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Research

# Mobility and Dispersal of Two Cosmopolitan Stored-Product Insects Are Adversely Affected by Long-Lasting Insecticide Netting in a Life Stage-Dependent Manner

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## Abstract

*Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) and *Trogoderma variabile* Ballion (Coleoptera: Dermestidae) are two stored-product insects that cause extensive damage to a variety of postharvest commodities. Long-lasting insecticide-incorporated netting (LLIN), commonly used to control vector-borne diseases in tropical regions, has only been recently studied in an agricultural setting. While prior research showed that LLIN was successful against stored-product beetles, little is known about differential susceptibility among stored-product insect life stages. The aim of this study was to evaluate LLIN efficacy against immature *T. castaneum* and *T. variabile* compared with adults. Movement and dispersal ability were evaluated after exposure to LLIN or an untreated, control netting. For the movement assay, video-tracking software recorded the postexposure effects of LLIN on distance traveled and velocity of the insects in 2-h trials. For the dispersal assay, insects were exposed to the netting then released into one end of a PVC pipe and allowed 48 h to disperse to a novel food patch located at the opposite end of the pipe. Our study found that movement and dispersal ability of *T. variabile* and *T. castaneum* are significantly reduced, often by multiple-fold, after LLIN exposure, with the larval stage of each species more tolerant to the insecticide netting than adults. These results indicate that LLIN is a promising tool for use in intercepting immigrating insects of different life stages in food facilities to protect stored products.

**Key words:** *Trogoderma variabile*, *Tribolium castaneum*, long-lasting insecticide netting, post-harvest, behavior

Every year, the United States produces and stores a large quantity of valuable commodities that contribute to global food security. For example, in 2017, the United States stored $1 billion USD in beans, $2 billion in rice, and $8 billion in wheat (National Agricultural Statistics Service 2019). As these commodities move through the postharvest supply chain from the farmer to the end user, they are vulnerable to insect infestation. Each link along the supply chain represents a new opportunity for insect infestation, with between 2 and 50% of commodities lost yearly after harvest (Davis 1991). This translates to roughly $100 billion worth of food products lost globally (Wacker 2018). Stored-product insect infestations are often treated with fumigation of commodities or structures. Historically, methyl bromide and phosphine have been the preferred fumigants for treating infested structures and commodities, respectively. The use of the former has been largely phased out because it was labeled an ozone-depleting substance by the Montreal Protocol (Fields and White 2002). Phosphine has remained the most common fumigant used for treating infested commodities, but resistance is becoming a worldwide problem (Huang et al. 2018, Schlipalius et al. 2018, Venkidusamy et al. 2018). Likewise, there has been increasing demand by consumers for low or no insecticide residues in the stored products throughout the postharvest supply chain (Batte et al. 2007). Since most grain and grain-based products start off free from insect infestation, avoidance of infestation by stored-product insects is a major focus of integrated pest management (IPM) programs.

A potential new IPM tactic is to use long-lasting insecticide-incorporated netting (LLIN) to serve as a barrier to prevent insect movement into food products (Morrison et al. 2018, Rumbos et al. 2018). Historically, LLIN has been used to reduce the spread of arthropod-borne diseases such as malaria by controlling mosquitoes and other vectors in tropical regions (Alonso et al. 1991). LLIN generally has multiyear efficacy in other systems and is relatively inexpensive (Martin et al. 2006). Recently, LLIN has been used as a kill mechanism in a trap (Kuhar et al. 2017) and management strategy

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in specialty crops (Fernández et al. 2017), as well as to address nuisance pest issues for homeowners (Bergh and Quinn 2018). If netting is to be used effectively as a barrier, it needs to affect insects quickly before they can move from the point of contact into the food product. Prior work evaluating efficacy against stored-product insects has found that even brief exposures to LLIN by adult red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae), and lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae), resulted in 2- to 3-fold reductions in movement as well as an inability to disperse to food patches that were in close proximity to the insects, including trivial distances of 25 cm (Morrison et al. 2018). However, it is unknown how other life stages such as immature stored-product insects respond after exposure to LLIN.

One widespread and cosmopolitan stored-product pest is *Trogoderma variabile* Ballion (Coleoptera: Dermestidae), commonly referred to as the warehouse beetle. As a secondary pest, *T. variabile* does not readily infest whole grains, but is an important pest of milled grains, processed grains, animal feed, pet food, spices, nuts, as well as animal carcasses and museum artifacts (Partida and Strong 1975, Hagstrum and Subramanyam 2006, Arthur and Kelley 2015). Adults of this species are highly mobile and can be captured in high numbers outside whenever conditions are favorable (Campbell and Arbogast 2004, McKay et al. 2017) and can also move considerable distances within a facility, even moving between floors (Campbell et al. 2002). Pheromone trap captures indicated that *T. variabile* occurrences inside and outside a food facility are influenced by seasonal changes, and less influenced by fumigation treatments (Campbell and Arbogast 2004). Moreover, a 10-yr study found that even after fumigation treatments decreased trap captures of *T. variabile*, captures of *T. variabile* quickly recovered inside a facility, suggesting that *T. variabile* readily immigrates into a facility from the outside (Gerken and Campbell 2019).

Another cosmopolitan, secondary pest in stored products is *T. castaneum* that feeds on a large variety of commodities and is an especially significant pest of flour (Hagstrum and Subramanyam 2006). Although adults tend to be less mobile than *T. variabile*, *T. castaneum* were found to move among floors within a flour mill (Semeao et al. 2013). *Tribolium castaneum* females making short or long dispersal flights have already mated multiple times, with additional male beetle encounters only increasing progeny production (Gurdasani et al. 2019). It is also known that *T. castaneum* can fly at least 300 m during a dispersal flight (Gurdasani et al. 2019). Healthy adult *T. castaneum* have been documented to walk 20–25 m in a 2-h period (Morrison et al. 2018). Prior work has found that the proportion of *T. castaneum* locating a novel food patch follows a distance-decay function, though conspecifics seem to be anemotactic (Romero et al. 2010).

While adult insects are the primary dispersers among food patches, the larvae are also capable of moving among food patches within and outside facilities in order to find favorable conditions (Campbell and Arbogast 2004), potentially spreading infestations. Some species such as *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae) undergo a wandering stage, which can result in spatially heterogenous populations (Campbell and Arbogast 2004, Mohandass et al. 2007). Larvae can cover considerable distance, with healthy immature *T. variabile* moving almost 15 m in a 2-h period, while *T. castaneum* larvae move 10 m in the same period (Morrison et al. 2019a). Developmental stages can often differ in their susceptibility to insecticides, so the stage dispersing might be differentially affected by exposure to treated surfaces. For example, wandering *P. interpunctella* larvae showed decreased adult emergence on methoprene-treated surfaces, whereas *P. interpunctella* eggs on the same surfaces showed little difference in adult emergence (Jenson et al. 2009). Additionally, stored-product psocids ranged from very susceptible to very tolerant to sulfuryl fluoride fumigation, depending on life stage (Athanassiou et al. 2012). Differences in susceptibility between larvae and adults can be due to a variety of factors including physiology and degree of contact with treated surfaces. Pupae of *Tribolium confusum* (Jacquelin du Val) (Coleoptera: Tenebrionidae) were less susceptible to the chemical than other life stages, particularly the adults and young larval stages (Saglam et al. 2013). Other species of postharvest insects, particularly *Trogoderma* spp., have larvae that were less susceptible to deltamethrin than adults because larvae are able to feed and presumably recover whereas adults typically do not feed during their short lifespan (Ghimire et al. 2017). Thus, in evaluating the benefits of using LLIN in IPM programs, it is important to also consider how immature life stages respond to LLIN exposure.

Even in cases where individual insects are not outright killed by insecticides, there may be sublethal effects on behavior that contribute to a compound successfully managing a population. These indirect effects of insecticides have been an understudied area of entomology (Guedes et al. 2017), but a comprehensive understanding of their effects is vital to determine the full effectiveness of a control tactic. Indirect effects may result in sublethal changes to an insect's migration, movement, reproduction, or other fundamental life process (Desneux et al. 2007), and may be mediated by sanitation in a food facility (Morrison et al. 2019b). For example, previous studies have shown that certain insecticides upregulate or attenuate a species' rate of reproduction by 1.8- to 2.3-fold (Kerns and Stewart 2000, Bao et al. 2008, Wang et al. 2008), while flight capacity of a species may decrease by 20–60% after sublethal exposure (Morrison et al. 2017). These effects determine the extent of future infestations and damage caused by the surviving insects. Importantly, indirect effects may be particularly relevant when considering tactics designed to stop the movement of immigrating insects between food patches such as LLIN in the postharvest environment. Therefore, the goals of this study were to evaluate the efficacy of LLIN against adult and immature *T. variabile* and as well as against immature *T. castaneum* (adult *T. castaneum* have been evaluated in an earlier study by Morrison et al. 2018) to determine whether there are life stage-specific differences in mortality, movement, and dispersal ability after exposure. Understanding how LLIN affects immatures will inform future management programs wanting to utilize this novel postharvest IPM approach.

## Materials and Methods

### Source Insects

Colonies of *T. castaneum* and *T. variabile* were obtained from the field in eastern Kansas in 2012 and 2016, respectively. They were reared in an environmental chamber under constant conditions (27.5°C, 60% RH, 14:10 [L:D] h). *Tribolium castaneum* were fed a prepared diet of 95% flour (100% organic, all-purpose, unbleached/unenriched flour) with 5% brewer's yeast, while *T. variabile* were fed ground dog food (Lamb & Chow, Purina One, St. Louis, MO) with a layer of oats and a moistened, crumpled paper towel on the surface.

To prepare immature individuals for use in each of the assays, the following procedure was performed. *Tribolium castaneum* larvae were raised on 40 g of prepared flour diet and kept in 118-ml jars. In each jar, 70–80 *T. castaneum* adults were added, then given 48 h to settle, mate, and lay eggs, before being removed using a #25 sieve (710 × 710 µm mesh, Fisher Scientific Co., Hampton, NH). Larvae were used in experiments 2.5–3 wk after removal of adults. A similar procedure was performed for *T. variabile* larvae, with 60 adults given 48 h to reproduce before being removed by a similar-sized sieve. Given that the larvae may go through supernumerary molts based on food availability and density it is difficult to determine instars, so *T. variabile* larvae were classified as large, given 7–8 wk to develop, or small, given 3–4 wk, which conforms to prior size classification schemes for *Trogoderma* spp. (Domingue et al. 2020).

To compare the relative susceptibility of larvae to adults, the effects of exposure to LLIN on mixed sex *T. variabile* adults were evaluated. Adult *T. variabile* were collected directly from colony jars immediately prior to experimentation and were no more than 7 d old. Adult *T. castaneum* were previously evaluated by Morrison et al. (2018). To facilitate comparisons with larval *T. castaneum* collected in this study, some results from that prior study for adult *T. castaneum* are included for life stage-specific comparisons but are not otherwise re-analyzed.

## Movement Assay

In order to assess mobility changes after exposure to netting, movement of adult or immature life stages for both species was tracked with a camera (GigE, Basler AG, Ahrenburg, Germany) centered and suspended 78 cm above the experimental arenas. Due to their size, small *T. variabile* larvae were unable to be video-tracked and thus are only used for the dispersal assay of this study. Prior to tracking movement, insects were exposed to LLIN (0.4% deltamethrin, D-Terrence, Vestergaard Inc., Lausanne, Switzerland) or control netting that was physically identical but lacked insecticide. Insects were exposed in a 24 × 24 cm square petri dish on a single layer of the netting for 1, 5, or 10 min. After exposure to netting, insect mobility was assessed immediately (approximately 1 min afterward), or the insects were held separately in 4 × 4 cm (H:D) plastic cups for 24, 72, or 168 h in an environmental chamber with conditions identical to those used for rearing. These postexposure durations were used to assess how quickly LLIN exposure causes behavioral effects in the insects and to determine whether changes in behavior persisted over time.

Six individuals were video-tracked simultaneously, in separate filter paper-lined (85 mm D, grade 1, GE Healthcare, Buckinghamshire, United Kingdom) petri dishes (100 × 15 mm D:H), with half consisting of control netting-exposed and half with LLIN-exposed individuals. Petri dishes and filter paper were discarded after use by each insect to avoid confounding effects from prior exposure. Average instantaneous velocity and total distance traveled over 2-h periods were calculated using Ethovision (v. 10.0.828, Noldus Inc., Leesburg, VA). At the end of this period, the health conditions of control and treated individuals were recorded as either alive, affected, or dead under a dissecting stereomicroscope (Nikon, Inc.). Affected individuals were noted as having slow, irregular, or uncoordinated movements, sometimes with twitching, and may be unable to right themselves when flipped on their back. Completely immobile individuals, even after prodding, were classified as dead. Only alive or affected individuals were used for the subsequent analysis of movement. In total, $n$ = 15 replicate individuals were tested per treatment combination for this assay, translating to a total of 360 individuals tested and 720 h of video per species and life stage.

## Dispersal Assay

To test dispersal capacity to new food patches, a dispersal apparatus was employed. Species- and life stage-specific cohorts of 20 larvae (*T. castaneum* and *T. variabile*) or adults (*T. variabile*) were exposed to LLIN or control netting for 5 min, then given 48 h to disperse across 10, 25, or 75 cm standardized sections of PVC pipe (3.175 cm ID). Similar to the movement assay, insects were held for 1 min (immediate release), 10 min, or 24 h after exposure before placing them in the dispersal apparatus. The ends of both sides of the PVC pipe were sealed with Parafilm to prevent escape. At the far end of the pipe, a hole (2.22 cm D) was drilled and centered over a glass jar (5 × 6.5 cm D:H) to create a pitfall trap design. The glass jar contained 20 g of flour, representing a novel food patch, to entice insects to disperse. At the end of the sampling period, the number of insects in the jar and the insects remaining in the PVC pipe was recorded. The condition of each insect as alive, affected or dead was also recorded. In total, $n$ = 12 replications each for *T. castaneum* larvae, *T. variabile* adults, and *T. variabile* larvae were completed per treatment combination, species, and life stage for this assay. In total, 3,600 *T. castaneum* and 3,960 *T. variabile* were tested in this experiment.

## Statistical Analyses

The total distance traveled and average instantaneous velocity from the movement assay were analyzed separately using a three-way factorial analysis of variance (ANOVA). The independent explanatory variables included the type of netting (LLIN or control), exposure time (1, 5, or 10 min), and postexposure holding duration (1 min, 24, 72, or 168 h). Separate models were used for each of the species and life stages. Residuals from each model were inspected to ensure that the assumptions of normality and homogeneity of variances were fulfilled. Where assumptions deviated, data were log-transformed, which corrected any issues. Upon a significant result from the overall model, Tukey HSD was employed for multiple comparisons. R Software was used (R Core Team 2019), and tests were considered significant at $\alpha = 0.05$, unless otherwise noted.

For the dispersal assay, the number of insects reaching the novel food source was used as the response variable in a three-way factorial ANOVA. The independent explanatory variables included the type of netting (LLIN or control), dispersal distance (10, 25, or 75 cm), and postexposure holding duration (1 min, 10 min, or 24 h). Separate models were used for each life stage and species. Residuals were inspected to ensure that assumptions were fulfilled, and data were log-transformed in cases where there was deviation. Upon a significant result from the overall model, Tukey HSD was used for multiple comparisons.

To compare the susceptibility of life stages after LLIN exposure, which may exhibit vastly different overall mobility from each other, raw response variables (distance moved, velocity, number dispersing to new patches) were transformed into a percent of total movement relative to the corresponding control netting-exposed individuals for each assay (e.g., individual LLIN-exposed response divided by control netting-exposed response multiplied by 100). In addition, the relative percent reduction in movement was calculated as 100 minus the relative percent movement to controls. This information was calculated for major variables in each experiment, as well as a global mean, parsed by life stage. To determine whether life stages varied in susceptibility, the values for larvae and adults were compared with a chi-squared test using the null hypothesis of equal movement and changes in movement between larvae and adults. A Bonferroni correction was used for the alpha threshold in these post hoc tests.

# Results

## Movement Assay: Distance Moved

Larval *T. castaneum* exposed to LLIN experienced significant reductions in distance moved compared with control-netting exposed individuals (ANOVA: $F = 69.8$; df = 1, 330; $P < 0.0001$; Fig. 1), with the former moving 3.6-fold less than the distance that controls moved (Fig. 1). Regardless of exposure time ($F = 2.55$; df = 2, 330; $P < 0.08$), LLIN-exposed *T. castaneum* larvae moved 2.7- to 10-fold less than control larvae. Postexposure holding duration also had a significant effect on the distance moved by larvae ($F = 41.9$; df = 3, 330; $P < 0.0001$). Immediately to 72 h after exposure, LLIN-exposed *T. castaneum* larvae moved 2.4- to 3.8-fold less far than the controls did. After 168 h, LLIN-exposed larvae moved 54-fold less than control larvae (Fig. 1). There was no two-way interaction between netting treatment and exposure time ($F = 0.79$; df = 2, 330; $P < 0.37$), but there was an interaction between exposure time and holding duration ($F = 9.31$; df = 3, 330; $P < 0.0001$), as well as a three-way interaction ($F = 12.2$; df = 6, 330; $P < 0.0001$), though these were likely quantitative and not qualitative.

The distance moved by *T. variabile* larvae was significantly reduced by exposure to LLIN compared with control netting (ANOVA: $F = 134$; df = 1, 336; $P < 0.0001$; Fig. 1), with LLIN-exposed larvae moving 3-fold less than the distance moved by controls. Likewise, exposure time to LLIN significantly reduced movement to a greater degree with increasing time ($F = 7.01$; df = 2, 336; $P < 0.001$); LLIN-exposed larvae moved 1.8-, 3.8-, and 6.1-fold less than the distance moved by their control netting-exposed counterparts at 1, 5, and 10 min, respectively. The movement of *T. variabile* larvae was more severely affected 24, 72, and 168 h later compared with immediately after exposure ($F = 21.8$; df = 3, 336; $P < 0.0001$). LLIN-exposed larvae moved 1.7-fold less than the distance moved by control netting-exposed individuals immediately after exposure, while they moved 3.1- to 11.6-fold less than the controls at 24, 72, and 168 h later (Fig. 1). There was a significant two-way interaction between netting type and exposure time ($F = 3.29$; df = 2, 336; $P < 0.05$), due to a change in the direction of the effect size. The two-way interaction between netting type and postexposure holding duration ($F = 1.08$; df = 3, 336; $P = 0.35$) and the three-way interaction between all variables ($F = 1.16$; df = 6, 336; $P = 0.33$) were not significant.

The distance that *T. variabile* adults moved was dramatically affected by exposure to LLIN ($F = 89.5$; df = 1, 176; $P < 0.0001$; Fig. 1) and was 9.2-fold less than the distance that the control netting-exposed adults moved. The exposure time to netting did not significantly affect the distance moved ($F = 0.08$; df = 2, 176; $P = 0.78$), demonstrating that even brief bouts of exposure were

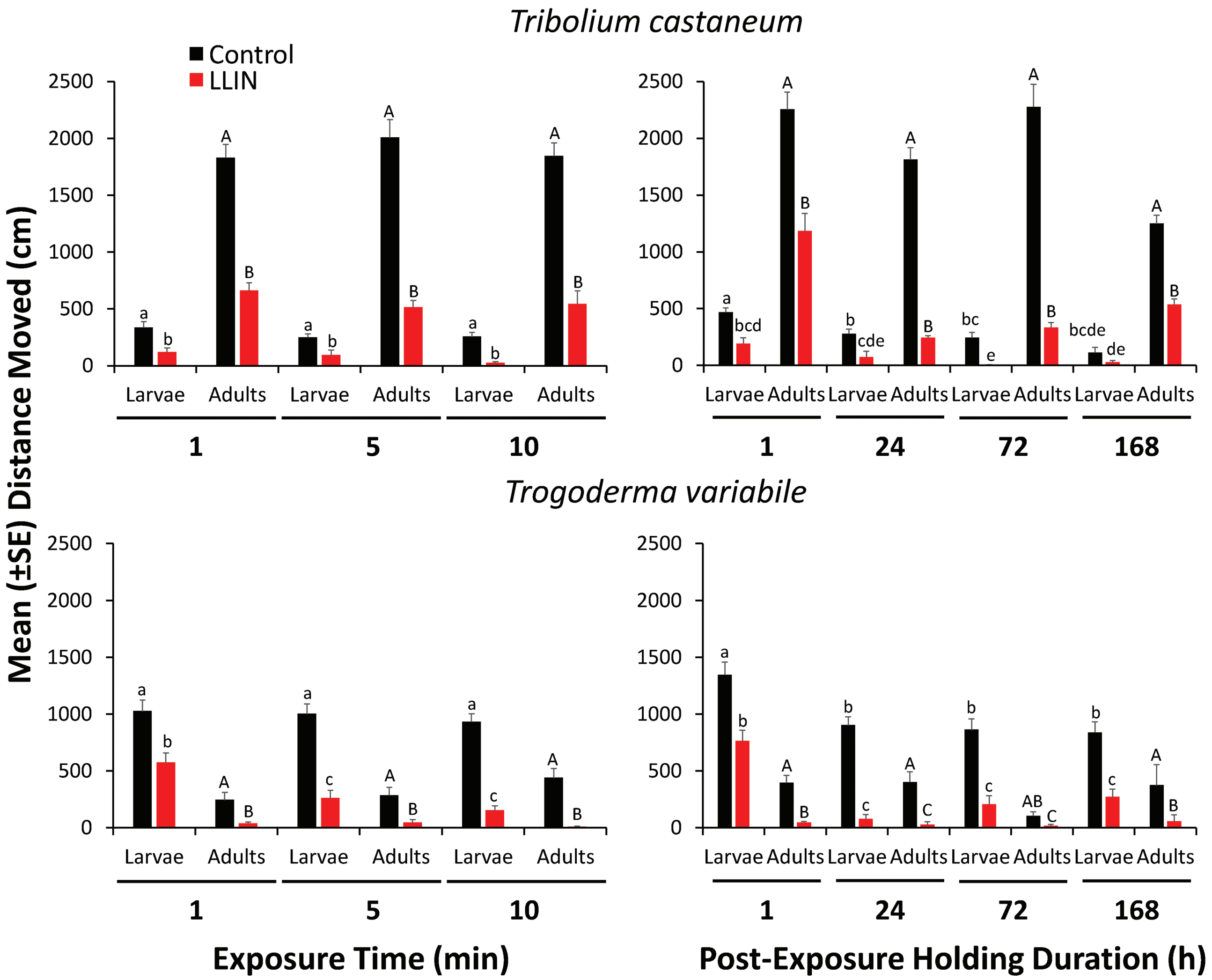

**Fig. 1.** The distance moved (± SE) by *Tribolium castaneum* (top) or *Trogoderma variabile* (bottom) adults and large larvae after varying exposure times (1–10 min, left column) and over time (1–168 h after exposure, right column) to control or long-lasting insecticide-incorporated netting during 2-h trials in the laboratory. Uppercase letters indicate pairwise comparisons among treatments for adults, while lowercase letters represent comparisons among treatments for larvae (Tukey HSD, $\alpha = 0.05$). Bars with shared letters are not significantly different from each other.

sufficient to elicit strong reductions in insect movement. At every postexposure holding duration, the movement of LLIN-exposed individuals were impaired compared with the controls ($F = 1.88$; df = 3, 176; $P = 0.13$); LLIN-exposed adults moved 6- to 14-fold less than the distance of the controls regardless of duration after exposure. None of the two-way interactions were significant (netting type × exposure time: $F = 1.80$; df = 2, 176; $P = 0.18$; netting type × postexposure holding duration: $F = 0.33$; df = 3, 176; $P = 0.81$; exposure time × postexposure holding duration: $F = 0.34$; df = 6, 176; $P = 0.91$), but the three-way interaction was significant ($F = 4.20$; df = 6, 176; $P < 0.01$).

Both LLIN-exposed *T. castaneum* larvae and adults moved 3.3-fold less than the distance that the control insects moved on average. There were no life stage-based differences in distance that insects moved by exposure time to LLIN except for 10-min exposure periods where movement of larvae was almost three times less than for adults (chi-squared test, Table 1). Importantly, the reduced movement between *T. castaneum* larvae and adults after exposure was similar, hovering at about 70% less than controls for each (Table 1). Larvae and adults moved similar distances relative to the controls immediately after exposure to LLIN, but larvae moved twice as much as adults at 24 h post-exposure, while by contrast they moved 8- to 1.6-fold less compared with adults at 72 and 168 h post-exposure, respectively (chi-squared tests, Table 2).

Overall, there was a more dramatic reduction in the distance that *T. variabile* adults moved than larvae, with adults moving almost three times less than larvae (chi-squared test, Table 1). Depending on exposure time, adults moved 1.6–8 times less than larvae after contact with LLIN (Table 1). Adults were more susceptible to LLIN immediately after exposure and 168 h later, where they moved five and two times less than larvae, respectively (chi-squared tests, Table 2).

## Movement Assay: Velocity

Larval *T. castaneum* exposed to LLIN exhibited a significantly reduced instantaneous velocity over 2-h trial periods ($F = 37.8$; df = 1, 330; $P < 0.0001$; Fig. 2), with insecticide-netting-exposed individuals moving 3.4-fold slower than controls. Exposure time of larvae on netting did not significantly affect velocity ($F = 1.89$; df = 2, 330; $P = 0.17$), indicating that in each case the velocity for LLIN-exposed larvae was 2.7- to 8.1-fold less than the controls. By contrast, the holding duration after exposure significantly affected velocity ($F = 26.0$; df = 3, 330; $P < 0.0001$), with velocity for LLIN-exposed larvae decreased by 2.3- to 55-fold compared with controls between 1 and 168 h after exposure. There was a significant two-way interaction between type of netting and exposure time ($F = 14.5$; df = 2, 330; $P < 0.001$), type of netting and postexposure holding duration ($F = 8.81$; df = 3, 330; $P < 0.0001$), and exposure time and postexposure holding duration ($F = 7.69$; df = 3, 330; $P < 0.0001$). The three-way interaction between all the variables on velocity was also significant ($F = 11.2$; df = 6, 330; $P < 0.0001$), but in each case, the LLIN-exposed individuals moved slower than controls.

The velocity of *T. variabile* larvae was significantly reduced when exposed to LLIN compared with control netting ($F = 142$; df = 1, 336; $P < 0.0001$), traveling 3-fold slower than the controls (Fig. 2). Exposure time to netting also significantly affected the velocity of larvae ($F = 7.42$; df = 2, 336; $P < 0.001$); individuals moved 1.8-fold slower than the controls after 1 min exposure to LLIN, while larvae moved 3.7- and 6.1-fold slower than controls after 5 and 10 min, respectively. The postexposure holding duration also significantly affected velocity of *T. variabile* larvae ($F = 33.2$; df = 3, 336; $P < 0.0001$). In particular, immediately after exposure to LLIN, larvae moved 1.8-fold slower than the controls, while at 24–168 h post-exposure, the velocity decreased by 3.1- to 12-fold compared with the controls. None of the interactions were significant (netting type × exposure time: $F = 3.20$; df = 2, 336; $P = 0.06$; netting type × postexposure holding duration: $F = 0.97$; df = 3, 336; $P = 0.42$; exposure time × postexposure holding duration: $F = 1.84$; df = 6, 336; $P = 0.08$; three-way interaction: $F = 0.99$; df = 6, 336; $P = 0.43$).

Similarly, the velocity of *T. variabile* adults was significantly reduced after contact with LLIN relative to control netting ($F = 94.8$; df = 1, 176; $P < 0.0001$), which traveled 10-fold slower. The exposure time to LLIN also significantly affected the velocity of adults ($F = 5.91$; df = 2, 176; $P < 0.01$). Specifically, adult velocity declined with LLIN exposure time; at 1, 5, and 10 min exposure, LLIN-exposed adults traveled 6.1-, 6.6-, and 48-fold slower than the control netting-exposed ones. The postexposure holding duration significantly affected the velocity of *T. variabile* adults ($F = 3.17$; df = 3, 176; $P < 0.05$), but regardless of time, the velocity for LLIN-exposed adults was decreased by 6- to 14-fold compared with controls. There was a significant two-way interaction between netting type and postexposure holding

**Table 1.** Relative susceptibility in distance moved by *T. castaneum* and *T. variabile* adults and larvae depending on exposure time to long-lasting insecticide-incorporated netting (LLIN) in the movement assay

| | LLIN-exposed larvae | | LLIN-exposed adults | |
|---|---|---|---|---|
| Exposure time | Relative % distance moved[a] | Relative % reduction in distance moved[b] | Relative % distance moved[a] | Relative % reduction in distance moved[b] |
| *T. castaneum* | | | | |
| 1 | 35.7a | 64.3A | 36.1a | 63.9A |
| 5 | 37.7a | 62.3A | 25.7a | 74.3A |
| 10 | 9.9b | 90.1A | 29.5a | 70.5A |
| Overall | 27.8a | 72.2A | 30.5a | 69.5A |
| *T. variabile* | | | | |
| 1 | 55.9a | 44.1B | 16.5b | 83.5A |
| 5 | 26.1a | 73.9A | 16.2a | 83.8A |
| 10 | 16.5a | 83.5A | 2.1b | 97.9A |
| Overall | 32.8a | 67.2A | 11.6b | 88.4A |

Lowercase letters represent comparisons between larvae and adults for percent distance moved compared with controls, whereas uppercase letters represent comparisons between larvae and adults in percent reduction in distance moved ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific exposure time.

[a] The distance that LLIN-exposed individuals moved as a percent of the distance moved by control netting-exposed individuals in the movement assay.

[b] The percent reduction in distance moved by LLIN-exposed individuals relative to control netting-exposed individuals in the movement assay.

**Table 2.** Relative susceptibility in distance moved by *T. castaneum* and *T. variabile* adults and larvae depending on holding duration after exposure to long-lasting insecticide-incorporated netting (LLIN) in the movement assay

| | Larvae | | Adults | |
|---|---|---|---|---|
| Postexposure holding duration | Relative % distance moved[a] | Relative % reduction in distance moved[b] | Relative % distance moved[a] | Relative % reduction in distance moved[b] |
| *T. castaneum* | | | | |
| 1 | 40.9a | 59.1A | 52.6a | 47.4A |
| 24 | 26.6a | 73.4A | 13.4b | 86.6A |
| 72 | 1.8b | 98.2A | 14.6a | 85.4A |
| 168 | 26.1b | 73.9A | 42.9a | 57.1A |
| Overall | 23.9a | 76.1A | 30.9a | 69.1A |
| *T. variabile* | | | | |
| 1 | 56.8a | 43.2B | 11.9b | 88.1A |
| 24 | 8.6a | 91.4A | 7.1a | 92.9A |
| 72 | 23.9a | 76.1A | 16.7a | 83.3A |
| 168 | 32.4a | 67.6A | 15.4b | 84.6A |
| Overall | 30.4a | 69.6A | 12.8b | 87.2A |

Lowercase letters represent comparisons between larvae and adults for percent distance moved compared with controls, while uppercase letters represent comparisons between larvae and adults in percent reduction in distance moved ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific postexposure holding duration.

[a] The distance that LLIN-exposed individuals moved as a percent of the distance moved by control netting-exposed individuals in the movement assay.

[b] The percent reduction in distance moved by LLIN-exposed individuals relative to control netting-exposed individuals in the movement assay.

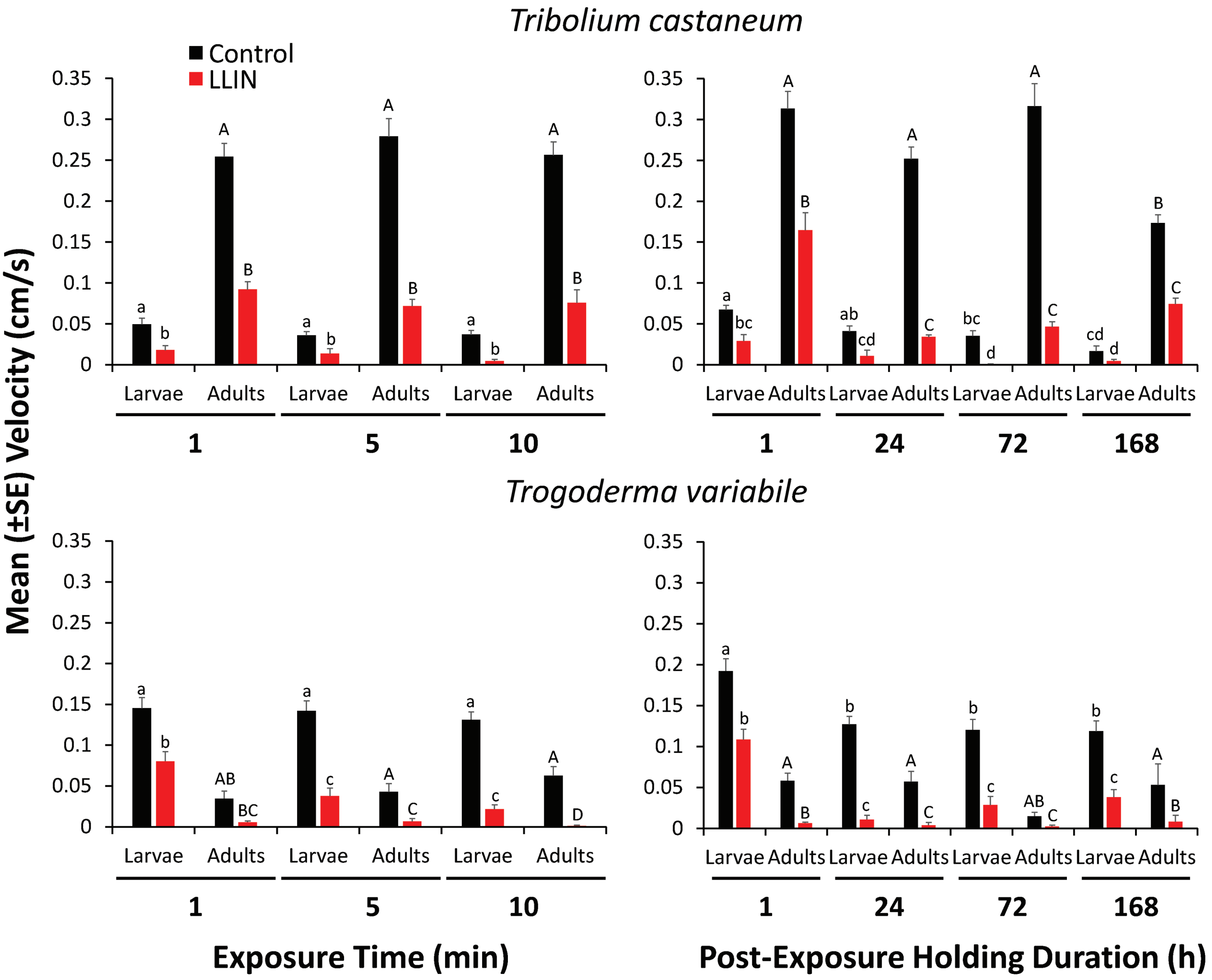

**Fig. 2.** The mean instantaneous velocity (± SE) by *Tribolium castaneum* (top) or *Trogoderma variabile* (bottom) adults and large larvae after varying exposure times (1–10 min, left column) and over time (1–168 h after exposure, right column) to control or long-lasting insecticide-incorporated netting during 2-h trials in the laboratory. Uppercase letters indicate pairwise comparisons among treatments for adults, while lowercase letters represent comparisons among treatments for larvae (Tukey HSD, $\alpha = 0.05$). Bars with shared letters are not significantly different from each other.

duration ($F = 25.6$; df = 3, 176; $P < 0.0001$), with a more pronounced decrease in velocity at 24 and 72 h than immediately or 168 h after exposure. There was also a significant netting type by exposure time interaction ($F = 17.7$; df = 2, 176; $P < 0.0001$); with a numerical, but nonsignificant decrease in velocity at 1-min exposure compared with significantly decreased velocity at 5 and 10 min. The interaction between exposure time and postexposure holding duration on velocity was not significant ($F = 2.65$; df = 6, 176; $P = 0.07$). Finally, the three-way interaction between all variables on velocity was also not significant ($F = 0.99$; df = 6, 176; $P = 0.43$).

Both *T. castaneum* larvae and adults were equally susceptible to LLIN exposure. For each life stage, the velocity was decreased by 3-fold compared with controls, with a corresponding reduction in movement of about 70% (chi-squared tests, Table 3). The life stages responded similarly to varying exposure time (Table 3), but differently for duration after exposure where larvae and adults behaved similarly 1 min and 168 h after LLIN exposure, while larvae moved two times faster than adults at 24 h after LLIN exposure and 8-fold slower at 72 h (chi-squared tests, Table 4).

*Trogoderma variabile* adults were much more susceptible to LLIN exposure than larvae, with adults moving 2.9-fold slower than larvae overall (Table 3). At 1 and 10 min of LLIN exposure, adults moved 3- and 8-fold slower than larvae, respectively (chi-squared tests, Table 3). After 1 min and 168 h after LLIN exposure, adults moved 5- and 2-fold slower than larvae, while velocity of the life stages was equivalent 24 and 72 h after exposure (Table 4).

## Dispersal Assay

Exposure to LLIN significantly affected the number of *T. castaneum* larvae reaching a novel food resource ($F = 89.9$; df = 1, 54; $P < 0.0001$; Fig. 3), with 2.2-fold fewer individuals

**Table 3.** Relative susceptibility in velocity of *T. castaneum* and *T. variabile* adults and larvae depending on exposure time to long-lasting insecticide-incorporated netting (LLIN) in the movement assay

| | LLIN-exposed larvae | | LLIN-exposed adults | |
|---|---|---|---|---|
| Exposure time | Relative % velocity[a] | Relative % reduction in velocity[b] | Relative % velocity[a] | Relative % reduction in velocity[b] |
| *T. castaneum* | | | | |
| 1 | 36.0a | 64.0A | 36.1a | 63.9A |
| 5 | 37.7a | 62.3A | 25.7a | 74.3A |
| 10 | 12.3b | 87.7A | 29.5a | 70.5A |
| Overall | 28.7a | 71.3A | 30.5a | 69.5A |
| *T. variabile* | | | | |
| 1 | 55.3a | 44.7B | 16.4b | 83.6A |
| 5 | 26.6a | 73.4A | 15.2a | 84.8A |
| 10 | 16.5a | 83.5A | 2.1b | 97.9A |
| Overall | 32.8a | 67.2A | 11.2b | 88.8A |

Lowercase letters represent comparisons between larvae and adults for relative percent velocity compared with controls, while uppercase letters represent comparisons between larvae and adults for percent reduction in velocity ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific exposure time.

[a] The velocity that LLIN-exposed individuals moved as a percent of the velocity moved by control netting-exposed individuals in the movement assay.

[b] The percent reduction in velocity moved by LLIN-exposed individuals relative to control netting-exposed individuals in the movement assay.

**Table 4.** Relative susceptibility in velocity of *T. castaneum* and *T. variabile* adults and larvae depending on holding duration after exposure to long-lasting insecticide-incorporated netting (LLIN) in the movement assay

| | Larvae | | Adults | |
|---|---|---|---|---|
| Postexposure holding duration | Relative % velocity[a] | Relative % reduction in velocity[b] | Relative % velocity[a] | Relative % reduction in velocity[b] |
| *T. castaneum* | | | | |
| 1 | 43.2a | 56.8A | 52.6a | 47.4A |
| 24 | 26.1a | 73.9A | 13.4b | 86.6A |
| 72 | 1.8b | 98.2A | 14.6a | 85.4A |
| 168 | 26.7a | 73.3A | 42.9a | 57.1A |
| Overall | 23.7a | 76.3A | 26.9a | 73.1A |
| *T. variabile* | | | | |
| 1 | 56.5a | 43.5B | 11.3b | 88.7A |
| 24 | 8.6a | 91.4A | 7.0a | 93.0A |
| 72 | 23.9a | 76.1A | 16.7a | 83.3A |
| 168 | 32.2a | 67.8A | 15.3b | 84.7A |
| Overall | 29.6a | 70.4A | 11.7b | 88.3A |

Lowercase letters represent comparisons between larvae and adults for relative percent velocity compared with controls, while uppercase letters represent comparisons between larvae and adults for percent reduction in velocity ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific postexposure holding duration.

[a] The velocity that LLIN-exposed individuals moved as a percent of the velocity moved by control netting-exposed individuals in the movement assay.

[b] The percent reduction in velocity moved by LLIN-exposed individuals relative to control netting-exposed individuals in the movement assay.

successfully dispersing after LLIN exposure compared with controls. The dispersal distance also significantly reduced dispersal of larvae ($F$ = 36.3; df = 2, 54; $P$ < 0.0001). Specifically, compared with controls, the percent of individuals dispersing was decreased by 1.3-, 2.5-, and 15-fold at 10, 25, and 75 cm, respectively. The postexposure holding duration, by comparison, did not significantly affect dispersal of *T. castaneum* larvae ($F$ = 3.01; df = 2, 54; $P$ = 0.06). There was a significant two-way interaction between type of netting and dispersal distance on the number of dispersing larvae ($F$ = 7.21; df = 2, 54; $P$ < 0.01), with equivalent numbers of larvae dispersing at 10 cm, but 2.5- and 15-fold fewer LLIN-exposed larvae dispersing compared with controls at 25 and 75 cm. The two-way interaction between type of netting and postexposure holding duration on dispersing larvae was not significant ($F$ = 0.36; df = 2, 54; $P$ < 0.70).

By contrast, the dispersal of small *T. variabile* larvae were not as strongly affected overall by exposure to LLIN netting ($F$ = 1.73; df = 1, 54; $P$ < 0.19; Fig. 4). Dispersal distance did significantly affect the number that successfully reached a novel food patch ($F$ = 16.6; df = 2, 54; $P$ < 0.0001), with 2.6- and 6.2-fold fewer small larvae making it across at 25 and 75 cm, respectively, compared with 10 cm. The postexposure holding duration also significantly affected dispersal ($F$ = 8.81; df = 2, 54; $P$ < 0.001); 3.5- and 2-fold fewer larvae were able to successfully disperse 10 min and 24 h after exposure, respectively, compared with immediately being given the opportunity to disperse. Importantly, the two-way interaction between type of netting and dispersal distance significantly affected the percentage of dispersing insects ($F$ = 3.38; df = 2, 54; $P$ < 0.05), with 6- and 4-fold fewer dispersing small *T. variabile* larvae after LLIN exposure compared with controls at 25 and 75 cm (Fig. 4). However, the two-way interaction between type of netting and postexposure holding duration was not significant ($F$ = 3.55; df = 2, 54; $P$ < 0.06).

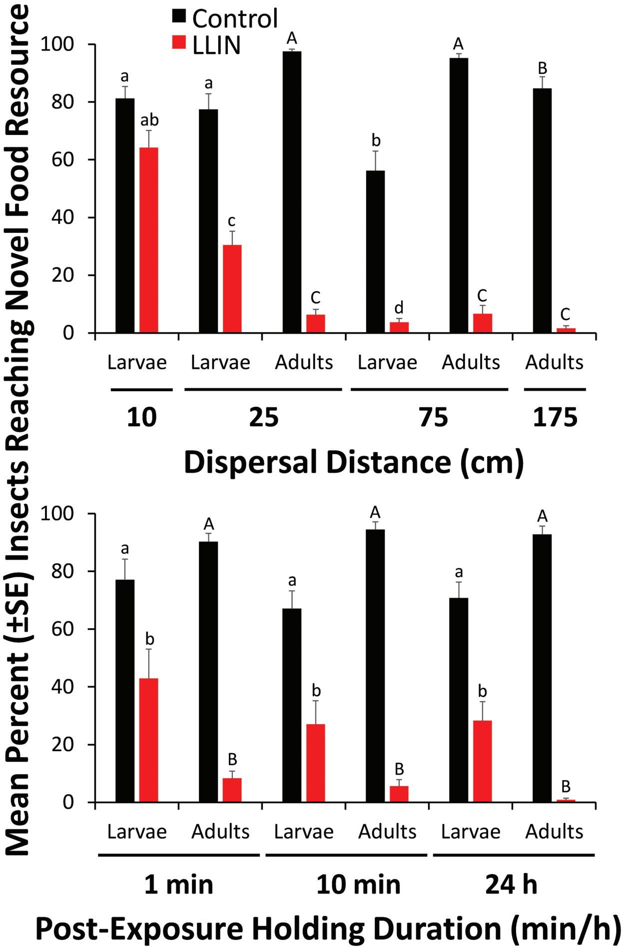

**Fig. 3.** The mean percentage of *Tribolium castaneum* adults and larvae reaching a novel food resource after 48 h, depending on distance that they had to travel (top) or the postexposure holding duration prior to dispersal opportunity (bottom) after cohorts of 20 adults were exposed for 5-min exposures to control or long-lasting insecticide-incorporated netting in environmental chambers at 30°C and 65% RH. Each bar represents the mean of $n$ = 12 (larvae) or $n$ = 18 (adults) replicates. Uppercase letters indicate pairwise comparisons among treatments for adults, while lowercase letters represent comparisons among treatments for larvae (Tukey HSD, α = 0.05). Bars with shared letters are not significantly different from each other. Larvae were given the opportunity to disperse at 10, 25, and 75 cm, whereas adults were given the opportunity to disperse at 25, 75, and 175 cm based on prior information about the dispersal capacity of each life stage.

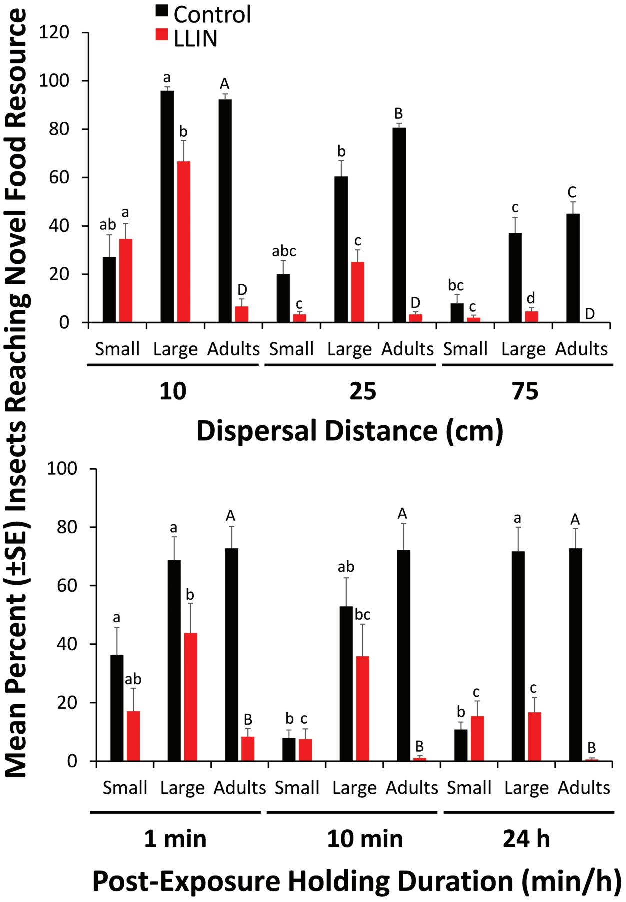

**Fig. 4.** The mean percentage of *Trogoderma variabile* adults and small or large larvae reaching a novel food resource after 48 h, depending on distance that they had to travel (top) or the postexposure holding duration prior to dispersal opportunity (bottom) after cohorts of 20 adults were exposed for 5-min exposures to control or long-lasting insecticide-incorporated netting in environmental chambers at 30°C and 65% RH. Each bar represents the mean of $n$ = 12 (small larvae, large larvae, or adult) replicates. Uppercase letters indicate pairwise comparisons among treatments for adults, while lowercase letters represent comparisons among treatments for larvae (Tukey HSD, α = 0.05). Bars with shared letters are not significantly different from each other.

Similar to *T. castaneum* larvae, the dispersal of large *T. variabile* larvae was significantly affected by LLIN exposure ($F$ = 73.5; df = 1, 54; $P$ < 0.0001; Fig. 4), with 2-fold fewer LLIN-exposed individuals dispersing compared with controls. The dispersal distance also significantly affected the number of large larvae reaching a novel food patch ($F$ = 87.5; df = 2, 54; $P$ < 0.0001). Two-fold and 4-fold large larvae reached a novel food patch when placed 25 and 75 cm away, respectively, compared with 10 cm (Fig. 4). While the postexposure holding duration also significantly affected dispersal of large larvae ($F$ = 4.48; df = 2, 54; $P$ < 0.05), there was an 8-fold reduction in the number reaching a novel food resource at 10 min and 24 h after exposure compared with if they were given the opportunity to disperse immediately. Importantly, the two-way interaction between type of netting and dispersal distance did not significantly affect the number of dispersing large larvae ($F$ = 0.22; df = 2, 54; $P$ = 0.80), with a 1.5-, 2.4-, and 8.1-fold reduction in the number of LLIN-exposed larvae reaching a novel food source compared with controls at 10, 25, and 75 cm, respectively. There was also a significant effect of the postexposure holding duration on the number of dispersing larvae ($F$ = 9.35; df = 2, 54; $P$ < 0.001), with fewer large larvae able to disperse immediately after exposure and 24 h later compared with 10 min later.

LLIN exposure to *T. variabile* adults had a pronounced negative effect on dispersal to novel food patches ($F$ = 1185; df = 1, 36; $P$ < 0.0001; Fig. 4), with a 22-fold decrease in the number of adults making it across after exposure compared with controls. Likewise, the increasing dispersal distances negatively affected the number successfully reaching a novel food patch ($F$ = 63.7; df = 2, 36; $P$ < 0.0001), with 1.2- and 2.2-fold fewer adults successful at 25 and 75 cm compared with 10 cm. The postexposure holding duration did not affect the number of dispersing adults ($F$ = 1.66; df = 2, 36; $P$ < 0.20). There was a significant interaction between type of netting and distance on the number of dispersing individuals ($F$ = 37.8; df = 2, 36; $P$ < 0.0001), but primarily manifested as a decreasing number of control netting-exposed adults reaching a novel food patch with increasing distance; importantly, there were 14- and 24-fold fewer LLIN-exposed adults that made it across compared with control netting-exposed adults at 10 and 25 cm, while not a single LLIN-exposed adult was able to reach the novel food patch at 75 cm. The two-way interaction between type of netting and postexposure holding duration on dispersal was not significant ($F$ = 1.46; df = 2, 36; $P$ < 0.25).

Overall, the number of dispersing *T. castaneum* adults after contact with LLIN was 8-fold lower than larvae (chi-squared test, Table 5). With increasing dispersal distance, adults maintained a relatively constant 93–98% reduction in movement, while the dispersal of LLIN-exposed larvae was progressively reduced by 21% at 10 cm to 93% at 75 cm (Table 5). Regardless of holding duration after exposure to LLIN, adults moved 6- to 44-fold less compared with larvae (Table 6).

Likewise, the number of dispersing *T. variabile* adults was 10-fold lower than large and small larvae (chi-squared test, Table 5). While there were 10-fold fewer adults that successfully dispersed to a novel food patch after LLIN exposure than larvae at 10 and 25 cm, not a single LLIN-exposed adult dispersed at 75 cm. Regardless of holding duration after LLIN exposure, 6- to 45-fold fewer adults than larvae dispersed to novel food patches (Table 6).

## Discussion

Prior work has documented the utility of pyrethroid-incorporated LLIN (e.g., trade name: Carifend , α-cypermethrin; d-Terrence, deltamethrin) in laboratory tests to induce mortality in multiple species of adult stored-product pests (Morrison et al. 2018, Rumbos et al. 2018, Paloukas et al. 2020). Adult *T. castaneum* and *R. dominica* exhibited multiple-fold decreases in movement and dispersal after exposure to deltamethrin-based LLIN (Morrison et al. 2018). Furthermore, traps comprised of the α-cypermethrin-based LLIN were effective at protecting the long-term storage of tobacco against adult insects in semifield and commercial facilities (Athanassiou et al. 2019). Notably, these prior studies have restricted their evaluation to adult stored-product insects. Ours is the first study to assess

**Table 5.** Relative susceptibility in dispersal ability of *T. castaneum* and *T. variabile* adults and larvae depending on dispersal distance after exposure to long-lasting insecticide-incorporated netting (LLIN) in the dispersal assay

| | Small larvae | | Large larvae | | Adults | |
|---|---|---|---|---|---|---|
| Dispersal distance | Relative % dispersal[a] | Relative % reduction in dispersal[b] | Relative % dispersal[a] | Relative % reduction in dispersal[b] | Relative % dispersal[a] | Relative % reduction in dispersal[b] |
| *T. castaneum* | | | | | | |
| 10 | — | — | 79.0[c] | 21.0[c] | — | — |
| 25 | — | — | 39.2a | 60.8B | 6.6b | 93.4A |
| 75 | — | — | 6.7a | 93.3A | 7.0a | 93.0A |
| 175 | — | — | — | — | 1.97[c] | 98.0[3] |
| Overall | — | — | 41.6a | 58.4B | 5.2b | 94.8A |
| *T. variabile* | | | | | | |
| 10 | 127.7a | –27.7C | 69.6b | 30.4B | 7.2c | 92.8A |
| 25 | 16.7b | 83.3A | 41.4a | 58.6B | 4.1c | 95.9A |
| 75 | 26.3a | 73.7B | 12.4b | 87.6AB | 0.0c | 100A |
| Overall | 72.0a | 27.3C | 41.1b | 58.9B | 3.8c | 96.2A |

Lowercase letters represent comparisons between larvae and adults for relative percent dispersal compared to controls, while uppercase letters represent comparisons between larvae and adults for percent reduction in dispersal ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific dispersal distance.

[a] The number of dispersing individuals that made it to novel food patches after LLIN exposure as a percent of the number of dispersing individuals after control netting-exposure in the dispersal assay.

[b] The percent reduction in successfully dispersing individuals after LLIN exposure relative to control netting-exposed individuals in the dispersal assay.

[c] Statistical comparisons not possible between these life stages for dispersal distance.

**Table 6.** Relative susceptibility in dispersal ability of *Tribolium castaneum* and *Trogoderma variabile* adults and larvae depending on postexposure holding duration after contact with long-lasting insecticide-incorporated netting (LLIN) in the dispersal assay

| | Small larvae | Large larvae | Adults | | | |
|---|---|---|---|---|---|---|
| Postexposure holding duration | Relative % Dispersal[a] | Relative % reduction in dispersal[b] | Relative % dispersal[a] | Relative % reduction in dispersal[b] | Relative % dispersal[a] | Relative % reduction in dispersal[b] |
| T. castaneum | | | | | | |
| 1 min | — | — | 55.7a | 44.3B | 9.2b | 90.8A |
| 10 min | — | — | 40.4a | 59.6B | 5.9b | 94.1A |
| 24 h | — | — | 40.0a | 60.0B | 0.9b | 99.1A |
| Overall | — | — | 45.3a | 54.7B | 5.3b | 94.7A |
| *T. variabile* | | | | | | |
| 1 min | 47.1a | 52.9BC | 63.6a | 36.4C | 11.5b | 88.5A |
| 10 min | 94.7a | 5.3C | 67.7b | 32.3B | 1.5c | 98.5A |
| 24 h | 14.2a | 42.3B | 23.3a | 76.7A | 0.8b | 99.2A |
| Overall | 72.7a | 27.3C | 51.5a | 48.5B | 4.6b | 95.4A |

Lowercase letters represent comparisons between larvae and adults for relative percent dispersal compared with controls, while uppercase letters represent comparisons between larvae and adults for percent reduction in dispersal ($\chi^2$, $\alpha = 0.05$). All comparisons are within a specific postexposure holding duration.

[a] The number of dispersing individuals that made it to novel food patches after LLIN exposure as a percent of the number of dispersing individuals after control netting-exposure in the dispersal assay.

[b] The percent reduction in successfully dispersing individuals after LLIN exposure relative to control netting-exposed individuals in the dispersal assay.

differential susceptibility to LLIN by stored-product insect larvae. Overall, we found that LLIN exposure more dramatically reduced adult movement and dispersal capacity compared with larvae.

Specifically, we have evaluated total distance moved and mean velocity as two measures of movement for adults and larvae after exposure to LLIN. These are important variables that mediate immigration into food facilities, foraging, mate-finding, dispersal, and other important biological processes. At 30°C, *T. castaneum* only spends 4% of its life cycle in the larval stage (Brown et al. 2009), while *T. variabile* spends 85% of its life cycle as larvae (Partida and Strong 1975). However, regardless of specific differences in life history, adults are considered the dispersing stage for both species (Hagstrum and Subramanyam 2006). Because adults of both species were generally more susceptible than larvae, targeting adults with LLIN thus has the potential to disrupt important events in the life cycle, including colonization of grain stores and food facilities from the landscape.

In a more realistic test of how LLIN may affect dispersal capacity to novel food patches, our data from the dispersal assay mimicked exposure to LLIN as insects enter a food facility with the potential food source being located a relatively trivial distance away. It is likely that in many cases inside food facilities the resources might be much further away because food facilities often represent sprawling complexes with multiple buildings. However, despite the trivial distance, both LLIN-exposed larvae and adults, but especially the latter, had difficulty reaching the novel food source even though many or most of the control netting-exposed individuals made it across. In prior work, although knockdown of stored-product adults was not immediate (D. S. Scheff, personal communication), LLIN-exposed *R. dominica* adults were found to be incapable of dispersing 25–175 cm in an equivalent assay (Morrison et al. 2018). Thus, it appears that the dispersal capacity of stored-product insects will be significantly impaired after brief contact with LLIN.

Differences in behavioral responses to LLIN after contact by insects may be species-specific and life stage-specific. For example, *T. variabile* adults appeared to experience greater reductions in mobility compared with *T. castaneum* adults (e.g., Tables 1 and 2). Other work has found that there may be species-specific responses to insecticides, e.g., with greater larval mortality for the khapra beetle, *Trogoderma granarium* (Everts) (Coleoptera: Dermestidae) than *T. variabile* after contact with an insecticide (Ghimire et al. 2017), despite the fact that both are closely related (Castalanelli et al. 2012). In addition, there may be life stage-specific differences in response to stressors. In this study, for example, we found *T. variabile* larvae were less susceptible than adults to LLIN exposure. Previous work in evaluating life stage-based susceptibility to other pyrethrin or pyrethroid products is consistent with our findings, but these studies typically provided a food source for the individuals after insecticide exposure (Kharel et al. 2014, Athanassiou et al. 2015, Ghimire et al. 2016). For example, Ghimire et al. (2016) found that *T. variabile* adults are over 3-fold more susceptible to deltamethrin, the active ingredient in the LLIN used in the present study, in residual contact trials compared with larvae. Similarly, Arthur and Fontenot (2012) determined that *T. castaneum* larvae were more susceptible than adults when exposed to chlorfenapyr residual on partially treated concrete arenas. However, those authors concluded that the mobility of life stages contributed to their differential susceptibility. Our study has shown that *T. variabile* larvae are actually more mobile than adults, while *T. castaneum* larvae are far less mobile than adults. Nevertheless, in the present study, the biological importance of these life stage-specific differences may be minimal because the dispersal capacities for both species were significantly and similarly impaired. Thus, it is likely for both species that the reduced movement will lead to reduced ability to infest stored products.

There are a variety of ways that LLIN may be integrated into the IPM programs of food facilities. For example, LLIN may be deployed over external vents, eaves, and openings of food facilities as a barrier against insects immigrating from the landscape, particularly adult stages. Our research shows that brief exposure to only a single layer of LLIN is sufficient to significantly affect dispersal ability. Additionally, LLIN may be deployed over windows, doorways, and other internal partitions inside food facilities. This LLIN deployment method will help localize the infestation to one area of the facility. Finally, pallets of product that are brought into warehouses may be wrapped with LLIN. This will prevent insect dispersal into and out of pallets of goods. However, future work should assess whether there are differences in efficacy associated with each of these deployment procedures. At a minimum, our current work provides insight into how dispersing life stages may be controlled by LLIN.

While we have demonstrated that the use of LLIN is very effective at reducing movement and dispersal of adults and even larvae, to a lesser extent, under controlled conditions, there are still several outstanding questions about the implementation. For example, it is currently unknown whether LLIN deployment method affects efficacy (e.g., on a vent, covering a pallet directly, etc.). In addition, there is little information about whether the systematic use of LLIN is able to intercept immigrating insects from the landscape to reduce commodity infestation in bins or infestation of structures such as warehouses or food facilities, though the data from this study and related work suggests that it is likely. Future work investigating these avenues will be relevant for determining the widescale applicability of LLIN for use in food facilities. In the meantime, LLIN is a promising new IPM tactic that warrants further consideration in the postharvest environment.

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