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GC-MS-MS profiling and toxicity of ethyl acetate extracts of some botanicals against rice weevil, *Sitophilus oryzae* (L.) in stored maize

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ABSTRACT

Ethyl acetate extracts of certain botanicals were evaluated in the laboratory for their contact and oral toxicity at 5% concentration against *Sitophilus oryzae* L. at Natural Pesticide Laboratory, Department of Agricultural Entomology, Agricultural College and Research Institute, Madurai, Tamil Nadu during 2019-21. The result revealed that all the botanicals were effective against *S. oryzae* compared to the control. Considering the contact toxicity, 5% ethyl acetate extract of *Mentha spicata* (86.67%) performed better 72 h after treatment. *Vitex negundo* and *Ocimum sanctum* caused (83.33%) mortality 72 h after treatment concerning oral toxicity. *Mentha spicata* caused 93.33% mortality after 15 days of treatment, whereas *Vitex negundo*, *Ocimum sanctum* and *Tagetes erecta* leaf caused 90.00% mortality, which was statistically at par with *Mentha spicata*. GC-MS-MS study revealed the presence of carvone (38.27%) at the retention time of 7.644, Hexadecanoic acid (2.13%) at the retention time of 27.600, Isopulegol (1.41%) at the retention time of 27.290 and phytochemicals with insecticidal potential in *Mentha spicata*.Whereas in *Ocimum sanctum*, Eugenol (59.72%) was the major compound at the retention time of 9.9. Hence, it was concluded that among the tested ethyl acetate extracts, *Mentha spicata* at 5% was the most effective as a contact and oral toxicant against *S. oryzae* in stored maize, which possesses the bioactive principle of carvone and it can be developed as an eco-friendly botanical formulation.

Key words: Botanicals, carvone, ethyl acetate, *Mentha spicata, Sitophilus oryzae*, toxicity

INTRODUCTION

Maize is a vital crop globally, but post-harvest losses from storage pests like *S. oryzae* pose a challenge, causing significant damage and reducing production. *S. oryzae* is highly destructive, infesting various grains during storage. Favourable conditions lead to rapid reproduction and extensive losses. Chemical pesticides have drawbacks, prompting the exploration of botanicals as an alternative solution. Botanical extracts contain bioactive phytochemicals with minimal non-target and environmental harm (Mahendra Mallah *et al.*, 2018). Addressing these issues is crucial for sustainable maize production (Thomas *et al.*, 2002; Park *et al.*, 2003; Batta, 2004; Mohale *et al.*, 2010).

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MATERIALS AND METHODS

Insect Collection, Rearing and Preparation of Extracts

Insect culture was maintained at the Natural Pesticide Laboratory in Madurai, Tamil Nadu, during 2019-21 under controlled temperature (30±2°C) and humidity conditions (70±5%) (Jayakumar *et al.*, 2017). Maize seeds were heat-treated at 40°C for 48 h to remove infestation. Adult weevils were reared and their F_1 generation was used for bioassays. Eleven botanicals, including *Acorus calamus*, were prepared as extracts for evaluation. The plant materials were processed, extracted with ethyl acetate, concentrated and stored for bioassays.

Contact Toxicity

Botanical extracts (0.5 g) were diluted to 5% with acetone. Circular filter paper discs (9cm diameter) were treated with 5% extracts or acetone as control. After drying, the discs were placed in Petri dishes with 10 weevils, and the lids were coated with vaseline. Each treatment was replicated three times, and per cent mortality was recorded after 24, 48 and 72 h (Kim and Ahn, 2001; Rani, 2012; Chaubey, 2016).

Oral Toxicity

Maize seeds (10 g) were coated with 5% botanical extracts and placed in plastic jars, with acetone as the control. Five pairs of adult weevils were released in each jar, and mortality was recorded after specific time intervals. Per cent mortality was calculated and the Abbott (1925) formula was used to correct for control mortality (Rani *et al.*, 2019).

Detection and Identification of Bioactive Compounds

For GC-MS/MS analysis, the crude ethyl acetate extracts of selected botanicals were prepared by diluting 0.2 ml of the extract with 1.5 ml of the respective solvent, filtering it and injecting it into a Gas Chromatography-Mass Spectrometry system. The analysis was conducted using a TQ 8040 instrument equipped with an Rxi-5 Sil MS column. The identification of compounds was performed by comparing the spectra of unidentified components with those stored in the NIST 17 mass spectral database. The retention time, per cent area, compound name, molecular formula and molecular weight were determined for each identified compound. The analysis was conducted according to the parameters mentioned by Jain *et al.* (2016).

Table 1. Contact toxicity of ethylacetate extract of certain botanicals against *S. oryzae* adults

Treatment	Cumulative contact toxicity (%)		
	24 h	48 h	72 h
T_1–*C.aurantium* leaf 5%	40.00±10.00 (39.14)^{e}	56.67±5.77 (48.84)^{d}	66.67±5.77 (54.78)^{d}
T_2–*C. longa* rhizome 5%	50.00±10.00 (44.99)^{cde}	56.67±5.77 (48.84)^{d}	63.33±5.77 (52.77)^{d}
T_3–*E. globulus* leaf 5%	53.33±15.28 (47.00)^{cde}	63.33±15.28 (53.06)^{cd}	70.00±10.00 (56.99)^{d}
T_4–*L. camara* leaf 5%	56.67±11.55 (48.93)^{cd}	63.33±15.28 (53.06)^{cd}	66.67±15.28 (55.07)^{d}
T_5–*M. spicata* leaf 5%	73.33±5.77 (59.00)^{b}	83.33±5.77 (66.14)^{ab}	86.67±5.77 (68.85)^{b}
T_6–*M. koenigii* leaf 5%	56.67±5.77 (48.84)^{cd}	60.00±0.00 (50.76)^{cd}	66.67±5.77 (54.78)^{d}
T_7–*O. sanctum* leaf5%	73.33±5.77 (59.00)^{b}	83.33±5.77 (66.14)^{ab}	83.33±5.77 (66.14)^{bc}
T_8–*R. communis* leaf 5%	40.00±10.00 (39.15)^{e}	56.67±5.77 (48.84)^{d}	70.00±10.00 (56.99)^{d}
T_9–*T. erecta* leaf 5%	60.00±10.00 (50.85)^{bc}	73.33±5.77 (59.00)^{bc}	73.33±5.77 (59.00)^{cd}
T_{10}–*T. erecta* flower 5%	43.33±5.77 (41.15)^{de}	53.33±5.77 (46.92)^{d}	60.00±0.00 (50.76)^{d}
T_{11}–*V. negundo* leaf 5%	73.33±5.77 (59.00)^{b}	80.00±0.00 (63.43)^{ab}	83.33±5.77 (66.14)^{bc}
T_{12}–*A. calamus* 1% (Standard check)	86.67±5.77 (68.85)^{a}	90.00±0.00 (71.56)^{a}	100.00±0.00 (89.50)^{a}
T_{13}–Acetone (Control)	0.00±0.00 (0.66)^{e}	0.00±0.00 (0.66)^{e}	0.00±0.00 (0.66)^{e}
T_{14}–Untreated check	0.00±0.00 (0.66)^{e}	0.00±0.00 (0.66)^{e}	0.00±0.00 (0.66)^{e}
Mean	50.48	58.57	63.57
S.Ed.	3.99	3.72	3.67

*Mean values of three replications are represented as mean±standard deviation. Figures in the parentheses are arcsine transformed values. In a column, the means followed by the same superscript are not significantly different from each other, DMRT (P ≤ 0.05). S. Ed: Standard error of the difference.

Table 2. Oral toxicity of ethyl acetate extract of different botanicals against adults of *Sitophilusoryzae*

Ethyl Acetate extracts	Cumulative oral mortality (%)*				
	3 DAT	5 DAT	7 DAT	9 DAT	15 DAT
T_1–*C. aurantium* leaf 5%	53.33±5.77 (46.92)^{e}	60.00±0.00 (50.76)^{d}	70.00±0.00 (59.79)^{d}	76.67±5.77 (61.21)^{cd}	76.67±5.7 (61.21)^{de}
T_2–*C. longa* rhizome 5%	56.67±5.77 (48.84)^{de}	60.00±0.00 (50.76)^{d}	70.00±0.00 (56.79)^{d}	73.33±5.77 (59.00)^{d}	73.33±5.77 (59.00)^{e}
T_3–*E. globulus*leaf 5%	66.67±5.77 (54.78)^{bcd}	70.00±0.00 (56.79)^{bc}	73.33±5.77 (59.00)^{cd}	80.00±0.00 (63.43)^{bcd}	86.67±5.77 (68.85)^{cd}
T_4–*L. camara* leaf 5%	60.00±10.00 (50.85)^{cde}	63.33±5.77 (52.77)^{cd}	73.33±5.77 (59.00)^{cd}	76.67±5.77 (61.21)^{cd}	80.00±10.00 (63.92)^{cde}
T_5–*M. spicata* leaf 5%	70.00±0.00 (56.79)^{bc}	76.67±5.77 (61.21)^{b}	83.33±5.77 (66.14)^{b}	86.67±5.77 (68.85)^{bc}	93.33±5.77 (77.37)^{b}
T_6–*M. koenigii* leaf 5%	66.67±5.77 (54.78)^{bcd}	70.00±0.00 (56.79)^{bc}	76.67±5.77 (61.21)^{bcd}	80.00±0.00 (63.43)^{bcd}	86.67±5.77 (68.85)^{cd}
T_7–*O. sanctum* leaf 5%	70.00±0.00 (56.79)^{bc}	73.33±5.77 (59.00)^{b}	80.00±0.00 (63.43)^{bc}	83.33±5.77 (66.14)^{bcd}	90.00±0.00 (71.56)^{bc}
T_8–*R. communis* leaf 5%	60.00±10.00 (50.85)^{cde}	63.33±5.77 (52.77)^{cd}	73.33±5.77 (59.00)^{cd}	76.67±5.77 (61.21)^{cd}	80.00±10.00 (63.92)^{cde}
T_9–*T. erecta* leaf 5%	70.00±0.00 (56.79)^{bc}	70.00±0.00 (56.79)^{bc}	76.67±5.77 (61.21)^{bcd}	80.00±0.00 (63.43)^{bcd}	90.00±0.00 (71.56)^{bc}
T_{10}–*T. erecta* flower 5%	56.67±5.77 (48.84)^{de}	60.00±0.00 (50.76)^{d}	70.00±0.00 (56.79)^{d}	73.33±5.77 (59.00)^{d}	73.33±5.77 (59.00)^{e}
T_{11}–*V. negundo* leaf 5%	73.33±5.77 (59.00)^{b}	76.67±5.77 (61.21)^{b}	76.67±5.77 (61.21)^{bcd}	83.33±5.77 (72.12)^{b}	90.00±0.00 (71.56)^{bc}
T_{12}–*A. calamus* 1% (Standard check)	83.33±5.77 (66.14)^{a}	86.67±5.77 (68.85)^{a}	100.00±0.00 (89.01)^{a}	100.00±0.00 (89.01)^{a}	100.00±0.00 (89.01)^{a}
T_{13}–Acetone (Control)	0.00±0.00 (0.91)^{f}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{f}
T_{14}–Untreated check	0.00±0.00 (0.91) ^{f}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{e}	0.00±0.00 (0.91)^{f}
Mean	56.19	59.29	65.95	69.29	72.86
S.Ed.	2.64	2.11	2.36	2.36	3.52

*Mean values of three replications are represented as mean±standard deviation. DAT–Days after treatment. Figures in the parentheses are arc sine transformed values. In a column, the means followed by the same superscript are not significantly different from each other. DMRT (P ? 0.05). S. Ed: Standard error of the difference.

Table 3. Phytochemical profile of ethyl acetate extract of *Mentha spicata* analyzed by GC-MS/MS

Peak	Retention time	% area	Compounds	Molecular formula	Molecular weight (Da)	Functional group
1	7.644	38.27	(-)-Carvone	$C_{10}H_{14}O$	150	Terpenoid
2	14.005	0.45	Calcifediol	$C_{27}H_{44}O_2$	400	Alcohol
3	17.825	0.58	Imidazole, 5-diethylamino-1-methyl-4-nitro-	$C_8H_{14}N_4O_2$	198	Diazole
4	18.439	0.51	3(2H)-Thiophenone, dihydro-, oxime, 1,1-dioxide	$C_4H_7NO_3S$	149	Ketone
5	19.051	1.72	Pentadecanoic acid	$C_{15}H_{30}O_2$	242	Fatty acid
6	19.273	0.42	2-Butene-1,4-diol, 2TMS derivative	$C_{10}H_{24}O_2Si_2$	232	Alcohol
7	19.458	0.82	6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one	$C_{11}H_{16}O_3$	196	Ketone
8	19.586	0.54	Behenyl chloride	$C_{22}H_{45}Cl$	344	Alkane
9	19.789	0.45	R-(+)-Methyl-2-isopropyl-5-oxohexanoate	$C_{10}H_{18}O_3$	186	Carboxylic acid
10	20.172	0.71	3,7-Dimethyloctyl acetate	$C_{12}H_{24}O_2$	200	Esters
11	20.307	41.13	Neophytadiene	$C_{20}H_{38}$	278	Diterpene
12	20.478	3.51	Sesquimustard	$C_6H_{12}C_{12}S_2$	218	Sulfur
13	26.948	2.39	Morphinan-4,5-epoxy-3,6-di-ol, 6-[7-nitrobenzofurazan-4-yl]amino-	$C_{26}H_{27}N_5O_6$	505	Alkane
14	27.29	1.41	Isopulegol	$C_{10}H_{18}O$	154	Terpenoids
15	27.435	0.77	Dodecanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5,5a-dihydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-11-oxo-1H-2,8a-methanocyclopenta[a	$C_{32}H_{50}O_6$	530	Fatty acid
16	27.6	2.13	Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester	$C_{35}H_{68}O_5$	568	Fatty acid, Ester
17	27.73	2.9	2-Undecenoic acid	$C_{11}H_{20}O_2$	184	Fatty acid

Table 4. Phytochemical profile of ethyl acetate extract of *Ocimum sanctum* analyzed by GC- MS/MS

S. No.	Retention time	% area	Compounds	Molecular formula	Molecular weight (Da)
Carboxylic acid					
1.	9.002	0.02	cis-5-Dodecenoic acid, dimethyl (3,3,3-trifluoropropyl) silylester	$C_{17}H_{31}F_3O_2Si$	352
2.	14.311	0.05	Azelaic acid	$C_9H_{16}O_4$	188
3.	9.876	0.05	8,11-Octadecadiynoic acid, methyl ester	$C_{19}H_{30}O_2$	290
		0.12			
Terpenoids					
4.	29.769	0.72	o-Mentha-1(7),8-dien-3-ol	$C_{10}H_{16}O$	152
5.	9.981	59.72	Eugenol	$C_{10}H_{12}O_2$	164
6.	11.486	14.03	Caryophyllene	$C_{15}H_{24}$	204
7.	12.311	1.43	Humulene	$C_{15}H_{24}$	204
8.	27.951	0.51	Sarreroside	$C_{30}H_{42}O_{10}$	562
9.	31.983	0.5	Milbemycin B, 6,28-anhydro-15-chloro-25-isopropyl-13-dehydro-5-O-demethyl-4-methyl-	$C_{33}H_{47}ClO_7$	590
10.	13.315	4.25	Azulene, 1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, [1R-(1.alpha.,3a.beta.,4.alpha., 7.beta.)]-	$C_{15}H_{24}$	204
		81.16			
Amine					
11.	6.591	0.09	Benzylamine, 2-hydroxy-N,N-di-[2-aminoethyl]-	$C_{11}H_{19}N_3O$	209
Alcohol					
12.	6.776	0.07	1-Methyl-4-[nitromethyl]-4-piperidinol	$C_7H_{14}N_2O_3$	174
13.	16.894	3.19	Neointermedeol	$C_{15}H_{26}O$	222
14.	21.817	0.14	Selin-6-en-4.alpha.-ol	$C_{15}H_{26}O$	222
		3.4			
Amide					
15.	7.093	0.02	Arachidonic amide, N-[5-hydroxy-n-pentyl]-	$C_{25}H_{43}NO_2$	389
16.	9.211	0.02	Arachidonoylethanolamide	$C_{22}H_{37}NO_2$	347
		0.04			
Amino acid					
17.	7.85	0.15	L-Methionine	$C_5H_{11}NO_2S$	149
18.	8.515	0.02	Glutaric acid, myrtenyl 2-methylpent-3-yl ester	$C_{21}H_{34}O_4$	350
		0.17			
Ester					
19.	8.055	0.06	Methyl 10,12-heptadecadiynoate	$C_{18}H_{28}O_2$	276
20.	14.889	0.07	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	436
		0.13			
Monoterpene					
21.	8.144	0.03	Isopulegol	$C_{10}H_{18}O$	154
22.	30.488	0.57	1-Methylverbenol	C11H18O	166
		0.6			
Ketone					
23.	8.541	0.04	2(1H)-Naphthalenone, 4a,5,8,8a-tetrahydro-4a-methyl-, trans-	$C_{11}H_{14}O$	162
Sesquiterpenoids					
24.	11.13	0.28	Santamarine	$C_{15}H_{20}O_3$	248
25.	5.337	0.02	Chlorozotocin	$C_9H_{16}ClN_3O_7$	313
26.	14.189	0.05	Caparratriene	$C_{15}H_{26}$	206
27.	33.36	0.16	(-)-Isolongifolol, TMS derivative	$C_{18}H_{34}OSi$	294
		0.51			
Sugars					
28.	11.903	0.01	Stevioside	$C_{38}H_{60}O_{18}$	804
Steroid (Fatty acid)					
29.	12.065	0.04	Danazol	$C_{22}H_{27}NO_2$	337
30.	20.614	2.26	Digitoxin	$C_{41}H_{64}O_{13}$	764
		2.3			
Diterpenoid					
31.	19.448	4.55	Neophytadiene	$C_{20}H_{38}$	278
32.	25.624	3.14	Phytol	$C_{20}H_{40}O$	296
		7.69			
Triterpenoid					
33.	21.385	0.2	Ergostane-3,5,6,12,25-pentol, 25-acetate, (3.beta., 5.alpha.,6.beta.,12.beta.)-	$C_{30}H_{52}O_6$	508

This methodology allowed for the detection and identification of bioactive compounds present in the extracts.

Statistical Analysis

The experiment was conducted in a completely randomized design (CRD). The per cent mortality was determined, transformed to arcsine values and statistically analyzed using SPSS for Windows (version 16) software to carry out an analysis of variance (ANOVA). The grouping of data was done by using Duncan's Multiple Range Test (DMRT) (Gomez and Gomez, 1984). The standard error of difference was calculated for each treatment effect to compare the mean difference among the treatments.

RESULTS AND DISCUSSION

The results revealed that all the botanicals exhibited toxicity against *S. oryzae* compared to untreated check and acetone control, while the treated check *A. calamus* 1% was more efficient than the other treatments.

Contact Toxicity of Ethylacetate Extracts on *Sitophilus oryzae*

Acorus calamus (1% concentration) exhibited 100% mortality after 72 h. *Mentha spicata* (5% concentration) showed 86.67% mortality, followed by *Vitex negundo* and *Ocimum sanctum* (both at 5% concentration) with 83.33% mortality (Table 1). Previous studies support the effectiveness of *Mentha spicata* and

Table 5. Phytochemical profile of ethyl acetate extract of *Vitex negundo* analyzed by GC- MS/MS

S. No.	Retention time	% area	Compounds	Molecular formula	Molecular weight (Da)
Sesquiterpenoid					
1.	15.51	6.19	(-)-Globulol	$C_{15}H_{26}O$	222
2.	25.164	1.45	Illudol	$C_{15}H_{26}O$	222
		7.64			
Carboxylic acid					
3.	20.175	1.29	2- Bromopropionic acid, octyl ester	$C_{11}H_{21}BrO_2$	264
		1.29			
Terpenoid					
4.	23.41	2.4	Flexibilide	$C_{20}H_{30}O_4$	334
5.	23.987	2.28	Kolavenol	$C_{20}H_{34}O$	290
6.	25.638	5.3	Phytol	$C_{20}H_{40}O$	296
7.	27.895	5.16	Neointermedeol	$C_{15}H_{26}O$	222
8.	28.085	1.34	Borneol, pentamethyldisilanyl ether	$C_{15}H_{32}OSi_2$	284
9.	28.89	2.5	Adamantane-1-carboxamide, N- (4-chlorobenzyl)-	$C_{18}H_{22}C_1NO$	303
10.	29.095	2.13	Azepine, 1-(dichloroboryl)-perhydro	$C_6H_{12}BC_{12}N$	179
11.	29.72	4.27	Isopulegol	$C_{10}H_{18}O$	154
		25.38			
Alkane					
12.	26.175	3.04	Cyclopentane, 2-n-octyl-	$C_{13}H_{24}$	180
Alcohol					
13.	26.289	16.35	3-Tetradecyn-1-ol	$C_{14}H_{26}O$	210
Amine					
14.	26.34	3.98	4-Chloro-3-n-hexyltetrahydropyran	$C_{11}H_{21}C_1O$	204
15.	26.408	7.13	4-Amino-6-morpholino-5-nitropyrimidine	$C_8H_{11}N_5O_3$	225
		11.11			
Ketone					
16.	26.365	3.72	12-Hydroxy-14-methyl-oxa-cyclotetradec-6-en-2-one	$C_{14}H_{24}O_3$	240
Ester					
17.	26.47	6.7	Phytyldecanoate	$C_{30}H_{58}O_2$	450
18.	28.541	1.84	Ethyl .alpha.-bromolaurate	$C_{14}H_{27}BrO_2$	306
		8.54			
Amino acid					
19.	28.505	1.09	Glutaric acid, monochloride, 3-m ethylpentyl ester	$C_{11}H_{19}C_1O_3$	234
Lactone					
20.	29.473	19.3	Milbemycin b, 13-chloro-5-dem ethoxy-28-deoxy-6,28-epoxy-5-(hydroxyimino)-25-(1-methylethyl)-, (6R,13R,25R)-	$C_{33}H_{46}C_1NO_7$	603

Vitex negundo against *S. oryzae* (Rahman *et al.*, 2003; Ainane *et al.*, 2019; Anandhabhairavi *et al.*, 2021; Anandhabhairavi *et al.*, 2022).

Oral Toxicity of Ethyl Acetate Extracts on *Sitophilus oryzae*

A. calamus (1% concentration) resulted in 100% mortality after seven days. *Mentha spicata* and *Vitex negundo* leaf extract exhibited high toxicity (93.33 and 90%, respectively) after 15 days at 5% concentration (Table 2). Previous studies confirm the efficacy of *Mentha arvensis*, *Ocimum sanctum* and *Vitex negundo* against *S. granarius*. LD_{50} values of *V. negundo* leaf extracts on *S. granarius* ranged from 2.490 to 3.727 ml/kg. Other studies demonstrated the toxic effects of *Ocimum basilicum* and *Eucalyptus saligna* on *S. oryzae* and *S. zeamais,* respectively.

GC-MS-MS Analysis

The ethyl acetate crude extract of *Mentha spicata* leaves was analyzed using GC-MS/MS, revealing 18 phytoconstituents, with (-)-Carvone as the most abundant compound (38.27%) (Table 3). (Aggarwal *et al.*, 2002; Shahbazi, 2015; Haouel-Hamdi *et al.*, 2021; Valcarcel *et al.*, 2021). Carvone exhibited toxic properties against *S. oryzae*, *Rhyzopertha dominica* and *Callosobruchus pusillus* (Pascual-Villalobos *et al.*, 2004; Garcia *et al.*, 2005). Hexadecanoic acid in *M. spicata* showed pesticide, nematicide and mosquito larvicide activity (Rahuman *et al.*, 2000; Aparna *et al.*, 2012). Isopulegol displayed highly toxic and persistent repellent effects (*Melo et al.*, 2021). Eugenol and caryophyllene were identified in *Ocimum sanctum* (Table 4). (Awasthi and Dixit, 2007). *Vitex negundo* contained bioactive compounds with insecticidal properties (Table 5). These findings are consistent with previous studies, emphasizing the potential of *M. spicata*, *O. sanctum* and *V. negundo* for insect control.

CONCLUSION

It was concluded that the ethyl acetate extract of *M. spicata* had contact and oral toxicity against *S. oryzae*, which possessed the bioactive principle of carvone. It shall be explored further for the development of botanical formulation.

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