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Research Article

Fumigant Activity of *Eruca sativa* Essential Oil against *Tribolium castaneum*

Kainaat Rana[1], Sidra Munir[1,2]* and Talha Ali Chohan[1]

[1]University of Lahore, Lahore, Pakistan; [2]Gulab Devi Educational Complex, Lahore, Pakistan.

Abstract | This study evaluated the fumigant toxicity of *Eruca sativa* essential oil against adult *Tribolium castaneum* under laboratory conditions. Beetles were exposed to four concentrations (25, 50, 75, and 100 μL/L air), and mortality was recorded after 24, 48, and 72 h. Mortality increased significantly with both concentration and exposure time, reaching a maximum of 89% at 72 h at the highest concentration. Probit analysis revealed a progressive decline in LC_{50} values with increased exposure duration, indicating enhanced toxicity over time. These results demonstrate that *E. sativa* essential oil exhibits notable fumigant activity against *T. castaneum* under controlled laboratory conditions.

Keywords | *Eruca sativa*, Fumigant activity, *Tribolium castaneum*, Botanical insecticide, Stored grain pests

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***Correspondence** | Sidra Munir, University of Lahore, Lahore, Pakistan; **Email:** sidra.munir@gdec.edu.pk
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INTRODUCTION

Stored grain losses caused by insect pests represent a persistent threat to global food security, particularly in developing countries where post-harvest losses may reach 10–30% due to inadequate storage infrastructure and ineffective pest management practices (Abass *et al*., 2018; Baoua *et al*., 2018). Among stored-product insects, *Tribolium castaneum* (Herbst), commonly known as the red flour beetle, is one of the most destructive and widely distributed pests. It infests a broad range of stored commodities, including wheat, maize, rice, and processed grain products, causing significant quantitative losses through feeding as well as qualitative deterioration resulting from contamination with frass, cast skins, and offensive odors (Attia *et al*., 2020; Islam, 2017).

Control of stored-product pests has traditionally depended on synthetic fumigants such as phosphine and methyl bromide, along with residual insecticides. Although these methods provide rapid and effective control, their prolonged and widespread use has resulted in serious limitations, including the development of insecticide resistance, risks to human health, and adverse environmental consequences (Isman, 2015; Khan, 2020; Wang *et al*., 2020; Sakka *et al*., 2020). Moreover, increasing regulatory restrictions and growing consumer demand for pesticide-free food products have intensified the search for safer and more sustainable pest management alternatives (Hikal *et al*., 2017; Singhano *et al*., 2020).

In this context, botanical essential oils have gained considerable attention as eco-friendly alternatives to conventional fumigants. These natural products contain diverse bioactive compounds that exhibit insecticidal, repellent, antifeedant, and growth-regulating properties, often acting on multiple physiological and behavioral targets in insects and thereby reducing the likelihood of resistance development (Isman, 2015; Hikal *et al*., 2017; Gaur and Kumar, 2020). Among botanicals, *Eruca sativa* (Mill.), a member of the family Brassicaceae, is of particular interest due to its high content of glucosinolates and their enzymatic hydrolysis products, especially isothiocyanates. These sulfur-containing compounds are known for their strong biological activity and have been reported to exert toxic effects against a range of insect pests (Bell *et al*., 2016; Gugliandolo *et al*., 2018; Rezaei *et al*., 2019).

Previous studies have demonstrated the fumigant efficacy of essential oils derived from plants such as *Eucalyptus* spp., *Withania somnifera*, and *Thymus vulgaris* against several stored-product insects (Sharma *et al*., 2016; Ebadollahi and Setzer, 2020; Gaur and Kumar, 2020). Even so, despite the documented insecticidal potential of *E. sativa* essential oil against certain insect species, including Blattella germanica (Rezaei *et al*., 2019), information regarding its fumigant toxicity against *T. castaneum* remains limited. In particular, systematic evaluations of dose- and time-dependent mortality responses and lethal concentration estimates for this species are scarce.

Thus, the present study aimed to evaluate the fumigant toxicity of *Eruca sativa* essential oil against adult *Tribolium castaneum* under controlled laboratory conditions, assess mortality responses at different concentrations and exposure durations, and determine LC_{50} values. This work seeks to contribute novel data on the potential of *E. sativa* as a botanical fumigant for the management of stored-product pests.

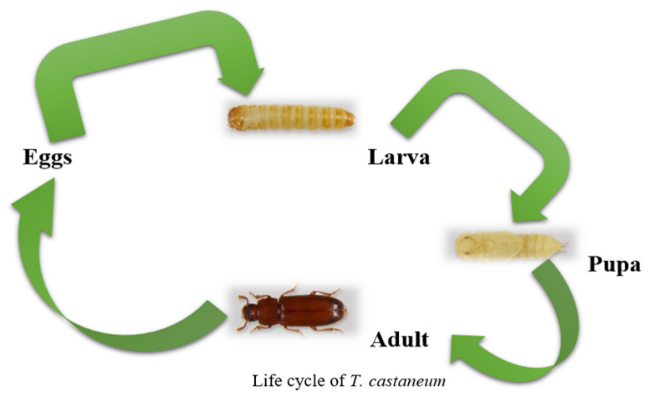

Figure 1: Insect life cycle.

MATERIALS AND METHODS

Insect culture

Adult *Tribolium castaneum* (Herbst) were obtained from a laboratory-maintained colony at the Department of Zoology insect rearing facility. The insects were reared on finely milled wheat flour supplemented with 5% brewer's yeast to provide essential nutrients for growth and reproduction (Islam, 2017). Cultures were maintained in glass jars covered with muslin cloth secured with rubber bands to allow adequate ventilation and prevent insect escape. All rearing and experimental insects were kept in a controlled environment chamber at 27 ± 2 °C, 65 ± 5% relative humidity (RH), and a 12:12 h light:dark photoperiod to ensure uniform physiological conditions (Attia *et al*., 2020). Only healthy, unsexed adults aged 7–14 days were used in bioassays to minimize variability in susceptibility (Sakka *et al*., 2020).

Essential oil extraction

Seeds of *Eruca sativa* were procured from a local agricultural supplier and visually inspected to ensure the absence of mold, debris, and insect damage. The seeds were thoroughly cleaned, air-dried, and ground into a coarse powder prior to extraction. Essential oil was extracted by hydro-distillation using a Clevenger-type apparatus for 3 h following the method described by Rezaei *et al*. (2019). The obtained oil was separated from the aqueous phase, dried over anhydrous sodium sulfate to remove residual moisture, and stored in airtight amber glass vials at 4 °C until use to minimize oxidation and loss of volatile constituents (Bell *et al*., 2016). The chemical composition of the essential oil was not analyzed in the present study, information regarding its constituents is inferred from previously published literature.

Fumigation bioassay

Fumigant toxicity assays were conducted following methods adapted from Sharma *et al*. (2016) and Ebadollahi and Setzer (2020). Circular filter papers (Whatman No. 1, 9 cm diameter) were treated with measured volumes of *E. sativa* essential oil to obtain final concentrations of 25, 50, 75, and 100 μL/L air. Treated filter papers were allowed to air-dry for approximately 2 min at room temperature to facilitate uniform volatilization and then attached to the underside of the lids of airtight glass containers (1 L volume).

Twenty unsexed adult *T. castaneum* were introduced into each container, and the lids were immediately sealed to prevent the escape of volatile compounds. Each concentration was replicated three times (n= 60 insects per concentration). Control treatments consisted of filter papers treated with solvent only and maintained under identical experimental conditions.

Mortality assessment

Insect mortality was assessed after 24, 48, and 72 h of exposure. Beetles were considered dead if they showed no movement when gently prodded with a fine camel hair brush (Umair *et al*., 2020). Mortality assessments were conducted in a blinded manner to reduce observer bias.

Data analysis

Observed mortality data were corrected for control mortality using Abbott's formula (Abbott, 1925). Corrected mortality percentages were subjected to probit analysis (Finney, 1971) using SPSS version 25.0 (IBM Corp., Armonk, NY, USA) to estimate median lethal concentrations (LC_{50}) and corresponding 95% confidence intervals. Prior to statistical analysis, mortality data were arcsine square root transformed to stabilize variances. One-way analysis of variance (ANOVA) was performed separately for each exposure period (24, 48, and 72 h) to evaluate the effect of concentration on mortality, followed by Tukey's HSD test for mean separation at a significance level of $p < 0.05$ (Gaur and Kumar, 2020).

RESULTS

Mortality of *Tribolium castaneum* increased significantly with increasing concentration of *Eruca sativa* essential oil and longer exposure duration ($p < 0.05$). A clear dose- and time-dependent response was observed across all treatments. Mortality levels were consistently higher at 48 and 72 h compared to 24 h for each concentration tested.

As shown in Table 1, at the lowest concentration, mortality increased from 40% after 24 h to 50% after 72 h, whereas at the highest concentration tested, mortality increased from 66% at 24 h to a maximum of 89% after 72 h. These results demonstrate that prolonged exposure enhanced the fumigant toxicity of *E. sativa* essential oil against adult *T. castaneum*.

Table 1: Percentage mortality of *Tribolium castaneum* after 24, 48, and 72 h of exposure to different concentrations of *Eruca sativa* essential oil.

Concentration	24 h (%)	48 h (%)	72 h (%)
5	40	44	50
10	50	60	61
15	58	71	74
20	62	76	82
25	66	82	89

Figure 2 illustrates the percentage mortality of *T. castaneum* at different concentrations of *E. sativa* essential oil after 24, 48, and 72 h of exposure. Mortality increased progressively with concentration at all exposure periods, confirming the fumigant activity of the oil. Error bars represent standard deviation among replicates.

Probit analysis indicated a gradual decline in LC_{50} values with increasing exposure time, reflecting enhanced toxicity following prolonged fumigation. The effects of concentration on mortality were statistically significant at each exposure period based on one-way ANOVA ($p < 0.05$), indicating a strong relationship between dose and insect mortality.

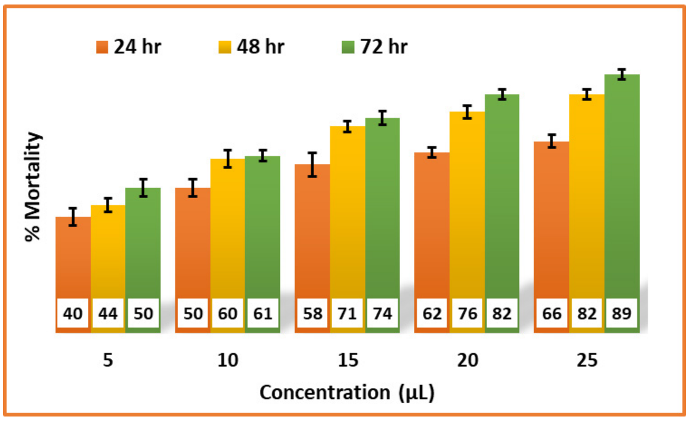

Figure 2: Illustrates the percentage mortality of *T. castaneum* at different concentrations of *E. sativa* essential oil after 24, 48, and 72 h of exposure. Mortality increased progressively with concentration at all exposure periods, confirming the fumigant activity of the oil. Error bars represent standard deviation among replicates.

DISCUSSION

The fumigant toxicity of *Eruca sativa* essential oil observed in the present study suggests that this plant possesses bioactive constituents capable of adversely affecting adult *Tribolium castaneum* under laboratory conditions. Although the chemical composition of the essential oil was not analyzed in this study, previous reports indicate that *E. sativa* seeds are rich in glucosinolates and their hydrolysis products, particularly sulfur-containing compounds such as isothiocyanates, which are known for their insecticidal properties (Bateni and Karimi, 2016; Bell *et al*., 2016). The insecticidal effects attributed to isothiocyanates in earlier studies include disruption of respiratory metabolism and interference with key enzymatic processes; however, such mechanisms are inferred here based solely on published literature rather than direct experimental evidence (Gupta *et al*., 2017).

The observed increase in mortality with prolonged exposure time indicates that fumigant efficacy of *E. sativa* essential oil is time dependent. This pattern is consistent with previous studies reporting enhanced toxicity of essential oils following longer exposure periods, likely due to increased penetration and accumulation of volatile compounds within insect tissues (Gaur and Kumar, 2020; Umair *et al*., 2020). Similar time-dependent effects have been documented for other botanical fumigants against *T. castaneum*, including essential oils of *Origanum majorana* and *Thymus* species (Sharma *et al*., 2016; Ebadollahi and Setzer, 2020). The progressive decline in LC_{50} values over time observed in this study further supports the influence of exposure duration on fumigant toxicity.

Botanical essential oils offer several advantages over conventional synthetic fumigants, including rapid biodegradation and reduced persistence in the environment, which may lower risks to non-target organisms and minimize residual contamination of stored products (Isman, 2015). These characteristics are particularly relevant given the widespread development of resistance to phosphine and increasing regulatory restrictions on chemical fumigants (Khan, 2020; Wang *et al*., 2020). However, the high volatility and sensitivity of essential oils to environmental factors such as temperature, humidity, and light may limit their consistency and residual effectiveness under practical storage conditions (Sakka *et al*., 2020).

To overcome these limitations, recent research has emphasized the importance of formulation strategies aimed at improving the stability and controlled release of botanical oils. Approaches such as nanoencapsulation and incorporation into biopolymer matrices have been shown to enhance the persistence and efficacy of essential oils in stored-product pest management systems (Rajkumar *et al*., 2020). In addition, combining botanical fumigants with other compatible control agents, including diatomaceous earths or entomopathogenic fungi, may offer synergistic effects and improve overall pest suppression, as demonstrated in previous integrated management studies (Wakil *et al*., 2020).

Although the results obtained under laboratory conditions indicate promising fumigant activity of *E. sativa* essential oil against *T. castaneum*, further investigations under semi-field and field storage conditions are necessary. Such studies should evaluate formulation stability, cost-effectiveness, effects on grain quality, and potential impacts on non-target organisms. Addressing these factors will be essential for determining the practical applicability of *E. sativa* essential oil as a component of integrated pest management programs for stored grain protection.

CONCLUSION

This study demonstrates that *Eruca sativa* essential oil exhibits significant fumigant toxicity against adult *Tribolium castaneum* under laboratory conditions, with mortality increasing in a concentration- and time-dependent manner. The findings suggest potential for further development of this oil as a botanical fumigant; however, additional studies under semi-field and field storage conditions are required to validate efficacy, stability, and practical applicability.

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NOVELTY STATEMENT

This study provides the first systematic evaluation of the time-dependent and dose-dependent fumigant toxicity of *Eruca sativa* essential oil specifically against *T. castaneum*.

AUTHOR'S CONTRIBUTION

SM: Formal Analysis and Manuscript write up; KR: Methodology and Data Collection; TAC: Conceptualization and Supervision.

Generative AI and AI assisted technology statement

The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

Conflict of interest

The authors have declared no conflict of interest.

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