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RESEARCH ARTICLE

Essential oil of garlic, *Allium sativum* L.: A promising alternative for the management of *Sitophilus oryzae* (L.)

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Abstract

The study evaluates the effectiveness of garlic essential oil as a fumigant and contact toxicant against adult rice weevils (*Sitophilus oryzae* L.) (Curculionidae: Coleoptera). Garlic essential oil demonstrated significant fumigant toxicity, causing 56.67% adult mortality at a concentration of 3 µl/96 cm^3 air on the first day, which increased to 99.13% by the fifth day after treatment (DAT). In addition, garlic essential oil exhibited contact toxicity, achieving up to 100% mortality at doses of 4 and 5 µl/40 g seeds. Even at lower concentrations (1 µl), mortality rates were as high as 78.33%. The calculated LC_{50} value was 2.58 µl/40 g of seeds. Garlic essential oil also had a considerable effect on reproduction, as no adults emergence was observed from seeds treated with 4 µl/40 g. GC-EAD analysis identified 39 compounds in garlic essential oil, with male and female *S. oryzae* exhibiting different antennal responses. Females displayed stronger reactions to alcohols and esters, while males were more responsive to alkenes and alkanes. The presence of chemical constituents in garlic essential oil that influence insect behaviour underscores its potential as a viable pest management solution against stored-product pests. Further exploration of these compounds for their insecticidal properties using GC-EAD studies and their development into the formulations could provide significant benefits to farmers and contribute to sustainable pest management practices.

Keywords

Allium sativum essential oil; fumigant and contact toxicity, GC-EAD; GCMS; *Sitophilus oryzae*

Introduction

Post-harvest losses during storage remain one of the primary obstacles to achieving food security in developing countries (1). Losses of up to 20% have been reported in major cereal crops (2), accounting for 53% in caloric terms. Maize (*Zea mays* L.), one of the most versatile cereal crops, is notable for its adaptability, various types and multifaceted uses, including food, fodder and fuel. It is the second most widely cultivated crop globally, following rice and is grown extensively in tropical and subtropical regions. The rice weevil (Curculionidae: Coleoptera) is a significant storage pest that infests maize as well as rice, sorghum, oats and barley (3). This pest is capable of penetrating and infesting undamaged kernels (4, 5) and can cause damage ranging from 85% to 100% in maize grains (6, 7, 8). In addition to direct damage, the presence of excreta and dead adults in stored maize raises serious

concerns. Moreover, heavy infestation often leads to the emergence of secondary pests and the development of harmful aflatoxins (9).

While synthetic insecticides can effectively reduce post-harvest losses, their use is increasingly discouraged due to their adverse effects on non-target organisms, the environment, the development of insecticide resistance and food residue contamination (10). Therefore, ecologically feasible and environmentally sustainable management strategies are essential for pest control. Plant-based products are gaining attraction as novel agents against storage pests due to their non-phytotoxic nature and easy biodegradability (11, 12). These botanicals exhibit a range of effects, including ovicidal, repellent, anti-feedant, sterilizing and toxic properties, while demonstrating broad-spectrum activity against various insect pests (12, 13).

Among botanicals, volatile essential oils derived from aromatic plants are being promoted as alternatives for pest management (14, 15). Essential oils disrupt insects' physiological, biochemical and behavioural processes due to their lipophilic nature, enabling both contact and fumigant toxicity (16). Many essential oils contain toxic compounds, such as monoterpenoids, which act as neurotoxicants, causing rapid knockdown or immobilization effects against specific insect pests (17, 18). They have been reported to exhibit neurotoxic, cytotoxic and mutagenic effects on storage pests (19, 20).

Steam distillates of garlic have demonstrated efficacy against various life stages of *Sitophilus zeamais* (21). The lethal and repellent effects of garlic essential oil on *Tenebrio molitor* have also been documented (22). Studies confirm the fumigant toxicity of garlic essential oil and its major components, such as diallyl disulfide, diallyl trisulfide and diallyl sulfide, against *Tribolium castaneum* (23, 24). Additionally, garlic essential oil has been shown to inhibit oviposition in *Sitotroga cerealella* (25). These studies highlight the diverse properties of garlic essential oil, including its repellent, fumigant and oviposition-deterrent effects.

The toxic compounds in garlic essential oil, such as diallyl disulfide and diallyl sulfide, have been shown to affect larval and pupal stages of *T. molitor*. Allicin, a compound in garlic essential oil, has been reported to alter insect locomotion and induce muscle contractions and paralysis (26). Furthermore, the high concentration of diallyl disulfide has been attributed to the mortality of the psyllid *Cacopsylla chinensis* (27). The fumigant toxicity of garlic essential oil has also been demonstrated against the Angoumois grain moth (*Sitotroga cerealella*), with diallyl trisulfide identified as a key contributor to its toxic effects (28).

This study is aimed to evaluate the fumigant and contact toxicities of garlic essential oil against *S. oryzae* and to investigate its effects on oviposition and adult emergence under laboratory conditions. Additionally, the compounds responsible for eliciting antennal responses in adult *S. oryzae* were analyzed using GC-EAD.

Materials and Methods

Insects

The rice weevil was cultured in the Central Instrumentation Laboratory, TNAU, Coimbatore, India. Adult *S. oryzae* were raised on fresh maize seeds that had been sun-dried to eliminate any residual insect stages. A 500 ml plastic container with approximately 400 g of rice grains, adequately infested with *S.oryzae*, was maintained at a moisture content of 12 percent. The culture was preserved under controlled environmental conditions of 27 ± 2 °C, 65 ± 5% relative humidity and a 12: 12 light-to-dark photoperiods. The container was covered with a muslin cloth to allow adequate ventilation while preventing the escape of insects (29, 30). Freshly emerged F1 adults from the culture were used for toxicity experiments, typically 39–45 days after the initial infestation. Subculturing was performed according to the above methodology to obtain a continuous supply of insects for experiments purposes.

Oil source

The essential oil was extracted following the methodology described by (31). About 1.5 kg of garlic cloves was oven-dried at 60^0C for three days. The dried cloves were powdered, yielding about 600g of material, which was subjected to extraction using a Soxhlet apparatus with acetone as the solvent in batches of 30 g. The powdered garlic sample (30g) was weighed, wrapped in Whatman No.1 filter paper and sealed securely. This prepared filter paper, containing with the powdered garlic, was placed in the thimble of the Soxhlet apparatus for the extraction process.

The sample-to- solvent ratio of 1: 10 was maintained for the extraction process, with 300 ml of acetone added to the round-bottom flask of the apparatus. The solvent was heated to a temperature of 50-60^0C and the extraction process was carried out for approximately 12 hours, ensuring the process was complete. The oil dissolved in the solvent was separated using a rotary evaporator, operating under reduced temperature and pressure. The extracted garlic essential oil was stored in a refrigerator at 4^0C for further studies.

For comparison purposes, the TNAU sweet flag (*Acorus calamus*) formulation was used as a standard reference, along with an untreated control to validate the experimental findings.

Fumigant toxicity

The fumigant toxicity of the essential oil against *S. oryzae* was evaluated following the methodology described by (32). Serial dilutions of the essential oil (1 µl, 1.5 µl, 2 µl, 2.5 µl and 3 µl) were prepared using acetone, based on prior range-finding tests. A Whatman No. 1 filter paper (1.5 cm in diameter) was impregnated with each test dose of the essential oil, adjusted to a final volume of 1.0 ml and subsequently placed in a Petri dish (9 cm in diameter, 1.5 cm in height and 96 cm^3in volume). The solvent was allowed to evaporate for 20 seconds before the insects were introduced.

Twenty unsexed adults of *S. oryzae* were released into each Petri plate. Filter papers treated with acetone (solvent) and untreated filter paper served as negative controls, while the TNAU sweet flag 6% EC formulation (0.8 ml, diluted to 1.0 ml with acetone) was used as positive control. All treatments were replicated thrice and left undisturbed under controlled condition at 27±2 °C and 65 ± 5% relative humidity. Insect mortality was

recorded daily for up to five days and the percentage mortality was corrected using Abbott's formula (33). The median lethal concentration (LC_{50}) was calculated based on the observed mortality across the different concentrations.

Contact toxicity

Adult mortality

The adult mortality due to the contact toxicity of essential oil against *S. oryzae* was assessed following the same methodology as described for fumigant toxicity studies. Serial dilutions of essential oil (1 µl, 2 µl, 3 µl, 4 µl and 5 µl) were prepared using acetone, based on prior range-finding tests. The essential oil was adjusted to a final volume of 1 ml with acetone and thoroughly mixed with 40 g of untreated maize grains (hybrid Co [H]M 8) in a glass jar. The mixture was stirred continuously for one minute manually to ensure even coating of the essential oil on the grain surface. The treated grains were left undisturbed for 20 sec to allow the solvent to evaporate completely.

Separate glass jars containing maize grains treated with acetone (solvent control) and untreated maize grain (negative control) were maintained, while grains treated with the TNAU sweet flag 6% EC formulation (0.8 ml diluted to 1.0 ml with acetone) served as positive control. All the treatments were replicated three times and stored under controlled conditions at 27±2 °C and 65 ± 5% relative humidity. Groups of 20 adult test insects, aged three to seven days, were introduced into each jar. The jars were covered with nylon mesh, secured with rubber bands, to prevent insect escape.

The percentage insect mortality was calculated using the appropriate formula and the median lethal concentration (LC_{50}) was determined based on the mortality observed at different concentrations.

$$\text{Corrected morality} = \frac{\text{Number of insects in treatment - Number of insects in control}}{\text{100 - Number of insects in control}} \times 100$$

Eqn. 1

Oviposition and adult emergence

The effect of garlic essential oil on oviposition and adult emergence of *S. oryzae* was evaluated following the methodology of (34). Maize grains (40 g) were treated with five different concentrations of garlic essential oil (1 µl, 2 µl, 3 µl, 4 µl and 5 µl), the TNAU sweet flag formulation (0.8 ml mixed with 0.2 ml acetone), acetone alone (1.0 ml) and untreated grains, following the same procedure as described in the contact toxicity studies. Each treatment was replicated three times.

After 20 min, when the solvent had evaporated completely, a pair of adult weevils aged 3 to 7days was introduced into each treatment setup. The adults were removed three days after their release. Oviposition was assessed on the fourth day by counting the number of egg plugs secreted over the eggs. Subsequently, adult emergence was recorded between days 38 and 44 to determine F2 progeny production.

Behavioural response

The essential oil formulation was subjected to GC-EAD analysis to identify its constituent compounds and to evaluate the behavioral response of *S. oryzae*. A gas chromatography (GC) system (Agilent 7890 B) equipped with a mass spectrometry detector (MSD) and coupled to a mass spectrometer (Agilent 5977) was employed for the analysis of garlic essential oil. A nonpolar HP-5 column with fused silica was used as the stationary phase and helium served as the carrier gas at a flow rate of 1 ml/min. The detector temperature was maintained at 300°C, while the injector temperature was set at 250°C. A 1 µl sample of the essential oil was injected in splitless GC-MS mode.

The oven temperature program began at 45°C (held for 1 minute), increased by 10°C per minute and was maintained at 280°C for 5 minutes. Compound identification was performed by comparing the obtained mass spectra with entries in the NIST 14 spectral database.

Electroantennogram (EAG) recordings were conducted on both male and female rice weevils to assess antennal responses. The heads of the insects were severed and placed on a reference electrode, ensuring proper alignment of the antennae. Ag-AgCl glass electrodes containing saline solution were used to facilitate signal recording. The saline solution was composed of sodium chloride, calcium chloride, potassium chloride, sodium bicarbonate, sodium orthophosphate and magnesium chloride dissolved in distilled water. A recording electrode was connected to a high-impedance DC amplifier (IDAC-4; Ockenfels Syntech, Buchenbach, Germany) for the experiment.

The analysis was further performed using a gas chromatograph (Agilent 7890 B GC) equipped with a flame ionization detector (FID) and a nonpolar HP-5 column. Nitrogen served as the carrier gas, with the injector and detector temperatures maintained at 250°C and 300°C, respectively. The oven temperature program mirrored that used in GC-MS analysis and a 2 µl sample was injected in splitless mode. The GC effluent was divided into two streams: one directed towards the detector and the other routed to the antennal preparation. A glass tube with humidified airflow was employed to transport the compounds from the GC column to the insect antenna.

Signals from the antenna and FID were recorded simultaneously and analyzed using specialized software provided by Ockenfels Syntech. Peaks from the GC column were deemed active if they elicited EAG responses in at least two out of three consecutive runs conducted during the experiment (35).

Statistical analysis

The data were corrected for control mortality using Abbott's formula. The toxicity data were analyzed using probit analysis to estimate the lethal concentrations (LC_{50} and LC_{95}), along with their 95% fiducial limits. The data on percentage mortality, egg-laying and percentage adult emergence were approximately transformed and statistically analyzed using one-way analysis of variance (ANOVA). All statistical analyses were done by using R software v.4.4.0. The compounds identified through GC-EAG were subjected to PCA biplot analysis to group them.

Results and Discussion

Fumigant toxicity

The garlic essential oil exhibited significant fumigant toxicity, with LC_{50} and LC_{95} values of 2.04 ppm and 4.71 ppm, respectively, against *S. oryzae* (Table 1). The fumigant toxicity of various plant essential oils has been documented by several authors in studies

Table 1. LC50 and LC95 of garlic essential oil against *S. oryzae* - Fumigant toxicity

Treatment	Chi square	Regression equation	LC_{50} (ppm)	95% Fiducial limit		LC_{95} (ppm)	95% Fiducial limit	
				LL	UL		LL	UL
Garlic oil	11.18	Y = 5.53 x + 3.207	2.04	1.85	2.25	4.71	3.40	6.51

LL – Lower limit; UL - Upper limit

targeting stored product pests. For instance, the essential oil of *Anethum anethoides* demonstrated an LC_{50} of 13.05 mg/L air against *Tribolium castaneum* (36). Similarly, the essential oil of *Mentha arvensis* exhibited an LC_{50} of 56.41 µL/L against *S. oryzae* and 47.03 µL/L against *Sitophilus zeamais* in fumigation assays (37). Studies conducted by (38) highlighted that eucalyptus oil showed excellent fumigant toxicity, with an LD_{50} of 28.94 µL/L air against *S. oryzae*. The fumigant toxicity of garlic essential oil against *T. castaneum* has been attributed to its major components, including diallyl disulfide, diallyl trisulfide and diallyl sulfide (39).

The adult mortality due to the fumigant toxicity of garlic essential oil is presented in Fig. 1. Significant mortality was observed when adult *S. oryzae* were exposed to filter paper discs impregnated with varying concentrations of the oil. After one day of exposure, the highest concentration (3 µL/96 cm^3 air) resulted in 56.67% adult mortality. A clear dose-dependent increase in mortality was noted with higher essential oil concentrations. By five days after treatment (DAT), adult mortality reached 99.13% at the concentration of 3 µL/96 cm^3 air. Conversely, the TNAU sweet flag formulation used as the positive control registered 68.33% adult mortality at 5 DAT. No mortality was observed in either the acetone-treated control or the untreated control even after five days of exposure (Fig. 1).

A similar trend of increased adult mortality with higher doses of garlic oil extracts has been observed against *T. castaneum* (40, 41). For instance, a study reported 50% mortality of *S. oryzae* adults with garlic essential oil at 5.45 µL/L after 24 hours of exposure (42). Another investigation revealed that garlic essential oil, either alone or in combination with 92% carbon dioxide, was toxic to the adult and pupal stages of *T. castaneum* and *Ephestia kuehniella*. Furthermore, fumigant activity and behavioral effects were observed in *Sitotroga cerealella* when rice grains were treated with garlic essential oil and its major components, diallyl disulfide and diallyl trisulfide (43).

These findings highlight that garlic essential oil exhibits exceptional insecticidal properties, as evidenced by laboratory studies.

Contact toxicity

The garlic essential oil exhibited significant contact toxicity, with LC_{50} and LC_{95} values of 2.58 ppm and 7.96 ppm, respectively, against *S. oryzae* (Table 2). The contact toxicity data are presented in Fig. 2. Complete (100%) adult mortality was observed at 5 days after treatment (DAT) with concentrations of 4 µL and 5 µL/40 g seeds. After five days of exposure, even the lowest concentration (1 µL/40 g seeds) resulted in considerable mortality, reaching 78.33%. In contrast, the TNAU Sweet Flag

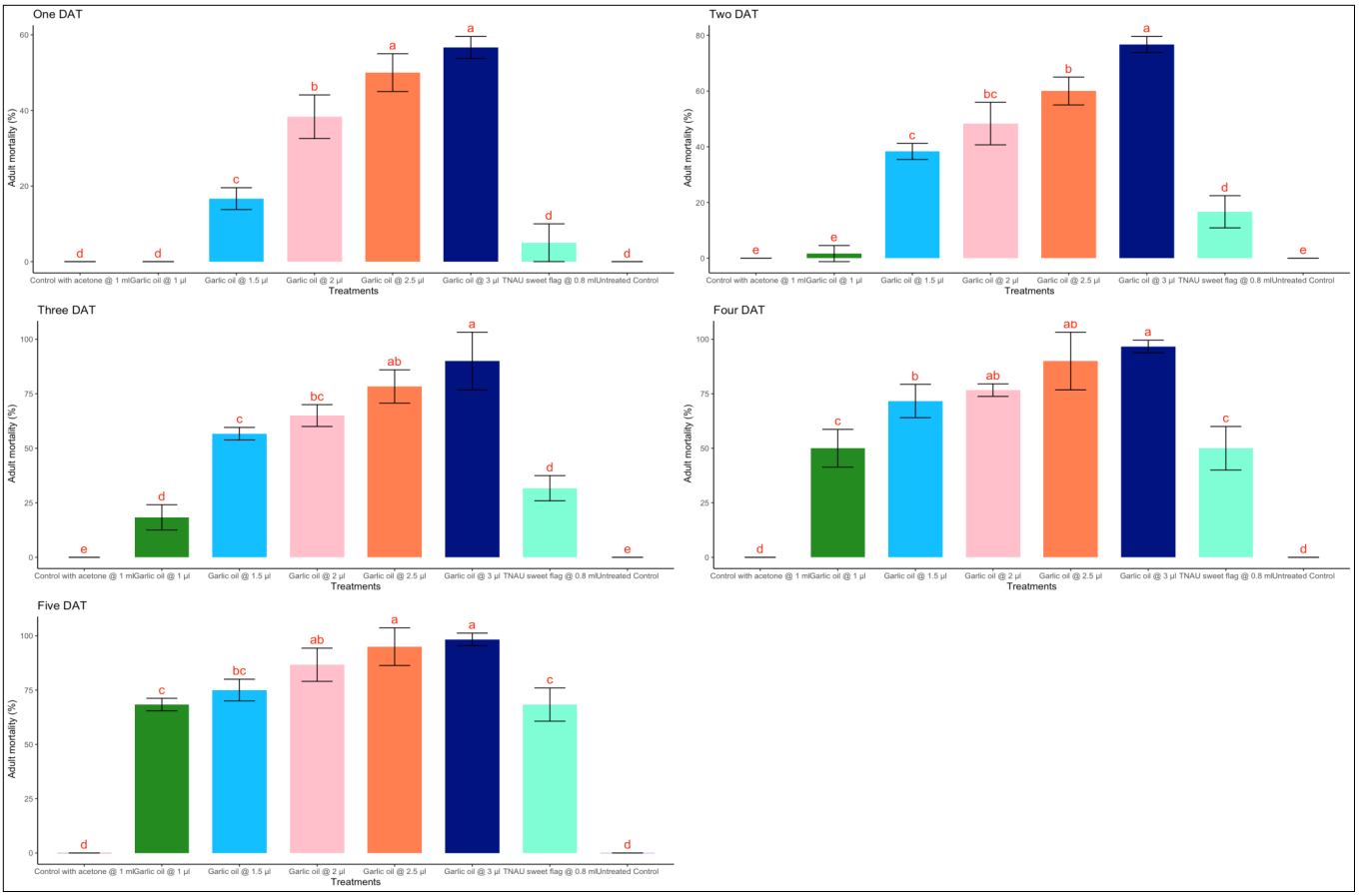

Fig. 1. Percent mortality of *S. oryzae* due to fumigant toxicity after exposure to garlic essential oil at different time intervals. Letters on the bars indicate significant differences between doses. Bars with same letter are not significantly different. Error bars indicate SD of means.

Table 2. LC_{50} and LC_{95} of garlic essential oil against *S. oryzae* - Contact toxicity

Treatment	Chi square	Regression equation	LC_{50} (ppm)	95% Fiducial limit		LC_{95} (ppm)	95% Fiducial limit	
				LL	UL		LL	UL
Garlic oil	2.64	Y = 3.976 x + 3.458	2.58	2.28	2.92	7.96	5.57	11.37

LL-Lower limit; UL-Upper limit

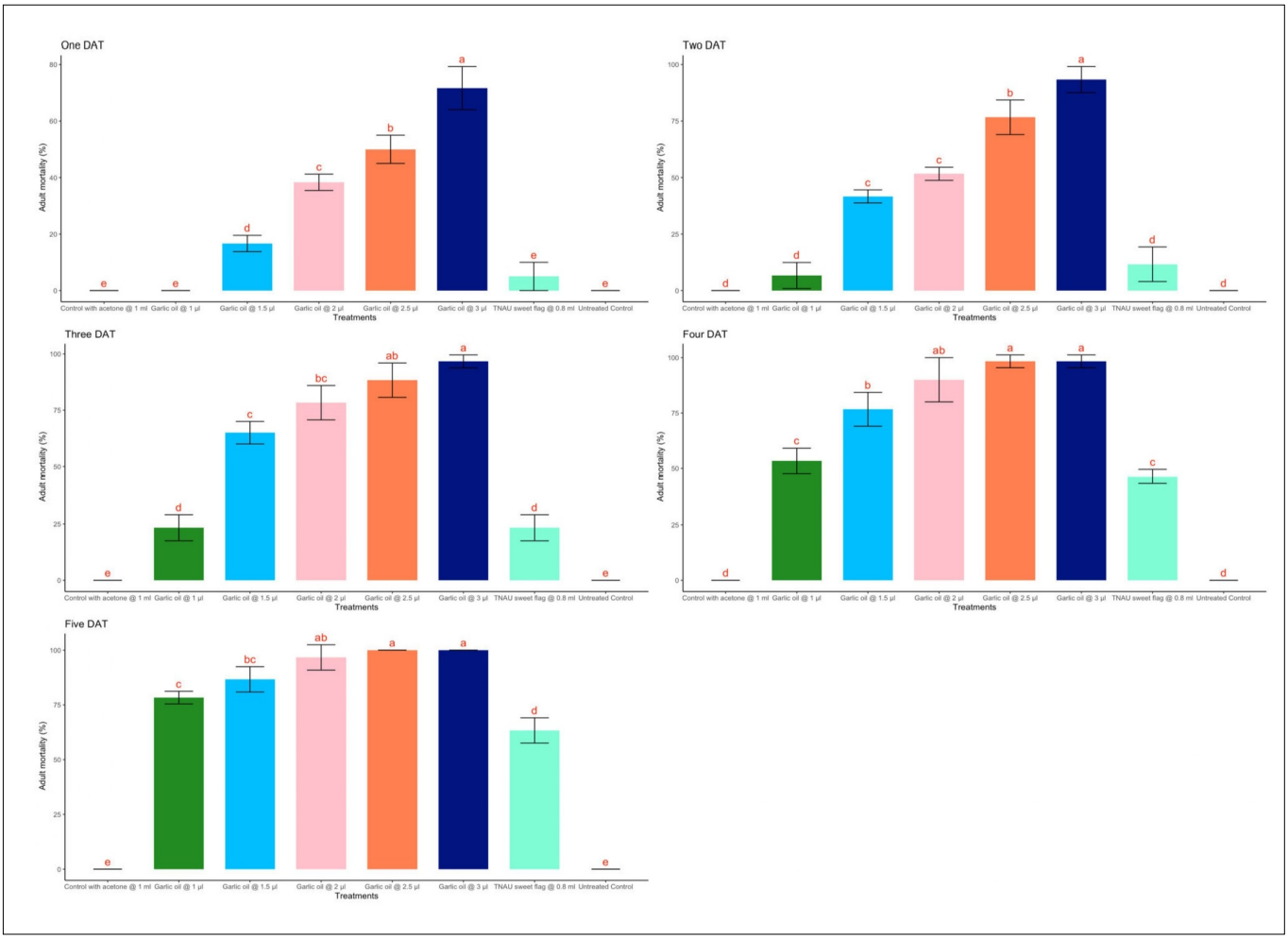

Fig. 2. Percent mortality of *S. oryzae* due to contact toxicity after exposure to garlic essential oil at different time intervals. Letters on the bars indicate significant differences between doses. Bars with same letter are not significantly different. Error bars indicate SD of means.

formulation achieved 63.33% mortality 5 DAT, while no mortality was observed in the untreated and solvent-treated control groups.

Previous studies have also demonstrated the toxicity of garlic essential oil against *S. zeamais* across all life stages in filter paper impregnation bioassays (21). Furthermore, an increase in essential oil concentration, combined with prolonged exposure durations, has been shown to enhance toxic effects on adult *S. oryzae*. For example, treating wheat grains with 0.15 mg/100 g of garlic oil resulted in 100% adult mortality (20).

Insects exposed to garlic essential oil exhibited impaired mobility and experienced muscular contractions, indicative of its neurotoxic action. The rapid knockdown and immobilization effects observed in insects highlight the oil's neurotoxicity. This phenomenon has been attributed to the inhibitory effects of certain compounds in garlic oil on the acetylcholinesterase enzyme (22).

Oviposition and adult emergence

The oviposition observed on seeds treated with varying concentrations of garlic essential oil ranged from 0.0 (at 4 µL) to 5.67 (at 1 µL) (Fig. 3). Notably, no adult emergence was recorded at a concentration of 3 µL/40 g of seeds, whereas adult emergence was 91.49% in the acetone-treated control and 97.16% in the untreated control. When seeds were treated with the TNAU Sweet Flag formulation, adult emergence ranged from 40.0% to 48.0%.

A 2.56-fold reduction in oviposition by *C. maculatus* was reported when seeds were treated with an 80% concentration of garlic essential oil (44). Similarly, approximately 90% suppression of egg hatchability and significant larval mortality were observed when garlic essential oil was combined with CO_2(45). The decline in adult emergence may also be due to reduced egg hatching and increased larval mortality.

Several studies have shown that adult emergence declines with increasing concentrations of essential oils (45). Essential oils with oviposition deterrent properties hold significant promise for managing storage pests, as they can substantially reduce pest infestations in stored food commodities.

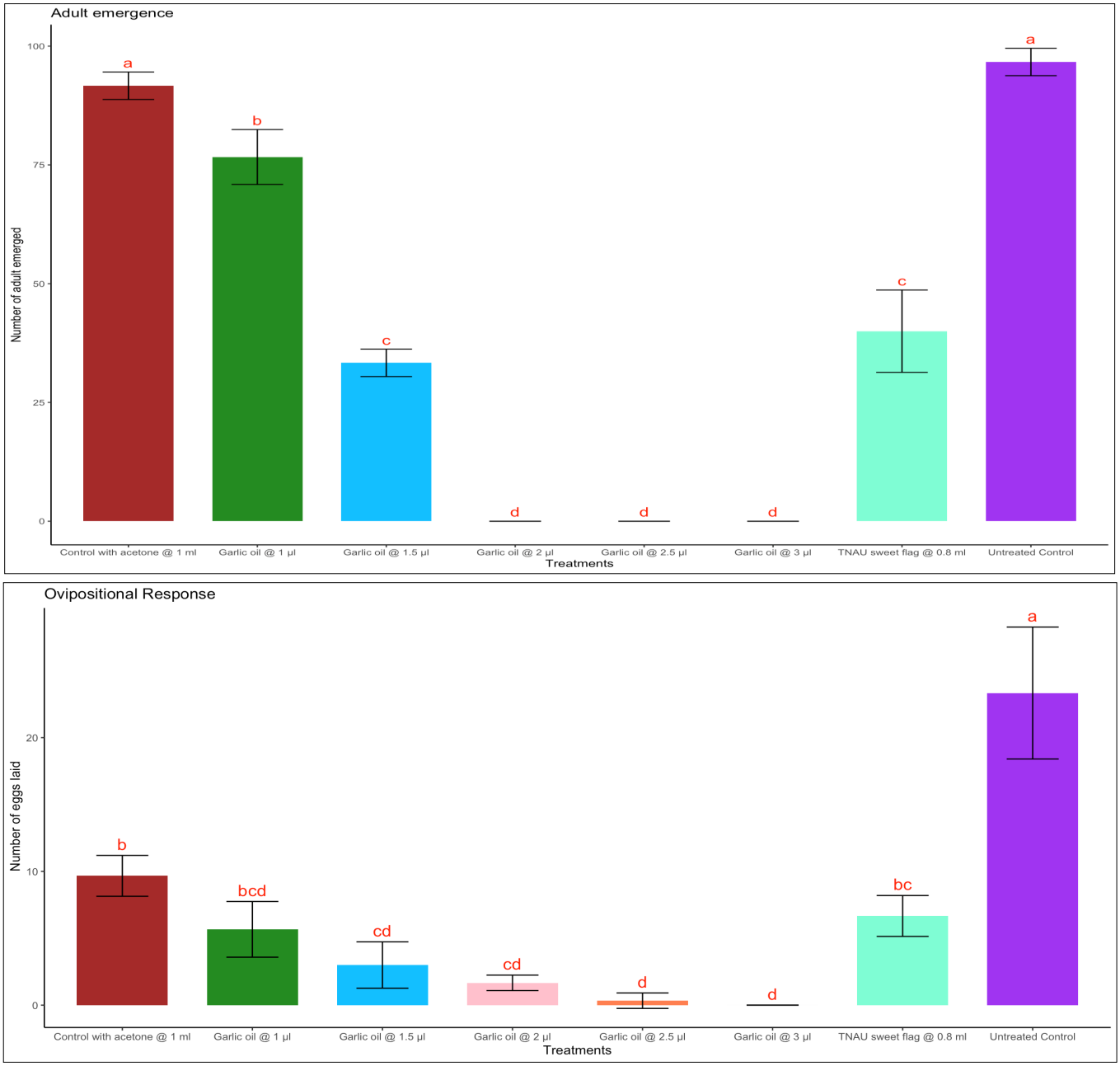

Fig. 3. Oviposition (Number of eggs laid) by *S. oryzae* adults and adult emergence after exposure to garlic essential oil. Letters on bars indicate significant differences between doses. Bars with same letter are not significantly different. Error bars indicate SD of means.

Compound identification and antennal response using GC-EAD

Gas Chromatography (GC) analysis detected 39 compounds in garlic essential oil, with a retention time ranging from 3.69 minutes to 30.83 minutes (Table 3). Male rice weevils responded to 13 compounds in Electroantennographic Detection (EAD) studies (Table 4), with most of these compounds being alkenes and alkanes, followed by alcohols, ketones and aldehydes. Female weevils exhibited strong antennal responses to alcohols and esters, followed by ketones and alkanes. Among the compounds identified, 5-Ethyl-5-methylheptadecane had the highest relative peak area of 8.94%. Another compound, 2-Decene, 2,4-dimethyl-, with a peak area of 4.83%, is recognized for its pesticidal properties and was previously identified in the acetone extract of *Azolla pinnata*, which exhibited larvicidal activity against *Aedes albopictus* (46).

A total of 15 components in garlic essential oil elicited antennal responses in females (Table 5). Among these, 3-Heptanone recorded the highest relative peak area of 3.013%, followed by 3-Hexanone with a peak area of 2.55%. In general, ketones and alcohols, such as 3-Hexanone, 1-Acetylcyclohexene, 1-Octen-3-yl-acetate, phenyl methyl dichlorosilane, 1-Dodecanol, 3,7,11-trimethyl- and 2-Hexadecanol, triggered responses in both sexes.

Many of these compounds demonstrated significant insecticidal properties. For example, chavicol, with a peak area of 4.09%, exhibits strong insecticidal activity. High concentrations of methyl chavicol have been found in extracts of *Ocimum basilicum* (47). When the vapor form of essential oil from *Ocimum selloi* was applied at a concentration of 1 mg/mL, it caused 100% mortality in *Spodoptera frugiperda* larvae within 24 hours. Similarly, higher concentrations of 1-Dodecanol and Octadecane from *Lantana camara* extract showed repellency against *Sitophilus zeamais* at all concentrations tested (48). Additionally, 3-Heptanone, with a relative peak area of 3.01%, was identified in *Alpinia officinarum* extract and exhibited feeding deterrent activity against adult *Tribolium castaneum*, with a repellent index of 18.21% (49).

Table 3. List of chemical compounds identified in the extracted garlic essential oil through Gas Chromatography

S.No.	RT (min)	Compounds	Area	Area (%)
1	3.69	Tetrahydropyran	13542085.24	9.73
2	4.06	3-Pentanol, 3-methyl-	551502.46	0.39
3	4.29	Hexane, 2,3-dimethyl-	860208.54	0.61
4	4.5	3-Hexanone	3547077.8	2.54
5	5.27	Cyclopentanol, 3-methyl-	2206829.36	1.58
6	6.08	Butyric acid, 3-hydroxy-3-methyl-, methyl ester	5367339.92	3.85
7	6.37	3-Heptanone	4195359.15	3.01
8	6.69	3-Heptanone, 2-methyl-	3572609.89	2.56
9	6.83	Butanoic acid, 3-methyl-, 2-propenyl ester	1797992.89	1.29
10	7.08	2-Decene, 2,4-dimethyl-	6722371.76	4.82
11	7.49	Formic acid, 5-methylhex-2-yl ester	1359186.02	0.97
12	8.5	n-Decane	281542.61	0.20
13	9.11	1-Acetylcyclohexene	1724995.71	1.23
14	9.45	2,5-Dimethylnonane	798610.79	0.57
15	9.69	Nonane, 4,5-dimethyl-	1423614.87	1.02
16	10.67	1-Octen-3-yl-acetate	365852.24	0.26
17	11.34	1,3-Diisopropyl-1,1,3,3-tetramethyldisiloxane	213929.28	0.15
18	11.52	Phenylmethyldichlorosilane	814603.15	0.58
19	12.98	Docosane, 9-octyl-	149898.68	0.10
20	14.84	Chavicol	5695542.58	4.09
21	15.08	17-Pentatriacontene	875366.26	0.62
22	15.85	Oleic acid, eicosyl ester	1362322.23	0.97
23	16.14	Oleic acid, 3-(octadecyloxy)propyl ester	2046794.03	1.47
24	17.06	1-Dodecanol, 3,7,11-trimethyl-	986516.55	0.70
25	17.81	1,3-Ditert-butyl-2-methoxy-5-methylbenzene	419893.92	0.30
26	19.32	Tetradecane, 2,6,10-trimethyl-	4214489.91	3.02
27	20.3	Butanoic acid, 3-methyl-, 3,7-dimethyl-2,6-octadienyl ester, (E)-	3758264.18	2.69
28	21.46	Octadecane, 3-ethyl-5-(2-ethylbutyl)-	994923.34	0.71
29	21.73	2-Hexadecanol	1877934.36	1.34
30	24.08	Succinic acid, 2-ethylhexyl but-3-en-1-yl ester	36193697.94	25.99
31	24.45	5-Ethyl-5-methylheptadecane	12453095.52	8.94
32	25.84	5-Octadecenal	527733.25	0.37
33	26.52	Cyclodecasiloxane, eicosamethyl-	1896590.68	1.36
34	27	5-Ethyl-5-methylnonadecane	650535.36	0.46
35	27.96	Methyl 2-hydroxy-heptadecanoate	3482518.12	2.50
36	28.35	Octadecane, 1-bromo-	1591142.4	1.14
37	29.13	11,14-Eicosadienoic acid, methyl ester	8649090.35	6.21
38	30.08	17β-Estradiol, 3-deoxy-	760283.31	0.54
39	30.83	cis-11-Eicosenoic acid	1305755.85	0.93
		TOTAL	**139238100.5**	**100**

RT (min)-Retention time in minutes

Table 4. List of active compounds eliciting antennal responses in adult male rice weevil (*S. oryzae*) through GC - EAG

RT (min)	Compounds	Chemical formula	Area (%)	Chemical group
4.5	3-Hexanone	$C_6H_{12}O$	2.54	Ketone
7.08	2-Decene, 2,4-dimethyl-	$C_{10}H_{20}$	4.82	Alkene
9.11	1-Acetylcyclohexene	$C_8H_{12}O$	1.23	Alkene
10.67	1-Octen-3-yl-acetate	$C_{10}H_{18}O_2$	0.26	Ester
11.52	Phenylmethyldichlorosilane	$C_8H_{10}Cl_2S$	0.58	Organosilane
12.98	Docosane, 9-octyl-	$C_{32}H_{66}$	0.10	Alkane
14.84	Chavicol	$C_9H_{10}O$	4.09	Phenol
15.08	17-Pentatriacontene	$C_{35}H_{70}$	0.62	Alkene
17.06	1-Dodecanol, 3,7,11-trimethyl-	$C_{15}H_{32}O$	0.70	Alcohol
21.73	2-Hexadecanol	$C_{16}H_{34}O$	1.34	Alcohol
24.45	5-Ethyl-5-methylheptadecane	$C_{20}H_{42}$	8.94	Alkane
25.84	5-Octadecenal	$C_{18}H_{34}O$	0.37	Aldehyde
27	5-Ethyl-5-methylnonadecane	$C_{20}H_{42}$	0.46	Alkane

RT (min)-Retention time in minutes

Table 5. List of active compounds eliciting antennal responses in adult female rice weevil (*S. oryzae*) through GC - EAG

RT (min)	Compounds	Chemical formula	Area (%)	Chemical group
4.06	3-Pentanol, 3-methyl-	$C_6H_{14}O$	0.39	Alcohol
4.5	3-Hexanone	$C_6H_{12}O$	2.54	Ketone
5.27	Cyclopentanol, 3-methyl-	$C_6H_{12}O$	1.58	Alcohol
6.37	3-Heptanone	$C_7H_{14}O$	3.01	Ketone
7.49	Formic acid, 5-methylhex-2-yl ester	$C_8H_{16}O_2$	0.97	Ester
8.5	n-Decane	$C_{10}H_{22}$	0.20	Alkane
9.11	1-Acetylcyclohexene	$C_8H_{12}O$	1.23	Alkene
10.67	1-Octen-3-yl-acetate	$C_{10}H_{18}O_2$	0.26	Ester
11.52	Phenylmethyldichlorosilane	$C_8H_{10}Cl_2S$	0.58	Organosilane
15.85	Oleic acid, eicosyl ester	$C_{27}H_{52}O_2$	0.97	Ester
16.14	Oleic acid, 3-(octadecyloxy)propyl ester	$C_{24}H_{46}O_3$	1.47	Ester
17.06	1-Dodecanol, 3,7,11-trimethyl-	$C_{15}H_{32}O$	0.70	Alcohol
21.46	Octadecane, 3-ethyl-5-(2-ethylbutyl)-	$C_{28}H_{58}$	0.71	Alkane
21.73	2-Hexadecanol	$C_{16}H_{34}O$	1.34	Alcohol
30.83	cis-11-Eicosenoic acid	$C_{20}H_{38}O_2$	0.93	Fatty acid

RT (min)-Retention time in minutes

Powders derived from *Sonchus oleraceus*, *Punica granatum L.* (pomegranate), *Thymus vulgaris L.* (thyme) and *Portulaca oleracea* (green purslane) were found to contain 2-Hexadecanol. These powders, when applied at a rate of 10 g/50 g of wheat grains, caused 70-90% adult mortality in *S. oryzae* within 14 days of treatment (50).

Principal Component Analysis (PCA) (Fig. 4) provided insights into the key characteristics of the active metabolites, while heatmap analysis illustrated the identified chemicals and their respective abundances through untargeted GC-MS profiling (Fig. 5). Interestingly, several compounds with smaller relative peak areas elicited substantial responses in rice weevils, whereas those with the highest peak areas did not (Fig. 6 and 7). An array of alcohols, esters and alkanes in the garlic essential oil extract contributed significantly to its insecticidal activity (51).

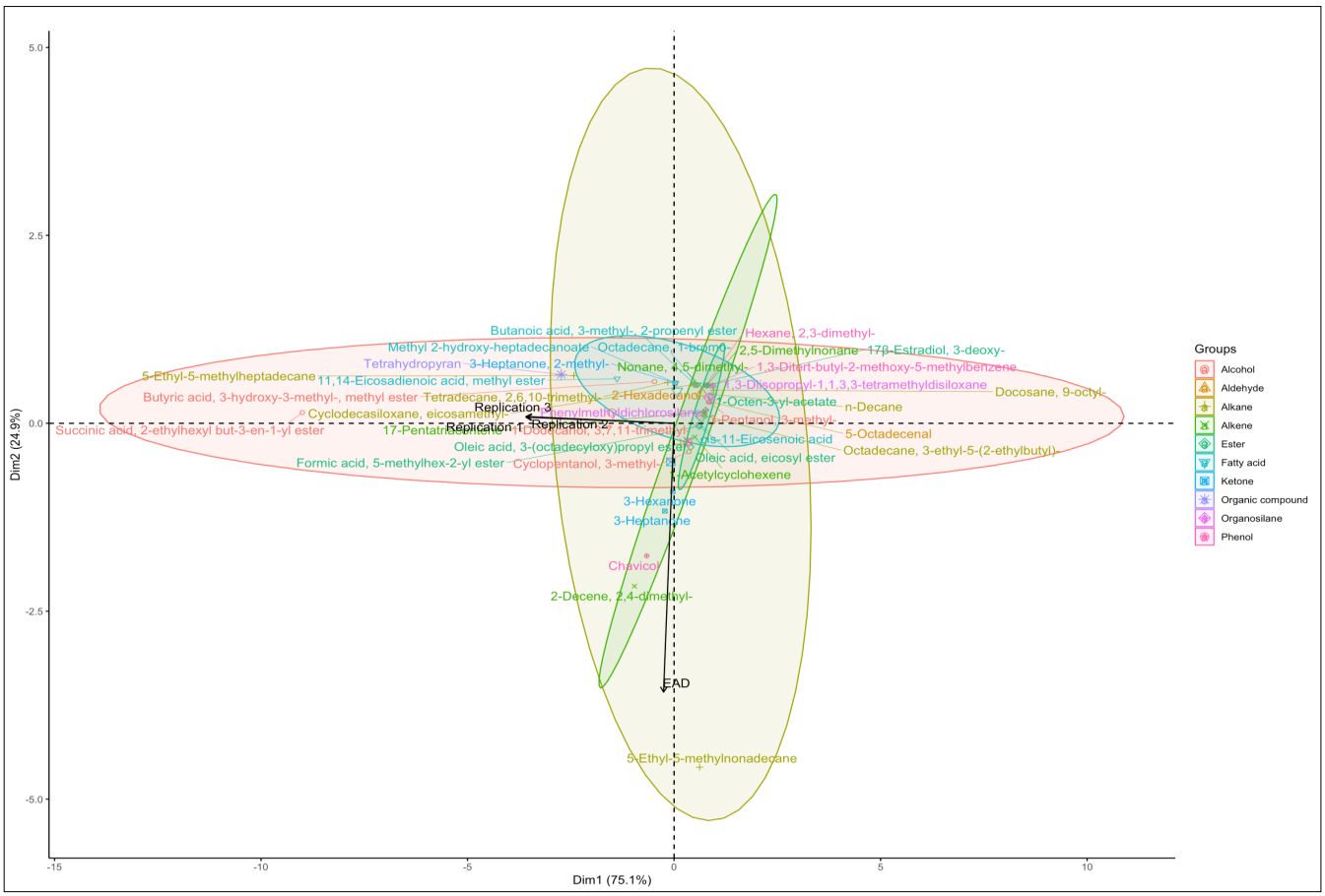

Fig. 4. PCA biplot revealing principal nature and compound group distribution.

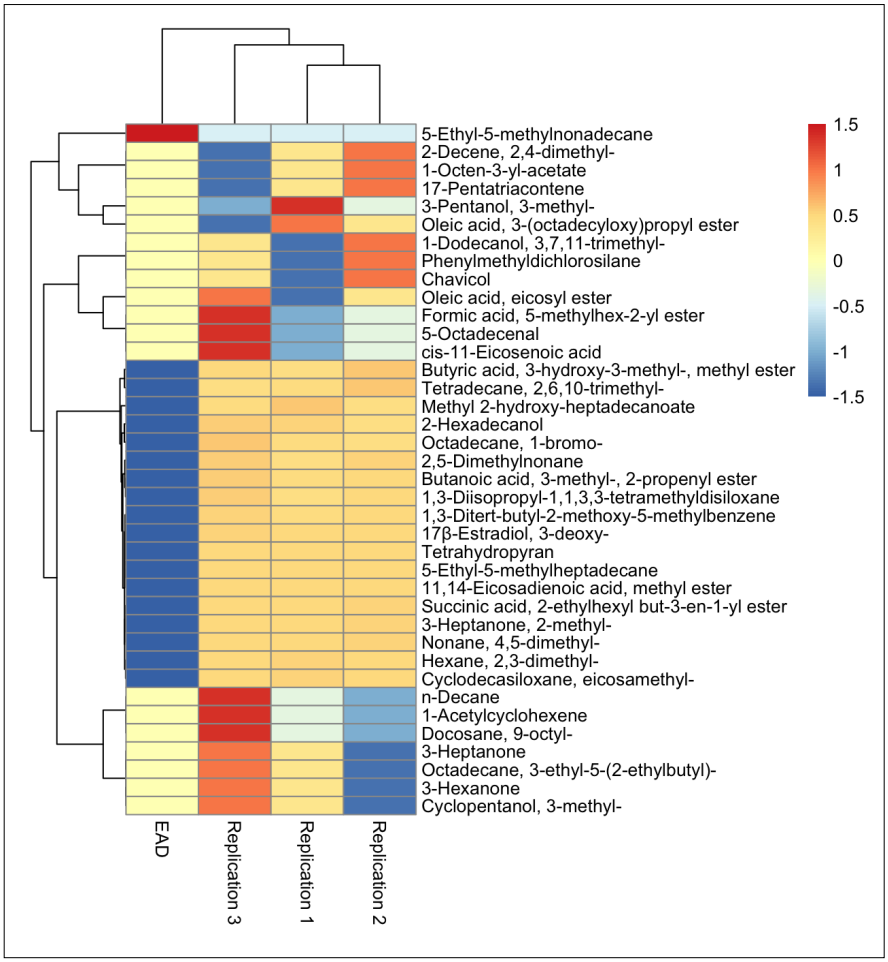

Fig. 5. Heatmap projecting compound distribution and their relative abundance.

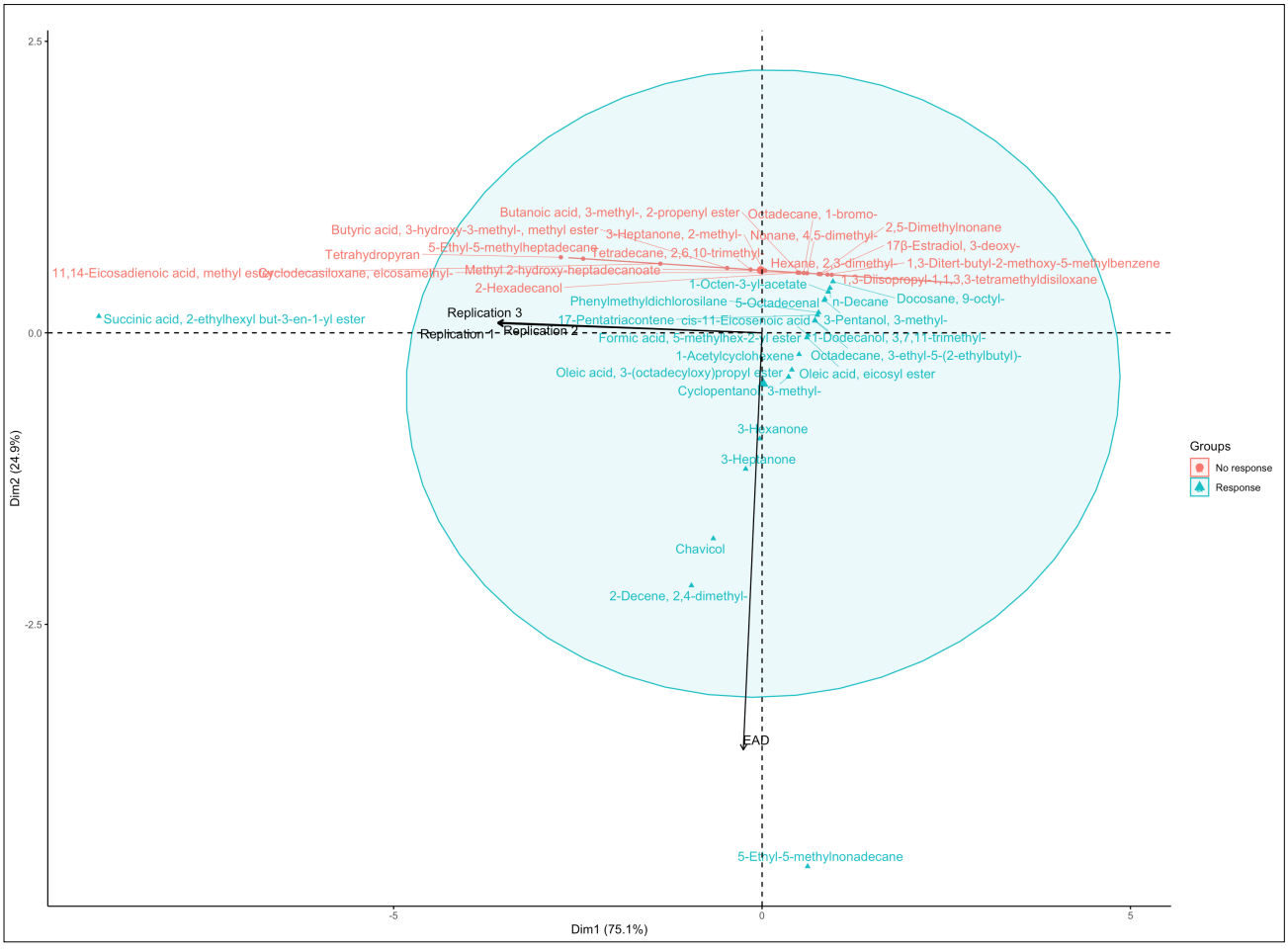

Fig. 6. This figure represents the comparative view of behavioural eliciting and non-eliciting compounds in conjunction with PCA biplot.

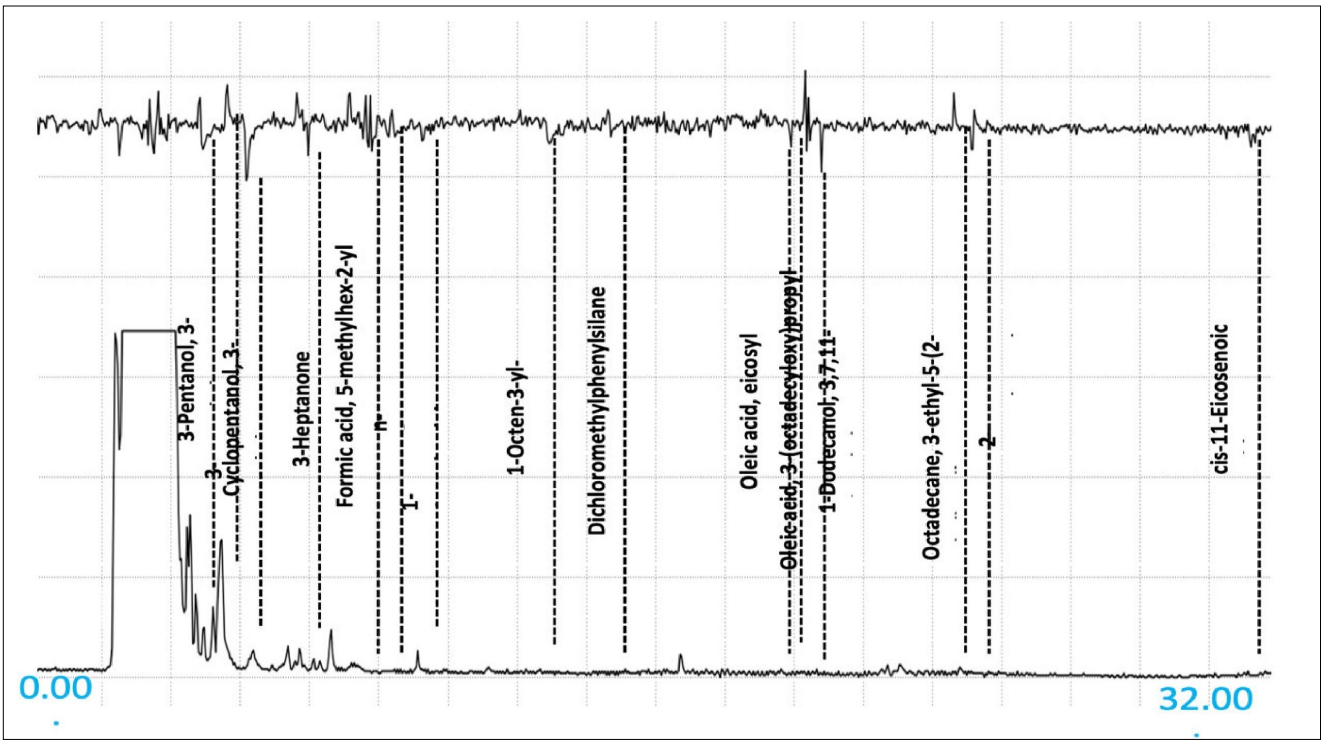

Fig. 7. Phytochemical analysis of garlic essential oil by GC-EAD revealed major compounds. 5-Ethyl-5-methyl heptadecane was most common (8.94%). 2-Decene, 2, 4-dimethyl-, followed (4.83%).

Conclusion

This research emphasizes the promising potential of garlic essential oil as a natural and environmentally friendly solution for the control of *S. oryzae* in stored grains. These findings underscore its significant fumigant and contact toxicity, as well as its ability to suppress oviposition and reduce F1 progeny emergence. Furthermore, the identification of specific chemical compounds influencing the behavior of *S. oryzae* provides valuable insights for the development of effective pest management strategies in the agricultural and food storage sectors. Given the potent insecticidal properties of garlic essential oil, compounds with repellent activity should undergo further validation through GC-EAG analysis. This will pave the way for the development of efficient formulations based on garlic essential oil for practical pest control applications.

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Authors' contributions

ST, NJM, KT, KG, PK, ET, SA, RA, PSS, BV, VPS, RR and MM all have participated in execution, design, statistical analysis of the experiment edited, revised and finalized the manuscript. All authors read and approved the final manuscript.

Compliance with Ethical Standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None

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