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## Evaluation of propylene oxide fumigation against *Ephestia cautella* (Walker, 1863) (Lepidoptera: Pyralidae) in dried figs and hazelnuts

Kuru incir ve fındıkta *Ephestia cautella* (Walker, 1863) (Lepidoptera: Pyralidae)'ya karşı propilen oksit fümigasyonunun değerlendirilmesi

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### ABSTRACT

This study aims to investigate the potential use of propylene oxide (PPO) for rapid control of the fig moth, *Ephestia cautella* (Walker, 1863) (Lepidoptera: Pyralidae). To this end, the biological efficacy of PPO (10 µl/l) against all biological stages of the fig moth was evaluated for a short exposure period (4 hours) under three different conditions: PPO alone (normal pressure), PPO+vacuum (100 mm Hg low pressure), and PPO+CO<sub>2</sub> (92% CO<sub>2</sub> concentration) in the absence and presence of dried figs and hazelnuts. In the absence of commodities, the biological tests showed 100% mortality rates in all biological stages of *E. cautella*, except for the pupa stage, when using the PPO+vacuum treatment. However, the PPO alone and PPO+CO<sub>2</sub> treatments did not achieve a 100% mortality rate for the biological stages of *E. cautella*. Conversely, in the presence of dried figs, the mortality rates for all biological stages of *E. cautella* ranged from 19.7% to 100% in the PPO+vacuum and PPO+CO<sub>2</sub> treatments. As for the shelled hazelnuts, all PPO treatments resulted in low mortality rates ranging from 0.7% to 10.6% with PPO+vacuum and PPO+CO<sub>2</sub> treatments. In conclusion, the study suggests that the PPO+vacuum treatment can have a viable potential for rapid insect control, particularly in dried figs, making it suitable for quarantine applications.

### INTRODUCTION

Infestation of storage pests during drying and storage of dried figs and hazelnuts can cause significant problems in the dried fig and hazelnut industry (Yıldız 2013). Addressing the problem of storage pests is crucial to ensure the sustainable growth and development of the dried fig and hazelnut

sector in Türkiye and to enable the country to maintain its status as a leading producer and exporter of dried figs and hazelnuts worldwide. The following pests can be mentioned as significant threats to stored dried figs and hazelnuts: Fig moth (*Ephestia cautella* Walk), Indian meal moth (*Plodia*

*interpunctella* Hübner; Pyralidae: Lepidoptera), dried fruit beetle (*Carpophilus* spp.; Nitidulidae: Coleoptera), sawtoothed grain beetle (*Oryzaephilus surinamensis* L.; Silvanidae: Col.) and dried fruit mite (*Carpoglyphus lactis* L.; *Carpoglyphidae*: Acari) (Turanlı 2003). In particular, the fig moth, *Ephesia cautella* (Walker) (Lepidoptera: Pyralidae), which is among the stored dried fruit pests and nuts, rapidly infests the products and causes significant damage by forming high populations (Tripathi 2018). Its larvae feed on the fruit, leading to a reduction in the fruit's quality (Küçüktopçu 2023). In addition, the presence of the pest's body residues, excreta, and secreted web-like substances further contributes to a significant reduction in the quality and commercial value of the product (Bilgili 2015, Celik et al. 2008, Ferizli and Emekci 2013). Effective measures to control these storage pests are essential to preserve the integrity of dried figs and hazelnuts during storage, transport, and export, thus safeguarding the reputation of the industry and its economic importance for the country.

Türkiye is known worldwide as the largest producer and exporter of dried figs (Özer 2020). With a significant share of 58% and a production volume of 85.500 tons in 2020-2021, it is at the forefront of global dried fig production (INC 2021). Such a significant production volume underlines dominant position of the country in the dried fig sector. Similarly, Türkiye, which accounts for about 80% of the world's hazelnut production, is the world's leading country in hazelnut production and export, producing 684 thousand tons of hazelnuts on 7.4 million acres of land in 2021 (Uzundumlu et al. 2022). In 2021, the hazelnut production area in Türkiye was 7.4 million da and the total hazelnut production was 684 thousand tons (Bayyurt and Kocakoç 2023). Hazelnuts are used as a basic ingredient in various food products such as chocolate, biscuits, confectionery desserts, cakes, ice cream, meals, and salads. This shows that hazelnut is a versatile material and plays an important role in the food industry. As a result, ensuring the sustainability and growth of the dried fig and hazelnut sectors has become a critical priority in supporting the agricultural and economic progress of Türkiye.

Methyl Bromide (MeBr) has been widely used in controlling stored product pests due to its broad-spectrum activity, low cost, and fast insect-killing ability (Fields and White 2002). However, its application has been restricted due to its ozone-depleting effect on the ozone layer (Schneider et al. 2003, Tütüncü and Emekci 2014). Under the Montreal Protocol, MeBr has been banned in developed countries since 2005 and in Türkiye since 2007, except for specific quarantine and pre-shipment applications. Given the restrictions on the use of MeBr and its impact on the environment, research

into possible methods of replacing MeBr is becoming increasingly crucial. The hard-shell fruit and nut industry relies mainly on phosphine as a chemical fumigant to prevent insect infestation after harvest. However, due to the carcinogenic effect of phosphine (Alavanja et al. 1990, Garry et al. 1990), its flammability (Ohtani et al. 1989), insect resistance (Benhalima et al. 2004, Daglish et al. 2014, Gautam et al. 2016, Sağlam et al. 2015) and the requirement for a longer exposure time (4 to 6 days or more), the use of phosphine is not suitable for quarantine applications (Isikber et al. 2006). The lack of readily available potentials that can quickly control insect infestations has led to significant adverse effects on the dried fig and hazelnut industry due to the loss of MeBr. Therefore, there is a critical need to develop new fumigants that can achieve rapid insect mortality (exposure periods of less than one day), especially for quarantine treatments.

Propylene oxide (PPO), a clear, colorless, volatile liquid fumigant, with an ether-like odor and 35 °C boiling point, is a significant organic chemical raw material (Weast et al. 1986). In the study by Meylan et al. (1986), the impact of PPO as a fumigant on human health and the environment was investigated. They reported that PPO poses significantly lower environmental risks compared to MeBr. They also observed that PPO has no ozone-depleting properties and rapidly converts to non-toxic propylene glycol in the soil and human stomach. The main disadvantage of PPO is its flammability in air, which ranges from 3% to 37%. Therefore, precautions need to be taken to reduce the fire risk to ensure the safe use of PPO. One such measure is using PPO gas under low pressure or in an atmosphere enriched with carbon dioxide (CO<sub>2</sub>). Recent studies have shown that PPO has an increased fumigation potential when used at low pressure (100 mm Hg) for a short exposure period (Isikber et al. 2006, Isikber et al. 2012, Navarro et al. 2004). Although previous laboratory studies have demonstrated that PPO can be effective against stored product pests at low pressure and enriched-CO<sub>2</sub> atmosphere for short-term exposure, there is limited research available in the literature regarding the efficacy of PPO with vacuum and CO<sub>2</sub> against stored product Lepidopteran insects in the presence of the commodity.

This study aimed to investigate the potential use of PPO for the rapid control of the fig moth as a potential fumigant to MeBr. For this reason, the biological efficacy of PPO alone at a concentration of 10 µl/l, 10 µl/l PPO+vacuum (100 mm Hg low pressure), and 10 µl/l PPO+CO<sub>2</sub> (92% CO<sub>2</sub> concentration) was investigated against all developmental stages of *E. cautella* (eggs, larvae, pupae and adults) in the absence and presence of dried figs and hazelnuts.

## MATERIALS AND METHODS

### *Products used in biological tests*

In the biological tests, sun-dried Sarılop (Calimyrna) (*Ficus carica* L.) (Moraceae) dried fig variety with  $21\% \pm 1$  moisture content and shelled Çakıldak hazelnut variety, which are hybrids of *Corylus avellana* L. and *Corylus maxima* Mill. (Betulaceae), containing 10-12% moisture content was utilized.

### *The source and rearing of insect culture used in biological tests*

In biological tests, all biological stages of *Ephestia cautella* were used. The main material of the *E. cautella* culture used in biological tests was obtained from Namık Kemal University, Faculty of Agriculture, Plant Protection Department Laboratory.

For rearing of *E. cautella* culture, a food mixture was prepared to consist of 350 g cornmeal for every 2 kg of wheat bran, along with 350-400 ml of glycerin, 450-500 ml of glucose syrup, and 1 teaspoon of inactive yeast. The wheat bran and cornmeal were kept in a deep freezer at  $-20\text{ }^{\circ}\text{C}$  for 3-4 days to prevent insect infestation. The ingredients were thoroughly mixed by hand and then processed in a mixer. To obtain the eggs of adult *E. cautella*, the adults were transferred from 3-L culture jars using the laboratory type of vacuum pump (KNF, Germany) to a 3-L culture glass jar, and the culture jar's mouth was covered with a mesh. After keeping the culture jar with mixed-sex adults in an air-conditioning cabinet for one day, the jar was inverted to allow the eggs to fall onto a piece of paper. The collected eggs ranging from 400 to 500 were added to 3-L jars containing 350-400 g of food and covered with a mesh that allowed air to pass through. The culture jars were placed in completely dark conditions within an incubator at a constant temperature of  $30\pm 1\text{ }^{\circ}\text{C}$  and  $65\pm 5\%$  humidity. Throughout the study, the insect culture was monitored daily, and these procedures were repeated to ensure continuity.

### *Fumigant*

The fumigant PPO was obtained from SERVA Electrophoresis GmbH company (Heidelberg, Germany) with  $>99\%$  purity (CAS no. 75569, Cat. no. 33715). PPO was transferred into a 100 ml glass bottle and securely sealed with a septum. During the treatment phase, a predetermined amount of PPO was drawn from the glass bottle using a gas-tight micro syringe (Hamilton, Switzerland).

### *Carbon dioxide (CO<sub>2</sub>) gas*

Carbon dioxide (CO<sub>2</sub>) used in biological tests has been supplied by Linde Gas (Ankara, Türkiye) Company in a pressurized steel cylinder with a purity of 99%.

### *Fumigation chamber*

The fumigation chamber consists of 3-L glass jars, each equipped with metal lids, a metal tube housing an inlet and an outlet hole. Two silicone flexible hoses, each with a length of 5 cm and a diameter of 0.62 cm, have been placed over the inlet and outlet metal tubes. These silicone hoses were securely attached using metal clamps to create a gas-tight environment. To ensure no gas leakage, silicone is carefully applied around the edges of the metal lids before closing them. This gas-tight system allows to use of vacuum and PPO safely without any leakage.

### *Biological tests conducted in a commodity-free atmosphere*

In biological experiments, 20 adults, pupae (1 to 2 days old), late-stage larvae (28 to 32 days old), and 50 eggs (1 to 2 days old) of *E. cautella* were used. Each developmental stage was carefully placed inside separate 50 ml glass vials. Food medium was added to the vials to meet the larvae's dietary needs, filling approximately 1/3 of their volume (equivalent to 10 g for 50 ml vials). To allow the PPO gas to enter the vials and prevent the insects from escaping, the mouths of the vials were covered with a fine muslin mesh and securely fastened with rubber bands. Afterwards, each insect vial was placed in a 3-L glass jar with a metal lid (fumigation chamber). Thus, the tested insects and PPO gas were kept in a gas-tight atmosphere in the fumigation chamber. To apply PPO under a low-pressure (vacuum) atmosphere, the vacuum pump (KNF, Germany) evacuated the air from the 3-L fumigation chamber, effectively reducing the pressure to 100 mm Hg. To ensure accurate monitoring of the low-pressure level inside the fumigation chamber, the low-pressure level was measured using a Celesco model SE-2000 vacuum gauge. After achieving the desired low-pressure level, PPO at 10  $\mu\text{l/l}$  was injected into the fumigation chamber using a 50  $\mu\text{l}$  gas-tight micro syringe (Hamilton Company, Bonaduz, Switzerland).

For applying PPO under a CO<sub>2</sub> atmosphere, we first established a low pressure of 60.8 mm Hg in the fumigation chamber. Subsequently, CO<sub>2</sub> gas was circulated within the fumigation chamber until the pressure returned to a normal atmospheric level. The CO<sub>2</sub> gas concentration inside the fumigation chamber was measured throughout this process using a precise CO<sub>2</sub>/O<sub>2</sub> measurement device (CheckPoint, PBI-Dansensor, Denmark). Once the desired 92% CO<sub>2</sub> level was achieved, PPO at a 10  $\mu\text{l/l}$  c was injected into the fumigation chamber using a 50  $\mu\text{l}$  micro syringe. In applying PPO alone (under normal pressure), 10  $\mu\text{l/l}$  concentration of PPO was directly injected into the fumigation chamber. In the biological tests, all stages of *E. cautella* were exposed to a combination of 10  $\mu\text{l/l}$  PPO alone, 10  $\mu\text{l/l}$  PPO with 92%

CO<sub>2</sub>, and a vacuum of 100 mm Hg for 4 hours. In addition, all developmental stages of *E. cautella* were exposed to separate treatments of vacuum (100 mm Hg low pressure) and CO<sub>2</sub> gas (92% CO<sub>2</sub> atmosphere) for 4 hours without PPO treatment. After completing the biological tests, the lids of the fumigation chamber were quickly closed and were kept in a completely dark climate chamber with a temperature of 26±1 °C and a relative humidity of 65±5% for 4 hours. To ensure the reliability and robustness of the results, each treatment of PPO was performed with 4 replicates and 4 control groups were included for each treatment.

*Biological tests conducted in the presence of commodity*

The biological tests conducted in the presence of products (figs and hazelnuts) were carried out following the same experimental procedures as those conducted without commodities. The only difference was using fumigation chambers formed within 3-liter glass jars with metal lids, where 1.3 kg of dried figs and shelled hazelnuts were placed.

*Data processing and analysis*

Following each treatment, the larvae, pupae, and adult insects were transferred to 200-milliliter jars containing standard diets. These containers were maintained at a temperature of 26 ± 1°C and a relative humidity of 70 ± 5% until they were inspected for mortality. The eggs, placed on Perspex slides, were also subjected to the same environmental conditions until the sites where they were laid were examined to determine egg hatch rates. Mortality counts for adults were made 4-5 d after exposure; for larvae, they were based on those insects that had failed to pupate 9 d after exposure; pupal mortality was based on those pupae that failed to produce adults 9 d after exposure; and egg hatch was counted 7 d after treatment.

Mortality data were corrected using Abbott's formula (Abbott 1925). All mortality data for each biological stage in PPO treatments were normalized using arcsine transformation. Subsequently, a two-way ANOVA was conducted using the GLM Procedure of SAS/STAT® 12.1 (SAS 2012), with PPO treatment and biological stage as the main factors. Mean mortality percentages for each biological stage and PPO treatment were separated using Tukey's HSD (Honestly Significant Difference) test.

**RESULTS**

*Mortality of life stages of Ephestia cautella exposed to Propylene oxide treatments in a commodity-free atmosphere*

The results showed that only PPO+vacuum achieved complete mortality (100%) of all life stages of *E. cautella* except its pupa stage, while PPO alone and PPO+CO<sub>2</sub> did not result in 100% mortality of life stages (Table 1). In contrast, PPO+vacuum exhibited significantly higher efficacy against all life stages than PPO alone and PPO+CO<sub>2</sub> (except the adult stage for PPO+CO<sub>2</sub>). Similarly, PPO+CO<sub>2</sub> achieved significantly higher efficacy against all life stages than PPO alone. PPO+CO<sub>2</sub> and PPO alone generally produced very low larva and pupa stage mortalities, ranging from 8 to 38%. These results indicated that the larva and pupa were the most tolerant stages for PPO treatments, whereas the adult and egg were the most susceptible.

It was observed that there was no statistically significant difference in mortality rates of all life stages of *E. cautella* exposed to only 92% concentration of CO<sub>2</sub>, 100 mm Hg vacuum and control treatments (Table 2). Only vacuum (100 mm Hg low pressure) and CO<sub>2</sub> gas (92% CO<sub>2</sub> concentration) treatment without PPO for 4 hours caused very low mortality levels of all life stages, ranging from 1.3% to 18.8%.

**Table 1.** Corrected percentage mortality (%) of life stages of *Ephestia cautella* exposed to PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> for 4 hours in a commodity-free atmosphere

PPO treatments	Corrected Percentage Mortality (%) ± Standard Error				F and P value
	Adult	Larva	Egg	Pupa	
PPO	74.7±3.4 Ba*	8.0±1.3 Cb	77.7±2.4 Ca	18.5±2.9 Cb	F <sub>3,12</sub> =146.2 P<0.0001
PPO+vacuum	100.0±0.0 Aa	100.0±0.0 Aa	100.0±0.0 Aa	87.7±4.4 Ab	F <sub>3,12</sub> =29.5 P<0.0001
PPO+CO <sub>2</sub>	97.3±1.5 Aa	14.7±0.0 Bd	89.0±0.5 Bb	38.5±2.5 Bc	F <sub>3,12</sub> =183 P<0.0001
Control	6.3±2.4	8.8±1.3	13.0±1.3	18.8±2.4	
F and P value	F <sub>2,9</sub> =37.1 P<0.0001	F <sub>2,9</sub> =2054.3 P<0.0001	F <sub>2,9</sub> =204.4 P<0.0001	F <sub>2,9</sub> =80.3 P<0.0001	For PPO treatment: F <sub>2,36</sub> =544.31, P<0.0001 For Biological stage: F <sub>3,36</sub> =275.06, P<0.0001 For PPO treatment*Biological stage: F <sub>6,36</sub> =52.08, P<0.0001

\*One-way ANOVA was applied to the mortality data for PPO treatments in each column and biological stages in each row. This means that a row with the same lower-case letter and a column with the same upper-case letter did not differ significantly (Tukey's HSD test at 5% level).

**Table 2.** Percentage mortalities (%) of all life stages of *Ephestia cautella* exposed to 92% CO<sub>2</sub>, 100 mm Hg vacuum alone and control treatment for 4 hours

PPO treatments	Percentage Mortality (%) ± Standard Error			
	Adult	Larva	Egg	Pupa
Control	3.8±1.3 A*	0.0±0.0 A	4.0±0.0 A	16.3±1.3 A
100 mm Hg vacuum	5.0±2.0 A	1.3±1.3 A	6.5±0.5 A	18.8±1.3 A
92% CO <sub>2</sub>	3.8±1.3 A	1.3±1.3 A	5.5±0.5 A	17.5±1.4 A
F and P value	F <sub>2,9</sub> =0.05 P=0.9491	F <sub>2,9</sub> =0.50 P=0.6224	F <sub>2,9</sub> =1.42 P=0.055	F <sub>2,9</sub> =0.90 P=0.4402

\*One-way ANOVA was applied to the mortality data for the treatments. This means that a column with the same upper-case letter did not differ significantly (Tukey's HSD test at 5% level).

*Mortality of all life stages of Ephestia cautella exposed to PPO treatments in the presence of dried figs*

Only PPO+vacuum treatment resulted in 100% mortality of *E. cautella* adults in the presence of dried figs (Table 3). PPO+vacuum achieved higher mortality rates of all life stages of *E. cautella* than PPO alone and PPO+CO<sub>2</sub> (except the egg for PPO+CO<sub>2</sub>). Similarly, PPO+CO<sub>2</sub> caused higher mortality rates in all life stages of *E. cautella* than PPO alone. PPO+CO<sub>2</sub> and PPO alone generally produced very low mortalities of larva and pupa stage, ranging from 1 to 40%, while they resulted in relatively high mortality rates of *E. cautella* adults and eggs, ranging from 64 to 94%. 10

ul/1 PPO+vacuum was insufficient to kill 100% of *E. cautella* larvae and pupae, even though it caused 100% or close to 100% mortality rates of *E. cautella* adults and eggs (Table 3).

*Mortality of all life stages of Ephestia cautella exposed to PPO treatments in the presence of shelled hazelnut*

PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> treatments for 4 hours of exposure in the presence of shelled hazelnuts caused very low mortality rates of all life stages of *E. cautella*, ranging from 0 to 10.6% (Table 4). None of the PPO treatments in the presence of shelled hazelnuts had fumigant toxicity to all life stages of *E. cautella*.

**Table 3.** Corrected percentage mortality (%) of life stages of *Ephestia cautella* exposed to PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> for 4 hours in a commodity-free atmosphere

PPO treatments	Corrected Percentage Mortality (%) ± Standard Error				F and P value
	Adult	Larva	Egg	Pupa	
PPO	64.0±2.6 Cb*	1.3±1.3 Cd	82.9±0.9 Ba	13.6±2.9 Cc	F <sub>3,12</sub> =168.33 P<0.0001
PPO+vacuum	100.0±0.0 Aa	36.8±2.1 Ad	93.6±1.5 Ab	65.2±2.9 Ac	F <sub>3,12</sub> =247.06 P<0.0001
PPO+CO <sub>2</sub>	86.6±1.5 Bb	19.7±1.3 Bd	94.1±1.0 Aa	40.9±2.9 Bc	F <sub>3,12</sub> =315.20 P<0.0001
Control	6.3±2.4	5.0±2.0	6.5±1.5	17.5±3.2	For PPO treatment: F <sub>2,36</sub> =277.40, P<0.0001 For Life stage: F <sub>3,36</sub> =622.39, P<0.0001
F and P value	F <sub>2,9</sub> =256.31 P<0.0001	F <sub>2,9</sub> =66.62 P<0.0001	F <sub>2,9</sub> =17.62 P=0.0008	F <sub>2,9</sub> =68.70 P<0.0001	For PPO treatment*Life stage: F <sub>6,36</sub> =14.76, P<0.0001

\*One-way ANOVA was applied to the mortality data for PPO treatments in each column and biological stages in each row. This means that a row with the same lower-case letter and a column with the same upper-case letter did not differ significantly (Tukey's HSD test at 5% level).

**Table 4.** Corrected percentage mortality (%) of all life stages of *Ephesia cautella* exposed to PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> for 4 hours in the presence of 1.3 kg of shelled hazelnut

PPO treatments	Corrected Percentage Mortality (%) ± Standard Error				F and P value
	Adult	Larva	Egg	Pupa	
PPO	0.0±0.0 Ac*	1.3±0.7 Abc	2.1±0.5 Aba	6.1±1.7 Aa	F <sub>3,12</sub> =10.67 P=0.0011
PPO+vacuum	1.4±0.8 Ab	1.9±0.6 Ab	4.8±0.6 Aba	10.6±2.9 Aa	F <sub>3,12</sub> =6.55 P=0.0072
PPO+CO <sub>2</sub>	0.7±0.7 Ab	1.3±0.7 Ab	5.9±0.9 Aa	7.6±1.5 Aa	F <sub>3,12</sub> =10.12 P=0.0013
Control	6.3±2.4	5.0±2.0	6.5±1.5	17.5±3.2	For PPO treatment: F <sub>2,36</sub> =3.68, P<0.0001 For Life stage: F <sub>3,36</sub> =25.35, P<0.0001 For PPO treatment*Life stage: F <sub>6,36</sub> =0.39, P<0.0001
F and P value	F <sub>2,9</sub> =1.29 P=0.3227	F <sub>2,9</sub> =0.28 P=0.7642	F <sub>2,9</sub> =9.42 P=0.0625	F <sub>2,9</sub> =0.90 P=0.4409	

\*One-way ANOVA was applied to the mortality data for PPO treatments in each column and biological stages in each row. This means that a row with the same lower-case letter and a column with the same upper-case letter did not differ significantly (Tukey's HSD test at 5% level).

## DISCUSSION AND CONCLUSION

PPO has shown promising results even in a short exposure period in the studies on its use in controlling some stored product pests (Isikber et al. 2002, Isikber et al. 2006, Navarro et al. 2004). However, it is known that various factors during the fumigation process, such as environmental conditions (temperature, relative humidity) during fumigation, the type of commodity used, and the performance of the equipment, can affect the efficacy of commercial fumigation. Understanding the insecticidal effectiveness of PPO and its penetration ability within the commodity is of great importance for using PPO gas as a commercial fumigant in the food industry.

In the present study, PPO applied at 10 µl/l concentration under 100 mm Hg low pressure was enough to obtain 100% mortality in the adult and egg stages of *E. cautella*, whereas it was not enough to reach complete mortality in its pupa stage. In parallel to these results, Isikber et al. (2004) reported that the LC<sub>99</sub> toxicities of PPO+vacuum for the adult, egg, larvae, and pupa of *E. cautella* were 5.7, 6.1, 13.0, and 14.4 µl/l, respectively. When PPO was applied under a vacuum or at a high CO<sub>2</sub> concentration, they had greater fumigant toxicity against all life stages of *E. cautella* than PPO alone. Similarly, Navarro et al. (2004) reported that when PPO was used in combination with 100 mm Hg vacuum and 92% concentration of CO<sub>2</sub>, the mortality rates of all life stages of *T. castaneum* except the egg stage were significantly higher compared to those of PPO applied alone. The results from our bioassay experiments, where we subjected all life stages to either low pressure or CO<sub>2</sub> alone for 4 hours, demonstrated minimal mortality rates akin to those observed in the control group. Consequently, our

findings assert that the combination of low pressure and CO<sub>2</sub> substantially heightened the potency of PPO against *E. cautella*. Notably, the utilization of low pressure or CO<sub>2</sub> in isolation exhibited negligible impact on the insects' well-being. Navarro et al. (2004) also reported that a 100 mm Hg vacuum and 92% concentration of CO<sub>2</sub> have a synergistic effect on the toxicity of PPO against insect pests. Previous studies conducted on several fumigants, particularly MeBr, and phosphine, have also demonstrated that insecticidal effectiveness could be enhanced by employing vacuum fumigation or blending with CO<sub>2</sub> (Calderon and Leesch 1983, Donahay and Navarro 1989, Monro et al. 1966).

Based on our results from a single concentration test, the larva and pupa were the most tolerant stages when exposed to PPO alone for 4 hours. Similarly, Isikber et al. (2017) reported that the eggs and adults of *E. cautella* were the most sensitive life stage to PPO with an LC<sub>99</sub> value of 16.52 and 18.91 µl/l for 4h, whereas pupae and larvae were the most tolerant with an LC<sub>99</sub> value of 134.06 and 48.72 µl/l, respectively. The eggs and pupae of stored-product insects are commonly recognized to exhibit greater tolerance compared to larvae and adults when exposed to MeBr (Athanassiou et al. 2015), phosphine (Aulicky et al. 2015), carbonyl sulfide (Plarre and Reichmuth 1996) and sulfuranyl fluoride (Athanassiou et al. 2012). Consequently, achieving effective control over the eggs of stored-product insects proves challenging using the majority of commonly employed fumigants and contact insecticides. Typically, significantly extended exposure periods are necessary to adequately manage the eggs. In contrast to phosphine, MeBr, and sulfuranyl fluoride, PPO is easy to kill *E. cautella* eggs during short exposure periods, which is particularly

important in providing a potential for fumigants that have a weak effect or a long exposure time on eggs.

There were significant differences in the toxicities of PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> treatments against all life stages of *E. cautella* in the presence of dried figs. PPO+vacuum achieved higher mortality rates of all life stages of *E. cautella* than PPO alone and PPO+CO<sub>2</sub> (except the egg for PPO+CO<sub>2</sub>). Even though 10 µl/l PPO+vacuum caused 100% or close to 100% mortality rates of *E. cautella* adults and eggs, it was not enough to kill 100% of *E. cautella* larvae and pupae. It shows that there is a need to increase PPO concentration to achieve the complete mortality of all life stages of *E. cautella* in the presence of dried figs. Generally, mortality rates of all life stages of *E. cautella* decreased when PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> were applied in the presence of dried figs. This decline could likely be attributed to the pronounced gas adsorption by the commodity, leading to a reduction in the accessible concentration of active gas. This phenomenon was also noted in tests of various fumigants on *Tribolium castaneum* (Herbst) (Punj 1969); the LC<sub>50</sub> values indicated a notable increase, ranging from 2.7 to 7.5 times, during fumigation exercises when paddy and groundnut kernels were present in comparison to treatments without these commodities.

On the other hand, in the presence of shelled hazelnuts, PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub>, and 100 mm Hg vacuum treatments gave very low mortality rates of all life stages of *E. cautella* ranging from 0 to 10.6%. There were dramatic decreases in mortality rates of all life stages of *E. cautella* when PPO alone, PPO+vacuum, and PPO+CO<sub>2</sub> were applied in the presence of hazelnuts. The reduction in mortality rates of all life stages was much higher in the presence of hazelnuts than in the presence of dried figs. This situation may be due to the higher PPO absorption rate of hazelnuts compared to those of dried figs. Isikber et al. (2006) documented significant sorption of PPO by oily products like peanuts, almonds, and walnuts following a 5-hour exposure period. The level of sorption was notably substantial ranging between 87% and 91% of the initial concentration. Zettler et al. (2003) also found that PPO sorption in almonds, pecans, and walnuts reached 97.3%, 99.2%, and 98.6% respectively, within 48 hours of the start of the fumigation process. In contrast to the nuts, Isikber et al. (2012) noted that PPO was absorbed to a lesser extent in dried figs (50% of the initial PPO concentration). These findings indicate that the toxicity of PPO to insects of stored products varies depending on the type of fumigated commodity, as the type of commodity can strongly influence the absorption of PPO. The results of this study show that the combination of PPO with 100 mm Hg low pressure and 92% CO<sub>2</sub> can be used as a potential alternative fumigant for rapid control of insect contamination, especially in dried figs. Nonetheless, additional investigations are imperative to

acquire comprehensive insights into PPO's ability to permeate commodity masses, its potential phytotoxicity, and its ramifications for commodity quality. To realize the practical application of PPO on a commercial scale, precise treatment protocols need to be established to effectively control insect pests that infest stored dried figs.

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## Author's Contributions

Authors declare the contribution of the authors is equal.

## Statement of Conflict of Interest

The authors have declared no conflict of interest.

## ÖZET

Bu çalışmanın temel amacı, propilen oksit (PPO)'in *Ephesia cautella*'nın hızlı kontrolü için potansiyel kullanımını araştırmaktır. Bu amaçla, PPO'nun (10 µl/l) incir güvesinin tüm biyolojik aşamalarına (yumurta, larva, pupa ve ergin) karşı biyolojik etkinliği, ürünlü (kuru incir ve fındık) ve ürünsüz ortamlarda üç farklı koşul altında [10 µl/l PPO tek başına (normal basınç), 10 µl/l PPO+vakum (100 mm Hg düşük basınç) ve 10 µl/l PPO+CO<sub>2</sub> (%92 CO<sub>2</sub> konsantrasyonu)] kısa maruz kalma süresince (4 saat) değerlendirilmiştir. Ürünsüz ortamda gerçekleştirilen biyolojik testlerde, PPO+vakum uygulamasında zararlının pupa dönemi hariç diğer tüm biyolojik dönemlerinde (ergin, yumurta ve larva) %100 ölüm oranı tespit edilirken, tek başına PPO ve PPO+CO<sub>2</sub> uygulamalarında *E. cautella*'nın biyolojik dönemlerinde hiçbir zaman %100 ölüm oranına ulaşamamıştır. Elde edilen bulgular sonucunda, *E. cautella*'ya karşı PPO+vakum uygulamasının tek başına PPO ve PPO+CO<sub>2</sub> uygulamalarına kıyasla daha yüksek insektisidal etkinlik gösterdiği tespit edilmiştir. Kuru incir kullanılan ortamda yürütülen biyolojik testlerde, PPO+vakum ve PPO+CO<sub>2</sub> uygulamalarında *E. cautella*'nın tüm biyolojik dönemlerinde meydana gelen ölüm oranları %19.7 ile %100 arasında değişiklik göstermiştir. Diğer yandan kabuklu fındık kullanılan ortamda yürütülen biyolojik testlerde, tüm PPO uygulamalarında %0.7 ile %10.6 arasında değişen düşük ölüm oranları gözlemlenmiştir. Bu bakımdan yapılan çalışma sonucunda, kuru incir ve kabuklu fındık bulunan ortamlarda yürütülen tüm PPO uygulamalarının *E. cautella*'ya karşı toksisitelerinde önemli farklılıklar tespit edilmiştir. Genel olarak yapılan bu çalışma, özellikle kuru incirlerde böcek kontaminasyonunu hızla kontrol etmek için umut vaat eden bir alternatif fümigant olarak PPO'nun potansiyelini ortaya koymuştur. Ancak, bu tür uygulamaların pratikte kullanılabilirliğini belirlemek için daha büyük ölçekli ticari deneylere ihtiyaç duyulmaktadır.

Anahtar kelimeler: incir güvesi, kuru incir, fındık, fumigant, propilen oksit

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