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Research

Stored-Product

Mobility of Phosphine-Susceptible and -Resistant *Rhyzopertha dominica* (Coleoptera: Bostrichidae) and *Tribolium castaneum* (Coleoptera: Tenebrionidae) After Exposure to Controlled Release Materials With Existing and Novel Active Ingredients

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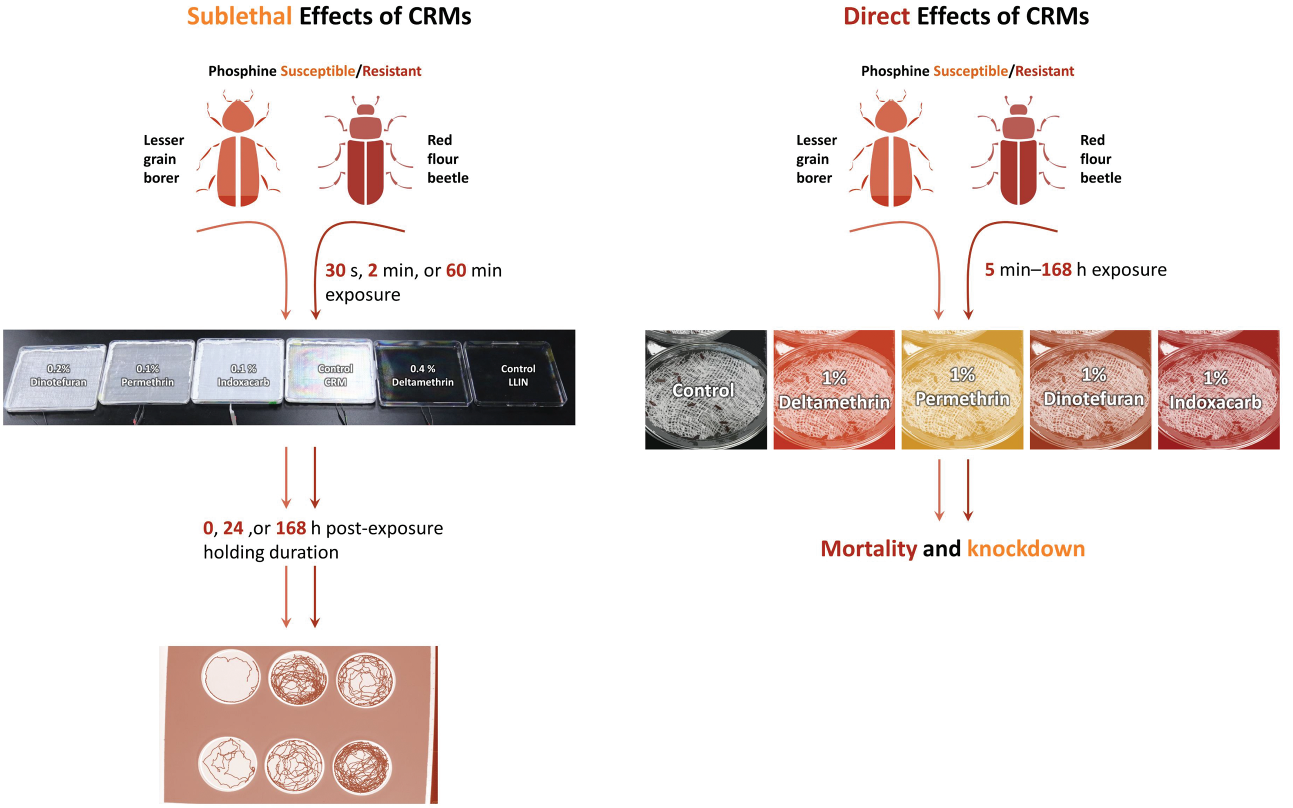
Abstract

There is interest in developing controlled release materials (CRMs) with novel modes of action to improve resistance management. Long-lasting insecticide-incorporated netting (LLIN) with deltamethrin has been effectively used against stored-product pests. Here, we evaluated the efficacy of different CRMs (LLIN or packaging) with each of four active ingredients (AI) (deltamethrin, permethrin, indoxacarb, and dinotefuran) and compared them to control CRMs in reducing movement and increasing mortality of phosphine-susceptible and -resistant *Rhyzopertha dominica* and *Tribolium castaneum*. Adults were exposed for 0.5, 2, or 60 min, and movement was assessed immediately or after 24, or 168 h using video-tracking and Ethovision software. We recorded total distance and velocity traveled by adults. Finally, we tested higher rates of each AI on surrogate netting material (e.g., standardized-sized cheesecloth) and varied exposure time to obtain median lethal time (LT_{50}) for each compound and susceptibility. Exposure to LLIN with deltamethrin significantly reduced the movement of both species compared to the other CRMs regardless of their susceptibility to phosphine. Deltamethrin was the most effective AI for both species, while dinotefuran and indoxacarb were the least effective for *R. dominica* and *T. castaneum* adults, respectively. Most AIs resulted in appreciable and approximately equivalent mortality at higher concentrations among phosphine-susceptible and -resistant strains. Our results demonstrate that CRMs can be an additional approach to combat phosphine-resistant populations of stored product insects around food facilities. Other compounds such as permethrin, dinotefuran, and indoxacarb are also effective against phosphine-resistant populations of these key stored product insects except indoxacarb for *T. castaneum*.

Key words: insecticide netting, stored products, phosphine resistance, red flour beetle, lesser grain borer

Graphical Abstract



Stored products include cereals grains, legumes, and other durable or processed commodities. Together, these products significantly contribute a high value to the economy as well as to global food security. In the United States, the production for rice, wheat, and corn alone represents a value of $62 billion USD (USDA-NASS 2019). As commodities move along the supply chain from farms to end consumers, insects may readily attack at each link. Insect damage causes economic losses by reducing the quality and quantity of food for human consumption. Annually, 2–50% of commodities are lost after harvest from insect infestation (Davis 1991), translating to over $100 billion in losses of food products globally (Wacker 2018). As a result of insect contamination, stored products may act as allergens or health hazards if not managed appropriately after harvest (Hubert et al. 2018). Therefore, it is critical to develop effective pest management strategies to mitigate losses.

The most common pest management tactic after harvest is fumigation in bulk storage of commodities. Historically, methyl bromide and phosphine were the most common fumigants used, but the former was phased out of usage in most applications due to its deleterious role in depleting the ozone layer in the atmosphere (Fields and White 2002). Phosphine is still widely used as the main fumigant after harvest but has experienced increasing problems with its use. For example, there has been a dramatic rise in phosphine-resistant populations of at least six stored product taxa around the world (Nayak et al. 2020). Furthermore, there has been a growing demand for low or no insecticide residues by consumers even after harvest (Batte et al. 2007). As a result, there has been an increasing push to diversify integrated pest management (IPM) programs for stored products.

Stored product insects regularly are moved around food facilities through two primary methods. First, stored product insects may be moved among facilities or different parts within a facility through human-mediated movement. This may periodically happen when receiving new commodities at a facility, when moving commodities to different parts of a facility internally, or when insects move independently in a facility after storage of new commodities (Campbell et al. 2002). Secondly, stored product insects may immigrate into food facilities from the surrounding landscape. It is well-known that stored product insects have wild refugia and hosts in the landscape, even far away from food facilities such as in prairie ecosystems (Jia et al. 2008). Additionally, in wooded and open sites, stored product insects such as the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae), may move 261–375 m in a short period (Mahroof et al. 2010) and the larger grain borer, *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae), is known for switching between wooded habitats and grain stores (Quellhorst et al. 2021). Indeed, others have called for insect movement to be explicitly considered in IPM programs (Jian 2019).

One way to prevent insect movement both by insects within a food facility and those immigrating from the landscape may be to use controlled release materials (CRMs). CRMs may be defined as formulated polymer matrices containing active ingredients (AIs, such as insecticides) that are designed to be effective for specific periods of time under specified conditions by affecting insect pests in one or all of the following ways: reducing fecundity, knocking down, and/or causing mortality (Calvert and Billingham 1979, Limm and Hollifield 1996, Focke and Van Pareen 2011, Gesta et al. 2015). A subset of CRMs is known as long-lasting insecticide-incorporated netting (LLIN), which have historically been used as bed nets to prevent the spread of arthropod-borne disease in tropical regions (Alonso et al. 1991, Barlow et al. 2001). More recently, however, LLINs are assessed in preharvest (Kuhar et al. 2017, Bergh and Quinn 2018) and postharvest agriculture (Morrison et al. 2018, Rumbos et al. 2018). LLINs have the added benefit of allowing airflow because of their larger mesh size, enabling them to be used on vents and in other areas where food dust accumulation may otherwise be a problem. Another subset of CRMs is insecticide-incorporated packaging, which acts as a barrier to insect immigration on a smaller scale (Paudyal et al. 2017, Scheff et al. 2021). The expanded use of CRMs may provide an additional tactic to diversify postharvest IPM programs.

Over the last four years, the efficacy of a deltamethrin-incorporated LLIN has been intensively investigated for both sublethal and lethal effects. Prior work has found that exposure to LLIN by multiple stored product insects resulted in movement that is reduced by 50–75%, while dispersal was reduced by 95–100% compared to controls (Morrison et al. 2018, Wilkins et al. 2020). Direct lethality for five out of eight stored product species ranged from 75% to 93% after a 5-minute exposure (Scheff et al. 2020), while multiple exposures to netting resulted in the same lethality as a single longer exposure (Gerken et al. 2021). LLIN has been shown to be effective for over a year (Scheff et al. 2020), while their deployment in pilot-scale warehouses, regardless of specific method, resulted in 93% fewer insects dispersing to a commodity as well as 99% fewer progeny produced after 6 weeks compared to a control (Wilkins et al. 2021). Thus, LLIN has exhibited great promise for diversifying IPM programs for stored products.

However, despite the positive results, one concern about using LLIN is its role in the resistance management of stored product insects at food facilities, including whether its use may promote resistance by stored product insects to its primary active ingredient, deltamethrin, and whether LLIN could be effective at controlling phosphine-resistant insects. There has been a dearth of studies testing CRMs with different AIs possessing other modes of action. In Greece, there have been tests using Carifend (BASF, Ludwigshafen, Germany), an LLIN that uses α-cypermethrin and which has shown good efficacy in the laboratory and field (Rumbos et al. 2018, Athanassiou et al. 2019, Paloukas et al. 2020). A recent study tested silica-coated nets for efficacy against adults of the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) and larvae of the confused flour beetle, *Tribolium confusum* du Val (Coleoptera: Tenebrionidae), and found that while mortality reached 100% for weevils, it never rose above 34% for the larvae (Agrafioti et al. 2020). Anaclerio et al. (2018), on the other hand, tested a permethrin-incorporated netting for control of *S. oryzae* and found it induced 98% mortality. However, most of these studies have assessed compounds with similar modes of action, and no study has assessed whether LLINs are effective at preventing movement by or inducing mortality of phosphine-resistant populations of stored product insects. Consequently, there is both a critical need to assess CRMs with new AIs, as well as assess how existing and new AIs may be effective against phosphine-resistant stored product insects. In particular, the incorporation of CRMs with novel modes of action might improve resistance management and significantly reduce the risk of phosphine-resistant populations at food facilities by providing multiple barriers to infestations.

Potential alternative AIs for consideration in CRMs include permethrin, dinotefuran, and indoxacarb. Permethrin is a type 1 pyrethroid, that has been shown to be less irritant than the currently used type 2 pyrethroid deltamethrin for operators when yarns are produced, and also when handled by rice mill warehouse workers in tropical climates such as Thailand (Bingham G.V. unpublished data), but may still provide the desired quick knockdown. Dinotefuran is an agonist against nicotinic acetylcholine receptors, and has been found to be highly effective against six species of stored product insects (Arthur and Fontenot 2013). Indoxacarb is a reduced-risk oxadiazine that has also been shown to have some efficacy against stored product insects (Khan 2020). Each of these may be good candidates for inclusion in CRMs to diversify the options of AIs available to stakeholders at food facilities.

Two critical and cosmopolitan stored product pest species for assessing LLIN-based management strategies are the red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae), and *R. dominica*, because phosphine-resistant populations for both are widespread in the United States and elsewhere (Afful et al. 2018, Nayak et al. 2020). These two species represent highly mobile species capable of dispersing far (Campbell et al. 2002, Mahroof et al. 2010), as well as two extremes in other life history parameters among stored product insects (Hagstrum and Subramanyam 2006). *R. dominica* is a pest that feeds and develops on kernels internally as larvae (Hagstrum and Subramanyam 2006, Morrison et al. 2020); it is also a strong flier (Edde and Phillips 2006) and long-distance disperser (Mahroof et al. 2010). By contrast, *T. castaneum* is an important pest of processing facilities, which feeds externally on damaged or broken grain (Hagstrum and Subramanyam 2006), is a strong walker (Morrison et al. 2019), but a comparatively weak flier that is largely confined to food facilities and areas around where grain is handled (Drury et al. 2009, Ridley et al. 2011). Thus, our aims in the current study were to: 1) evaluate the efficacy of different CRMs with alternative AIs, including a high concentration of deltamethrin, indoxacarb, permethrin, or dinotefuran mimicking that found in commercially available CRMs compared to untreated control CRMs using movement of *R. dominica* and *T. castaneum* as a surrogate measure of efficacy, and 2) determine the efficacy of the CRMs against phosphine-susceptible and resistant strains of *R. dominica* and *T. castaneum*.

Materials and Methods

Experimental Insects

Phosphine-susceptible and -resistant strains of *T. castaneum* and *R. dominica* were used in this study. For both, four to eight-week-old adults were used. Cultures of susceptible strains of *T. castaneum* and *R. dominica* have been maintained in the laboratory since 1958 and 1972, respectively, at the USDA Center for Grain Animal Health Research in Manhattan, KS. Resistant strains of both species were derived from field populations collected in Enid, Garfield County, Oklahoma, USA in 2009. The resistance ratios for *R. dominica* and *T. castaneum* were 1,519 and 119 based on LC_{99} (Opit et al. 2012). *T. castaneum* was reared on a mixture of 95% unbleached, organic flour and 5% brewer's yeast, while *R. dominica* was reared on organic whole wheat. All the strains were maintained at 25–27.5°C, 65% RH, and a photoperiod of 14:10 or 16:8 (L:D) h.

Treatments

Two different CRMs were used, including insecticide netting and insecticide-incorporated packaging. In total, there were six treatments, including packaging (Vestergaard SA., Lausanne, Switzerland) with either 0.1% (w/w AI) indoxacarb, 0.1% permethrin, or 0.2% dinotefuran, or no AI but identical physical properties, and long-lasting insecticide-incorporated polyethylene netting (2 × 2 mm mesh, Vestergaard SA., Lausanne, Switzerland) with 0.4% deltamethrin, or control netting without insecticide but otherwise identical in physical properties. These were used with the movement assay.

In order to evaluate whether the CRMs were more effective at higher concentrations of each AI, we used cheesecloth (100 % cotton, Loins Services, Inc. Charlotte, NC) as a common CRM surrogate material sprayed with technical grade AIs dissolved in acetone. We prepared solutions (containing 1% of each AI above) with each technical grade insecticide in acetone (Supp Table 1 [online only]) and sprayed 2 ml of each insecticide solution uniformly on glass Petri dishes (5 cm diameter) containing a 4.8 cm diameter piece cheesecloth by using an artist's airbrush sprayer (Badger 100 series, Badger Corporation, Franklin Park, IL) which is commonly used in stored product insect research (e.g., Arthur and Morrison 2020, Morrison et al. 2021). A 1% concentration of AI was used to approximate the far higher concentration of AI in commercially available incorporated long-lasting materials compared to the concentration typically used in direct spray applications. The insecticide-treated cheesecloth was allowed to dry at room temperature overnight (~18 h) inside a fume hood

Sublethal Movement Assay for CRMs

Mixed-sex adult beetles were exposed to the CRMs mentioned above. Cohorts of 5–20 adults were exposed for 0.5, 2, or 60-min interval on CRMs affixed to a 24 × 24 cm^2 Petri dish in the laboratory. After exposure, effects of the insecticides on adults were assessed either immediately or after being held for 24 or 168 h in Petri dishes under the same environmental chamber conditions as the colonies but without supplemental food, and then assayed using the video-tracking system described below. The movement of adults was tracked in six individual Petri dishes (100 × 15 mm D: H) with a piece of filter paper (85 mm D, Grade 1, GE Healthcare, Buckinghamshire, United Kingdom) lining the bottom for 1 h using a network camera (GigE, Basler AG, Ahrenburg, Germany) affixed 80 cm above the dishes. The Petri dishes were backlit using a LED light box (42 × 30 cm W:L, LPB3, Litup, Shenzhen, China) to increase contrast and affixed in place with white foam board.

Video was streamed to a computer and processed in Ethovision (v.14.0, Noldus Inc., Leesburg, VA). The program automatically calculated the total distance moved (cm) and the mean instantaneous velocity (cm/s) over the 1-h period for each adult. An input filter was created that specific distance was only accumulated if it was less than the length of two beetles (~8 mm) per 0.03 s to avoid cursor bounce. Each adult was considered a replicate and was never used more than once. Only adults classified as alive or affected (as defined in Morrison et al. 2018) were used in the assay. Briefly, alive adults were defined as moving with normal speed and activity and able to right themselves if flipped, while affected adults exhibited sluggish or drunken movements, could not right themselves if flipped, and some or all of their limbs exhibited twitching. Dead adults were completely immobile. In total, 15 replicates were performed per treatment combination, translating to 97,200 min of video for a total of 1,620 adults tested for each species.

Time to Lethal Exposure Assay with Higher Concentrations on Cheesecloth

Cohorts of 20 mixed-sex (~1:1 M: F sex ratio) adult beetles were exposed continuously up to 96 h or 1 week in the laboratory on cheesecloth (e.g., as an absorptive CRM surrogate material) at constant conditions (27.5 ± 0.1°C, 65% RH, a photoperiod of 14:10 [L:D] h) in an environmental chamber. Exposure times were added iteratively at the same 1% concentration to yield a sufficient number of points to calculate median lethal time (LT_{50}) up to 1 week (Supp Table 2 [online only]). At each time point, the condition of insects was checked, and rated as alive, affected, or dead according to the definitions in Morrison et al. (2018) for each of the different treatments.

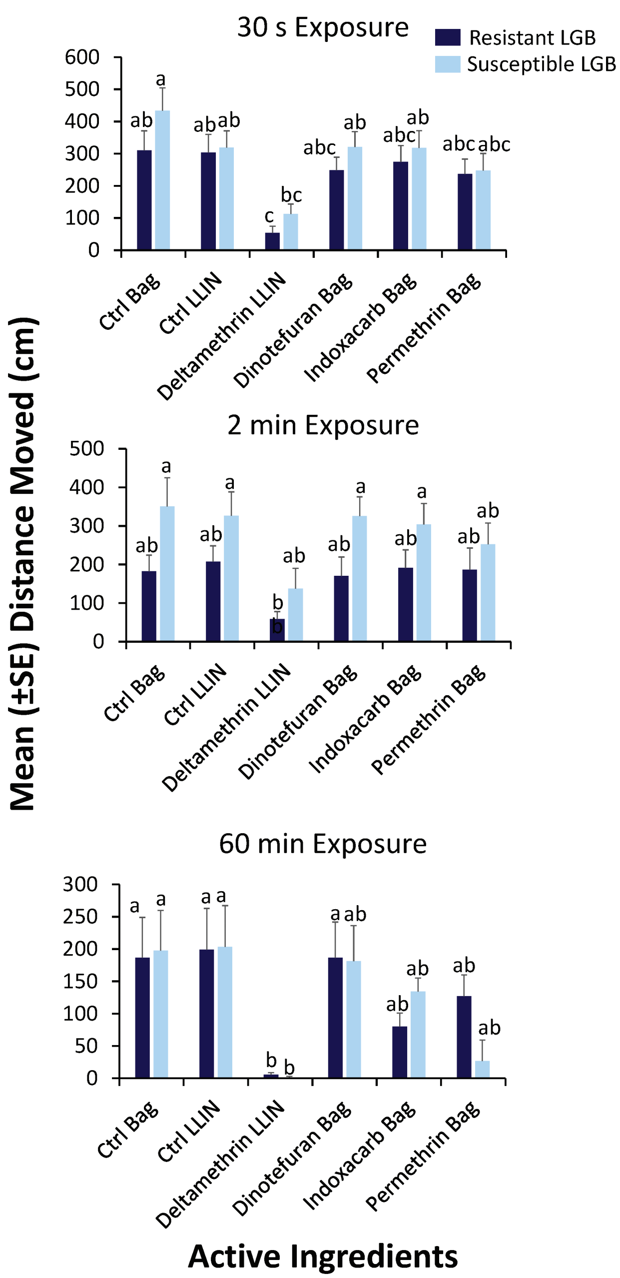


Fig. 1. Distance moved (±SE, cm) by susceptible (light shade) and resistant (dark shade) *R. dominica* (LGB) after varying exposure for 30 s (top panel), 2 min (middle panel), or 60 min (bottom panel) to different controlled release materials in the sublethal movement assay during tracking with a video camera coupled with Ethovision for 60-min periods in the laboratory. Bars with shared letters are not significantly different from each other (Tukey HSD, α = 0.05). Letters illustrate multiple comparisons for the interaction among exposure time, susceptibility, and controlled release materials. Abbreviations: Ctrl—control (no insecticide), and LLIN—long-lasting insecticide-netting.

Dead insects were completely immobile, even after prodding, and were removed from the tested arenas, but retained in analyses. There were 4 replicate cohorts for each treatment combination of insecticide, exposure, phosphine susceptibility, and species.

Statistical Analysis

First, the total distance moved, and instantaneous velocity were analyzed as separate response variables with an overall linear mixed model, using run date as a random variable. Exposure time (30 s, 2, or 60 min), CRMs (netting or packaging), AIs (control netting, 0.4% deltamethrin netting, control packaging, 0.1% indoxacarb packaging, 0.1% permethrin packaging, or 0.2% dinotefuran packaging), phosphine susceptibility of insects (susceptible or resistant), and postexposure holding duration (0, 24, or 168 h) were fixed explanatory variables. Separate models were run for each species. Each model had the same form as above. Assumptions of normality and homogeneity in variances were checked using residuals and histograms, and to correct issues, log-transformation was used.

Table 1. Summary of statistical model results for the log of the distance moved by *R. dominica* after exposure to controlled release materials in the sublethal movement assay at the Center for Grain and Animal Health Research in Manhattan, KS over a 60-min period

				Exposure time-specific models[a]					
	Overall model			60 min[b]		2 min[c]		30 s[d]	
Variable	Df	F	*P*	F	*P*	F	*P*	F	*P*
susceptibility	1	0.71	0.40	7.05	0.01	0.85	0.36	0.13	0.71
treatment	5	0.61	0.69	1.54	0.18	1.19	0.31	1.69	0.14
postexposure	2	4.79	0.01	7.07	0.00	16.54	0.00	1.75	0.18
exposuretime	2	0.14	0.87	–	–	–	–	–	–
susceptibility:treatment	5	1.17	0.32	2.14	0.06	0.78	0.56	0.56	0.73
susceptibility:postexposure	2	1.44	0.24	0.95	0.39	5.42	0.00	0.66	0.52
treatment:postexposure	10	9.84	0.00	20.66	0.00	12.58	0.00	20.15	0.00
exposuretime:postexposure	4	3.69	0.01	–	–	–	–	–	–
susceptibility:exposuretime	2	0.51	0.60	–	–	–	–	–	–
treatment:exposuretime	10	0.64	0.78	–	–	–	–	–	–
susceptibility:treatment:postexposure	10	3.54	0.00	1.89	0.04	8.62	0.00	6.18	0.00
susceptibility:treatment:exposuretime	10	0.97	0.47	–	–	–	–		–
susceptibility:exposuretime:postexposure	4	0.26	0.91	–	–	–	–		–
treatment:exposuretime:postexposure	20	5.40	0.00	–	–	–	–		–
susceptibility:treatment:exposuretime:postexposure	20	4.65	0.00	–	–	–	–		–
Residuals	1,523								

[a]Models specific to each exposure time do not include exposure time as a fixed, explanatory variable in the model structure, and thus entries containing exposure time have been left blank.

[b]Residual df is 518.

[c]Residual df is 498.

[d]Residual df is 507.

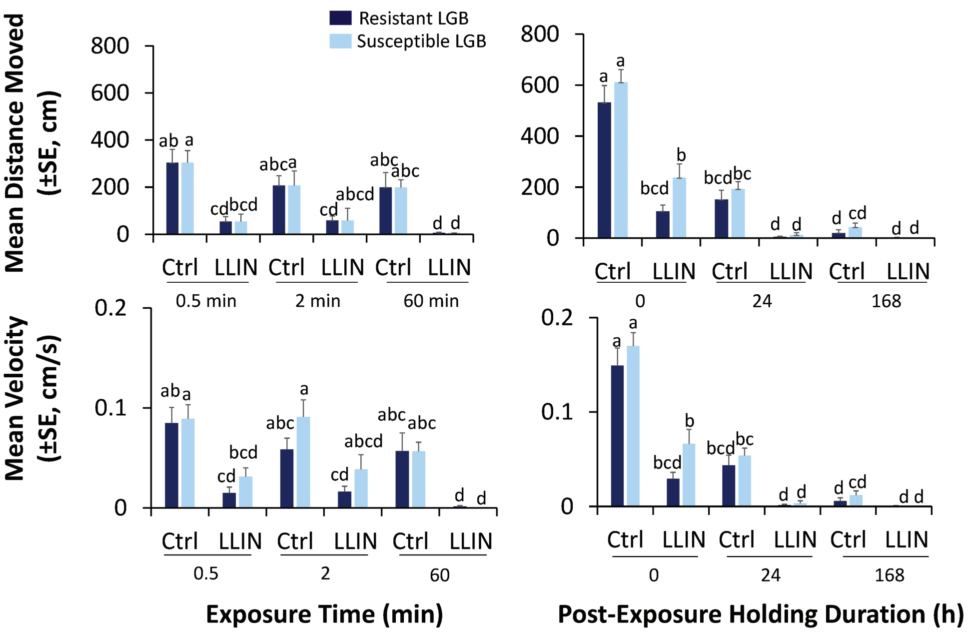


Fig. 2. Distance moved (±SE) (top panels) or velocity (±SE) (bottom panels) by susceptible (light shade) and resistant (dark shade) *R. dominica* (LGB) after exposure to long-lasting 0.4% deltamethrin-incorporated netting (LLIN) or control netting (Ctrl) in the sublethal movement assay during tracking for 60-min periods on Ethovision after various exposure times (left) and varying postexposure holding durations (right). Bars with shared letters are not significantly different from each other (Tukey HSD, $\alpha = 0.05$). Letters represent multiple comparisons between the interaction among exposure time, susceptibility, and LLIN (left column) or among postexposure holding duration, susceptibility, and LLIN (right column).

Table 2. Summary of the specific statistical model results for the log distance moved by *R. dominica* after exposure to 0.4% long lasting deltamethrin-incorporated netting in the sublethal movement assay over a 60-min period

	Overall model		
Variable	Df	F	P
susceptibility	1	5.51	0.02
TREATMENT	1	0.59	0.44
postexposure	2	0.80	0.45
Exposuretime	2	1.82	0.16
susceptibility:treatment	1	1.36	0.24
Susceptibility:postexposure	2	7.22	0.00
Treatment:postexposure	2	29.70	0.00
Exposuretime:postexposure	4	9.82	0.00
Susceptibility:exposuretime	2	4.75	0.01
Treatment:exposuretime	2	0.22	0.81
Susceptibility:treatment:postexposure	2	18.85	0.00
Susceptibility:treatment:exposuretime	2	3.76	0.02
Susceptibility:exposuretime:postexposure	4	7.10	0.00
Treatment:exposuretime:postexposure	4	14.77	0.00
Susceptibility:treatment:exposuretime:postexposure	4	11.40	0.00
Residuals	506		

Multiple comparisons were not performed with the overall models, because the goal of the overall model was to determine where significant interactions may be occurring and to parse appropriately. Movement analyses were run using R software (R Core Team 2020) and tests were considered significant at α = 0.05.

Based on significant interactions with exposure time in the overall models for distance moved and instantaneous velocity by each strain and species, we separately ran simplified models for each exposure time (30 s, 2, or 60 min). Each model used AIs (control netting: 0.4% deltamethrin netting; control packaging: 0.1% indoxacarb packaging, 0.1% permethrin packaging, or 0.2% dinotefuran packaging), phosphine susceptibility of insects (susceptible or resistant), and postexposure holding duration (0, 24, or 168 h) as fixed, explanatory variables. As a result, Tables with these summaries of these model results lack entries for any terms with exposure time (e.g., Tables 2, 4, 6, and 8). Assumptions of normality and homoscedastic variances were checked using residuals and histograms, and log-transformation were used to correct issues where appropriate. Tukey HSD test was used for multiple comparisons upon a significant result from the model through the *HSD.test* function in the *agricolae* package (de Mendiburu and Yaseen 2020). In order to assess the importance of phosphine susceptibility, 2-way, 3-way, and 4-way interactions with this factor and others were examined.

Based on the initial results of the overall model with all the CRMs showing the greatest effect with the deltamethrin-LLIN, a follow-up analysis was performed that focused solely on this CRM and its interaction with phosphine-susceptible or -resistant strains of each species. Again, the total distance moved, and instantaneous velocity were analyzed as separate response variables within a linear mixed model framework, using run date as a random variable. The following were included as fixed, explanatory variables: exposure time (30 s, 2, or 60 min), treatment (control netting, or 0.4% deltamethrin netting), and postexposure holding duration (0, 24, or 168 h). Separate models were run for each strain (e.g., phosphine-susceptible or -resistant) and species. Assumptions of normality and homogeneity in variances were checked using residuals and histograms, and were corrected with log-transformation where deviations

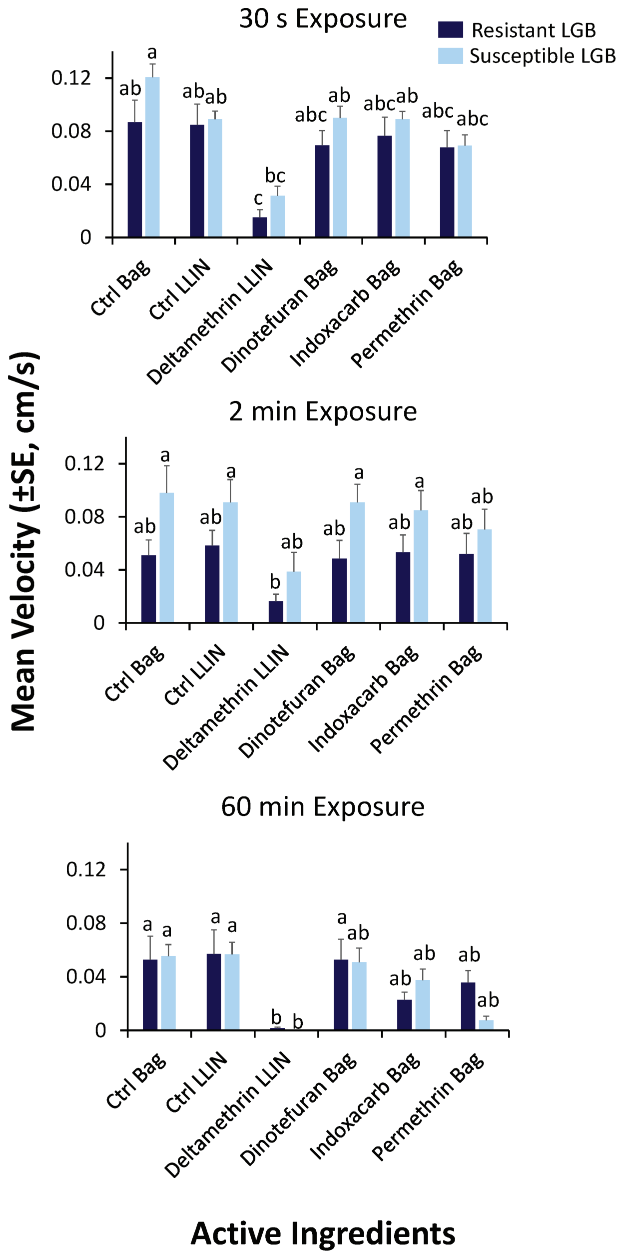


Fig. 3. Velocity (±SE, cm/s) by susceptible (light shade) and resistant (dark shade) *R. dominica* (LGB) after varying exposure for 30 s (top panel), 2 min (middle panel), or 60 min (bottom panel) to different controlled release materials in the sublethal movement assay during tracking with a video camera coupled with Ethovision for 60-min periods in the laboratory. Bars with shared letters are not significantly different from each other (Tukey HSD, α = 0.05). Letters illustrate multiple comparisons for the interaction among exposure time, susceptibility, and controlled release materials. Abbreviations: Ctrl—control (no insecticide), and LLIN—long-lasting insecticide-netting.

were detected in assumptions. Upon a significant result from the model, Tukey HSD test was used for multiple comparisons and was implemented with the function *HSD.test* from the *agricolae* package (de Mendiburu and Yaseen 2020). In order to assess the importance of phosphine susceptibility, 2-way, 3-way, and 4-way interactions with this factor and others were examined.

To determine the LT_{50}, the total insects, and the number of dead insects were broken down for each strain, species, and AI, then analyzed by probit analysis using the procedure PROC PROBIT from SAS software (SAS Institute Inc 2018).

Results

Movement Assay: *R. dominica*

Overall Distance Moved Model

Overall, neither the phosphine susceptibility nor the CRMs significantly affected the distance moved by *R. dominica* (Fig. 1; Table 1). Further, exposure time also did not significantly affect the distance moved by adults. However, postexposure holding duration significantly affected the distance moved by *R. dominica*, with adults moving only 30 and 5% of the distance after 24 and 168 h, respectively, compared to the adults moving immediately after exposure (0 h). None of the two-way interactions between the susceptibility and the other factors were significant (Table 1). However, the interaction between treatment and postexposure holding duration significantly affected the distance moved. Adults exposed to deltamethrin LLIN moved 36, 6, and 7% of the distance moved by those exposed to other CRMs with postexposure holding durations of 0 h, 24 h, and 168 h, respectively. Similarly, exposure time by postexposure holding duration also significantly affected the distance moved by *R. dominica*. The 3-way interactions among susceptibility, treatment, and postexposure holding duration as well as treatment, exposure time, and postexposure holding duration significantly affected distance moved by *R. dominica*. Finally, the 4-way interaction between all the factors also significantly affected the distance moved by *R. dominica* (Table 1).

LLIN-Specific Distance Moved Model

As the deltamethrin-incorporated insecticide netting was more effective than other CRMs, we focused our results more on this particular CRM. Overall, neither treatment nor postexposure holding duration and exposure time significantly affected the distance moved by *R. dominica* (Fig. 2; Table 2). However, the phosphine susceptibility did significantly affect the distance moved by *R. dominica*, but in most pairwise comparisons of resistant to susceptible strain, there was no significant difference in distance moved by adults (Fig. 2). The two-way interaction between the susceptibility and treatment was not significant. Similarly, the interaction between treatment and exposure time was also not significant. However, all the other two-way interactions between postexposure holding duration and the other factors significantly affected the distance moved by *R. dominica*. Similarly, the interaction between the susceptibility and exposure time was significant. Importantly, the 3-way interactions between all factors and the 4-way interaction between all the factors significantly affected the distance moved by *R. dominica* (Fig. 2; Table 2). These were mostly quantitative interactions. For example, the distance moved by *R. dominica* immediately after exposure was 3–5-fold less than those exposed to control netting, but this was to 16–30-fold less 24 h after exposure but equilibrated to no effect by 168 h later because of the general lack of movement, even in the controls (Fig. 2). Adult *R. dominica* exposed for 30 s moved 6-fold less compared to control netting exposed individuals, while those exposed for 2 min and 60 min moved 4- and 35-fold less, respectively.

Overall Velocity Model

The velocity of *R. dominica* movement was neither affected by the susceptibility nor the CRMs (Fig. 3; Table 3). Additionally, the exposure time also did not significantly affect the velocity by adults. However, postexposure holding duration significantly affected the velocity of *R. dominica* movement, with adults moving only 31 and 5% the distance after 24 and 168 h compared with adults moving immediately after exposure. The two-way interaction between the susceptibility and the other factors was not significant. However, the interaction between the treatment and postexposure holding

Table 3. Summary of statistical model results for the log of the velocity moved by *R. dominica* after exposure to controlled release materials in the sublethal movement assay at the Center for Grain and Animal Health Research in Manhattan, KS over a 60-min period

				Exposure time-specific models[a]					
	Overall model			60 min[b]		2 min[c]		30 s[d]	
Variable	Df	F	*P*	F	*P*	F	*P*	F	*P*
Susceptibility	1	1.06	0.30	6.78	0.01	0.98	0.32	0.07	0.79
Treatment	5	1.73	0.12	2.38	0.04	1.10	0.36	6.71	0.00
Postexposure	2	13.75	0.00	12.28	0.00	11.35	0.00	1.49	0.19
Exposuretime	2	0.58	0.56	–	–	–	–	–	–
Susceptibility:treatment	5	1.12	0.35	1.15	0.33	1.80	0.11	0.42	0.83
Susceptibility:postexposure	2	0.60	0.55	0.82	0.44	2.92	0.06	1.28	0.28
Treatment:postexposure	10	9.13	0.00	4.23	0.00	3.28	0.00	27.29	0.00
Exposuretime:postexposure	4	3.01	0.02	–	–	–	–	–	–
Susceptibility:exposuretime	2	1.85	0.16	–	–	–	–	–	–
Treatment:exposuretime	10	0.52	0.87	–	–	–	–	–	–
Susceptibility:treatment:postexposure	10	5.42	0.00	1.80	0.06	9.91	0.00	12.50	0.00
Susceptibility:treatment:exposuretime	10	1.47	0.14	–	–	–	–	–	–
Susceptibility:exposuretime:postexposure	4	2.51	0.04	–	–	–	–	–	–
Treatment:exposuretime:postexposure	20	6.85	0.00	–	–	–	–	–	–
Susceptibility:treatment:exposuretime:postexposure	20	6.25	0.00	–	–	–	–	–	–
Residuals	1,523								

[a]Models specific to each exposure time do not include exposure time as a fixed, explanatory variable in the model structure, and thus entries containing exposure time have been left blank.

[b]Residual df is 518.

[c]Residual df is 498.

[d]Residual df is 507.

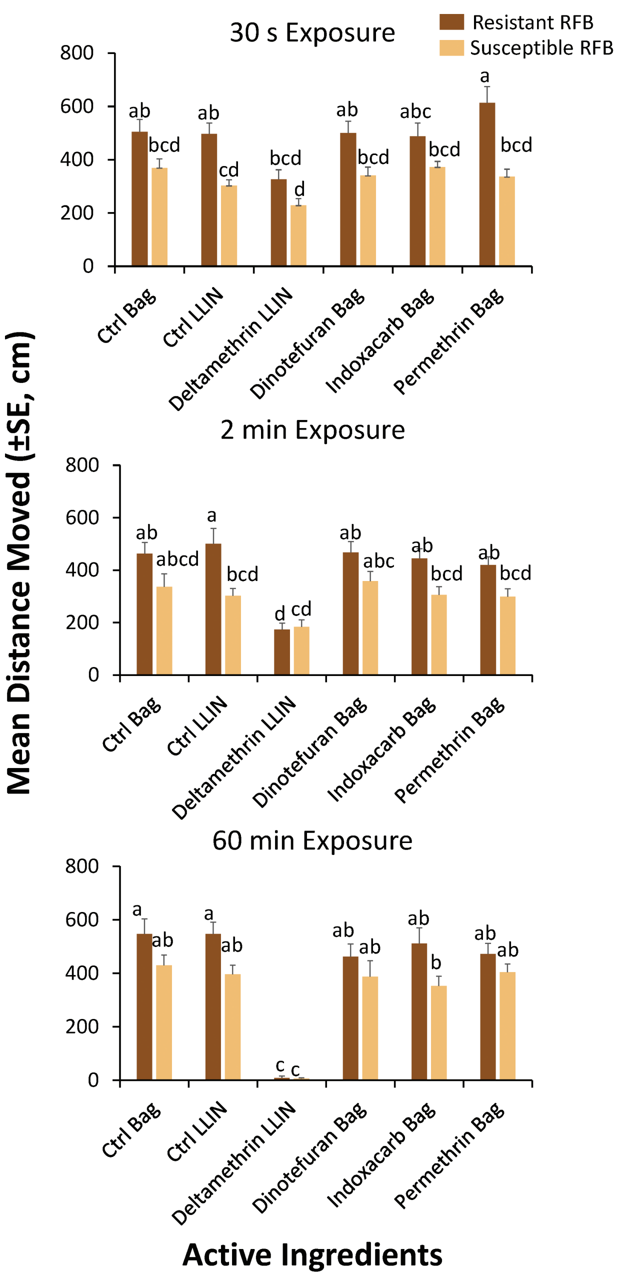


Fig. 4. Distance moved (±SE, cm) by susceptible (light shade) and resistant (dark shade) *T. castaneum* (RFB) after varying exposure for 30 s (top panel), 2 min (middle panel), or 60 min (bottom panel) to different controlled release materials in the sublethal movement assay during tracking with a video camera coupled with Ethovision for 60-min periods in the laboratory. Bars with shared letters are not significantly different from each other (Tukey HSD, α = 0.05). Letters illustrate multiple comparisons for the interaction among exposure time, susceptibility, and controlled release materials. Abbreviations: Ctrl—control (no insecticide), and LLIN—long-lasting insecticide-netting.

duration significantly affected the velocity. Adults exposed to deltamethrin LLIN moved with velocities that were only 36, 6, and 7% of the velocity moved by the adults exposed to other CRMs at 0, 24, and 168 h, respectively. Similarly, exposure time by postexposure holding duration also significantly affected the velocity of *R. dominica* movement. The 3-way interaction among the susceptibility, treatment, and the exposure time was not significant. However, the other 3-way interactions affected velocity of *R. dominica* movement, along with the 4-way interaction (Table 3).

Table 4. Summary of the specific statistical model results for the log velocity moved by *R. dominica* after exposure to 0.4% long lasting deltamethrin-incorporated netting in the sublethal movement assay over a 60-min period

	Overall model		
Variable	Df	F	*P*
Susceptibility	1	5.51	0.02
Treatment	1	0.00	0.96
Postexposure	2	2.05	0.13
Exposuretime	2	1.07	0.34
Susceptibility:treatment	1	3.35	0.07
Susceptibility:postexposure	2	4.14	0.02
Treatment:postexposure	2	6.25	0.00
Exposuretime:postexposure	4	19.04	0.00
Susceptibility:exposuretime	2	3.53	0.03
Treatment:exposuretime	2	1.08	0.34
Susceptibility:treatment:postexposure	2	14.87	0.00
Susceptibility:treatment:exposuretime	2	3.22	0.04
Susceptibility:exposuretime:postexposure	4	10.08	0.00
Treatment:exposuretime:postexposure	4	11.98	0.00
Susceptibility:treatment:exposuretime:postexposure	4	9.48	0.00
Residuals	506		

LLIN-Specific Velocity Model

The velocity of *R. dominica* movement was neither affected by postexposure holding duration nor exposure time (Fig. 2; Table 4). Further, the treatment also did not affect the velocity. However, the susceptibility did significantly affect the velocity of *R. dominica*. The two-way interaction between the susceptibility and treatment on the velocity of adults was not significant nor was the interaction between the treatment and exposure time. However, all the other two-way interactions between postexposure holding duration and the other factors significantly affected the velocity of *R. dominica*. Likewise, the interaction between the susceptibility and exposure time was significant. Importantly, all the 3-way interactions and the 4-way interactions between all the factors significantly affected the velocity of *R. dominica* (Fig. 2; Table 4). In particular, LLIN-exposed adults moved 3–5-fold to 143–177-fold slower than the control netting-exposed individuals immediately after 0–24 h, but there was no effect by 168 h later because movement was almost nonexistent (Fig. 2). Exposing *R. dominica* to LLIN for 30 s to 2 min resulted in adults moving 2–6-fold slower than the control netting exposed individuals, while those exposed for 60 min moved 57-fold less.

Movement Assay: *T. castaneum*

Overall Distance Moved Model

The distance moved by *T. castaneum* was neither affected by the phosphine susceptibility nor by the exposure time (Fig. 4; Table 5). However, postexposure holding duration significantly affected the distance moved by the adults, with them moving 94% of the distance after 24 h compared to the distance adults moved initially and 12% farther after 168 h compared to initially. Similarly, the CRMs also significantly affected the distance moved by *T. castaneum*. Adults exposed to deltamethrin-LLIN moved 2.8-, 2.7-, and 2.8-fold less than the adults exposed to packaging materials that were incorporated with dinotefuran, indoxacarb, and permethrin respectively. None of the two-way interactions between susceptibility and the other factors were significant. Further, exposure time by postexposure holding duration also did not significantly affect the distance moved

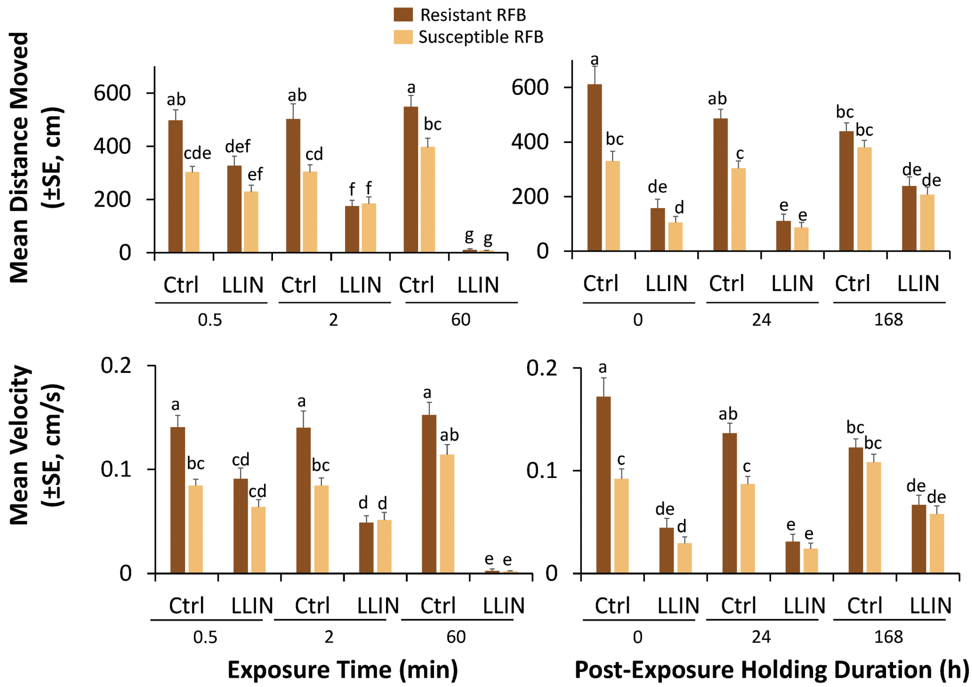


Fig. 5. Distance moved (±SE) (top panels) or velocity (±SE) (bottom panels) by susceptible (light shade) and resistant (dark shade) *T. castaneum* (RFB) after exposure to long-lasting 0.4% deltamethrin-incorporated netting (LLIN) or control netting (Ctrl) in the sublethal movement assay during tracking for 60-min periods on Ethovision after various exposure times (left) and varying postexposure holding durations (right). Bars with shared letters are not significantly different from each other (Tukey HSD, $\alpha = 0.05$). Letters represent multiple comparisons between the interaction among exposure time, susceptibility, and LLIN (left column) or among postexposure holding duration, susceptibility, and LLIN (right column).

Table 5. Summary of statistical model results for the log of the distance moved by *T. castaneum* after exposure to controlled release materials in the sublethal movement assay at the Center for Grain and Animal Health Research in Manhattan, KS over a 60-min period

				Exposure time-specific models[a]					
	Overall model			60 min[b]		2 min[c]		30 s[d]	
Variable	Df	F	P	F	P	F	P	F	P
Susceptibility	1	3.50	0.06	0.37	0.54	1.43	0.23	2.24	0.13
Treatment	5	4.71	0.00	6.72	0.00	1.54	0.18	2.19	0.05
Postexposure	2	4.47	0.01	0.67	0.51	2.83	0.06	1.32	0.27
Exposuretime	2	2.21	0.11	–	–	–	–	–	–
Susceptibility:treatment	5	1.82	0.11	2.52	0.03	0.15	0.98	0.88	0.49
Susceptibility:postexposure	2	2.27	0.10	0.10	0.91	1.50	0.22	0.80	0.45
Treatment:postexposure	10	2.58	0.00	1.09	0.36	1.42	0.17	1.26	0.25
Exposuretime:postexposure	4	1.21	0.31	–	–	–	–	–	–
Susceptibility:exposuretime	2	0.24	0.78	–	–	–	–	–	–
Treatment:exposuretime	10	2.90	0.00	–	–	–	–	–	–
Susceptibility:treatment:postexposure	10	2.64	0.00	4.24	0.00	1.35	0.20	1.70	0.08
Susceptibility:treatment:exposuretime	10	0.94	0.49	–	–	–	–	–	–
Susceptibility:exposuretime:postexposure	4	0.81	0.52	–	–	–	–	–	–
Treatment:exposuretime:postexposure	20	1.09	0.35	–	–	–	–	–	–
Susceptibility:treatment:exposuretime:postexposure	20	0.90	0.58	–	–	–	–	–	–
Residuals	1,454								

[a]Models specific to each exposure time do not include exposure time as a fixed, explanatory variable in the model structure, and thus entries containing exposure time have been left blank.
[b]Residual df is 491.
[c]Residual df is 492.
[d]Residual df is 471.

by *T. castaneum*. However, the treatment and postexposure holding duration significantly affected the distance moved. Adults exposed to deltamethrin LLIN moved 32, 25, and 49% of the distance moved by the adults exposed to other AIs at 0, 24, and 168 h, respectively. Similarly, the treatment and exposure time did significantly affect the distance moved. Adults exposed to deltamethrin LLIN moved 63, 47, and 2% of the distance moved by adults exposed to other AIs at 0.5, 2, and 60 min, respectively (Fig. 4). The interactions between

Table 6. Summary of the specific statistical model results for the log distance moved by *T. castaneum* after exposure to 0.4% long lasting deltamethrin-incorporated netting in the sublethal movement assay over a 60-min period

	Overall model		
Variable	Df	F	*P*
Susceptibility	1	0.17	0.68
Treatment	1	10.13	0.00
Postexposure	2	5.43	0.00
Exposuretime	2	2.70	0.07
Susceptibility:treatment	1	0.37	0.55
Susceptibility:postexposure	2	1.01	0.36
Treatment:postexposure	2	8.35	0.00
Exposuretime:postexposure	4	1.26	0.28
Susceptibility:exposuretime	2	0.42	0.65
Treatment:exposuretime	2	3.22	0.04
Susceptibility:treatment:postexposure	2	0.85	0.43
Susceptibility:treatment:exposuretime	2	1.13	0.32
Susceptibility:exposuretime:postexposure	4	0.12	0.97
Treatment:exposuretime:postexposure	4	3.67	0.01
Susceptibility:treatment:exposuretime:postexposure	4	0.49	0.74
Residuals	750		

susceptibility, treatment, and postexposure holding duration significantly affected the distance moved by *T. castaneum* (Table 4).

LLIN-Specific Distance Moved Model

Similar to the results for *R. dominica*, the deltamethrin incorporated insecticide netting was also more effective in reducing *T. castaneum* movement compared to other CRMs. Neither the susceptibility nor the exposure time significantly affected the distance moved by *T. castaneum* (Fig. 5; Table 6). However, the treatment affected the distance moved by adult *T. castaneum*. LLIN-exposed *T. castaneum* moved 35% of the distance moved by the adults exposed to the control netting. Further, the postexposure holding duration also significantly affected the distance moved by *T. castaneum*. At 24 h, adults exposed to deltamethrin LLIN moved 75% of the distance moved by the adults exposed at day of treatment and at 168 h postexposure holding duration moved 69% farther. The two-way and three-way interactions among susceptibility and the other factors did not significantly affect the distance moved by *T. castaneum*. The only significant interactions were between the treatment and postexposure holding duration, the treatment and exposure time, as well as the three-way interaction among treatment, exposure time, and postexposure holding duration.

Overall Velocity Model

Overall, the main effects of the phosphine susceptibility, postexposure holding duration, and exposure time did not significantly affect the velocity of *T. castaneum* movement (Fig. 6; Table 7). However, the type of CRM significantly affected the velocity moved. Adults exposed to deltamethrin LLIN, for example, moved with velocity that was 2.8-, 2.7-, and 2.8-fold less than the adults exposed to packaging materials that incorporated dinotefuran, indoxacarb, and permethrin, respectively. None of the two-way interactions between the susceptibility and the other factors were significant. Further, the exposure time by postexposure holding duration also did not significantly affect the velocity of *T. castaneum* movement. Adults exposed to deltamethrin LLIN moved with a velocity 32, 25, and 50% of the velocity moved by the adults exposed to other CRMs

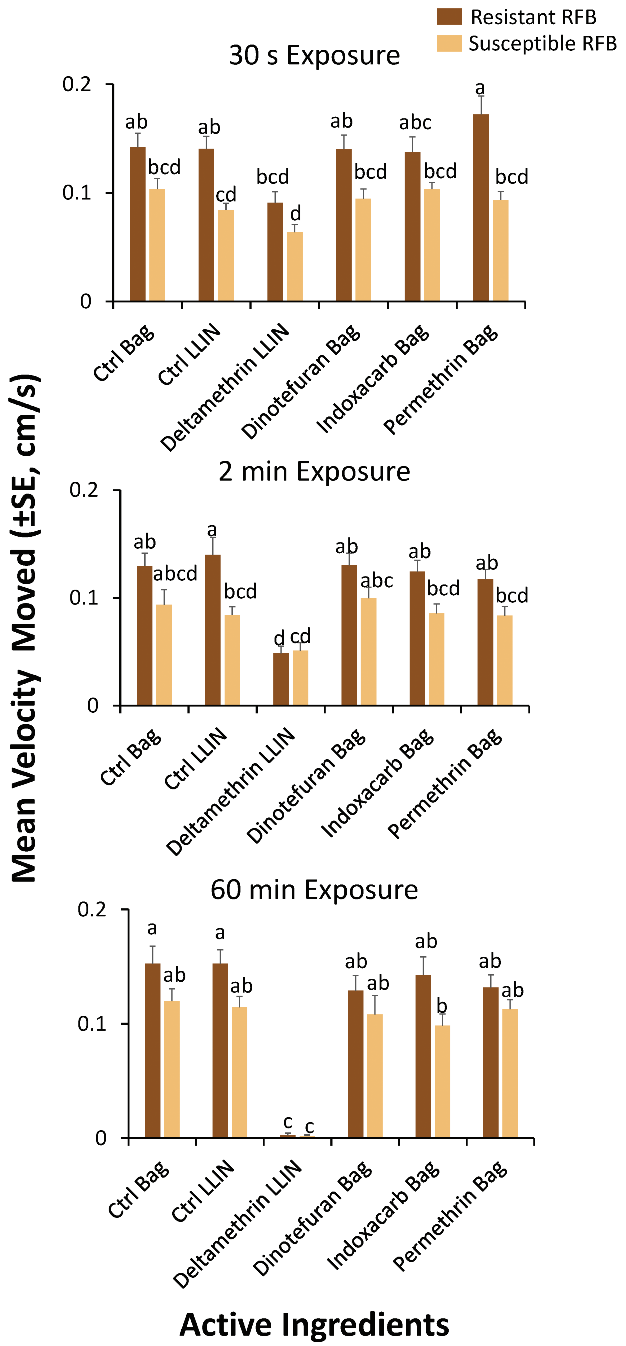


Fig. 6. Velocity (±SE, cm/s) by susceptible (light shade) and resistant (dark shade) *T. castaneum* (RFB) after varying exposure for 30 s (top panel), 2 min (middle panel), or 60 min (bottom panel) to different controlled release materials in the sublethal movement assay during tracking with a video camera coupled with Ethovision for 60-min periods in the laboratory. Bars with shared letters are not significantly different from each other (Tukey HSD, $\alpha = 0.05$). Letters illustrate multiple comparisons for the interaction among exposure time, susceptibility, and controlled release materials. Abbreviations: Ctrl—control (no insecticide), and LLIN—long-lasting insecticide-netting.

at immediately after exposure, 24, and 168 h, respectively. Similarly, the treatment and exposure time significantly affected the velocity moved. Adults exposed to deltamethrin LLIN moved with a velocity of 63, 47, and 2% of the velocity moved by adults exposed to other treatments at 0.5, 2, and 60 min, respectively (Fig. 6). The only significant 3-way interactions were among the susceptibility, treatment,

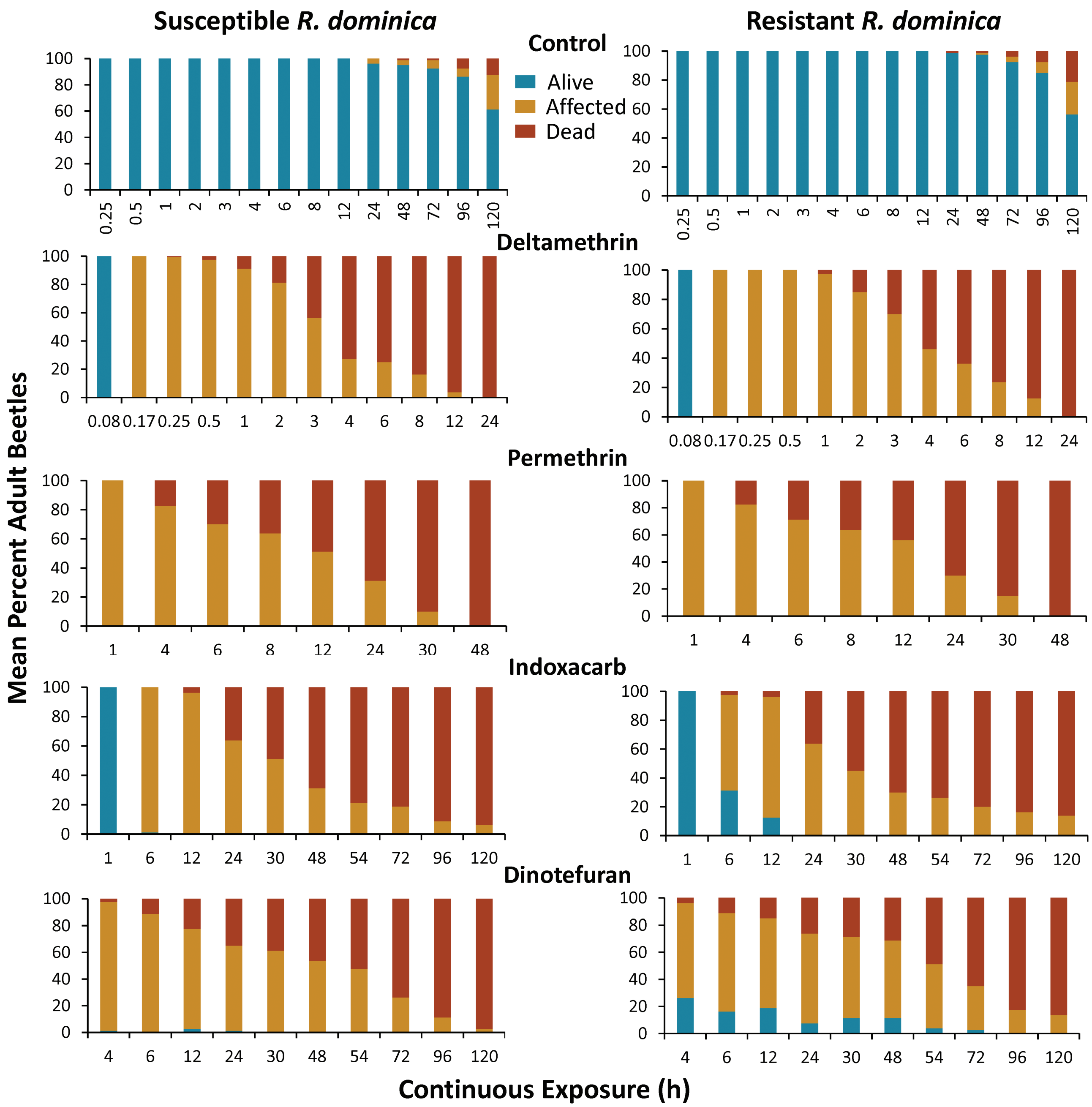


Fig. 7. The percentage of susceptible (left) and resistant (right) *R. dominica* adults that were alive (medium shade), affected (light shade), or dead (dark shade) after exposure to treated cheesecloths with 1% concentration of AI for deltamethrin, permethrin, dinotefuran, indoxacarb, or solvent only control (e.g., acetone) after various exposure times.

and postexposure holding duration, which significantly affected the velocity of *T. castaneum* movement (Table 7).

LLIN-Specific Velocity Model

Overall, neither the susceptibility nor the exposure time significantly affected the velocity of *T. castaneum* movement (Table 8), but the treatment did. Adults exposed to deltamethrin-LLIN moved 35% slower than the adults exposed to the control netting. Further, postexposure holding duration significantly affected the velocity of *T. castaneum* movement. At 24 h, adults exposed to deltamethrin LLIN moved 75% of the distance moved by the adults exposed at day of treatment and at 168 h postexposure holding duration moved 69% farther. The two-way and three-way interactions between the susceptibility and the other factors did not significantly affect *T. castaneum* movement. However, the interactions between the treatment and the postexposure holding duration significantly affected the velocity of *T. castaneum* movement. The interactions between the treatment, exposure time, and postexposure holding duration significantly affected the velocity of *T. castaneum* movement, while the 4-way interactions did as well.

Lethality Assay on Cheesecloth: *R. dominica*

The results of LT_{50} analysis indicated that the deltamethrin killed both phosphine-susceptible and -resistant *R. dominica* relatively quickly and equally (Fig. 7; Table 9). The fastest acting AI was deltamethrin with an LT_{50} value that was 9.8–17.2-fold less than

Table 7. Summary of overall statistical model results for the log of the velocity moved by *T. castaneum* after exposure to various controlled release materials with AI concentrations 0.1–0.4% in the sublethal movement assay over a 60-min period

				Exposure time-specific models[a]					
	Overall model			60 min[b]		2 min[c]		30 s[d]	
Variable	Df	F	*P*	F	*P*	F	*P*	F	*P*
Susceptibility	1	1.63	0.20	0.99	0.32	0.83	0.36	3.77	0.05
Treatment	5	5.41	0.00	5.89	0.00	2.62	0.02	2.42	0.03
Postexposure	2	2.69	0.07	0.50	0.61	1.59	0.20	2.30	0.10
exposuretime	2	2.71	0.07	–	–	–	–	–	–
Susceptibility:treatment	5	0.82	0.54	3.96	0.00	0.22	0.96	0.84	0.53
Susceptibility:postexposure	2	0.99	0.37	0.38	0.69	0.80	0.45	1.57	0.21
Treatment:postexposure	10	2.48	0.01	2.39	0.01	1.37	0.19	1.44	0.16
Exposuretime:postexposure	4	1.98	0.10	–	–	–	–	–	–
Susceptibility:exposuretime	2	0.86	0.42	–	–	–	–	–	–
Treatment:exposuretime	10	2.96	0.00	–	–	–	–	–	–
Susceptibility:treatment:postexposure	10	1.32	0.21	2.73	0.00	0.96	0.47	1.88	0.05
Susceptibility:treatment:exposuretime	10	1.53	0.12	–	–	–	–	–	–
Susceptibility:exposuretime:postexposure	4	1.64	0.16	–	–	–	–	–	–
Treatment:exposuretime:postexposure	20	1.15	0.29	–	–	–	–	–	–
Susceptibility:treatment:exposuretime:postexposure	20	0.98	0.48	–	–	–	–	–	–
Residuals	1,454								

[a]Models specific to each exposure time do not include exposure time as a fixed, explanatory variable in the model structure, and thus entries containing exposure time have been left blank.
[b]Residual df is 491.
[c]Residual df is 492.
[d]Residual df is 471.

that of dinotefuran. In each case, the LT_{50} value was separated by a negligible amount for the phosphine-susceptible and -resistant strains of *R. dominica*. Based on the LT_{50} values of the four AIs against *R. dominica*, deltamethrin showed the highest efficacy followed by permethrin, indoxacarb, and dinotefuran. The control mortality was 7.5% at 96 h for both the phosphine-susceptible and -resistant strains of *R. dominica* (Fig. 7).

Lethality Assay on Cheesecloth: *T. castaneum*

Similar to the results for *R. dominica*, LT_{50} analysis indicated that the deltamethrin killed both the phosphine-susceptible and resistant *T. castaneum* relatively quickly and equally (Fig. 8; Table 10). The fastest acting AI was deltamethrin with an LT_{50} that was 16.1–23.3-fold less than dinotefuran. Based on the LT_{50} values of the four AIs against *T. castaneum*, deltamethrin showed the highest efficacy followed by permethrin, dinotefuran, and indoxacarb. *T. castaneum* was not susceptible to indoxacarb, because the mortality caused by the indoxacarb at 168 h for both phosphine-susceptible and -resistant strains of *T. castaneum* was only 7.5%. Thus, we didn't calculate an LT_{50} for indoxacarb. The control mortality was 1.25% for phosphine-susceptible and 0% for resistant strain of *T. castaneum* at 168 h (Fig. 8).

Discussion

This is the first study to assess the mobility of phosphine-susceptible and -resistant strains of *R. dominica* and *T. castaneum* and after exposure to LLIN and other CRMs with different AIs. Overall, we demonstrated that deltamethrin LLIN is the most effective CRM among those tested in this study at reducing the movement of *R. dominica* and *T. castaneum*, but other CRMs were also effective at inducing mortality at higher concentrations. Further, we found deltamethrin and other CRMs at higher concentrations successfully and equally affected phosphine-susceptible and -resistant strains for both species. Additionally, we found that deltamethrin caused mortality faster than permethrin, dinotefuran, and indoxacarb regardless of phosphine susceptibility.

We found that the distance moved and velocity was reduced for both phosphine-susceptible and -resistant *R. dominica* and *T. castaneum* after exposure to the AIs tested in the study. These are the crucial movement variables and they mediate immigration into food facilities, foraging, mating, egg-laying, and dispersal (Jian 2019). Indirect toxicity was observed through reduced movement, especially when exposed to deltamethrin LLIN. Further, the sublethal effects from LLIN exposure for both species were present immediately after exposure and lasted up to 168 h. This indicates that exposure to deltamethrin LLIN has both immediate and lasting effects on the movement of both species regardless of their susceptibility to phosphine. Previous work has reported that deltamethrin-incorporated LLIN was successful at reducing the movement of *R. dominica* and *T. castaneum* (Morrison et al. 2018) and other life stages in the laboratory and field (Wilkins et al. 2020, 2021), but had not determined how it may change with phosphine susceptibility. We found that deltamethrin LLIN was effective at reducing movement in highly phosphine-resistant strains of *R. dominica* and *T. castaneum* from Enid, Oklahoma (Opit et al. 2012). Thus, along with other work demonstrating the utility of pyrethroid-incorporated netting to manage stored product insects (Morrison et al. 2018, Rumbos et al. 2018, Paloukas et al. 2020), our data suggests LLIN is a valuable additional tool to manage phosphine-resistant populations of stored product insects.

At low concentrations (e.g., 0.1–0.2%), the alternative CRMs did not significantly affect the movement or mortality of either *R. dominica* or *T. castaneum*, but the movement and mortality of both species were dramatically affected at higher concentrations (e.g., 0.4–1%) when tested on a surrogate CRM. In this study, two

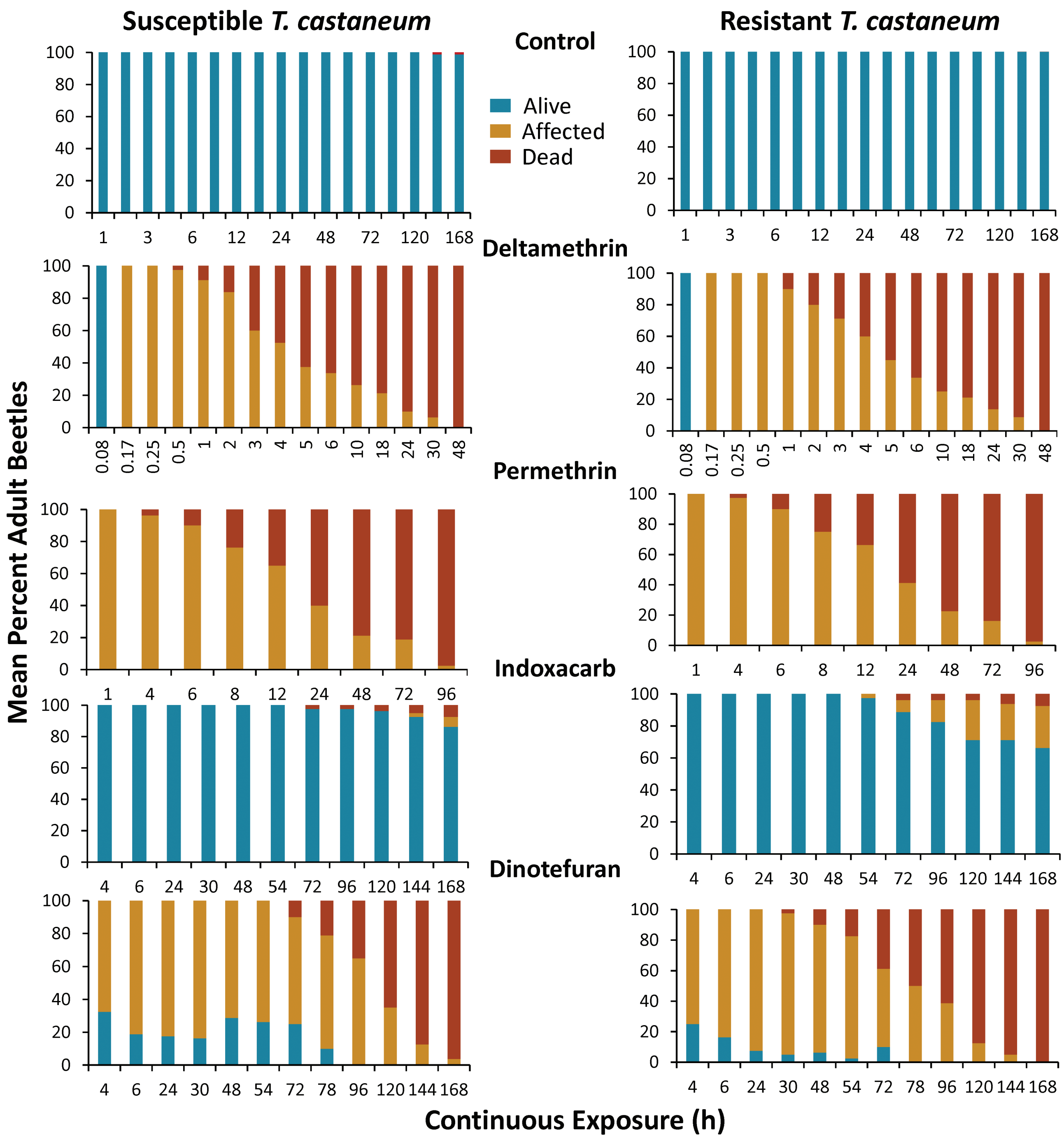


Fig. 8. The percentage of susceptible (left) and resistant (right) *T. castaneum* adults that were alive (medium shade), affected (light shade), or dead (dark shade) after exposure to treated cheesecloth with various 1% concentration of AI for deltamethrin, permethrin, dinotefuran, indoxacarb, or solvent only control (e.g., acetone) after various exposure times.

different types of CRMs were used: netting and packaging materials. AIs included deltamethrin, permethrin, indoxacarb, and dinotefuran. We observed that deltamethrin-incorporated LLIN was the most effective, followed by permethrin for both species and they were equally effective for both phosphine-susceptible and -resistant strains. While this was followed in order by indoxacarb and dinotefuran in effectiveness for *R. dominica*, the order was reversed for *T. castaneum*. Insecticide efficacies may depend on various factors like environmental, biological, and physical factors including specific insecticide and formulation, modes of action, actual application rate (Arthur et al. 2020), the time interval insects are exposed to the pesticide (Morrison et al. 2021), target insect species (Arthur and Morrison 2020), and surface substrate (Zettler and Arthur 2000). Deltamethrin-incorporated LLIN, the most effective CRM, used a label rate of 0.4%, which was twice as high as the other CRMs with other AIs. In order to produce a fair comparison and assess a higher concentration, the same surrogate substrate (cheesecloth) and the same concentration (1%) was used. When this was performed, we found that continuous exposure to many of the alternative AIs were suitable alternatives to deltamethrin at inducing mortality in

R. dominica and *T. castaneum*, resulting in equally low LT_{50} in both phosphine-susceptible and -resistant insects. It is interesting to note that in commercially labeled LLIN, the deltamethrin concentration is 4,000 ppm, which is a far higher rate than what would typically be used in a residual or grain protectant spray with the same compound (Arthur and Morrison 2020); this was the partial justification for using a higher rate of AIs in this study. We found that *T. castaneum* was not susceptible to indoxacarb, causing only 7.5% mortality by 168 h. Our results were similar to Daglish and Nayak (2012), which showed indoxacarb was effective as a grain protectant against *R. dominica* but not against *T. castaneum* even at a high dose. Overall, permethrin, dinotefuran, and to a lesser extent, indoxacarb, appear suitable for inclusion in CRMs targeting stored product insects.

Rotation of insecticides with different modes of action is required for the successful stewardship of insecticidal products, and this is required to limit the development of insecticide-resistance at food facilities. Relying on a single active ingredient or mode of action will increase the probability of developing resistant populations over time. In this study, deltamethrin and permethrin are pyrethroids that act on the insect's central nervous system by altering the gating kinetics of voltage-gated sodium channels (Soderlund 2010). When insects are exposed to these two compounds, it typically causes tremors, involuntary extremity movements, and reduced coordination, which results in insect mortality (Narahashi 1971). Dinotefuran is a neonicotinoid that acts agonistically on the nicotinic acetylcholine receptor, disrupting synapses in the central nervous system (Tomizawa and Yamamoto 1993), while indoxacarb is an oxadiazine that blocks neuronal sodium channels in insects (Lapied et al. 2001). All the compounds tested in this study resulted in significant numbers of affected and dead insects, except for indoxacarb in the case of *T. castaneum*. It has been reported that indoxacarb bioactivates very slowly for some insects, which may cause the slow onset of symptoms (Wing et al. 2000), making it less useful for inclusion in CRMs.

The present study has provided information about important alternative compounds with a different mode of action for potential inclusion in CRMs to manage stored product insects. The inclusion of new AIs in CRMs will support appropriate resistance management programs at food facilities and their inclusion in comprehensive IPM program to prevent future resistance issues. With increasing phosphine resistance among stored product insects around the world (Zettler et al. 1989, Pimentel et al. 2009, Opit et al. 2012, Nguyen et al. 2016, Cato et al. 2017), tools that are effective against both

Table 8. Summary of the specific statistical model results for the log velocity moved by *T. castaneum* after exposure to 0.4% long lasting deltamethrin-incorporated netting in the sublethal movement assay over a 60-min period

	Overall model		
Variable	Df	F	*P*
Susceptibility	1	0.02	0.89
Treatment	1	5.87	0.02
Postexposure	2	5.68	0.00
Exposuretime	2	2.73	0.07
Susceptibility:treatment	1	0.08	0.78
Susceptibility:postexposure	2	1.74	0.18
Treatment:postexposure	2	13.11	0.00
Exposuretime:postexposure	4	2.39	0.05
Susceptibility:exposuretime	2	0.56	0.57
Treatment:exposuretime	2	2.73	0.07
Susceptibility:treatment:postexposure	2	1.19	0.30
Susceptibility:treatment:exposuretime	2	2.84	0.06
Susceptibility:exposuretime:postexposure	4	0.70	0.59
Treatment:exposuretime:postexposure	4	7.85	0.00
Susceptibility:treatment:exposuretime:postexposure	4	7.03	0.00
Residuals	750		

Table 9. Comparison of model results for time when 50% of phosphine-susceptible and –resistant *R. dominica* adults have been killed when continuously exposed to cheesecloth treated with 1% concentration of each AI and held under constant conditions (27.7 ± 0.01°C, 68.6 ± 0.05% RH) in the laboratory

Treatment	Susceptibility	LT_{50} (h)	95% CI	Slope ± SE	R^2	χ^2	df	*P*
Deltamethrin	Susceptible	3.37	3.00–3.73	1.07 ± 0.10	0.86	13.92	18	0.73
	Resistant	4.47	4.03–4.99	1.05 ± 0.06	0.95	5.62	18	0.99
Permethrin	Susceptible	11.29	9.91–12.90	0.77 ± 0.05	0.92	14.63	22	0.88
	Resistant	11.98	0.46–13.81	0.74 ± 0.04	0.94	9.42	22	0.99
Dinotefuran	Susceptible	43.98	38.56–49.97	0.88 ± 0.05	0.86	15.92	22	0.94
	Resistant	58.01	52.11–65.27	0.92 ± 0.09	0.82	15.49	22	0.84
Indoxacarb	Susceptible	31.67	27.02–35.57	0.99 ± 0.08	0.91	5.76	18	1.00
	Resistant	30.44	24.97–34.80	0.88 ± 0.12	0.76	13.22	18	0.78

Table 10. Comparison of model results for time when 50% of phosphine-susceptible and –resistant *T. castaneum* adults have been killed when continuously exposed to cheesecloth treated with 1% concentration of each AI and held under constant conditions (27.7 ± 0.01°C, 68.6 ± 0.05% RH) in the laboratory

Treatment	Susceptibility	LT_{50} (h)	95% CI	Slope ± SE	R^2	χ^2	df	*P*
Deltamethrin	Susceptible	4.46	3.81–5.13	0.64 ± 0.08	0.72	34.28	26	0.13
	Resistant	4.98	4.34–5.7	0.68 ± 0.06	0.82	22.26	26	0.67
Permethrin	Susceptible	19.18	15.88–22.78	0.64 ± 0.03	0.95	4.32	18	0.9996
	Resistant	19.19	15.96–22.73	0.65 ± 0.04	0.94	6.57	18	0.99
Dinotefuran	Susceptible	104.02	99.49–108.98	2.49 ± 0.30	0.79	21.83	18	0.24
	Resistant	80.00	75.69–84.48	1.98 ± 0.16	0.89	15.86	18	0.60

phosphine-susceptible and -resistant populations are necessary. Our study demonstrated that the use of deltamethrin-incorporated netting reduces the mobility of both *R. dominica* and *T. castaneum* adults regardless of their phosphine susceptibility, while deltamethrin and other AIs are effective against phosphine-susceptible and resistant populations of both species. A future active ingredient worth testing extensively is alpha-cypermethrin against a variety of stored product insects, as it has demonstrated promise in prior work (Rumbos et al. 2018, Athanassiou et al. 2019, Paloukas et al. 2020, Agrafioti et al. 2021). Overall, our study suggests a variety of new AIs are effective against phosphine-resistant populations of key stored product insects, and LLIN may be used as an additional tool to combat phosphine resistance.

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Supplementary Data

Supplementary data are available at *Journal of Economic Entomology* online.

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