

Research Article

Pesticidal Potential of Essential Oils Against *Callosobruchus maculatus* Fab. (Coleoptera: Bruchidae) under Laboratory Conditions

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Authors' Contributions

ZP conducted the experiments and write initial draft. AAG, LBR and JH conceived the idea and design the experiment. NK help in conduct of experiments. MIK and SAM help analyse and presentation of data. AAG help in designing the study and finalized the manuscript.

Keywords

Bruchid beetle, *Callosobruchus*, Mortality, Repellence, Stored grain



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Abstract | Pulse beetle, *Callosobruchus maculatus* Fab. is a serious, multi-voltine and most destructive cosmopolitan pest in Asian and African countries on various pulses. Considering the target specificity of botanicals and less hazardous than synthetic insecticides, essential oils of orange (*Citrus sinensis* L.), lemongrass (*Cymbopogon citratus* Stapf.), eucalyptus (*Eucalyptus globulus* Labill.) and peppermint (*Mentha piperita* L.) were evaluated against *C. maculatus* adults. Each oil was applied at 0.1, 0.5, and 1.0ml doses on the filter paper. A long glass cylinder divided into three (A, B, and C) sections, supplied with 20g cowpea seeds at ends was used in the study. Ten freshly emerged male and female *C. maculatus* adults were released separately in the middle of B with the help of aspirator as treated (with oil) and untreated filter were placed at A and C sections, respectively. The number of adults were counted at A and C after 24 and 48 h to calculate the repellent efficacy and mortality percentage. All the essential oils showed pesticidal potential with orange and lemongrass found to be more effective in causing mortality of both male and female *C. maculatus*, whereas the peppermint and eucalyptus were found to be more repellent. The pesticidal potential of all oils increased with increasing doses and exposure timings. Maximum mortality and repellency of females after 48 h was recorded in orange oil (100.00±0.00%) and peppermint oil (90.00±0.00%), respectively, both applied at rate of 1.0ml dose, whereas maximum mortality and repellency of males after 48 h was also recorded in orange oil (96.67±3.33%) and peppermint (90.00±0.00%), respectively, applied at rate of 1.0ml dose. After 48 h, the lowest (0.008) and highest (0.095) LD₅₀ values against *C. maculatus* female were recorded for orange and eucalyptus essential oils, respectively. The lowest (0.030 ml) and highest (0.057) LD₅₀ values against *C. maculatus* males were also recorded for orange and eucalyptus essential oils, respectively. Thus, all the essential oils were found equally effective against both male and female *C. maculatus*. Therefore, it is suggested that the essential oils of orange or peppermint may be applied in the warehouses against *C. maculatus* to restrict their damage.

Novelty Statement | The essential oils of orange, lemongrass, eucalyptus, and peppermint were evaluated at 0.1, 0.5, and 1.0ml doses against adult male and female *C. maculatus*. All the essential oils were found effective to cause mortality and repellence in both the sexes with maximum mortality and repellency recorded in orange and peppermint oils, respectively. Pesticidal potential of all oils increased with increasing doses and exposure time against both the sexes of *C. maculatus*.

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Introduction

The pulse beetle, *Callosobruchus maculatus* Fab. (Coleoptera: Bruchidae) is commonly known as

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cowpea weevil. It is a serious, multi-voltine and most destructive cosmopolitan pest of pulses like cowpea, lentils, black gram, green gram, and chickpea in Asian and African countries (Boeke *et al.*, 2004; Muntaha *et al.*, 2017). The damage of *C. maculatus* starts from the standing crop in the field and the same will carry with the produce even during the storage in godowns (Ahuchaogu and Ojiako, 2020; Seth *et al.*, 2017). During storage, *C. maculatus* can cause 100% damage of pulse seeds (Gbaye *et al.*, 2011), whereas its average losses can range between 20-30 and 5- 10% in tropical and temperate countries, respectively (Kiradoo and Srivastava, 2010). Muntaha *et al.* (2017) reported that *C. maculatus* causes up to 10% damage to stored chickpea and up to 90% loss to stored gram. Pulse beetle infestation also invites secondary organisms such as rot organisms mostly fungi which leads to mycotoxin contamination, which create further degradation of legume grains. Seeds infested by pulse beetle thus become unhealthy for the consumption with poor germination percentage due to quantity and quality losses and fungal growth (Bhalla *et al.*, 2008). Various management tools such as physical, biological, botanical, and chemical methods are utilized to minimize bruchids infestation, hence reduce grain losses (Rahman and Sabiha, 2018). However, to overcome the bruchid's infestation and protect stored legumes, insecticides are the most common method. Despite the direct and rapid control of bruchids by synthetic pesticides, many reasons i.e., pest resurgence, resistance, persistence, effects on non-target organisms, high cost, residual environmental impacts make insecticides an unsuitable option (Elhag, 2000). Additionally, stored legumes should not be treated with insecticides as detrimental residues of toxic chemicals may cause serious effects on human health during consumption. Therefore, many other conventional and plant-based control methods are considered more safer and secure than insecticides (Weaver and Subramanyam, 2000). Natural insecticides that are extracted from plants contain several bio-active compounds and are generally known as bio-insecticides (Bai *et al.*, 2019; Buxton *et al.*, 2020; Peeyush *et al.*, 2011). Plants produced huge variety of chemicals i.e., phenolics, steroids, terpenoids and alkaloids that are used by plants for their protection and also possessed important insecticidal and medicinal properties (Hamada *et al.*, 2018; Hamad *et al.*, 2019; Hussein *et al.*, 2017). Various essential oils have repellent compounds that repel targeted insects by affecting their touch, taste, and smell senses. Generally, repellent effect of essential oils prevent pest from achieving their targeted site (Choochote *et al.*, 2007; Haq *et al.*, 2021; Nerio *et al.*, 2010). Therefore, present study was conducted to evaluate the efficacy of some botanical oils i.e., lemongrass oil (*Cymbopogon citratus*), eucalyptus oil (*Eucalyptus globulus*), peppermint oil (*Mentha piperita*) and orange oil (*Citrus sinensis*) against *C. maculatus*.

Materials and Methods

Study location

The experiments were carried out at Stored Grain Research Laboratory, Department of Entomology, Sindh Agriculture University, Tandojam during 2022.

Collection and rearing of *Callosobruchus maculatus*

Callosobruchus maculatus were obtained from the culture that was already managed at the laboratory as further rearing was done in cowpea seeds. The F1 generation adults obtained from the culture were then utilized for the experiment to determine the repellence and mortality of various essential oils against them.

Essential oils

Four essential oils were purchased from Chiltan pure international, an ISO certified company and used in the study mentioned below:

1. Lemongrass (*Cymbopogon citratus* Stapf.)
2. Eucalyptus (*Eucalyptus globulus* Labill.)
3. Peppermint (*Mentha piperita* L.)
4. Orange (*Citrus sinensis* L.)

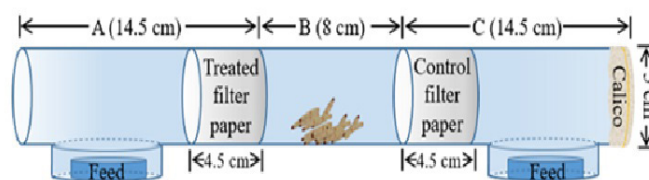


Figure 1: Systematic diagram of cylinder traps for evaluating pesticidal efficacy of essential oils against *Callosobruchus maculatus*.

Experimental set-up, data collection and analysis

Following the study of Lee *et al.* (2020) and Park *et al.* (2018), a long glass cylinder (500 ml, 32 cm height, 2.75 cm diameter) trap was manufactured with slight modifications (Figure 1). The cylinder connected with feeding dishes divided into three sections (A, B, C), where the one end was covered with muslin cloth for ventilation and prevention of *C. maculatus* from escaping during the entire duration of experiment. Method of Jo *et al.* (2013) and Khaskheli *et al.* (2021) was used to evaluate repellency of the of essential oils. In this experiment, three doses of essential oils i.e., 0.1, 0.5, and 1.0ml were used against both adult males and females of *C. maculatus*. The respective dose of the individual oil was applied on a filter paper, thereafter, it was then allowed to air-dry for 20 minutes. Afterwards, the treated filter paper with respective dose of a particular oil was placed between section A and B of the glass cylinder, whereas control filter paper without any oil was kept between B and C (Figure 1). Ten freshly emerged F1 adults (males or females) were released separately in the middle of section B with the help of an aspirator. The face of glass cylinder was closed with a fine muslin cloth

at the end of section C to ensure the ventilation inside it. The entire experiment was conducted at the temperature of $30 \pm 2^\circ\text{C}$ with $55 \pm 5\%$ R.H. The completely randomized design was used to setup the experiment where three replications were managed for each essential oil treatment. A separate glass cylinder was used for each treatment, whereas cylinder was thoroughly cleaned and wiped using tissue paper after performing each replication.

The data were collected by counting the number of insects at sections C and A after 24 and 48 h of the application of essential oils.

The percentage repellency of the essential oils was calculated using the equation given below:

$$\text{Repellent efficacy \%} = \frac{N_c - N_a}{N_c + N_a} \times 100$$

Where, N_c is the number of insects in C section, and N_a is the number of insects in section A.

The percentage of mortality was calculated by using the following equation:

$$\text{Mortality (\%)} = \frac{\text{Number of dead adults}}{\text{Total number of adults released}} \times 100$$

Analysis of variance was used to analyse the obtained data on mortality and repellency of essential oils for *C. maculatus* males and females, whereas means with significant differences were separated using the Least Square Difference (LSD) at 5% probability. All the analysis were performed using STATISTIX 8.1 computer software. Moreover, SPSS version 21, IBM Corp. was used to perform Probit analysis for the calculation of LD_{50} values for various essential oils for their insecticidal potential against *C. maculatus*.

Results and Discussion

Insecticidal potential of essential oils against *Callosobruchus maculatus* females

Figure 2 shows the results regarding the percentage mortality of *C. maculatus* females after 24 h application of various essential oils applied at various doses. A highly significant difference was recorded among various essential oils applied at various doses ($F = 9.17$, $P < 0.001$) to elicit the *C. maculatus* female mortality. According to the results, significantly the highest mortality ($86.67 \pm 3.33\%$) after 24 h of application was recorded in orange oil applied at 1.0ml dose, followed by 83.33 ± 3.33 and $10.00 \pm 0.00\%$ mortality recorded in lemongrass and peppermint essential oils, respectively, both applied at 1.0ml dose. Moreover, in eucalyptus essential oil treatment, the highest mortality ($20.00 \pm 5.77\%$) of *C. maculatus* females was recorded at

0.5ml dose, whereas the lowest mortality ($6.67 \pm 3.33\%$) was recorded in 0.1 ml peppermint oil.

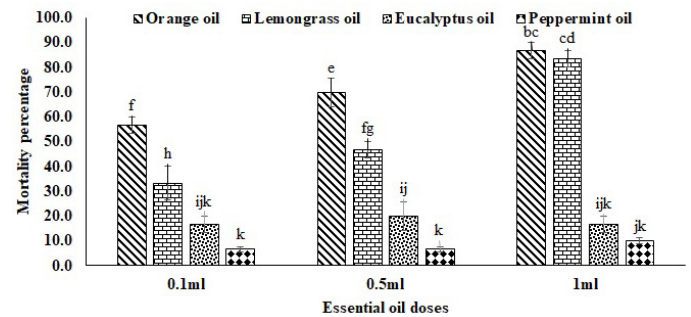


Figure 2: Effect of various essential oils on percentage mortality of female *Callosobruchus maculatus* after 24-h

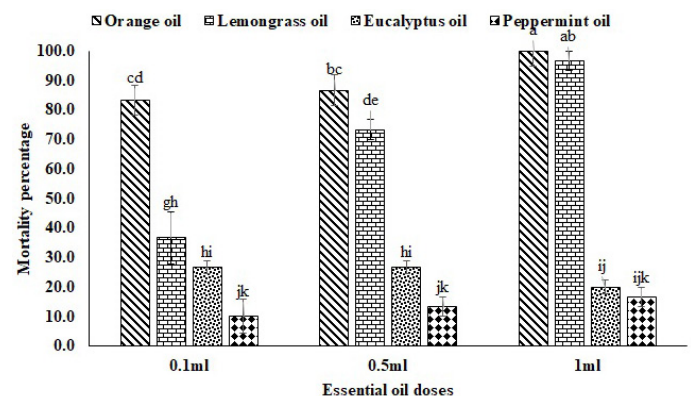


Figure 3: Effect of various essential oils on percentage mortality of female *Callosobruchus maculatus* after 48-h.

The mortality percentage of *C. maculatus* showed an increasing trend due to the application of various doses of essential oils after 48 h, exhibiting highly significant ($F = 12.98$, $P < 0.001$) difference among various treatments (Figure 3). According to the results, significantly the highest ($100.00 \pm 0.00\%$) mortality percentage of *C. maculatus* after 48 h at 1.0ml dose was recorded in orange oil, followed by lemongrass ($96.67 \pm 3.33\%$), whereas the lowest mortality was recorded in peppermint ($10.00 \pm 5.77\%$) at 0.1ml dose. Moreover, the maximum mortality of female *C. maculatus* in orange oil ($86.67 \pm 3.33\%$) and lemongrass ($73.33 \pm 3.33\%$) was recorded at 0.5ml dose.

Figure 4 shows the results regarding the repellency percentage of *C. maculatus* females due to the application of different doses i.e., 0.1, 0.5 and 1.0ml after 24 h. Different essential oils showed significant difference ($F = 16.76$, $P < 0.001$) in their potential to repel *C. maculatus* females. The highest repellency percentage was recorded in peppermint ($83.33 \pm 3.33\%$) and eucalyptus ($63.33 \pm 3.33\%$), whereas the maximum repellency of the females in orange oil ($26.67 \pm 6.67\%$) was recorded at 0.1ml dose and lemongrass oil ($40.00 \pm 5.77\%$) at 0.5ml dose. Moreover, orange ($13.33 \pm 3.33\%$) and lemongrass ($16.67 \pm 3.33\%$) oils exhibited the lowest repellency when applied at 1.0ml dose.

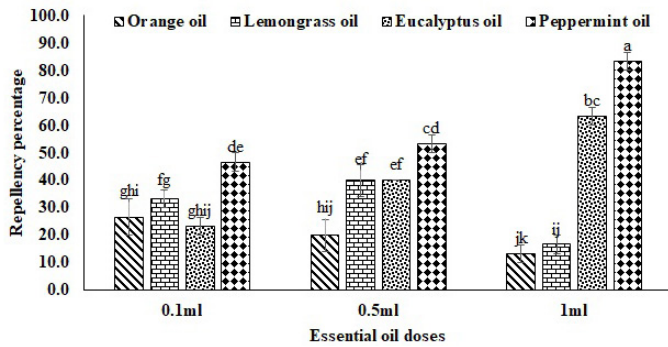


Figure 4: Effect of various essential oils on percentage repellency of female *Callosobruchus maculatus* after 24-h.

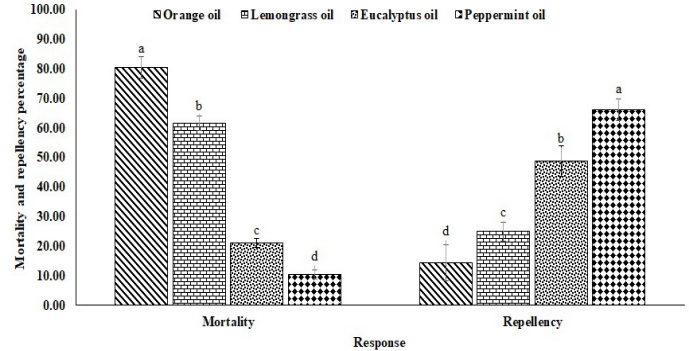


Figure 6: Overall mortality and repellency percentage of various essential oils against female *Callosobruchus maculatus*.

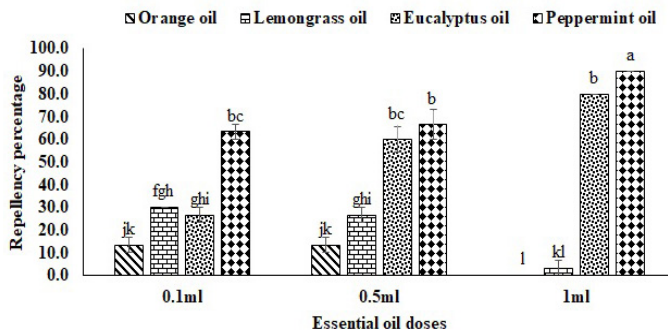


Figure 5: Effect of various essential oils on percentage repellency of female *Callosobruchus maculatus* after 48-h.

An increase in the repellency of *C. maculatus* was recorded at 48 h of the application of various doses of essential oils, however, there was a significant difference ($F = 26.19$, $P < 0.001$) among various oils applied at various doses (Figure 5). The highest repellency ($90.00 \pm 0.00\%$) was recorded in peppermint and eucalyptus ($80.00 \pm 0.00\%$) at 1.0ml dose, whereas the maximum repellency in lemongrass and orange oils was recorded as 30.00 ± 0.00 and $13.33 \pm 3.33\%$, respectively, all applied at 0.1ml dose. Moreover, the lowest repellency was recorded in lemongrass (3.33 ± 3.33) due to rise in mortality percentage at 1.0ml dose.

Overall repellency and mortality of *C. maculatus* females due to the application of various doses of different essential oils are given in Figure 6. The results confirmed a highly significant difference among various essential oils regarding their potential to cause mortality ($F = 394.77$, $P < 0.001$) and repellency ($F = 220.21$, $P < 0.001$) of *C. maculatus* females. Significantly, the highest overall mortality of female *C. maculatus* ($80.56 \pm 3.57\%$) was recorded in orange oil, whereas the mortality percentage recorded in lemongrass ($61.67 \pm 6.12\%$), eucalyptus ($21.11 \pm 1.59\%$), and peppermint ($10.56 \pm 1.51\%$) was significantly different from each other. Moreover, significantly the highest ($66.11 \pm 3.63\%$) repellency of *C. maculatus* females was recorded in peppermint oil, followed by 48.89 ± 5.11 and $25.00 \pm 3.16\%$ repellency observed in eucalyptus and lemongrass oils, respectively. Moreover, the lowest ($14.44 \pm 2.46\%$) repellency percentage of *C. maculatus* females was recorded in orange oil treatments.

Insecticidal potential of essential oils against Callosobruchus maculatus males

Figure 7 shows the results regarding the percentage mortality of *C. maculatus* males after 24 h application of various essential oils applied at various doses. A highly significant difference was recorded among various essential oils applied at various doses ($F = 7.33$, $P = 0.0002$) to elicit the *C. maculatus* male mortality. According to the results, significantly the highest mortality ($86.67 \pm 3.33\%$) after 24 h of application was recorded in orange oil applied at 0.5 and 1.0ml doses. Moreover, the maximum mortality in lemongrass, and peppermint oils was recorded as $43.33 \pm 3.33\%$ and $20.00 \pm 0.00\%$, respectively, both applied at 1.0ml dose, whereas in eucalyptus, the highest mortality ($20.00 \pm 0.00\%$) was recorded at 0.5 and 1.0ml doses. Overall, the lowest mortality i.e., $6.67 \pm 3.33\%$ was recorded in peppermint oil applied at 0.1ml dose.

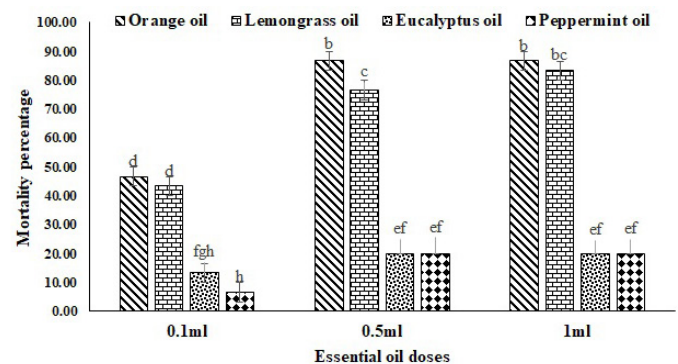


Figure 7: Effect of various essential oils on percentage mortality of male *Callosobruchus maculatus* after 24-h.

The mortality percentage of *C. maculatus* showed an increasing trend due to the application of various doses of essential oils after 48 h, with highly significant ($F = 16.19$, $P < 0.001$) difference among various treatments (Figure 8). According to the results, significantly the highest ($96.67 \pm 3.33\%$) mortality percentage of *C. maculatus* males after 48 h was recorded in orange oil applied at 1.0ml dose, followed by lemongrass oil ($86.67 \pm 3.33\%$) when applied at 0.5 and 1.0ml doses. Moreover, the maximum mortality of *C. maculatus* males in eucalyptus oil ($23.33 \pm 3.33\%$) was

recorded at 0.1ml dose. Overall, the lowest mortality was recorded in peppermint (10.00±0.00%) at 0.1ml dose.

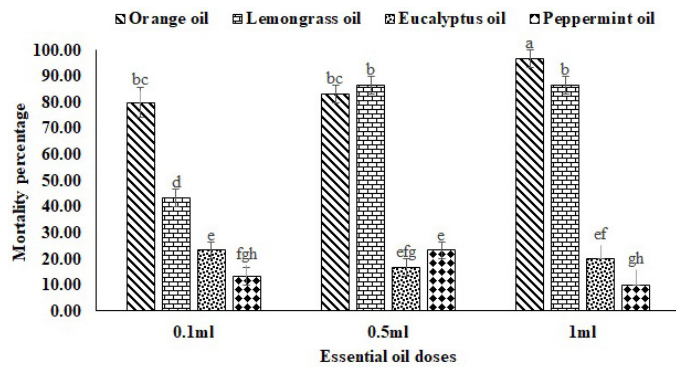


Figure 8: Effect of various essential oils on percentage mortality of male *Callosobruchus maculatus* after 48-h.

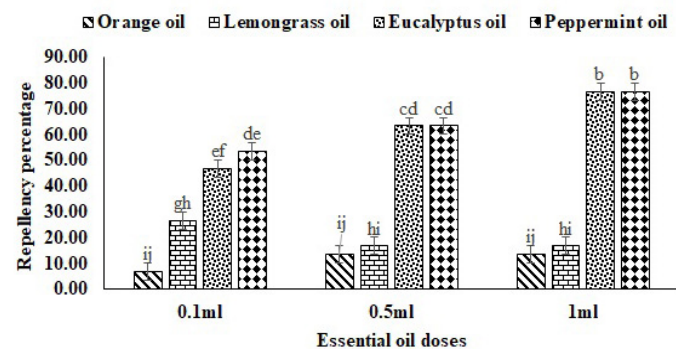


Figure 9: Effect of various essential oils on percentage repellency of male *Callosobruchus maculatus* after 24-h.

Figure 9 shows the results regarding the repellency percentage of *C. maculatus* males due to the application of 0.1, 0.5, and 1.0ml doses of essential oils after 24 h. Different essential oils showed significant difference ($F=4.37$, $P=0.0041$) in their potential to repel *C. maculatus* males as the highest ($76.67 \pm 3.33\%$) repellency percentage was recorded in peppermint and eucalyptus at 1.0ml dose. Moreover, the maximum repellency of the males in orange oil ($13.33 \pm 3.33\%$) and lemongrass ($16.67 \pm 3.33\%$) when recorded at 0.5 and 1.0ml doses. Overall, the orange oil exhibited the lowest repellency ($6.67 \pm 3.33\%$) when applied at 0.1ml dose and lemongrass ($16.67 \pm 3.33\%$) when applied at 0.5 and 1.0ml doses.

An increase in the repellency of *C. maculatus* was recorded at 48 h of the application of various doses of essential oils with a significant difference among various oils ($F=21.07$, $P<0.001$) applied at various doses (Figure 10). The highest repellency ($90.00 \pm 0.00\%$) of *C. maculatus* males was recorded in peppermint and eucalyptus oil ($80.00 \pm 0.00\%$), applied at 1.0ml dose. The maximum mortality in lemongrass and orange oils was recorded as 36.67 ± 3.33 and $6.67 \pm 3.33\%$, respectively, both applied at 0.1ml dose. Overall, the lowest repellency was recorded in orange oil ($3.33 \pm 3.33\%$) at 1.0ml and lemongrass ($10.00 \pm 0.00\%$) at 0.5ml dose due to rise in mortality

percentage in the respective treatment oils.

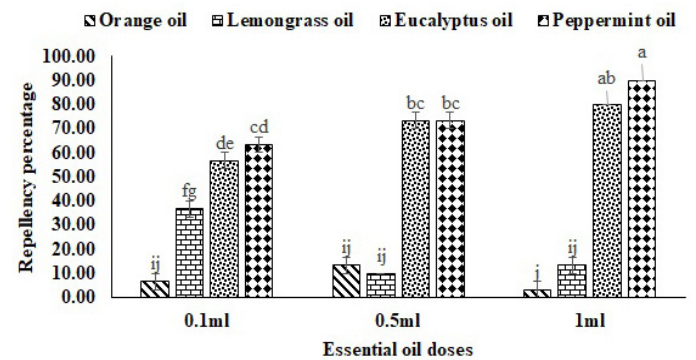


Figure 10: Effect of various essential oils on percentage repellency of male *Callosobruchus maculatus* after 48-h.

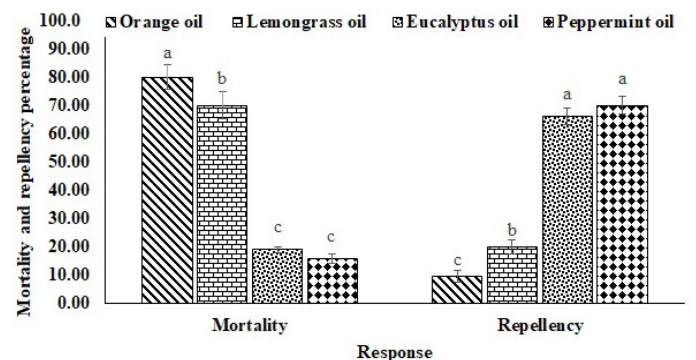


Figure 11: Overall mortality and repellency percentage of various essential oils against male *Callosobruchus maculatus*.

Overall repellency and mortality of *C. maculatus* males due to the application of various doses of different essential oils are given in Figure 11. The results confirmed a highly significant difference among various essential oils regarding their potential to cause mortality ($F=563.90$, $P<0.001$) and repellency ($F=418.71$, $P<0.001$) of *C. maculatus* males. Significantly, the highest overall mortality of *C. maculatus* ($80.00 \pm 4.20\%$) was recorded in orange oil, whereas the mortality percentage recorded in lemongrass ($70.00 \pm 4.78\%$), eucalyptus ($18.89 \pm 1.11\%$), and peppermint ($15.56 \pm 1.66\%$) was significantly different from each other. Moreover, significantly the highest ($70.00 \pm 3.02\%$) mortality of *C. maculatus* males was recorded in peppermint oil, followed by eucalyptus ($66.11 \pm 3.04\%$) and lemongrass ($20.00 \pm 2.43\%$) essential oils. Overall, the lowest repellency ($9.44 \pm 2.06\%$) of *C. maculatus* males was recorded in orange essential oil.

Overall effect of essential oils on mortality of *C. maculatus* male and female

The comparative analysis of various essential oils indicated all the oils were found equally effective ($F=1.12$, $P=0.2918$) to cause mortality of both male and female *C. maculatus*. However, orange oil and eucalyptus were found relatively more toxic against females, whereas lemongrass and peppermint were more lethal against males (Figure 12).

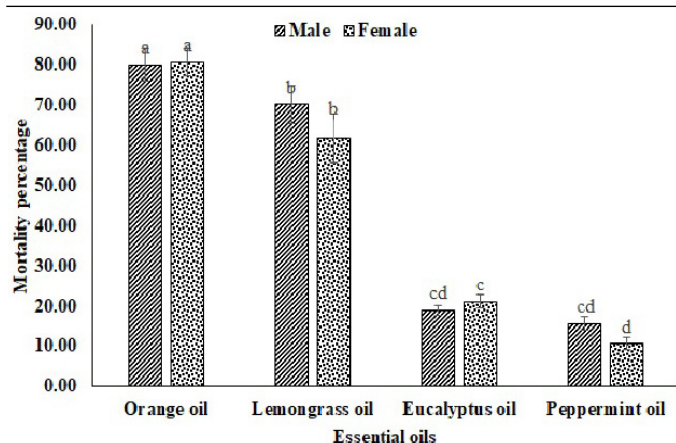


Figure 12: Overall mortality percentage of various essential oils against male and female *Callosobruchus maculatus*.

Overall effect of essential oils on repellency of C. maculatus male and female

The comparative analysis of various essential oils indicated that all the oils were found equally effective ($F = 1.47$, $P = 0.2268$) to cause repellency of both male and female *C. maculatus*. However, peppermint oil and eucalyptus showed relatively more repellent potential against males, whereas lemongrass and orange were more effective against females (Figure 13).

*Lethal dose (LD_{50}) of essential oils against *Callosobruchus maculatus* females and males*

Table 1 describes the results regarding the LD_{50} values calculated for various essential oils used against female of *C.*

maculatus. It was evident from the results that LD_{50} values of all the essential oils were time dependent, instead of dose dependent as LD_{50} values decrease at the data observation of 48 h from 24 h. Among all the essential oils, the lowest LD_{50} values after 24 h were recorded in orange [0.012 ml (0.000-0.046)], followed by lemongrass [0.070 ml (0.002-0.147)] and peppermint [0.108 ml (0.002-0.238)], whereas the eucalyptus showed the highest LD_{50} values of 0.198 ml (0.071-0.337) against *C. maculatus* females. After 48 h, the lowest [0.008 (0.001-0.019)] and highest [0.095 ml (0.054-0.136)] LD_{50} values were recorded for orange and eucalyptus essential oils, respectively. Moreover, LD_{50} values recorded for lemongrass and peppermint treatment were 0.068 ml (0.029-0.097) and 0.033 ml (0.000-0.133), respectively.

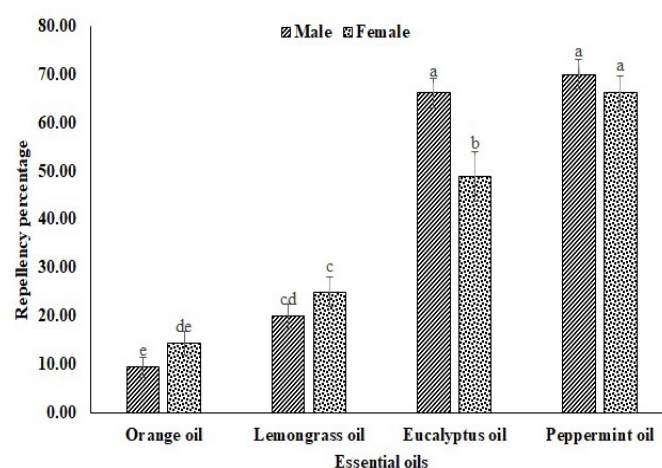


Figure 13: Overall repellency efficacy of various essential oils against male and female *Callosobruchus maculatus*.

Table 1: LD_{50} values of various essential oils against *Callosobruchus maculatus* females.

Essential oil	H	LD_{50} (ml)	FL 95%	Slope \pm SE	χ^2	Probability
Eucalyptus	24-h	0.198	0.071-0.337	1.010 \pm 0.105	35.684	< 0.001
	48-h	0.095	0.054-0.136	1.885 \pm 0.139	27.762	< 0.001
Lemon grass	24-h	0.070	0.002-0.147	1.629 \pm 0.138	94.411	< 0.001
	48-h	0.068	0.029-0.097	2.536 \pm 0.254	33.723	< 0.001
Peppermint	24-h	0.108	0.002-0.238	1.012 \pm 0.108	66.581	< 0.001
	48-h	0.033	0.000-0.113	1.128 \pm 0.133	89.602	< 0.001
Orange	24-h	0.012	0.000-0.046	0.996 \pm 0.152	25.692	0.001
	48-h	0.008	0.001-0.019	1.142 \pm 0.206	13.719	0.056

Table 2: LD_{50} values of various essential oils against *Callosobruchus maculatus* males.

Essential oil	H	LD_{50} (ml)	FL 95%	Slope \pm SE	χ^2	Probability
Eucalyptus	24-h	0.139	0.089-0.187	0.767 \pm 0.103	11.453	0.120
	48-h	0.057	0.024-0.093	0.669 \pm 0.105	6.175	0.520
Lemon grass	24-h	0.051	0.019-0.083	1.721 \pm 0.164	23.351	0.001
	48-h	0.032	0.002-0.067	1.674 \pm 0.201	33.095	< 0.001
Peppermint	24-h	0.069	0.023-0.117	1.343 \pm 0.123	26.913	< 0.001
	48-h	0.040	0.005-0.076	1.816 \pm 0.200	35.515	< 0.001
Orange	24-h	0.094	0.064-0.121	2.999 \pm 0.258	30.619	< 0.001

Table 2 describes the results regarding the LD₅₀ values calculated for various essential oils used against male of *C. maculatus*. It was evident from the results that LD₅₀ values of all the essential oils were time dependent, instead of dose dependent as LD₅₀ values decrease at the data observation of 48 h from 24 h. Among all the essential oils, the lowest LD₅₀ values after 24 h was recorded in lemongrass oil [0.051 ml (0.019–0.083)], followed by peppermint [0.069 ml (0.023–0.117)] and orange [0.094 ml (0.064–0.121)], whereas the eucalyptus showed the highest LD₅₀ values of 0.139 ml (0.089–0.187) against *C. maculatus*. After 48 h, the lowest [0.030 ml (0.001–0.064)] and highest [0.057 ml (0.024–0.093)] LD₅₀ values were recorded for orange and eucalyptus essential oils, respectively. Moreover, LD₅₀ values recorded for lemongrass and peppermint treatment were 0.032 ml (0.002–0.067) and 0.040 ml (0.005–0.076), respectively.

The current study was undertaken on the insecticidal potential of four essential oils i.e., orange oil, lemongrass, eucalyptus, and peppermint at three different doses i.e., 0.1, 0.5, and 1.0 ml to determine their mortality and repellent efficacy against adult male and female *C. maculatus*. The obtained results indicated that essential oils of orange and lemongrass were found more effective to cause mortality of both male and female *C. maculatus*, whereas the peppermint and eucalyptus were found to be repellent towards both adults. Moreover, the mortality or repellence of *C. maculatus* adults increased with increasing dose of essential oils and their exposure timings. Among essential oils, orange oil showed highest mortality in females and males followed by lemongrass, whereas the highest repellence of males and females was recorded in peppermint oil, followed by eucalyptus. Moreover, among all tested doses of various essential oils, 1.0 ml was proved to be more effective as compared to 0.1 and 0.5 ml, as the highest mortality or repellence of *C. maculatus* males and females was recorded after 48 h of the exposure.

Many studies have been reported to determine the insecticidal activity of essential oils against *C. maculatus*. De Souza *et al.* (2019) investigated the essential oil of lemongrass (*C. citratus*) against *C. maculatus*. The changes in the behaviour and high mortality were observed in male and female *C. maculatus*. Behavioural changes has led to reduction in the acetylcholinesterase activities in both sexes, β -esterase in females and decrease triacylglycerol in mated and unmated females. Kéita *et al.* (2001) also found males more susceptible than females when treated with essential oils of *Ocimum basilicum* and *O. gratissimum*. Similarly, Ojebode *et al.* (2016) investigated the potency of the essential oils i.e., *A. indica*, *C. sinensis*, and *C. citratus* against *C. maculatus* and other stored product pests. The highest mortality was observed in *C. sinensis* on first day of application followed by *A. indica* and *C. citratus*. *Cymbopogon citratus* also caused hundred percent mortality

of adult within one h of exposure, whereas *C. sinensis* showed 100 percent mortality after three h of exposure. According to the study of Jayakumar *et al.* (2017) five essential oils i.e., nutmeg, eucalyptus, camphor, rosemary, and wintergreen showed 100 percent mortality against *C. maculatus*. The highest mortality of *C. maculatus* after 96 h of exposure was observed in citrodora (96 percent) and lemongrass oil (92 percent) at 5% concentration (Raja and William, 2008). Omotoso *et al.* (2020) found that essential oil of lemongrass has strong mortality and repellent effect against *C. maculatus* as reduction in egg laying, 80 percent mortality and 66 percent repellency was recorded on treated cowpea within 24 h of exposure. However, the beetle mortality and repellency were time and concentration dependent. Similarly, essential oil of lemongrass was capable to cause 100 percent mortality after 1-h of exposure, whereas orange oil (*Citrus sinensis*) exhibited 100 percent mortality after 3rd h of exposure against *C. maculatus* (Ojebode *et al.*, 2016). Saeidi (2015) evaluated the efficacy of plant extract of *Eucalyptus camaldulensis* and *Eucalyptus globulus* and found that *E. globulus* was more effective against *C. maculatus* after 72-h of exposure. Ekeh *et al.* (2013) study reported that application of *Citrus sinensis* oil suppress *C. maculatus* population for moderate time of storage. While evaluating the insecticidal potential of peppermint oil the obtained results reveal that the most susceptible stage of *C. maculatus* was egg. Moreover, the application of peppermint oil has greater effect on male as compared to female and can significantly decrease the fecundity, survivorship, and mating frequency of next generation (El-Nagar *et al.*, 2012).

Pandey *et al.* (2014) while evaluating thirty-five essential oils of medicinal and aromatic plants against *C. maculatus* and *C. chinensis* found that *Adhatoda vasica* and *Chenopodium ambrosioides* L. showed 100% repellency for both insects, whereas *Chenopodium* sp. oil also caused 100% death of all tested organisms of the two species. Results also confirmed that application of 0.29 and 0.58 μ l/ml of *Chenopodium* oil also showed a promising feeding deterrence with reduced seed damage for the two species. According to Satongrod *et al.* (2021) essential oil from piperaceae plant family also showed repellent effect against many stored grain pests. Similarly, when Oliveira *et al.* (2017) treat the *Vigna unguiculata* seeds with *Piper hispidinervum* (Piperaceae), they found that it repels insects away from the seeds and resulted in significantly less seed damage. Manju *et al.* (2019) also evaluated the repellent efficacy of twelve botanicals against *C. maculatus* and found that *P. nigrum* and *A. indica* extracts were more effective to control its population than others in green gram storage. Similarly, oils of castor, neem, and eucalyptus on *Cajanus cajan* seeds showed longer emergence period as well as no adult emergence, while the application of sunflower take longer time than control (Dinesh and Raj, 2012). Idoko and Ileke (2020) found that essential oil

from *Aframomum melegueta* as the most effective among five oils they evaluated against the pulse beetle. Moreover, the effectiveness of citrus fruit peel extracts (Harshani and Karunaratne, 2021) and *Gnidia kraussiana* (Kosini *et al.*, 2021) was also evaluated against *C. maculatus* and were found to significantly affect its oviposition and larvae.

It has been mentioned that essential oils of lemongrass contain majority of terpenes and terpenoids such as 10.120- β -pinene and β -myrcene; 20.182- Citral (geranial); 19.766-citral (geranial) and citronellal; 20.435- Epoxy-linalooloxide (Furanoid); 22.733- Lavandulyl acetate; 23.338- Neric acid, whereas 10.073- β -pinene and β -myrcene; 10.577- Octanal; 11.812- Limonene; 14.455- Linalool; 13.474- 1-Octanol; 17.516- Decanal were reported as major terpenes and terpenoids in orange oil (Ojebode *et al.*, 2016). Thus, the presence of such huge quantities of terpenes and terpenoids in orange and lemongrass essential oils may be responsible for their more toxic potential against *C. maculatus* adults. Moreover, Myint *et al.* (2021) identified eight main constituents in peppermint with menthol being the most abundant (43.29%) and the same may be responsible for its more repellent potential against *C. maculatus* adults. Myint *et al.* (2021) also reported comparatively less LD₅₀ values of 0.018 (0.017–0.019) for peppermint essential oil against *C. maculatus* than essential oil of *Syzygium aromaticum*.

A recent review on the use of essential oils against *C. maculatus* indicated that essential oils from 121 plant species belonging to 26 families have shown their insecticidal potential against *C. maculatus*. Among the plant families, Lamiaceae (30 species) and Asteraceae (22 species) are the two most widely tested families against the *C. maculatus* as generally terpenoids and sesquiterpenoids classes of chemicals present in these plant essential oils are responsible for the insecticidal properties (Mssillou *et al.*, 2022).

Therefore, in continuation of above studies, all essential oils elicit promising mortality and repellent effect against *C. maculatus*, but essential oils of orange and lemongrass were found more effective to cause mortality of both male and female *C. maculatus*, whereas the peppermint and eucalyptus were found to be repellent. Moreover, the mortality or repellence of *C. maculatus* adults increased with increasing dose of essential oils and their exposure timings.

Conclusions and Recommendations

All the essential oils i.e., orange, lemongrass, eucalyptus, and peppermint exhibited pesticidal potential against male and female *C. maculatus* by causing their mortality and repellency. Orange and lemongrass oils were found to be more effective and lethal towards both males

and females, whereas peppermint and eucalyptus oils were found to be more repellent. The pesticidal potential of all oils increased with increasing doses and exposure timings as the lowest LD₅₀ value was recorded for orange essential oil. Therefore, essential oils of either orange or peppermint may be applied in the warehouses against *C. maculatus* to mitigate their population growth and grain damage.

Conflict of interest

The authors have declared no conflict of interest.

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