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RESEARCH ARTICLE

Five natural compounds of botanical origin as wheat protectants against adults and larvae of *Tenebrio molitor* L. and *Trogoderma granarium* Everts

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Abstract

The botanical substances constitute valuable alternatives to synthetic insecticides. In the last decades, numerous substances of natural origin have been tested against stored-product insects, mostly as fumigants or for contact toxicity, while there is limited knowledge on the efficacy of plant secondary metabolites if used as grain protectants. In the present study, we evaluated the lethal activity of 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal and (*E*, *E*)-2,4-decadienal as wheat protectants for the management of larvae and adults of two important storage pests, *Tenebrio molitor* (Coleoptera: Tenebrionidae) and *Trogoderma granarium* (Coleoptera: Dermestidae). 2-undecanone caused 98.9% mortality to the exposed *T. molitor* adults at 1000 μl/kg wheat 7 days post-exposure, while acetic acid and furfural followed providing 94.4% and 92.2% mortality respectively. 2-Undecanone and (*E*)-2-decenal caused the highest mortalities to *T. molitor* larvae (i.e., 87.8% and 80.0% respectively) exposed to 1000 μl/kg wheat for 7 days. All *T. granarium* adults were dead at 1000 μl (*E*)-2-decenal or acetic acid/kg wheat 5 or 7 days post-exposure respectively. Complete (100%) mortality was assessed for larvae exposed to (*E*, *E*)-2,4-decadienal and (*E*)-2-decenal at 1000 μl/kg wheat after 4 and 6 days respectively. Our findings report for the first time that 2-undecanone, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal are effective new candidate control agents of different developmental stages of *T. molitor* and *T. granarium*.

Keywords Botanical compounds · Grain protectants · Yellow mealworm beetle · Khapra beetle · Developmental stages

Introduction

Stored products are attacked by the remarkable numbers of 1663 insect and 280 mite species (Hagstrum et al. 2013) while grain losses worldwide reach 10–20% due to insect infestations (Islam et al. 2021). The damages of food commodities, in terms of quality and quantity, caused by stored-product pests consist a major issue for food-related industries (Rajendran and Sriranjini 2008). Also, the existence of pests in stored products is linked to the elevated risk of food safety and public health (e.g., spread of pathogens, production of allergens) making their effective management challenging (Hill 2003; Mason and McDonough 2012; Djekic et al. 2019).

The yellow mealworm beetle, *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) infests a wide spectrum of food materials having plant and animal origin (Hill 2003; Robinson 2005; Hagstrum et al. 2013; Guo et al. 2014). The detoxifying cytochrome P450 enzymes of this species have been reported of markedly high activity, leading to reduced pesticide sensitivity (Pedersen et al. 2020). Previous reports have documented that *T. molitor* is tolerant to contact insecticides such as alpha-cypermethrin, thiamethoxam, diatomaceous earths (DEs), deltamethrin, and spinosad (Trewin and Reichmuth 1997; Mewis and Ulrichs 2001; Athanassiou et al. 2015; Kavallieratos et al. 2019b). However, the efficacy of insecticides against *T. molitor* is regulated by abiotic (temperature,

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relative humidity (RH)) and biotic factors (life stage, type of treated grain commodity) (Kavallieratos et al. 2019b, 2021).

The khapra beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae), is one of the 100 most important invasive species worldwide and it has been recently reported from Greece (Lowe et al. 2000; Athanassiou et al. 2015). This beetle affects a wide range of stored products, survives in extreme abiotic conditions, and is considered one of the most harmful insect pests of stored agricultural products and foodstuffs (Lindgren et al. 1955; Hill 2003; Athanassiou et al. 2016; Kavallieratos et al. 2017b, 2019a; Papanikolaou et al. 2019). The fact that larvae of this species undergo diapause for several years has favored its widespread in several countries of Africa, Asia, and Europe (Aitken 1975; Myers and Hagstrum 2012; Athanassiou et al. 2016, 2019; EPPO 2019). Recent studies have revealed that the management of *T. granarium* meets certain difficulties due to elevated tolerance to several contact insecticides as surface treatments or grain protectants (e.g., alpha-cypermethrin, beta-cyfluthrin, chlorfenapyr, cypermethrin, deltamethrin, DE, pyriproxyfen, spinosad, thiamethoxam) (Athanassiou et al. 2015, Kavallieratos et al. 2016, 2017a; Ghimire et al. 2017).

The Common Agricultural Policy has highlighted the environmental, food safety, and animal welfare standards and proposed the use of alternatives for the management of stored-product insects (Schillhorn van Veen 1999). The non-synthetic plant protection products are novel effective tools that have the potential to be used alone or in mixtures with synthetic insecticides in the storage environment (Athanassiou et al. 2014). Edible biopesticides of botanical origin may be considered low-risk alternatives according to EC 1107/2009 Articles 22, 47 (EUR Lex 2009), necessitating a smaller experimental dataset for registration purposes thus lower regulatory barrier. Plant secondary metabolites with insecticidal activities (Isman 2006, 2008) are usually selected from libraries of thousands of compounds, originating in biodiversity hotspots like the Mediterranean Basin, to be developed individually into plant protection products. Crude botanicals, representing complex clusters of plant secondary metabolites, may have equally significant or even better plant protection properties, a selective mode of action and help to avoid the emergence of resistant strains of pest species due to the wide variety of secondary metabolites (Isman 2006; Gupta and Birdi 2017; Ntalli and Caboni 2017). Natural botanicals exhibit repellent, antifeedant, sterilizing, ovicidal, or toxic effects on insects (Isman 2006). As members of the botanical insecticides, essential oils (EOs) exhibit a significant range of pesticidal activities (Ntalli et al. 2010a, 2011; Benelli and Pavela 2018a, b; Benelli et al. 2019; Kavallieratos et al. 2020b; Pavela et al. 2019, 2020) and can be prepared in a "green", easy, and cost-effective way, not employing organic solvents or sophisticated extraction procedures. Among the EO components, the monoterpenes have drawn the greatest attention (Coats et al. 1991). Considering the mode of action of monoterpenes, Houghton et al. (2006) proved that they inhibit acetylcholinesterase (AChE) enzyme activity, although in several cases, there was a lack of direct correlation between insect toxicity and AChE inhibition (Lee et al. 2001; López and Pascual Villalobos 2010, 2014). Monoterpenes have been also reported to affect the cytochrome P4502B1-dependent enzymes in rats that are involved in the activation of genotoxic substances (De Oliveira et al. 1997), as well as to inhibit the α-amylase enzyme activity in the red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) (Huang et al. 1999).

The evaluation of the toxicity of various botanical compounds against the stored-product insects has been mainly conducted by applying them as fumigants. For example, according to Regnault Roger and Hamraoui (1995) the oxygenated monoterpenes are more toxic compounds than the non-oxygenated monoterpenes as fumigants against the bean weevil, *Acanthoscelides obtectus* (Say) (Coleoptera: Chrysomelidae). In another fumigation assay, Lee et al. (2003) reported that all adults of the rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), *T. castaneum*, and the sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) died when exposed to cineole, *l*-fenchone and pulegone at 50 mg/ml air 14 h post-exposure. Similarly, Erler (2005) postulated that vapors of γ-terpinene and terpinen-4-ol provided 100% mortality to adults of the confused flour beetle, *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) at 92.4 and 184.8 mg/l air after 4 days of exposure. The latter concentration had the same effect on the larvae of the Mediterranean flour moth, *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae). The vapors of carvone at 972 ppm and *trans*-anethole at 880 ppm killed all exposed adults of the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae) 1 day post-exposure (López et al. 2008). The use of *DL*-camphor or estragole as fumigants in combination with low temperature (19 °C) and low pressure (50 mm Hg) successfully controlled the diapausing larvae of the Indianmeal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae) (Mbata et al. 2012). Mbata and Payton (2013) reported that there was variable susceptibility among different life stages of the cowpea weevil, *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae) when fumigated with the monoterpenes *trans*-anethole, estragole, *S*-carvone, *L*-fenchone, geraniol, γ-terpinene, and *DL*-camphor. However, there is little knowledge regarding the use of botanical substances as grain protectants (Islam et al. 2010; Mbata et al. 2012; Osman et al. 2016; Kavallieratos et al. 2020a,b; Pavela et al. 2020) and a paucity of information concerning other than EO originating natural molecules against stored-products insects. Herein, we tested botanical molecules belonging to different chemical groups to delineate for insecticidal activity, while they have already been proved of significant nematicidal activity in the recent years. In specific, we study *trans*-anethole (phenolic monoterpene) and 2-undecanone (dialkyl ketone) representing EO ingredients (Ntalli et al. 2010a, 2011); (*E*)-2-decenal (dec-2-enal) and (*E*, *E*)-2,4-decadienal (polyunsaturated fatty aldehydes) constituting *Ailanthus altissima* (Mill.) Swingle (Sapindales: Simaroubaceae) organic extracts (Caboni et al. 2012; Ntalli et al. 2016a); and furfural (aldehyde) and acetic acid (monocarboxylic acid) as ingredients of the fruits of *Melia azedarach* L. (Sapindales: Meliaceae) (Ntalli et al. 2010b; Caboni et al. 2012; Ntalli et al. 2020). Scanning and transmission electron microscopy studies documented that the acetic acid, (*E*)-2-decenal, and 2-undecanone provoke irreversible ultrastructural modifications on *Meloidogyne incognita* (Kofold & White) Chitwood (Rhabditida: Meloidogynidae) J2 (Ntalli et al. 2016b). Interestingly, the insecticidal efficacy of *trans*-anethole, 2-undecanone, and furfural has been already reported on other insects. In specific, through contact toxicity trials: e.g., *trans*-anethole vs. *T. castaneum* (Mondal and Khalequzzaman 2010), 2-undecanone vs. *Aedes albopictus* (Skuse) (Diptera: Culicidae) (Liu et al. 2014), furfural vs. *S. oryzae* and the cigarette beetle, *Lasioderma serricorne* (F.) (Coleoptera: Ptinidae) (Urrutia et al. 2021). To the best of our knowledge, no data exist to date on the activity of the aforementioned molecules for the management of *T. granarium* and *T. molitor*. Thus, the objective of the present study was to evaluate the lethal activity of 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal as wheat protectants against larvae and adults of *T. granarium* and *T. molitor*.

Materials and methods

Commodity

Hard wheat, *Triticum durum* Desf. (var. Claudio), free from pests and pesticides, was used in the trials. Wheat was sieved to remove impurities and kept at subzero temperatures for several months. Prior to experimentation, it was warmed under room temperature. Its moisture content was 12.6% as determined by a moisture meter (mini GAC plus, DICKEY-john Europe S.A.S., Colombes, France).

Insects

The insects used in the trials were obtained from colonies that are kept in the Laboratory of Agricultural Zoology and Entomology, Agricultural University of Athens, since 2014. The founding individuals have been collected from Greek storage facilities. We used unsexed adults of *T. granarium*, < 24 h old; small larvae of *T. granarium* < 3 mm long (Kavallieratos and Boukouvala 2019); unsexed adults of *T. molitor*, < 2 weeks old; and small larvae of *T. molitor* < 10 mm long (Kavallieratos et al. 2019b). *Trogoderma granarium* was cultured on whole wheat, at 30 °C and 65% RH in continuous darkness (Kavallieratos and Boukouvala 2019). *Tenebrio molitor* was reared on oat bran with slices of potatoes as a source of moisture (De Vosjoli 2007), at 30 °C and 65% RH in continuous darkness (Kavallieratos et al. 2019b).

Test compounds

The compounds 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal (Fig. 1), all over 98% purity, were obtained from Sigma–Aldrich (Buchs, Switzerland).

Bioassays

Each test substance was tested at 1000 μl/kg and 500 μl/kg wheat. The test concentrations were selected on the basis of preliminary efficacy tests on both insect species. For the test concentration of 1000 μl/kg wheat, a volume of 250 μl of test substance was dissolved in 6 ml pure ethanol and the solution was applied on 0.25 kg wheat. Similarly, 125 μl of test substance were dissolved in 6 ml pure alcohol to treat 0.25 kg wheat, representing the test concentration of 500 μl/kg wheat. Spraying was performed with the airbrush AG-4 (Mecafer S.A., Valence, France) on wheat lots that were laid out on different trays. Additional lots of 0.25 kg wheat treated with same volumes (6 ml) of (a) pure ethanol and (b) water served as controls. After spraying the lots of wheat were transferred separately in 1-l glass containers and were shaken manually for 10 min to achieve equal distribution of test solution on the entire wheat mass. Three samples of 10 g each were obtained per treated lot or control and placed inside small glass vials (7.5 × 12.5 cm diameter and height) with a different scoop. A Precisa XB3200D (Alpha Analytical Instruments, Gerakas, Greece) compact balance was used to weigh the portions of 10 g of wheat on a thin layer. A new layer was used for each weighing. The lid of each vial bore a 1.5-cm diameter hole in the center covered with muslin so as to permit adequate ventilation. The escape of insects was prohibited by coating the upper internal parts of the vials with polytetrafluoroethylene (60 wt % dispersion in water) (Sigma-Aldrich, Chemie GmbH, Taufkirchen, Germany). Coating was conducted with a swap, 24 h before the placement of individuals in the vials. Subsequently, 10 adults of *T. granarium* or 10 larvae of *T. granarium* or 10 adults of *T. molitor* or 10 larvae of *T. molitor* were separately placed inside each vial. Next, all vials were transferred in incubators set at 30 °C and 65% RH. Mortality of adults or larvae was evaluated under an Olympus stereomicroscope (SZX9, Bacacos S.A., Athens, Greece) at ×

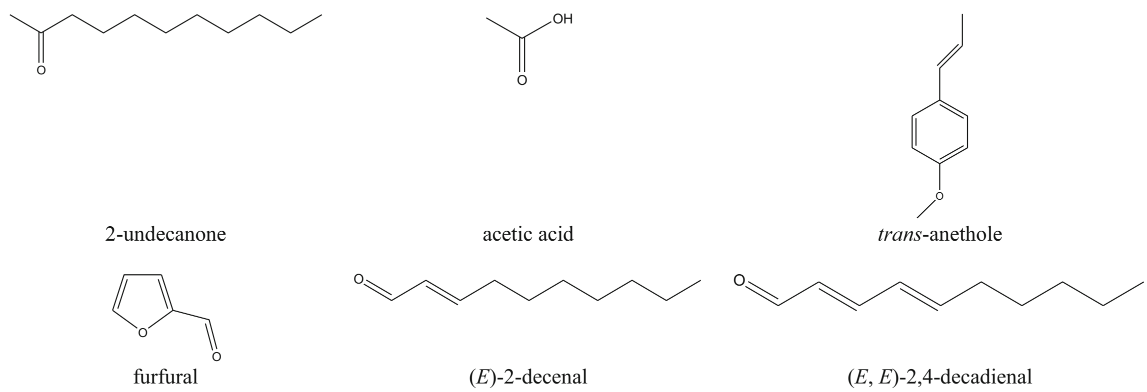

Fig. 1 Test plant secondary metabolites against *T. molitor* and *T. granarium* larvae and adults

57 total magnification by pushing gently each individual with a brush (Cotman 111 No 000, Winsor and Newton, London, UK) to inspect movements after 4, 8, and 16 h and 1, 2, 3, 4, 5, 6, and 7 days of exposure. Different brushes were used for each treatment and controls. All tests were repeated three times for test substances and controls, by preparing new wheat lots and new vials each time.

Statistical analysis

Mortality in the controls, pure alcohol and water, was low (< 5%) for all tested species and life stages. Therefore, no correction was necessary for the mortality values. Before conducting analyses, the mortality data were log $(x + 1)$ transformed to normalize variance (Zar 2014; Scheff and Arthur 2018). Statistical analyses were carried out separately for each tested species and life stage by following the repeated measures model (Sall et al. 2001). The repeated factor was the exposure interval while mortality was the response variable. Concentration and compound were the main effects. The associated interactions of the main effects were considered during analysis. The JMP 14 software (SAS Institute 2018) was used to analyze all data. Means were separated by the Tukey-Kramer honestly significant difference (HSD) test at the 0.05 probability (Sokal and Rohlf 1995).

Results

Effectiveness against *T. molitor*

Between and within exposure intervals, all main effects and related interactions were significant ($P < 0.05$) for both *T. molitor* adults and larvae, except for the interaction exposure × compound × concentration for *T. molitor* larvae which was not significant (Table 1). Concerning *T. molitor* adults, furfural was the most effective compound at 500 µl/kg wheat, followed by *trans*-anethole causing 41.1% and 21.1% mortalities, respectively, after 7 days of exposure (Table 2). The overall mortality provided by the other compounds did not exceed 13.3%. (*E*, *E*)-2,4-decadienal was the least effective compound for both concentrations since it killed 5.6% and 41.1% of the exposed adults at 500 µl/kg and 1000 µl/kg wheat respectively. After 3 days of exposure, 2-undecanone caused 60.0% mortality at 1000 µl/kg wheat, while it reached 98.9% 7 days post-exposure. The compound acetic acid and furfural provided 94.4% and 92.2% mortalities after 7 days of exposure respectively. Mortality caused by *trans*-anethole and (*E*)-2-decenal reached the moderate levels of 64.4% and 56.7%, respectively, after 7 days of exposure.

Regarding *T. molitor* larvae, the overall mortality at 500 µl/kg wheat did not exceed 47.8% after 7 days of exposure for 2-undecanone and (*E*)-2-decenal (Table 3). The compound acetic acid killed 7.8% while both *trans*-anethole and furfural killed 11.1% of the exposed larvae at 500 µl/kg wheat 7 days post-exposure. At 1000 µl/kg wheat, furfural provided the highest larval mortality (i.e., 66.7%) after 5 days of exposure, while 2-undecanone and (*E*)-2-decenal caused the highest mortalities to larvae (i.e., 87.8% and 80.0% respectively) 7 days post-exposure. In contrast, the lowest mortality (i.e., 16.7%) was caused by *trans*-anethole after 7 days of exposure at 1000 µl/kg wheat.

Effectiveness against *T. granarium*

Between and within exposure intervals, all main effects and related interactions were significant ($P < 0.05$), for both *T. granarium* adults and larvae, except the interaction compound × concentration for *T. granarium* adults which was not significant (Table 1). Concerning *T. granarium* adults, > 95% of the exposed individuals were dead on wheat treated with (*E*)-2-decenal 5 days post-exposure at 500 µl/kg wheat. Complete (100%) mortality of *T. granarium* adults was reached by (*E*)-2-decenal after 7 days of exposure at 500 µl/kg wheat or after 5 days at 1000 µl/kg wheat. Similarly, acetic acid killed all adults 7 days post-exposure at 1000 µl/kg wheat (Table 4). (*E*, *E*)-2,4-decadienal killed the same percentage of adults (i.e., 94.4%) after 7 days of exposure at both

Table 1 MANOVA parameters about the main effects and associated interactions for mortality levels of *T. molitor* and *T. granarium* adults and larvae between and within exposure intervals (in all cases error *df* = 96)

Source	*T. molitor* adults			*T. molitor* larvae			*T. granarium* adults			*T. granarium* larvae		
Between exposure intervals												
Source	*df*	*F*	*P*	df	*F*	*P*	df	*F*	*P*	df	*F*	*P*
Intercept	1	940.9	<0.01	1	822.1	<0.01	1	7410.1	<0.01	1	1994.1	<0.01
Compound	5	8.2	<0.01	5	15.7	<0.01	5	13.6	<0.01	5	52.3	<0.01
Concentration	1	133.5	<0.01	1	60.8	<0.01	1	58.6	<0.01	1	93.6	<0.01
Compound × concentration	5	5.9	<0.01	5	6.2	<0.01	5	1.1	0.39	5	3.0	0.02
Within exposure intervals												
Exposure × compound	45	3.6	<0.01	45	3.7	<0.01	45	3.3	<0.01	45	3.3	<0.01
Exposure × concentration	9	21.9	<0.01	9	6.7	<0.01	9	8.4	<0.01	9	7.6	<0.01
Exposure × compound × concentration	45	2.4	<0.01	45	1.4	0.06	45	1.8	<0.01	45	2.1	<0.01

concentrations. 2-undecanone provided 94.4% mortality to *T. granarium* adults 6 days post-exposure at 1000 μl/kg wheat. The mortalities caused by all other compounds ranged between 64.4 and 81.1% at 500 μl/kg wheat, or between 82.2 and 92.2% at 1000 μl/kg wheat after 7 days of exposure.

As far as *T. granarium* larvae are concerned, no compound was lethal to all individuals even after 7 days of exposure at 500 μl/kg wheat (Table 5). (*E*)-2-decenal was the most effective tested compound at 500 μl/kg wheat since it provided 87.8% mortality at the end of the experiment followed by (*E*, *E*)-2,4-decadienal which caused 80.0% mortality. At 1000 μl/kg wheat, (*E*, *E*)-2,4-decadienal killed 90.0% of the larvae on wheat after only 2 days of exposure, while all individuals were dead 4 days post-exposure. (*E*)-2-decenal was also highly effective because it killed 94.4% of larvae after 4 days of exposure. Two days later, all exposed larvae were dead on wheat treated with (*E*)-2-decenal at 1000 μl/kg wheat. 2-undecanone, acetic acid, and furfural provided moderate mortalities, ranging between 11.1 and 46.7% at 500 μl/kg wheat or between 54.4 and 76.7% at 1000 μl/kg wheat 7 days post-exposure. The compound *trans*-anethole caused the lowest mortality scoring 8.9% at 500 μl/kg wheat and 28.9% at 1000 μl/kg wheat after 7 days of exposure.

Discussion

The published information on the potential of substances of botanic origin as effective grain protectants is sporadic. For instance, the binary combinations of the DE SilicoSec with cinnamaldehyde or eugenol on wheat exhibited synergistic activity by causing higher mortalities to adults of *S. oryzae* than the DE or the monoterpene alone (Islam et al. 2010). Mbata and Payton (2013) showed that mated female individuals of *C. maculatus* did not lay eggs on beans that had been treated with *E*-anethole, estragole, *S*-carvone, *L*-fenchome, geraniol, γ-terpinene, and *DL*-camphor. The exposure of the 4th instar larvae of *T. granarium* on wheat treated with 0.5 ml diluted caraway oil or 2 ml diluted carvone for 2 days led to serious histological decays of their midgut. Also, the midgut and the ovarioles of the emerged adult individuals suffered several histological abnormalities (Osman et al. 2016). Recently, Kavallieratos et al. (2020a) found that the sesquiterpene isofuranodiene is highly effective against adults of *T. granarium* after 3 days of exposure at 1000 μl/kg wheat.

According to our results, the lethality of *T. molitor* adults, after exposure to 1000 μl/kg wheat of test substances for 7 days, ranged between 56.7 and 98.9%. Regarding *T. molitor* larvae, the assessed lethality was between 16.7 and 87.8%. This fact demonstrates that adults are more susceptible to the test compounds than larvae. Previous studies have documented that the adult stage of *T. molitor* is more susceptible than larval stage to several contact insecticides as grain protectants. For instance, Kavallieratos et al. (2019b) reported that deltamethrin, pirimiphos-methyl, SilicoSec, and spinosad killed more adults than small or large larvae on wheat, barley, and maize. Recently, Kavallieratos et al. (2021) documented that pirimiphos-methyl provided significantly higher mortality to *T. molitor* adults than small and large larvae under different combinations of temperature (20, 25, 30 and 35 °C) and RH levels (55 and 75%) on stored barley. In contrast, the monoterpenes eugenol, caryophyllene oxide, α-humulene, α-phellandrene, and α-pinene were more lethal to larvae than adults of *T. molitor* in toxicity tests 2 days post-exposure (Martínez et al. 2018). The authors hypothesized that differences in penetration and metabolization of the compounds into the bodies of adults and larvae could explain the different levels of the observed toxicity. Whether the monoterpenes tested by Martínez et al. (2018) remain more lethal to larvae than adults if applied as grain protectants merits further investigation. The method of application, i.e., application of the molecules directly as liquids on the thorax, application via fumigation, and application on grains, may affect insects differently. In fact, according to our results, the

Table 2 Mean mortality (% ± SE) of *T. molitor* adults over selected exposure intervals on wheat, treated with 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal

Exposure	4 h	8 h	16 h	1 day	2 days	3 days	4 days	5 days	6 days	7 days		
Concentration: 500 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 B	0.0 ± 0.0 B	0.0 ± 0.0 B	0.0 ± 0.0 B	3.3 ± 1.7 AB	3.3 ± 1.7 AB	3.3 ± 1.7 ABb	6.7 ± 2.4 Ab	6.7 ± 2.4 Ab	6.7 ± 2.4 Ab	3.6	<0.01
acetic acid	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 BC	4.4 ± 1.8 ABC	6.7 ± 2.4 ABC	7.8 ± 2.8 ABCab	8.9 ± 2.6 ABab	8.9 ± 2.6 ABb	10.0 ± 2.4 Ab	5.5	<0.01
trans-anethole	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 D	2.2 ± 1.5 CD	4.4 ± 2.4 BCD	10.0 ± 5.3 ABCD	13.3 ± 6.2 ABCab	16.7 ± 6.5 ABCab	17.8 ± 6.2 ABab	21.1 ± 5.6 Aab	7.3	<0.01
furfural	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 D	5.6 ± 1.8 C	15.6 ± 3.8 BC	24.4 ± 5.0 ABa	32.2 ± 4.0 ABa	37.8 ± 4.3 Aa	41.1 ± 3.9 Aa	45.4	<0.01
(*E*)-2-decenal	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 C	3.3 ± 1.7 BC	7.8 ± 2.2 ABab	8.9 ± 2.0 ABab	10.0 ± 2.4 ABab	13.3 ± 2.4 Aab	12.2	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 BC	1.1 ± 1.1 BC	2.2 ± 1.5 BC	2.2 ± 1.5 BCb	4.4 ± 1.8 ABb	4.4 ± 1.8 ABb	5.6 ± 1.8 Ab	2.6	<0.01
F	-	-	-	1.1	1.3	2.3	3.8	3.9	4.9	6.1		
P	-	-	-	0.39	0.28	0.06	<0.01	<0.01	<0.01	<0.01		
Concentration: 1000 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 E	0.0 ± 0.0 E	5.6 ± 1.8 Da	10.0 ± 1.7 Ca	38.9 ± 3.1 Ba	60.0 ± 4.1 ABa	72.2 ± 5.7 ABa	86.7 ± 5.3 Aa	95.6 ± 2.4 Aa	98.9 ± 1.1 Aa	120.1	<0.01
acetic acid	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 Cb	2.2 ± 1.5 Cb	24.4 ± 4.7 Bab	55.6 ± 4.7 Aa	81.1 ± 3.9 Aa	88.9 ± 2.6 Aa	92.2 ± 2.8 Aa	94.4 ± 2.4 Aa	157.2	<0.01
trans-anethole	0.0 ± 0.0 E	1.1 ± 1.1 E	2.2 ± 1.5 DEab	6.7 ± 2.9 CDEab	11.1 ± 3.5 CDbc	16.7 ± 5.3 BCbc	36.7 ± 5.8 ABab	51.1 ± 7.0 Aab	57.8 ± 7.0 Abc	64.4 ± 7.3 Ab	28.2	<0.01
furfural	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 Db	0.0 ± 0.0 Db	18.9 ± 3.5 Cabc	31.1 ± 3.9 Bab	52.2 ± 5.2 ABab	67.8 ± 5.7 Aab	82.2 ± 6.6 Aab	92.2 ± 3.2 Aa	218.7	<0.01
(*E*)-2-decenal	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 Db	0.0 ± 0.0 Db	0.0 ± 0.0 Dd	10.0 ± 2.4 Cc	23.3 ± 3.3 Bb	40.0 ± 5.3 ABb	48.9 ± 6.8 Acd	56.7 ± 6.7 Abc	142.6	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 E	0.0 ± 0.0 E	1.1 ± 1.1 Eb	2.2 ± 1.5 DEb	5.5 ± 1.8 CDEcd	10.0 ± 2.9 CDc	14.4 ± 4.1 BCc	16.7 ± 3.3 ABCc	32.2 ± 4.9 ABd	41.1 ± 6.3 Ac	18.0	<0.01
F	-	1.0	4.4	7.5	14.6	10.0	13.2	17.4	15.4	16.2		
P	-	0.43	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		

Within each column, means followed by the same lowercase letter are not significantly different; $df = 5, 53$; Tukey-Kramer HSD test at $P = 0.05$. Within each row, means followed by the same uppercase letter are not significantly different; $df = 9, 89$; Tukey-Kramer HSD test at $P = 0.05$. Where no letters exist, no significant differences were recorded. Where dashes exist, no analysis was conducted

Table 3 Mean mortality (% ± SE) of *T. molitor* larvae over selected exposure intervals on wheat, treated with 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal

Exposure	4 h	8 h	16 h	1 day	2 days	3 days	4 days	5 days	6 days	7 days		
Concentration: 500 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 D	0.0 ± 0.0 D	2.2 ± 1.5 D	5.6 ± 2.4 CD	13.3 ± 2.9 BCa	23.3 ± 5.0 ABa	26.7 ± 4.7 ABa	31.1 ± 5.9 ABa	38.9 ± 6.1 Aa	47.8 ± 4.3 Aa	27.9	<0.01
acetic acid	0.0 ± 0.0 B	0.0 ± 0.0 B	1.1 ± 1.1 AB	3.3 ± 1.7 AB	3.3 ± 1.7 ABab	4.4 ± 1.8 ABb	4.4 ± 1.8 ABbc	5.6 ± 1.8 ABbc	6.7 ± 2.4 ABd	7.8 ± 2.2 Ab	2.6	0.01
trans-anethole	0.0 ± 0.0 B	0.0 ± 0.0 B	0.0 ± 0.0 B	0.0 ± 0.0 B	1.1 ± 1.1 Bb	2.2 ± 1.5 Bb	2.2 ± 1.5 Bc	4.4 ± 1.8 ABc	8.9 ± 2.0 Abcd	11.1 ± 2.6 Ab	8.0	<0.01
furfural	0.0 ± 0.0 B	0.0 ± 0.0 B	1.1 ± 1.1 AB	2.2 ± 1.5 AB	3.3 ± 1.7 ABab	3.3 ± 1.7 ABb	4.4 ± 1.8 ABbc	5.6 ± 1.8 ABbc	7.8 ± 2.8 ABcd	11.1 ± 3.1 Ab	3.0	<0.01
(*E*)-2-decenal	0.0 ± 0.0 D	0.0 ± 0.0 D	1.1 ± 1.1 D	1.1 ± 1.1 D	2.2 ± 1.5 Db	5.6 ± 2.4 CDab	11.1 ± 2.6 BCab	15.6 ± 3.4 BCabc	30.0 ± 4.1 ABab	47.8 ± 5.5 Aa	23.2	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 D	2.2 ± 1.5 CD	2.2 ± 1.5 CDb	5.6 ± 1.8 BCab	10.0 ± 1.7 ABab	22.2 ± 3.6 Aab	25.6 ± 4.1 Aabc	34.4 ± 4.4 Aa	23.1	<0.01
F	-	-	0.7	1.4	3.5	3.9	7.4	5.5	7.2	10.2		
P	-	-	0.61	0.25	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		
Concentration: 1000 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 F	1.1 ± 1.1 EF	4.4 ± 1.8 DEab	8.9 ± 2.6 Dab	20.0 ± 2.9 Cab	31.1 ± 4.6 BCab	44.4 ± 4.8 ABCa	64.4 ± 4.1 ABa	82.2 ± 3.2 ABa	87.8 ± 3.2 Aa	57.9	<0.01
acetic acid	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 Db	2.2 ± 1.5 CDb	3.3 ± 1.7 CDc	6.7 ± 2.4 Ccd	10.0 ± 3.3 BCb	18.9 ± 3.1 ABb	30.0 ± 4.1 Ab	36.7 ± 2.9 Ab	26.8	<0.01
trans-anethole	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 Cb	2.2 ± 1.5 BCb	3.3 ± 1.7 ABCc	4.4 ± 1.8 ABCd	6.7 ± 2.4 ABCb	10.0 ± 2.4 ABc	13.3 ± 3.3 Ac	16.7 ± 4.1 Ac	7.0	<0.01
furfural	1.1 ± 1.1 D	3.3 ± 1.7 D	11.1 ± 2.0 Ca	24.4 ± 3.8 BCa	42.2 ± 5.5 ABa	52.2 ± 6.2 ABa	57.8 ± 5.2 ABa	66.7 ± 5.3 Aa	66.7 ± 5.3 Aab	68.9 ± 5.6 Aab	53.5	<0.01
(*E*)-2-decenal	0.0 ± 0.0 D	1.1 ± 1.1 D	1.1 ± 1.1 Db	2.2 ± 1.5 Db	4.4 ± 2.4 CDc	14.4 ± 4.1 BCbcd	32.2 ± 6.2 ABa	50.0 ± 8.2 Aa	65.6 ± 7.3 Aab	80.0 ± 6.2 Aab	37.8	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 E	2.2 ± 1.5 DE	4.4 ± 1.8 DEab	5.6 ± 2.4 DEb	8.9 ± 3.1 CDbc	21.1 ± 4.2 BCabc	37.8 ± 5.2 ABa	55.6 ± 7.1 ABa	63.3 ± 6.0 ABab	74.4 ± 4.8 Aab	29.2	<0.01
F	1.0	1.4	9.0	8.0	12.5	9.2	14.6	18.5	17.2	16.3		
P	0.43	0.25	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		

Within each column, means followed by the same lowercase letter are not significantly different; $df = 5, 53$; Tukey-Kramer HSD test at $P = 0.05$. Within each row, means followed by the same uppercase letter are not significantly different; $df = 9, 89$; Tukey-Kramer HSD test at $P = 0.05$. Where no letters exist, no significant differences were recorded. Where dashes exist, no analysis was conducted

Table 4 Mean mortality (% ± SE) of *T. granarium* adults over selected exposure intervals on wheat, treated with 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal

Exposure	4 h	8 h	16 h	1 day	2 days	3 days	4 days	5 days	6 days	7 days		
Concentration: 500 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	1.1 ± 1.1 F	2.2 ± 1.5 EF	7.8 ± 2.8 DE	15.6 ± 2.4 CDa	24.4 ± 2.9 BCab	35.6 ± 1.8 ABCb	44.4 ± 2.9 ABb	61.1 ± 2.0 ABbc	68.9 ± 3.9 ABbc	73.3 ± 4.1 Abc	42.0	<0.01
acetic acid	1.1 ± 1.1 C	2.2 ± 1.5 C	5.6 ± 2.9 BC	12.2 ± 3.6 Bab	32.2 ± 5.2 Aab	47.8 ± 6.0 Aab	54.4 ± 6.3 Ab	62.2 ± 5.2 Abc	68.9 ± 5.9 Abc	81.1 ± 5.1 Aabc	40.2	<0.01
trans-anethole	0.0 ± 0.0 D	1.1 ± 1.1 D	3.3 ± 1.7 CD	7.8 ± 2.8 Cab	17.8 ± 2.8 Bb	32.2 ± 4.0 ABb	41.1 ± 4.2 ABb	48.9 ± 5.1 ABc	52.2 ± 5.7 Ac	64.4 ± 6.9 Ac	40.2	<0.01
furfural	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 C	2.2 ± 1.5 Cb	14.4 ± 3.4 Bb	33.3 ± 5.8 Ab	40.0 ± 6.2 Ab	51.1 ± 6.8 Ac	61.1 ± 6.3 Ac	68.9 ± 6.3 Ac	83.0	<0.01
(*E*)-2-decenal	2.2 ± 1.5 C	4.4 ± 1.8 C	7.8 ± 2.8 BC	16.7 ± 4.1 Ba	50.0 ± 8.0 Aa	72.2 ± 4.9 Aa	88.9 ± 3.5 Aa	95.6 ± 1.8 Aa	98.9 ± 1.1 Aa	100.0 ± 0.0 Aa	38.8	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 C	8.9 ± 4.2 Bab	44.4 ± 5.8 Aa	63.3 ± 6.2 Aa	80.0 ± 4.4 Aa	83.3 ± 4.7 Aab	91.1 ± 2.6 Aab	94.4 ± 2.4 Aab	104.6	<0.01
F	1.1	2.0	1.8	3.6	5.4	8.8	13.9	9.9	9.9	7.1		
P	0.39	0.10	0.12	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		
Concentration: 1000 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 D	1.1 ± 1.1 Dab	13.3 ± 2.4 Ca	28.9 ± 3.1 B	51.1 ± 3.9 ABabc	70.0 ± 3.7 Aab	78.9 ± 3.5 Aab	91.1 ± 2.0 Aa	94.4 ± 2.4 Aa	94.4 ± 2.4 Aa	152.2	<0.01
acetic acid	4.4 ± 2.4 C	7.8 ± 2.8 Ca	22.2 ± 2.8 Ba	38.9 ± 4.6 AB	70.0 ± 6.0 Aab	84.4 ± 4.4 Aa	88.9 ± 3.9 Aab	96.7 ± 1.7 Aa	98.9 ± 1.1 Aa	100.0 ± 0.0 Aa	40.9	<0.01
trans-anethole	0.0 ± 0.0 D	0.0 ± 0.0 Db	11.1 ± 3.5 Cab	24.4 ± 5.8 BC	48.9 ± 7.5 ABbc	63.3 ± 7.8 Ab	74.4 ± 5.6 Ab	84.4 ± 5.6 Aa	88.9 ± 3.5 Aa	92.2 ± 3.6 Aab	68.8	<0.01
furfural	0.0 ± 0.0 E	0.0 ± 0.0 Eb	3.3 ± 1.7 Db	21.1 ± 3.5 C	36.7 ± 2.9 BCc	40.0 ± 3.3 BCc	45.6 ± 3.8 ABc	60.0 ± 5.5 ABb	75.6 ± 6.5 ABb	82.2 ± 5.5 Ab	133.3	<0.01
(*E*)-2-decenal	3.3 ± 2.4 C	7.8 ± 3.2 Cab	18.9 ± 4.2 Ba	36.7 ± 5.3 AB	74.4 ± 6.3 Aa	93.3 ± 3.3 Aa	98.9 ± 1.1 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	38.1	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 D	2.2 ± 1.5 Dab	7.8 ± 2.2 Cab	27.8 ± 5.5 B	54.4 ± 5.0 ABabc	76.7 ± 5.3 Aab	85.6 ± 4.1 Aab	91.1 ± 3.1 Aa	93.3 ± 2.9 Aa	94.4 ± 2.4 Aa	88.6	<0.01
F	2.6	3.6	4.8	1.9	5.3	14.6	19.5	12.8	6.7	4.7		
P	0.04	<0.01	<0.01	0.11	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		

Within each column, means followed by the same lowercase letter are not significantly different; *df* = 5, 53; Tukey-Kramer HSD test at *P* = 0.05. Within each row, means followed by the same uppercase letter are not significantly different; *df* = 9, 89; Tukey-Kramer HSD test at *P* = 0.05. Where no letters exist, no significant differences were recorded. Where dashes exist, no analysis was conducted

Table 5 Mean mortality (% ± SE) of *T. granarium* larvae over selected exposure intervals on wheat, treated with 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal

Exposure	4 h	8 h	16 h	1 day	2 days	3 days	4 days	5 days	6 days	7 days		
Concentration: 500 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 D	0.0 ± 0.0 D	3.3 ± 1.7 D	10.0 ± 2.4 Ca	16.7 ± 3.3 BCa	24.4 ± 4.1 ABCab	28.9 ± 3.5 ABab	37.8 ± 5.2 ABab	41.1 ± 5.4 ABab	46.7 ± 4.4 Aab	33.0	<0.01
acetic acid	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 C	4.4 ± 1.8 BCab	5.6 ± 1.8 BCabc	11.1 ± 3.5 ABbc	14.4 ± 4.1 ABb	18.9 ± 3.5 Ab	22.2 ± 4.0 Ab	24.4 ± 4.1 Abc	16.0	<0.01
trans-anethole	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	1.1 ± 1.1 BCb	1.1 ± 1.1 BCbc	3.3 ± 1.7 ABc	4.4 ± 2.4 ABc	5.6 ± 3.4 ABc	7.8 ± 3.2 Ac	8.9 ± 4.2 Ad	2.8	<0.01
furfural	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 C	0.0 ± 0.0 Cb	0.0 ± 0.0 Cc	2.2 ± 1.5 BCc	2.2 ± 1.5 BCc	5.6 ± 1.8 ABc	8.9 ± 2.6 ABc	11.1 ± 3.1 Acd	8.1	<0.01
(*E*)-2-decenal	0.0 ± 0.0 D	0.0 ± 0.0 D	0.0 ± 0.0 D	1.1 ± 1.1 Db	8.9 ± 2.6 Cab	33.3 ± 4.4 Ba	52.2 ± 2.8 ABa	68.9 ± 3.1 ABa	82.2 ± 2.2 Aa	87.8 ± 2.2 Aa	143.6	<0.01
(*E*, *E*)-2,4-decadienal	0.0 ± 0.0 C	1.1 ± 1.1 C	2.2 ± 1.5 C	5.6 ± 2.4 BCab	8.9 ± 2.6 Bab	26.7 ± 4.1 Aab	46.7 ± 5.3 Aa	55.6 ± 5.8 Aab	65.6 ± 5.8 Aab	80.0 ± 5.5 Aa	46.1	<0.01
F	-	1.0	1.9	5.1	7.5	13.6	24.0	24.9	18.9	20.4		
P	-	0.43	0.11	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		
Concentration: 1000 µl/kg wheat												
Compounds											*F*	*P*
2-undecanone	0.0 ± 0.0 Fb	1.1 ± 1.1 EFab	5.6 ± 1.8 DEab	13.3 ± 3.7 CDab	24.4 ± 5.3 BCbc	46.7 ± 5.5 ABabc	61.1 ± 6.8 Aabc	70.0 ± 5.3 Aab	73.3 ± 5.0 Aa	75.6 ± 5.0 Aa	45.1	<0.01
acetic acid	0.0 ± 0.0 Db	0.0 ± 0.0 Db	1.1 ± 1.1 Dbc	3.3 ± 1.7 Dbc	14.4 ± 3.4 Cc	31.1 ± 3.9 Bbc	42.2 ± 5.5 ABbc	61.1 ± 5.1 ABab	70.0 ± 4.7 ABa	76.7 ± 4.4 Aa	88.7	<0.01
trans-anethole	0.0 ± 0.0 Cb	0.0 ± 0.0 Cb	0.0 ± 0.0 Cc	1.1 ± 1.1 BCc	2.2 ± 1.5 BCd	6.7 ± 3.3 ABCd	11.1 ± 4.2 ABd	16.7 ± 5.3 Ac	23.3 ± 6.2 Ab	28.9 ± 7.7 Ab	8.7	<0.01
furfural	0.0 ± 0.0 Db	0.0 ± 0.0 Db	0.0 ± 0.0 Dc	2.2 ± 1.5 Dc	14.4 ± 4.1 Cc	25.6 ± 4.8 BCc	32.2 ± 3.2 ABc	41.1 ± 3.5 ABb	51.1 ± 3.9 ABa	54.4 ± 3.8 Aa	60.7	<0.01
(*E*)-2-decenal	0.0 ± 0.0 Db	1.1 ± 1.1 Dab	5.6 ± 1.8 Cab	27.8 ± 5.7 Ba	66.7 ± 7.5 Aab	83.3 ± 6.9 Aab	94.4 ± 3.4 Aab	98.9 ± 1.1 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	81.6	<0.01
(*E*, *E*)-2,4-decadienal	3.3 ± 1.7 Ca	5.6 ± 2.4 Ca	15.6 ± 3.4 Ba	33.3 ± 4.1 Ba	90.0 ± 2.9 Aa	98.9 ± 1.1 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	100.0 ± 0.0 Aa	45.7	<0.01
F	4.0	3.3	10.9	15.0	20.4	21.6	23.7	18.9	12.6	9.9		
P	<0.01	0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01		

Within each column, means followed by the same lowercase letter are not significantly different; $df = 5, 53$; Tukey-Kramer HSD test at $P = 0.05$. Within each row, means followed by the same uppercase letter are not significantly different; $df = 9, 89$; Tukey-Kramer HSD test at $P = 0.05$. Where no letters exist, no significant differences were recorded. Where dashes exist, no analysis was conducted

monoterpene *trans*-anethole was more toxic to adults than larvae as wheat protectant. In the same frame, the pyrethroid insecticides exhibit different performance towards adults and larvae according to the method of application, i.e., surface treatment vs. grain protectants (Athanassiou et al. 2015, Kavallieratos et al. 2019b, 2021). The problem of performance of insecticides and substances that exhibit insecticidal properties within the developmental stage of *T. molitor* could be faced up with the combination of compounds that exhibit elevated mortality levels to movable stages of this species which are responsible for the infestations of stored products. Further experimentation is needed to clarify this issue.

Our study shows that the overall mortality of *T. granamum* adults ranged between 82.2 and 100% at 1000 μl/kg wheat after 7 days of exposure. In fact, (*E*)-2-decenal killed all exposed adults and larvae 6 days post-exposure. This is one of the most important findings since it is well documented that the larva is the most harmful stage of *T. granarium* due to the rapid population growth on numerous grains (Athanassiou et al. 2016; Kavallieratos et al. 2017b) or non-grain commodities (Kavallieratos et al. 2019a). Furthermore, larvae, contrary to adults, are tolerant to several insecticides of chemical or natural origin either as grain protectants (i.e., cypermethrin, deltamethrin, pirimiphos-methyl, SilicoSec, *s*-methoprene, spinosad) (Kavallieratos et al. 2017a) or as surface treatments (i.e., alpha-cypermethrin, deltamethrin, thiamethoxam) (Athanassiou et al. 2015; Kavallieratos and Boukouvala 2018). So far, only few chemical insecticides have provided elevated management of *T. granarium* larvae and adults such as chlorfenapyr, pirimiphos-methyl and a mixture of acetamiprid plus d-tetramethrin plus piperonyl butoxide (Kavallieratos et al. 2017a, Kavallieratos and Boukouvala 2018, 2019). Surprisingly, there is limited knowledge regarding the efficacy of botanicals as effective grain protectants against *T. granarium*. For example, a recent study Kavallieratos et al. (2020a) showed that although isofuranodiene provided > 96% mortality to *T. granarium* adults, it killed < 38% of the exposed larvae 7 days post-exposure. However, similar findings to the current study have been reported by Kavallieratos et al. (2020b) who found that the EO of *Mentha longifolia* (L.) Huds. (Lamiales: Lamiaceae) completely (100%) suppressed larvae and adults of *T. granarium* after 2 days and 16 h of exposure, respectively, at 1000 μl/kg wheat. In the same study, the EO of *Dysphania ambrosioides* (L.) Mosyakin and Clemants (Caryophyllales: Amaranthaceae) was also highly effective by killing all adults of *T. granarium* and > 95% of larvae at the same concentration 2 and 4 days post-treatment. Therefore, it becomes evident that botanicals, either as single compounds or as constituents of EOs, are effective agents against this species and compatible to synthetic insecticides.

The tested substances exhibit a safe profile within mammalian systems, an issue that may enable them to be endorsed as components of a biorational strategy in the frame of integrated management of infestations and losses of stored food commodities. In specific, furfural, *trans*-anethole, 2-undecanone, and (*E*)-2-decenal are EU Food Improvement Agents (EUR Lex 2012) as well as food additives Generally Recognized as Safe (GRAS) in the USA (USDHHS 2021). 2-undecanone is a natural flavor ingredient that is not genotoxic while it does not meet safety issues regarding sensitization of the skin (Api et al. 2019). The compound acetic acid is a normal body metabolite in mammals and can be found naturally in a wide variety of foods. It is frequently used as a preservative and its commonest use is as vinegar (Pravasi 2014). (*E*, *E*)-2,4-decadienal is used as a synthetic flavoring and fragrance material. Toxicity studies on certain strains of male and female rats and mice determined the no-observed-adverse-effect level at 100 mg/kg body weight. Moreover, (*E*, *E*)-2,4-decadienal was not mutagenic in vitro or in vivo (Chan 2011). (*E*, *E*)-2,4-decadienal and (*E*)-2-decenal are safe at maximum use level of 5 mg/kg feed for all animal species. No safety concern would arise for the consumer from the use of these compounds up to the highest safe levels in feed (Bampidis et al. 2019).

Our study gives the first insight into the use of 2-undecanone, acetic acid, *trans*-anethole, furfural, (*E*)-2-decenal, and (*E*, *E*)-2,4-decadienal as possible natural insecticides against different developmental stages of *T. molitor* and *T. granarium* infesting stored wheat. Among these substances, 2-undecanone was the most effective against *T. molitor* adults and larvae while (*E*)-2-decenal was 100% lethal for both life stages of *T. granarium* followed by (*E*, *E*)-2,4-decadienal. Additional studies are necessary to clarify the spectrum of efficacy of the tested substances by including more stored-product insect species and food commodities under different levels of temperature and RH levels. Currently, we are in the process of testing the toxicity of mixtures of the compounds to demonstrate any synergic, antagonistic, or additive effects, along with the ultrastructural deformations on stored-product insects.

Availability of data and materials The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Author contribution NN and NGK conceived and designed research. AS, EPN, and MCB conducted the experiments. NGK and EPN analyzed data. NN, NGK, and EPN drafted the manuscript. All authors read, edited, and approved the manuscript.

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Declarations

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

Consent to participate Not applicable

Consent for publication Not applicable

Conflict of interest The authors declare no competing interests.

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