining the effect of feeding on contaminated cockroaches alongside a clean diet. The results demonstrated that pyriproxyfen could be delivered to the colony and reach the queens, resulting in reduced oocyte size. However, the presence of other active ingredients in the tested baits could cause queen intoxication before they could acquire sufficient pyriproxyfen to disrupt oocyte development. In comparison, fipronil demonstrated rapid elimination of queens and brood, which would significantly impact colony growth (Edwards 1986, Passera 1994, Boonen and Billen 2017, Rajakumar et al. 2024). Previous studies have also identified fipronil as a strong candidate for managing Pharaoh ant infestations, both as a residual spray and in bait formulations (Lee 2000, Buczkowski et al. 2005). In cases where both Pharaoh ant and German cockroach infestations are present, fipronil-based cockroach gel baits could inadvertently control Pharaoh ant infestation, in addition to reducing German cockroaches.

Acknowledgements

The authors thank Naoki Yamanaka (University of California, Riverside) for providing fluorescence microscopy, Daiki Fujinaga (University of California, Riverside) for valuable suggestions on DAPI staining, and Allen Y. Zhao and Monique Arviso for their assistance with data collection.

Author Contributions

Xingwei Feng (Conceptualization [supporting], Data curation [lead]), Formal analysis [lead], Investigation [lead], Methodology [equal], Validation [lead], Visualization [lead], Writing—original draft [lead], Writing—review & editing [lead]), Dong-Hwan Choe (Supervision [supporting], Writing—original draft [supporting], Writing—review & editing [supporting], Michael Rust [Supervision [supporting], Writing—original draft [supporting], Writing—review & editing [supporting]), and Chow-Yang Lee (Conceptualization [equal], Funding acquisition [lead], Investigation [equal], Methodology [equal], Project administration [lead], Resources [lead], Supervision [lead], Writing—original draft [supporting], Writing—review & editing [supporting])

Supplementary Material

Supplementary material is available at *Journal of Economic Entomology* online.

Funding

This study was supported by the UC Riverside Urban Entomology Endowed Chair Research Fund.

Conflicts of Interest

All authors declare no conflict of interest.

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

References

Alcamo IE, Frishman AM. 1980. The microbial flora of field-collected cockroaches and other arthropods. *J. Environ. Health* 42:263–266. http://www.jstor.org/stable/44536571

Ameen A, Wang C, Kaakeh W, et al. 2005. Residual activity and population effects of noviflumuron for German cockroach (Dictyoptera: Blattellidae) control. *J. Econ. Entomol.* 98:899–905. https://doi.org/10.1603/0022-0493-98.3.899

Appel AG, Rust MK. 2021. Management using baits. In: Wang C, Lee CY, Rust MK, editors. *Biology and management of the German cockroach*. 1st ed. CSIRO Publishing. p. 213–230.

Berenbaum MR. 1994. *Bugs in the system: insects and their impact on human affairs*. 1st ed. Addison-Wesley.

Boase CJ, Rupes V. 1996. An optimized sulfluramid bait formulation for control of both Pharaoh's ants *Monomorium pharaonis* (L.) (Hymenoptera: Formicidae) and German cockroaches *Blattella germanica* (L.) (Dictyoptera: Blattellidae). In: Wildey KB, editor. Proceedings of the Second International Conference on Insect Pests in the Urban Environment. Organising Committee. p. 153–162.

Boonen S, Billen J. 2017. Caste regulation in the ant *Monomorium pharaonis* (L.) with emphasis on the role of queens. *Insect. Soc.* 64:113–121. https://doi.org/10.1007/s00040-016-0521-z

Børgesen LW. 2000. Nutritional function of replete workers in the Pharaoh's ant, *Monomorium pharaonis* (L.). *Insectes Soc.* 47:141–146. https://doi.org/10.1007/PL00001692

Børgesen LW, Jensen PV. 1995. Influence of larvae and workers on egg production of queens of the Pharaoh's ant, *Monomorium pharaonis* (L.). *Ins. Soc.* 42:103–112. https://doi.org/10.1007/BF01245702

Buczkowski G, Schal C. 2001. Emetophagy: fipronil-induced regurgitation of bait and its dissemination from German cockroach adults to nymphs. *Pestic. Biochem. Physiol.* 71:147–155. https://doi.org/10.1006/pest.2001.2572

Buczkowski G, Scharf ME, Ratliff CR, et al. 2005. Efficacy of simulated barrier treatments against laboratory colonies of Pharaoh ant. *J. Econ. Entomol.* 98:485–492. https://doi.org/10.1093/jee/98.2.485

Buczkowski G, Scherer CW, Bennett GW. 2008. Horizontal transfer of bait in the German cockroach: indoxacarb causes secondary and tertiary mortality. *J. Econ. Entomol.* 101:894–901. https://doi.org/10.1093/jee/101.3.894

Burden GS, Smittle BJ. 1975. *Blattella germanica*, *Periplaneta americana*, and *Monomorium pharaonis*: control with insecticidal baits in an animal building and in insectaries. *J. Med. Entomol.* 12:352–353. https://doi.org/10.1093/jmedent/12.3.352

Choe DH, Rust MK. 2008. Horizontal transfer of insecticides in laboratory colonies of the Argentine ant (Hymenoptera: Formicidae). *J. Econ. Entomol.* 101:1397–1405. https://doi.org/10.1093/jee/101.4.1397

Dingha B, Jackai L, Monteverdi RH, et al. 2013. Pest control practices for the German cockroach (Blattodea: Blattellidae): a survey of rural residents in North Carolina. *Fla. Entomol.* 96:1009–1015. https://doi.org/10.1653/024.096.0339

Durier V, Rivault C. 2000. Secondary transmission of toxic baits in German cockroach (Dictyoptera: Blattellidae). *J. Econ. Entomol.* 93:434–440. https://doi.org/10.1603/0022-0493-93.2.434

Eason CT, Milne L, Potts M, et al. 1999. Secondary and tertiary poisoning risks associated with brodifacoum. *N. Z. J. Ecol.* 23:219–224. https://www.jstor.org/stable/24054775

Edwards JP. 1975. The effects of a juvenile hormone analogue on laboratory colonies of Pharaoh's ant, *Monomorium pharaonis* (L.) (Hymenoptera, Formicidae). *Bull. Entomol. Res.* 65:75–80. https://doi.org/10.1017/S0007485300005782

Edwards JP. 1986. The biology, economic importance, and control of the Pharaoh's ant, *Monomorium pharaonis* (L.). In: Vinson SB, editor. *Economic impact and control of social insects*. 1st ed. Praeger. p. 257–271.

Edwards JP, Abraham L. 1990. Changes in food selection by workers of the Pharaoh's ant, *Monomorium pharaonis*. *Med. Vet. Entomol.* 4:205–211. https://doi.org/10.1111/j.1365-2915.1990.tb00278.x

Edwards JP, Baker LF. 1981. Distribution and importance of the Pharaoh's ant *Monomorium pharaonis* (L) in National Health Service Hospitals in England. *J. Hosp. Infect.* 2:249–254. https://doi.org/10.1016/0195-6701(81)90045-1

Edwards JP, Clarke B. 1978. Eradication of Pharaoh's ants with baits containing the insect juvenile hormone analogue methoprene. *Int. Pest Control* 20:8–10.

Eow AG, Lee CY. 2007. Comparative nutritional preferences of tropical pest ants, *Monomorium pharaonis*, *Monomorium floricola* and *Monomorium destructor* (Hymenoptera: Formicidae). *Sociobiology* 49:165–186.

Fardisi M, Gondhalekar AD, Ashbrook AR, et al. 2019. Rapid evolutionary responses to insecticide resistance management interventions by the German cockroach (*Blattella germanica* L.). *Sci. Rep.* 9:8292. https://doi.org/10.1038/s41598-019-44296-y

Fotedar R, Shriniwas UB, Verma A. 1991. Cockroaches (*Blattella germanica*) as carriers of microorganisms of medical importance in hospitals. *Epidemiol. Infect.* 107:181–187. https://doi.org/10.1017/S0950268800048809

Gliniewicz A, Sawicka B, Czajka E. 2003. Occurrence of insect pests in hospitals in Poland. *Prz. Epidemiol.* 57:329–334.

Gliniewicz A, Sawicka B, Mikulak E. 2006. Pest control and pesticide use in hospitals in Poland. *Indoor Built Environ.* 15:57–61. https://doi.org/10.1177/1420326X06062235

Hamilton JA, Wada-Katsumata A, Schal C. 2023. Cockroaches as trojan horses for control of cockroach aggregations with baits. *J. Econ. Entomol.* 116:529–537. https://doi.org/10.1093/jee/toad018

Hoare JM, Hare KM. 2006. The impact of brodifacoum on non-target wildlife: gaps in knowledge. *N. Z. J. Ecol.* 30:157–167. https://www.jstor.org/stable/24056337

Jones SC, Bryant JL. 2012. Contact toxicity and residual efficacy of Indoxacarb against the European earwig (Dermaptera: Forficulidae). *Insects* 3:593–600. https://doi.org/10.3390/insects3030593

Joseph SV. 2021. Transovarial effect of novaluron: persistence and residual effects on *Stephanitis pyrioides* (Hemiptera: Tingidae). *Can. Entomol.* 153:702–713. https://doi.org/10.4039/tce.2021.33

Kim C-W, Song J-S, Choi S-Y, et al. 2007. Detection and quantification of Pharaoh ant antigens in household dust samples as newly identified aeroallergens. *Int. Arch. Allergy Immunol.* 144:247–253.

Klotz JH, Hansen L, Pospischil R, et al. 2008. *Urban ants of North America and Europe identification, biology and management*. Cornell University Press. p. 196.

Kopanic RJ, Jr, Schal C. 1999. Coprophagy facilitates horizontal transmission of bait among cockroaches (Dictyoptera: Blattellidae). *Environ. Entomol.* 28:431–438. https://doi.org/10.1093/ee/28.3.431

Lee CY. 2000. Performance of hydramethylnon-and fipronil-based containerized baits against household ants in residential premises. *Trop. Biomed.* 17:45–48.

Lee CY, Wang C. 2021. German cockroach infestations in the world and their social and economic impacts. In: Wang C, Lee CY, Rust MK, editors. *Biology and management of the German cockroach*. 1st ed. CSIRO Publishing. p. 1–16.

Leong MKH, Grace JK. 2008. Occurrence and distribution of ants (Hymenoptera: Formicidae), cockroaches (Blattodea), centipedes (Chilopoda), and wasps (Hymenoptera: Vespidae) of public health importance on the island of Oahu. *Proc. Hawaiian Entomol. Soc* 40:33–49.

Lim SP, Lee CY. 2005. Effects of juvenile hormone analogs on new reproductives and colony growth of Pharaoh ant (Hymenoptera: Formicidae). *J. Econ. Entomol.* 98:2169–2175. https://doi.org/10.1093/jee/98.6.2169

Ma Z, Fu J, Zhang Y, et al. 2024. Toxicity and behavior-altering effects of three nanomaterials on red imported fire ants and their effectiveness in combination with indoxacarb. *Insects* 15:96. https://doi.org/10.3390/insects15020096

Man LS, Lee CY. 2014. Structure-invading pest ants in healthcare facilities in Singapore. *Sociobiology* 59:241–249. https://doi.org/10.13102/sociobiology.v59i1.681

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025

Menasria T, Moussa F, El-Hamza S, et al. 2014. Bacterial load of German cockroach (*Blattella germanica*) found in hospital environment. *Pathog. Glob. Health*. 108:141–147. https://doi.org/10.1179/2047773214Y.0000000136

Mendenhall VM, Pank LF. 1980. Secondary poisoning of owls by anticoagulant rodenticides. *Wildl. Soc. Bull.* 8:311–315. https://www.jstor.org/stable/3781183

Page K, Hughes VS, Bennett GW, et al. 2006. German cockroach proteases regulate matrix metalloproteinase-9 in human bronchial epithelial cells. *Allergy* 61:988–995. https://doi.org/10.1111/j.1398-9995.2006.01103.x

Passera L. 1994. Characteristics of tramp species. In: Williams DF, editor. *Exotic ants: biology, impact, and control of introduced species*. 1st ed. Westview Press. p. 23–43.

Peacock AD, Sudd JH, Baxter AT. 1955. Studies in Pharaoh's ant, *Monomorium pharaonis* (L.). 11. Colony foundation. *Entomol. Mon. Mag.* 91:125–129.

Pitts-Singer TL, Barbour JD. 2017. Effects of residual novaluron on reproduction in alfalfa leafcutting bees, Megachile *r*rotundata F.(Megachilidae). *Pest Manag. Sci.* 73:153–159. https://doi.org/10.1002/ps.4356

Rajakumar A, Pontieri L, Li R, et al. 2024. From egg to adult: a developmental table of the ant *Monomorium pharaonis*. *J. Exp. Zool. B Mol. Dev. Evol.* 342:557–585. https://doi.org/10.1002/jez.b.23278

Schal C. 2011. Cockroaches. In: Hedges S, Moreland D, editors. *Mallis handbook of pest control*. 10th ed. G I E Pub. p. 150–291.

Schal C, DeVries ZC. 2021. Public health and veterinary importance. In: Wang C, Lee CY, Rust MK, editors. *Biology and management of the German cockroach*. 1st ed. CSIRO Publishing. p. 17–52.

Stanley MC. 2004. Review of the efficacy of baits used for ant control and eradication. Landcare Research Contract Report: LC0405/044.

Tang Q, Bourguignon T, Willenmse L, et al. 2019. Global spread of the German cockroach, *Blattella germanica*. *Biol. Invasions* 21:693–707. https://doi.org/10.1007/s10530-018-1865-2

Tay JW, Lee CY. 2014. Influences of pyriproxyfen on fecundity and reproduction of the Pharaoh ant (Hymenoptera: Formicidae). *J. Econ. Entomol.* 107:1216–1223. https://doi.org/10.1603/EC14030

Tay JW, Lee CY. 2015. Induced disturbances cause *Monomorium pharaonis* (Hymenoptera: Formicidae) nest relocation. *J. Econ. Entomol.* 108:1237–1242. https://doi.org/10.1093/jee/tov079

Tay JW, Neoh KB, Lee CY. 2014. The roles of the queen, brood, and worker castes in the colony growth dynamics of the Pharaoh ant, *Monomorium pharaonis* (Hymenoptera: Formicidae). *Myrmecol. News* 20:87–94. https://doi.org/10.25849/myrmecol.news_020:087

Vail KM. 1996. Foraging, spatial distribution, and control of the Pharaoh ant, *Monomorium pharaonis* (L.) [PhD dissertation]. University of Florida.

Vail KM, Williams DF. 1995. Pharaoh ant (Hymenoptera: Formicidae) colony development after consumption of pyriproxyfen baits. *J. Econ. Entomol.* 88:1695–1702. https://doi.org/10.1093/jee/88.6.1695

van den Brink NW, Elliott JE, Shore RF, et al. 2018. Anticoagulant rodenticides and wildlife: Concluding Remarks. In: van den Brink NW, Elliott JE, Shore RF, Rattner BA, editors. *Anticoagulant rodenticides and wildlife*. Springer. p. 379–386. https://doi.org/10.1007/978-3-319-64377-9_14

Wetterer JK. 2010. Worldwide spread of the Pharaoh ant, *Monomorium pharaonis* (Hymenoptera: Formicidae). *Myrmecol. News* 13:115–129. https://doi.org/10.25849/myrmecol.news_013:115

Yap HH, Lee CY. 1994. A preliminary study on the species composition of household ants on Penang Island. *Malaysia. J. Biosci.* 5:64–66.

Downloaded from https://academic.oup.com/jee/article/118/5/2536/8238419 by University of California, Riverside user on 27 October 2025