Insecticidal activity of essential oil from *Jasminum* sambac (L.) Aiton against *Callosobruchus maculatus* (Fabricius, 1775) (Coleoptera: Bruchidae)

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Abstract: Mungbean seeds [*Vigna radiata* (L.)] are easily damaged by insects during storage, and essential oils (EOs) have proved effective for controlling insect infestation. This research study assessed the bioactivity of EO from *Jasminum sambac* (L.) Aiton against *Callosobruchus maculatus* (F.). The chemical composition of EO was analysed by a gas chromatograph-mass spectrometer (GC-MS). All experiments were conducted following a completely randomised design (CRD) with four replications of the residual contact and seed dressing bioassays. Results indicated that *J. sambac* EO contained 35 compounds (97.587%), with the main component 3-hexen-1-ol benzoate. Residual contact toxicity LC₅₀ values of this EO on adult *C. maculatus* at 24, 48, and 72 h were 5.01, 4.00, and 3.13 μL/mL, respectively. *J. sambac* EO at 8 μL/L showed the highest residual contact efficacy against *C. maculatus*, killing up to 100% in 72 hours. The highest concentration of *J. sambac* EO (1 mL/kg) gave optimal protection against *C. maculatus* within 4 and 5 days with 100% mortality. This concentration showed 100% inhibition of adult F1 progeny emergence, with no seed damage and weight loss of *V. radiata* L. after 30 days of exposure, and not significantly different from 0.8 mL/kg. Results indicated that *J. sambac* EO effectively prevented *C. maculatus* infestation and it could be used as an alternative to commercial chemical insecticides.

Keywords: chemical compositions; toxicity; flowers; essential oils; storage insect pests

Vigna radiata (L.) R. Wilczek, commonly known crop in South and Southeast Asia. Legume crops as mungbean, is an economically important legume have a short life cycle of about 60 days and are usually

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grown as monocultures under rainy conditions with nitrogen-fixing ability and low water requirements. Legumes have complex root systems that maximise exploration for water and nutrients and are drought-resistant (Kumar & Sharma 2009). Legumes are grown under crop rotation and inter-cropping with cereals (Wang et al. 2022). They produce edible dry seeds as an excellent human source of protein, carbohydrates, minerals, and vitamins (Nair et al. 2013; Karthikeyan et al. 2014). Legumes are cultivated worldwide on about 7.3 million ha, mainly in Asia, with recent increases in Australia, America, Canada, and Africa (Nair et al. 2012, 2019).

Mungbean is a protein-rich staple containing 24–25% digestible low-flatulence protein, large amounts (54-56%) of carbohydrates, dietary fibre, minerals, vitamins, polyphenols, flavonoids, amino acids and carotenoids (Gopalan et al. 1989). However, mungbean yield is low due to drought, pests, diseases, and poor agricultural practices. Postharvest losses are significantly impacted both qualitatively and quantitatively due to insect infestation. Insect pests pose a formidable threat to mungbean production in Asia, including Thailand. Mungbean is susceptible to infestation by 65 recorded insect pest species in the field and during storage. These insects cause serious damage to crops (Raghu et al. 2016). Gosh and Durbey (2003) estimated that 40-50% of mungbean postharvest losses result from insect infestation during storage. Pest infestations are difficult to detect when harvesting. Two undetected pairs of adult insects per ton of grain would result in severe damage after six months of storage (Emery & Nayak 2007).

Bruchids in the genus Callosobruchus of the Bruchidae Family inflict severe damage on Species belonging to *Vigna* genus, particularly *V. radiata*, under storage conditions (Gahukar & Reddy 2018; Mishra et al. 2018; Mariyammal et al. 2019). Callosobruchus maculatus Fabricius (Coleoptera: Chrysomelidae: Bruchinae) is the most common insect species that attacks mungbean during storage, causing severe seed damage. In field conditions, bruchid infection is mild, with oviposition on the pod surface (Tripathi et al. 2016; Chawe et al. 2019). One insect-infested seed is a potential site for bruchid population development under storage. Bruchid damage greatly reduced the commercial and nutritional values of the grain (Mofunanya & Namgbe 2016; Hamdi et al. 2017; Sreedhar et al. 2020). Insects also contaminate grains through excretions, adversely impacting consumer acceptance (Biancolillo et al. 2019). Infected seeds are not appropriate for human consumption and are unsuitable for agriculture.

Chemical and fumigant pesticides are the most often used as the most convenient and economical method to prevent outbreaks in storage systems (Cao et al. 2019) and control insect pests. However, these pesticides have serious effects on human health and the environment. Repeated use of synthetic pesticides over time hinders the biological control of natural enemies, leading to infestation by other insects, and also increases pesticide resistance (Hill et al. 2017; Kim et al. 2017; Hawkins et al. 2019). Therefore, alternative approaches are urgently required that are environmentally friendly and harmless to non-targeted species. Natural plants and their derivatives have proven viable alternatives, with more than 2 000 natural plant species documented as having insecticidal properties with low health risks (Pavela 2016; Jerbi et al. 2021).

Essential oils (EOs) are volatile secondary compounds naturally produced by plants that protect stored grains from pest attacks (Omar 2020). EOs contain many chemical constituents and cause toxicity by interfering with various insect physiology and biochemistry aspects. They inhibit the acetylcholinesterase activity of insects and protect stored products from insect infestation (Kiran et al. 2017; Kavallieratos et al. 2021; Kavallieratos et al. 2022). Many EOs show insecticidal properties. Several previous studies demonstrated the potential of natural compounds on the mortality and repellency of insects in stored grains including fumigant toxicity and bioactivity of EO of Wedelia trilobata (L.) Hitchc. on C. maculatus (Satongrod et al. 2021), extracts of Piper nigrum in the control of C. maculatus (Wanna 2021) and EO of Plectranthus amboinicus (Lour.) Spreng. controls C. maculatus (Wanna & Kwang-Ngoen 2019; Satongrod & Wanna 2020). Using natural products as an alternative way to control C. maculatus infestation is safer than spraying with chemical pesticides.

The *Jasminum* genus contains around 600 species of small trees and vines in the Oleaceae. These glabrous twining shrubs are widely grown in gardens and commonly found in forests throughout tropical Asia and warm temperate regions in Europe and Africa. Their flowers and leaves have multipurpose uses. The flowers have long been used in Asian traditional medicines to treat diarrhoea, fever, conjunctivitis, abdominal pain, dermatitis, asthma, abscess, breast can-

cer, uterine bleeding, and toothache, while in China, the leaves are used to treat quadriplegia gall, dysentery, and stomachache. J. sambac (L.) Aiton. Has high medical value and is cultivated in many Asian countries, including Thailand (Kunhachan et al. 2012). Some phytoconstituents of the plant are iridoid glycosides (Zhang et al. 1995), linalyl 6-O-malonyl-β-Dglucopyranoside, benzyl 6-O-β-D-xylopyranosyl-β-D-glucopyranoside (β-primeveroside), 2-phenylethyl β-primeveroside, 2-phenylethyl 6-O-α-L-rham-nopyranosyl-β-D-glucopyrano-side (β-rutinoside) (Inagaki et al. 1995), dotri-acontanoic acid, dotriacontanol, oleanolic acid, daucosterol, and hesperidin (Zhang et al. 2004). Volatile plant components include benzyl acetate, indole, E-E- α -farnesene, Z-3-hexenyl benzoate, benzyl alcohol, linalool, and methyl anthranilate (Edris et al. 2008), and EOs have antibacterial activity (Rath et al. 2008).

Copious information exists on the medicinal activity of *J. sambac* extracts. This study investigated the chemical composition of *J. sambac* EO (locally called Ma-li-son), a local variety commonly found in Thailand and insecticidal activity by residual contact toxicity bioassay against *C. maculatus*. The efficacy of *J. sambac* EO for the protection of mungbean seeds from *C. maculatus* infestation laboratory conditions was also assessed.

MATERIAL AND METHODS

Insect rearing. Adult *C. maculatus* were collected from naturally infested mungbean seed hosts under storage in Kantarawichai District, Maha Sarakham Province, Thailand. The seeds were stored in a hot air oven at 55 °C for 4 h to kill any pests or contaminants, following the method of Mookherjee et al. (1968). The sterilised mungbean seeds (150 g) were then placed in a 500 mL plastic container, and 25 pairs of *C. maculatus* adults were released. The container was covered with a perforated muslin cloth lid to allow ventilation and prevent the escape of insects. The C. maculatus were allowed to breed and lay eggs for seven days under laboratory conditions (30 ± 5 °C and 70 \pm 5% relative humidity). The container remained in the laboratory until the adult progeny emerged. Newly adult *C. maculatus* (aged 1–3 days) were used for all further bioassays. All experiments were performed using a completely randomised design with four replications under the same laboratory condition described for insect rearing.

Essential oil. Pure EO from *J. sambac* was purchased in an amber bottle from Botanicessence Essential Oil [Location M-Square Plaza, B1 Floor, Maleenon Building (Channel 3), Rama 4 Road, Bangkok, Thailand]. The oil was stored in the dark at 4 °C until required for analyses and bioassays.

Chemical composition analysis. The chemical composition of J. sambac EO was determined according to the method of Satongrod and Wanna (2020) using a gas chromatograph-mass spectrometer (GC-MS) series Clarus 680 (PerkinElmer, Akron, OH, USA). Separation was achieved on an Elite-5MS capillary column (5% phenylmethyl polysiloxane stationary phase, 30 m, 0.32 mm i.d., 0.25 µm film thickness; PerkinElmer, USA). A 0.5 µL of the sample was injected with split mode (split ratio of 1:10 v/v). The carrier gas was helium with a 1.0 mL/min flow rate, and the injector temperature was maintained at 250 °C. The oven temperature was initially 60 °C and held for 0 min, then increased to 246 °C at a rate of 3 °C/min, operating in electron impact mode of 70 eV. A mass analyser was used as a quadrupole, and the temperature detector was set at 200 °C. Spectra were scanned (m/z) from 50 to 550 amu.

The identification of *J. sambac* EO constituents was carried out based on retention index (RI), determined with reference to homologous series of n-alkanes (C6-C23), MS Library search (NIST and WILEY), and by comparing RI and mass spectral data with the literature (Adams 2007). The relative amounts of individual components were calculated based on the relative % peak areas without using a correction factor.

Residual contact bioassay. Residual contact toxicity of J. sambac EO on adult C. maculatus was investigated by bioassays according to the applied methods of Kim et al. (2003) and Usha Rani and Rajasekharareddy (2010). A filter paper (7 cm diameter, surface area 23.77 cm²) was added with 0.3 mL of EO solution at different concentrations diluted with acetone at 0, 2, 4, 6 and 8 μL/L, respectively and placed in a petri dish (7 cm diameter). The control treatment was prepared using acetone only. Acetone was allowed to evaporate for 10 min before releasing five pairs of C. maculatus adults into each petri dish and covering them with a lid. All treatments were repeated four times following a completely randomised design. All petri dishes were kept under laboratory conditions (30 ± 5 °C and 70 ± 5% relative humidity). After 24, 48 and

72 h of treatment, adult mortality of *C. maculatus* was observed and recorded. The insects were defined as dead when no leg or antennal movements were detected (Wanna et al. 2021).

Seed dressing application. Mungbean seeds used in the bioassay were clean, healthy, and uninfected. J. sambac EO was diluted with acetone and mixed with 100 g of mungbean seeds at 0.2, 0.4, 0.6, 0.8 and 1 mL/kg seeds. A 500 mL flask containing 100 g of mungbean seeds and EO solution was shaken manually for about 2 min until the seeds were uniformly coated with the oils (Talukder & Howse 1994). The treated mungbean seeds were removed from the flask and air-dried for 1 h to completely evaporate the solvent. Then, 100 g of mungbean seeds were placed in a small jute bag (12×10 cm). Five pairs of C. maculatus adults were released into the bag, placed in a plastic container (17 \times 12 × 7 cm) and sealed with a perforated lid of muslin cloth to allow ventilation and prevent other insects from entering. The control was prepared using acetone only. The experiment was replicated four times following a completely randomised design. All containers were stored under laboratory conditions $(30 \pm 5 \, ^{\circ}\text{C} \text{ and } 70 \pm 5\% \text{ relative humidity}).$

Adult mortality of *C. maculatus* from each bag was observed and recorded every 24 hours for five days. The insects were determined as dead when no leg or antennal movements were detected (Wanna et al. 2021). After seven days, the *C. maculatus* adults were removed from each bag and stored for 30 days under the same laboratory conditions. The number of F1 adult emergence was observed from each bag after 30 days, and a percentage reduction in adult emergence or inhibition rate (% IR) was calculated. Percentages of seed damage and weight loss were recorded at the end of the experiment.

Data analysis. Adult mortality data were adjusted for control mortality according to Abbott's (1925) formula. Mortality in the control treatment ranged between 5 and 20%. Residual contact toxicity of *J. sambac* EO on *C. maculatus* adults was assessed for concentration-mortality response using probit analysis (Finney 1971) to provide median lethal concentration (LC_{50}) values and related parameters. Data from adult mortality, F1 progeny emergence, inhibition rate, seed damage, and weight loss were analysed using the F-test by oneway ANOVA. Mean values were compared at a 0.05 significance level by the least significant difference test (LSD) using Statistix (version 9.0).

RESULTS

Chemical composition. The GC-MS analysis of *J. sambac EO* from leaves led to the identification by retention index, as determined on an Elite-5MS column using the homologous series of n-hydrocarbons. 35 chemical components, covering 97.59% of peak areas, were detected within the time range of 3.483 and 60.135 min. Their relative importance varies from 0.10% to 22.88% in terms of peak areas (Table 1). The main compound was 3-hexen-1-ol benzo-

Table 1. Chemical composition of EO obtained from *Jasminum sambac*

No.	Compound	Retention	Peak area
		time (min)	(%)
1	3-hexen-1-ol	3.483	0.69
2	4-hexen-1-ol, acetate	6.895	1.08
3	benzyl alcohol	8.304	5.86
4	cis-linaloloxide	9.785	0.36
5	benzoylacetonitrile	10.026	1.15
6	linalool	10.551	10.98
7	phenylethyl alcohol	10.824	0.21
8	1,6-heptadiyne	10.891	0.44
9	benzyl carbamate	13.041	12.95
10	methyl salicylate	13.986	1.36
11	linalyl acetate	16.511	6.36
12	acetic acid, 2-phenylethyl ester	16.596	0.36
13	indole	18.337	3.83
14	methyl anthranilate	20.457	12.82
15	1H-azepine-1-carboxylic acid, 2-methyl- methyl ester	22.689	0.10
16	α-farnesene	26.817	2.07
17	γ-muurolene	26.902	0.13
18	isoledene	27.172	0.27
19	3-hexen-1-ol benzoate	29.810	22.88
20	methyl jasmonate	31.996	0.64
21	α-cadinol	32.369	0.17
22	andrographolide	33.027	0.11
23	oplopanonyl acetate	35.209	0.36
24	benzyl benzoate	36.513	2.89
25	α-vetivol	38.369	0.25
26	4-benzyloxybenzoic acid	39.858	0.60
27	trans-geranylgeraniol	44.955	2.38
28	1-hexadecanol	46.544	0.15
29	9,12,15-Octadecatrienal	47.081	0.14
30	9-Nonadecene	49.418	0.09
31	1,7-Hexadecadiene	51.625	0.15
32	1-Heneicosanol	52.516	5.33
33	1,9-eicosadiene	57.678	0.11
34	1-cyclohexyl-1-pentyne	59.624	0.19
35	hexadecanoic acid, phenylme- thyl ester	60.135	0.13
Tota	1		97.59

Table 2. Residual contact LC50 values of Jasminum sambac EO against adults of Callosobruchus maculatus

Time (h)	п	LC ₅₀ (95% CL) (μL/mL)	LC ₉₀ (95% CL) (μL/mL)	Regression equation	r^2
24	240	5.01 (4.07-5.82)	9.06 (7.85–10.20)	y = 9.875x + 0.50	0.98
48	240	4.00 (3.00-5.08)	7.60 (6.40-8.98)	y = 11.125x + 5.50	0.97
72	240	3.13 (1.99-3.66)	6.43 (4.56–6.50)	y = 12.125x + 12.00	0.94

n – the number of adult *Callosobruchus maculatus* tested; CL – the confidence limit; r^2 – the correlation coefficient; LC₅₀ and LC₉₀ – the concentration of EO *Jasminum sambac*, which is lethal to 50% and 90% of *C. maculatus* exposed during the testing times, respectively

Table 3. Residual contact toxicity of *Jasminum sambac* EO against *Callosobruchus maculatus* after 24, 48 and 72 h exposure

Concen-	Mean mortality (%) ± SE				
tration (μL/mL)	24 h	48 h	72 h		
0	0.00 ± 0.00^{d}	0.00 ± 0.00^{d}	$0.00 \pm 0.00^{\rm e}$		
2	20.00 ± 8.16^{c}	32.50 ± 9.57^{c}	47.50 ± 5.00^{d}		
4	37.50 ± 9.57^{b}	50.00 ± 8.16^{b}	65.00 ± 5.77^{c}		
6	67.50 ± 5.00^{a}	80.00 ± 0.00^{a}	90.00 ± 0.00^{b}		
8	75.00 ± 5.77^{a}	87.50 ± 9.57^{a}	100.00 ± 0.00^{a}		
F-test	非非	非非	非非		
CV (%)	16.46	12.51	5.65		
LSD	9.9214	9.3314	5.1479		

^{**}a significant difference at P < 0.01

Means followed by the same letter(s) in the same column are not significantly different (P > 0.05) from each other using the least significant difference test

ate (22.88%), followed by benzyl carbamate (12.95%), methyl anthranilate (12.82%), linalool (10.98%), linalyl acetate (6.36%), benzyl alcohol (5.86%), 1-heneicosanol (5.33%), indole (3.83%), benzyl benzoate (2.89%),

trans-geranylgeraniol (2.38%), α -farnesene (2.07%), methyl salicylate (1.36%), benzoylacetonitrile (1.15%) and 4-hexen-1-ol, acetate (1.08%).

Residual contact bioassay. *J. sambac* EO caused 75.00% mortality after 24 h exposure at the highest concentration of 8 μ L/mL with LC₅₀ and LC₉₀ values of 5.01 and 9.06 μ L/mL. The residual contact toxicity at 48 h showed 87.50% mortality with LC₅₀ of 4.00 and LC₉₀ of 7.60 μ L/mL. The 100% mortality was found at 72 h with residual exposure toxicity of 3.13 and 6.43 μ L/mL, respectively. Mortalities of *C. maculatus* at 6 μ L/mL after 24 and 48 h were not significantly different from 8 μ L/mL (Tables 2 and 3).

Seed dressing application. The mortality of *C. maculatus* reached 100% at 1 mL/kg after four days of exposure and was not significantly different from 0.8 mL/kg. After five days of exposure, the 0.8 mL/kg dose rate presented 100% mortality. Mortality at the lowest concentration of 0.2 mL/kg significantly differed from the control throughout the exposure time (Table 4).

Adult emergence of *C. maculatus* was highest in the control treatment at 12.75 ± 0.96 . At the highest con-

Table 4. Mortality percentage of *Callosobruchus maculatus* adults after 1, 2, 3, 4, and 5 days on mungbean seeds treated with different concentrations of *Jasminum sambac* EO

D (1 : 1	Insect mortality [mean (%) ± SE] Residual contact					
Dose of <i>Jasminum sambac</i>						
EO (mL/kg)	day 1	day 2	day 3	day 4	day 5	
Control	$0.00 \pm 0.00^{\rm e}$	$0.00 \pm 0.00^{\rm e}$	$0.00 \pm 0.00^{\rm e}$	0.00 ± 0.00^{d}	$0.00 \pm 0.00^{\rm e}$	
0.2	25.00 ± 5.77^{d}	37.50 ± 5.00^{d}	55.00 ± 5.77^{d}	70.00 ± 8.16^{c}	80.00 ± 0.00^{d}	
0.4	30.00 ± 8.16^{d}	40.00 ± 8.16^{d}	55.00 ± 5.77^{d}	$72.50 \pm 5.00b^{c}$	$87.50 \pm 5.00^{\circ}$	
0.6	40.00 ± 0.00^{c}	$52.50 \pm 5.00^{\circ}$	$67.50 \pm 5.00^{\circ}$	80.00 ± 0.00^{b}	95.00 ± 5.77^{b}	
0.8	47.50 ± 5.00^{b}	62.50 ± 5.00^{b}	77.50 ± 5.00^{b}	92.50 ± 9.57^{a}	100.00 ± 0.00^{a}	
1	60.00 ± 0.00^{a}	77.50 ± 5.00^{a}	87.50 ± 5.00^{a}	100.00 ± 0.00^{a}	100.00 ± 0.00^{a}	
F-test	非非	非非	赤赤	赤赤	非非	
CV (%)	13.52	11.71	8.51	7.99	4.05	
LSD	6.7807	7.8297	7.2186	8.2118	4.6321	

^{**}significant difference at P < 0.01

Means followed by the same letter(s) in the same column are not significantly different (P > 0.05) from each other using the least significant difference test

Table 5. Adult emergence and seed losses after seed treatment of *Jasminum sambac* EO against *Callosobruchus maculatus*

Dose of <i>Jasminum sambac</i> EO (mL/kg)	F1 adult emergence	Inhibition rate (%)	Seed damage (%)	Weight loss (%)
Control	12.75 ± 0.96^{a}	_	0.53 ± 0.06^{a}	3.73 ± 0.04^{a}
0.2	7.25 ± 0.96^{b}	42.95 ± 8.41^{c}	0.24 ± 0.03^{b}	1.59 ± 0.09^{b}
0.4	6.00 ± 0.82^{c}	52.75 ± 7.62^{bc}	0.19 ± 0.03^{c}	1.39 ± 0.09^{c}
0.6	5.00 ± 0.82^{c}	60.33 ± 8.86^{b}	0.16 ± 0.00^{c}	1.16 ± 0.09^{d}
0.8	0.50 ± 1.00^{d}	96.16 ± 7.69^{a}	0.01 ± 0.03^{d}	$0.02 \pm 0.05^{\rm e}$
1.0	0.00 ± 0.00^{d}	100.00 ± 0.00^{a}	0.00 ± 0.00^{d}	$0.00 \pm 0.00^{\rm e}$
F-test	ગ્રેલ્ ગ્રેલ્	赤米	非非	非非
CV (%)	15.87	10.36	17.21	5.18
LSD	1.2380	11.002	0.0479	0.1013

^{**}a significant difference at P < 0.01

Means followed by the same letter(s) in the same column are not significantly different (P > 0.05) from each other using the least significant difference test

centration of 1 mL/kg, no adult emergence of *C. maculatus* progeny was recorded, with 100% inhibition. This was not significantly different from the 0.8 mL/kg treatment that showed a number of progenies at 0.50 ± 1.00 and 96.16% inhibition rates, respectively. Progeny production significantly decreased with increased dosage of *J. sambac* EO (Table 5).

C. maculatus causes seed damage and weight loss in stored mungbean. Seed damage and weight loss were highest in the control treatment at 0.53% and 3.73%, respectively, with no seed damage and weight loss at the highest 1 mL/kg concentration of *J. sambac* EO (Table 5).

DISCUSSION

J. sambac EO contained many important compounds, including 3-hexen-1-ol benzoate, methyl anthranilate, linalool, benzyl alcohol, indole, benzyl benzoate and à-farnesene. Results were consistent with Ahmed et al. (2016) who reported the main chemical constituents of J. sambac EO as linalool, benzyl acetate and benzyl benzoate, while Edris et al. (2008) identified benzyl acetate, indole, E-Eα-farnesene, Z-3-hexenyl benzoate, benzyl alcohol, linalool, and methyl anthranilate in EO from J. sambac flower. Jirovetz et al. (2007) reported the chemical constituents of J. grandiflorum L. from GC and GC-MS analysis as benzyl acetate, benzyl benzoate, phytol, linalool, isophytol, geranyl linalool, methyl linoleate and eugenol. Younis et al. (2011) determined the chemical compositions of J. sambac EO from Pakistan obtained at the closed bud and open flower stages. The major identified compounds were citronellol, phenyl ethyl alcohol, geranial, eugenol, farnesol, geranyl acetate, citrinyl acetate, 2-phenyl ethyl acetate, citral and benzyldehyde. The composition of the volatile fraction of Egyptian *J. sambac* flowers was studied using GC-MS. The main volatile constituents were benzyl acetate, indole, E-E- α -farnesene, Z-3-hexenyl benzoate, benzyl alcohol, linalool and methyl anthranilate (Edris et al. 2008).

For residual contact toxicity bioassay, higher concentrations of J. sambac EO resulted in greater mortality of C. maculatus. J. sambac EO was LC₅₀ of 3.13 µL/mL and at 8 µL/mL gave the highest efficiency protection against adults of C. maculatus with 100% mortality within 72 h after exposure. Benzyl benzoate in J. sambac EO increases agricultural yield by combating pests in agricultural production. Benzyl benzoate has been reported to be acaricidal (McDonald & Tovey 1993) and insecticidal (Jantan et al. 2005). Veeraphant et al. (2011) reported that EO from J. sambac flower at a concentration of 0.1% can kill 100% of Dermatophagodes pteronyssinus within 10 min, and its LC₅₀ values at 24 and 48 h were 0.0079 and 0.0072 mL/mL, respectively. The most active J. sambac EO contains 2.27% benzyl benzoate, but it is very active against *D. pteronyssinus*; it may be due to the effect of the other minor compounds which has strong activity against D. Pteronyssinus, with LC50 of benzyl benzoate at 24 and 48 h with 0.0003 and < 0.0001 mL/mL, respectively. Massango et al. (2017) explained that insects are exposed to plant toxins through contact with joint membranes. EOs from plants have an inhibitory effect on the activity of glutathione S-transferases, an

enzyme that helps to eliminate insect toxins and ultimately causes the insects to die. El-Wakeil (2013) stated that plant extract toxins enter the insect body through the spiracle, impacting cellular respiration by inhibiting or disrupting NADH-coenzyme ubiquinon reductase (complex I) activity and the mitochondrial electron transport system. As a result, the insect is deprived of oxygen and dies. Rani (2012) concluded that plant toxins interfere with acetylcholinesterase (AChE) function and disrupt the entry-exit of ions into the nerve impulses, leading to paralysis and insect death. EOs are produced from renewable, botanical, biodegradable products. They act at low doses, are economical, have low environmental impact and are often undetectable. Kalaiselvi et al. (2011) reported that the methanolic extract of J. sambac flower through acute and subchronic toxicity studies in mice reported that it is quite safe and can be used to treat chronic diseases like diabetes without toxicity.

Using *J. sambac* EO at 1 mL/kg gave optimal 100% efficiency for C. maculatus control over four days and inhibited adult F1 emergence of C. maculatus at 100%. Plant EOs act as a repellent, insecticide, antifeedant, oviposition deterrent, and growth regulators (Koul et al. 2008). Rotimi and Evbuomwan (2012) and Upadhyay (2012) reported that the abdomen of female insects contains an ovipositor which contains chemosensillar. At the appropriate place or location on the seed, insects use their ovipositor to pierce a hole in which to lay their eggs and then cover them with a sticky substance. If the area or position is unsuitable, the insect will not lay eggs and continue searching for another laying location. Obembe and Ojo (2018) explained that plant toxins affect the chemical sensory neurons of the insect for oviposition. If the seed surface is unsuitable or poisonous, the insect stops touching the spawn and retreats without damage to the mung bean seeds.

Our results demonstrated that a commercially pure EO product of *J. sambac* was an effective residual contact method with significant toxicity against *C. maculatus* under storage conditions of mungbean seeds. This EO demonstrated potential as an alternative to synthetic insecticides for population control of *C. maculatus* during mungbean storage. This information will be useful for farmers in developing countries to control stored product pests, including *C. maculatus*. It would also be interesting to determine the exact compounds responsible for the biocidal activity, their mecha-

nisms of action on the targets, and their effects on consumers and subsidiaries. Moreover, for the practical application of EOs as novel insecticides, further studies on the safety of EOs to humans and the development of formulations are necessary to improve the efficacy and stability and reduce cost.

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