THE POTENTIAL USE OF TERMITE'S SEMIOCHEMICALS FOR SAFEGUARDING DATE PALMS AGAINST TERMITE

ALOTAIBI, N. J.

Department of Biology, Faculty of Science, Taif University, Taif 21974, Saudi Arabia
(e-mail: n.alotaibi@tu.edu.sa; ORCID: 0000-0002-7719-3643)

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Abstract. Termites are responsible for the significant loss of date palm production. Limitations and hazardous side effects of chemical pesticides pushed toward finding eco-friendly alternatives to control pests. Using semiochemicals with natural enemies has grown in recent years, making biological control agents and semiochemicals crucial for integrated pest management of insect pests. The current study has revealed a chemical interaction between three organisms, date palm termites-ants. This study simplifies this complex relationship among date palms, termites and ants, essential for effective pest management. This study investigated the impact of termite attacks and the presence of ants on the volatile compounds profile released into the chemosphere of the community consisting of the date palm *Phoenix dactylifera*, the harvester termite *Anacanthotermes ochraceus*, and the ant *Messor cf. wasmanni*. This study aims to analyze the volatile compounds emitted by the date palm and surrounding insects in response to these intricate interactions. The extraction of volatile organic compounds (VOCs) was carried out using closed-loop stripping (CLS) and then analyzed via gas chromatography-mass spectrometry (GC-MS) and Canonical analysis of principle coordinates (CAP) was performed. Termites and ants were collected from Taif City and molecularly identified based on the mitochondrial cytochrome oxidase I. The date palm was exposed to different types and order of treatments, including non-infested palm, termite-infested palm and termite-infested palm + ants. Infested date palms displayed distinct emissions of camphene, (+)-4-careneand, 3-carene, guaiene, and (-)-aristolene compared to non-infested plants. The characteristic VOC profile was the share of infested date palms in the presence of ants, with γ-terpinene, ketone, aromadendrene, isobornyl acetate and bornyl acetate. These semiochemicals play multiple functions, such as repellent or killing activities. Termite-killing or repelling semiochemicals can be integrated alone or with biological control agents for better and more effective control of termites affecting palm trees. Moreover, alarming or defensive semiochemicals of termites can be counteracted to suppress their defense mechanisms.
Keywords: *termites, ants, pest management, volatile organic compounds, gas chromatography-mass spectrometry*

Introduction

Date palm (*Phoenix dactylifera* L.) is an important crop in several Middle Eastern countries, particularly Saudi Arabia. In addition to its nutritional value (Hussain et al., 2020), it contributes significantly to the national economy of the Kingdom (Allbed et al., 2017). Based on MEWA reports (MEWA, 2021), the Kingdom has more than 31 million date palm trees with an annual production of over 1.5 m tons to occupy the second global producer of dates after Egypt. However, Saudi Arabia was ranked first in date export in 2021 (Alotaibi et al., 2023). In addition, the Kingdom comprises a diverse palm cultivar (> 400 cultivars), which prefer to grow in arid and semiarid regions (Almadini et al., 2021). Despite the increased production of date palms in Saudi Arabia, the average production yield is low compared to other countries. This is attributed to poor management practices, insect pests and plant diseases (Jonoobi et al., 2019; Krueger, 2021; Salomón-Torres et al., 2021). Insect pests and other plant diseases may account for a 30% loss in palm date production (El-Juhany, 2010). Insect pests such as red palm weevil (*Rhynchophorus ferrugineus*), old world date mite (*Oligonychus afrasiaticus*), lesser date moth (*Batrachedra amydraula*) and termites are the significant

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constraints on date palm production in Kingdom of Saudi Arabia (Alotaibi et al., 2023; Latifian and Rad, 2022).

Termites are a large group of insects (>2600 species) widely flourishing in tropic and subtropic regions. They are eusocial insects with the division of labour among workers, soldier's queens and larvae (Eggleton, 2011; Legendre et al., 2013). Based on phylogenetic studies, termites can be categorized into lower termites (e.g., Hodotermitidae, Kalotermitidae, Mastotermitidae, and Termopsidae) and higher termites (Termitidae) (Krishna et al., 2013). In Saudi Arabia, 19 species of termites have been recognized within four families: Kalotermitidae, Hodotermitidae, Rhinotermitidae, and Termitidae (Sharaf et al., 2021). Of these species, the harvester termite (*Anacanthotermes ochraceus*) is an important pest of date palms in Saudi Arabia and other countries such as Kuwait, Oman, Egypt, Turkey and the United Arab Emirates (Cheraghi et al., 2012; El-Shafie et al., 2017; Kaakeh, 2005; Sharaf et al., 2021). In nature, ants are termites' primary predators and can destroy their nests. For example, *Megaponera* ants can discover escape channels of *Macrotermes subhylinus*, and Ponerines can eat thousands of termites within a day (Frank et al., 2017). The predator-prey relationships involve predatory tactics developed by ants and the defensive strategies of termites (Latifian and Rad, 2022).

Individuals of termite colonies communicate via several chemically identified intra- or inter-caste pheromones. These pheromones are required to perform tasks assigned to workers, soldiers and pheromones for egg laying, aggregation, royal recognition, queen pheromones, trail-following and alarming pheromones (Mitaka and Akino, 2021). Interestingly, termitophagous predators (e.g., ants) can eavesdrop and mimic pheromones produced by termites (Rosa et al., 2018; Wen et al., 2017). For example, *Odontoponera transversa* can track trail-following pheromones of *Macrotermes yunnanensis* and *Ancistrotermes dimorphus* (Wen et al., 2017). Due to the importance of pheromones in attracting natural predators, recent studies focus on integrating them into insect management strategies (e.g., biological control) (Sharma et al., 2019). Integrated pest management programs use semiochemicals or pheromones to engage in chemical communications between plants, insect pests, parasitoids, and predators (Guerrero and Reddy, 2023). In this regard, several studies reported the significance of incorporating semiochemicals such as synthetic tricosane, Terpenoids, (E, E)-α-farnesene, methyl salicylate, (Z)-3-hexenyl acetate, and (E)-2-octen-1-ol, nepetalactol, monoterpenes and oxygenated monoterpenes in biological control system against many insects (Sharma et al., 2019).

In Saudi Arabia, termites are one of the significant arthropods affecting date palm production. The current study analyzed the intricate interplay and chemical signaling in the tri-trophic community of termites (important date pests palm), ants (predators of termites) and date palm trees. Three treatments were adopted to analyze the tripartite interactions including non-infested palm trees, palm trees infested with termites and the addition of black ants to the bitrophic community. Chemical compounds from three organisms were analyzed by GC-MS and their possible contribution to biological control were discussed.

Materials and methods

Site description and insect sampling

Insects (termites and ants) were collected from infected date palm trees (*Phoenix dactylifera*) growing in Al-Hawiyah, Taif, Saudi Arabia (N21.428299, E40.473267).

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The soil in the collection is sandy with small rocks, including desert plants, high temperature weather, and a cement plant. The insects were collected in plastic boxes, removed from dust with a soft brush and transported to the laboratory. A total number of termites (n = 500/palm) and ants (n = 200/palm) individuals were used in the experiment. Black ants were maintained in plastic boxes and fed on sugar until they were used in the experiment.

Molecular identification

DNA extraction from all insects (termites and ants) was attempted using the QIAamp® DNA Mini Kit (QIAGEN, Germany) following the manufacturer's instructions. PCR was performed to amplify the mitochondrial *COI* gene using the primer set (LCO1490) (F-5'-GGTCAACAAATCATAAAGATATTGG-3') and (R HCO2) (R-5'-TAAACTTCAGGGTGACCAAAAAATCA-3') (Folmer et al., 1994). The PCR reactions were performed in 20 µl volume; nuclease-free H_2O (12.8 µl), genomic DNA (2 µl), primers (0.6 µl of each) and 5X FIREPol® Ready to LoadMaster Mix (Solis BioDyne) (4 µl). The cycling conditions were initial denaturation at 95°C for 5 min, 35 cycles of denaturation at 95°C for 40, annealing at 47°C for 15 and, extension at 72°C for 1 min, and a final extension step at 72°C for 5 min. Amplicons corresponding to the COI gene of each insect were retrieved from the gel using Illustra^{TM} GFX PCR DNA (GE Healthcare). DNA sequencing was performed on both strands using a 3130xl Genetic Analyzer (Biosystems; Thermo Fisher Scientific, USA). The raw sequence data were edited and assembled using the EditSeq program of the Lasergene software package, version 3.18 (DNAStar, Madison, Wisc., USA). The BLAST tool was used to identify the assembled sequences (Syngai, 2013).

Experiment workflow

The experiment was designed to study the interactions between three organisms (palm trees, termites, and black ants) by analyzing the released volatile organic compounds (VOCs) *Figure 1*. Six pots of date palm trees, *P. dactylifera,* (age: 2 years, length: 100 cm, diameter: 20 cm), were brought to the laboratory for experimental purposes. The palm trees were maintained for one week under a 12-h-light and 12-h-dark cycle. A day before the volatile collection, palm trees were thoroughly washed with tap water, air dried and completely enclosed within PolyVinyl Carbonate (PVC) to prevent contamination. Small pores were made at the above side of PVC for ventilation and the soil surrounding the palm trees was coated with aluminum foil to avoid chemical soil interference. Three treatments were established to analyze the tripartite interactions: i) Treatment 1 (T_1): included a noninfested palm tree (*Phoenix dactylifera*), ii) treatment 2 (T_2): included palm trees infested with termites (*A. ochraceus*) (500 workers/palm), and iii) treatment 3 (T_3): included the addition of 200 black ants/palm (*Messor cf. wasmanni*) were added to T_2 to collect VOCs after 24 h of exposure. Each group was subjected to Six independent biological replicates.

Headspace analysis

Airborne metabolites collections

Headspace extraction was performed on the different groups by using closed-loop stripping (CLS) (Chehab et al., 2010; Kunert et al., 2009), using 3 mg of activated

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charcoal filter (activated charcoal suitable for GC, crystal, 05112, particle size (0.3 - 0.5 mm)) connected to a pump (model no. DC6/18 F; Fürgut GmbH, Tannheim, Germany). The charcoal filters were cleaned before use by rinsing them with methanol (1.5 ml), followed by ethyl acetate (1.5 ml) and drying them at 80°C for 2 h. After sampling, charcoal traps were eluted using 3 x 20 µL of ethyl acetate and the elute was collected in a 1 mL glass insert, kept in a 1.5 mL vial, and closed tightly with a cap fitted with PTFE-butyl-PTFE septa (VWR, Darmstadt, Germany). The vials were stored at -80°C till to be used for GC-MS analysis.

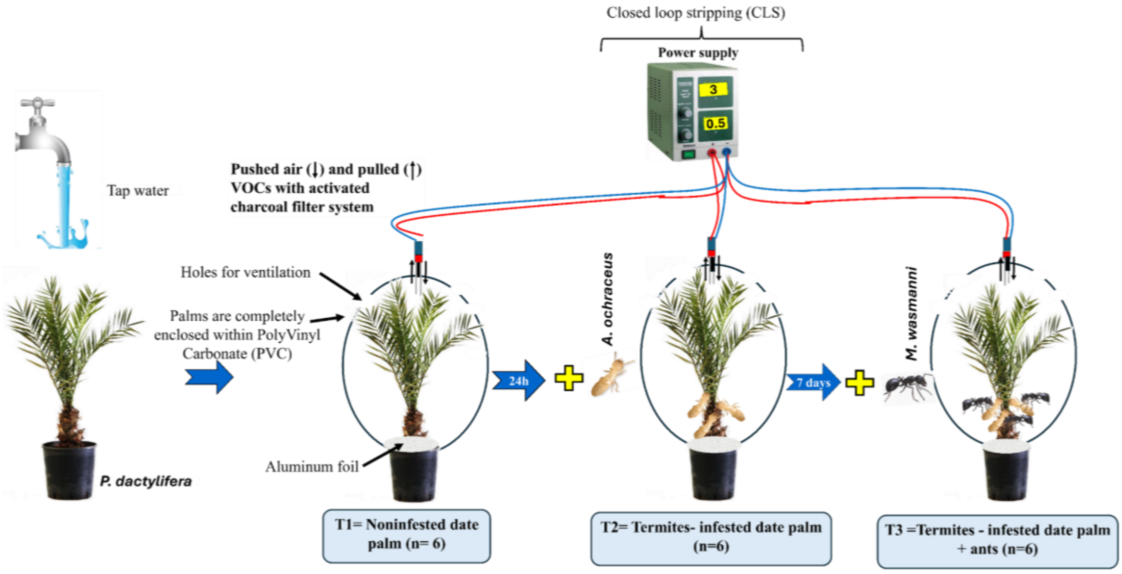

Figure 1. *Experimental setup using closed-loop stripping (CLS) to collect VOCs from the chemosphere of three treatments (T1. Noninfested date palm, T2. Termites-infested date palm and T3. Termites-infested date palm + ants)*

Data analysis and identification of chemosphere compounds by GC-MS

Samples were analyzed using a GC-MS Agilent 7890A (Agilent Technologies, Santa Clara, USA). The GC was equipped with a nonpolar DB capillary column (30 m × 250 µm, 0.25 µm film thickness, Agilent Technologies) using helium as carrier gas at a constant 1 mL/min flow rate. The GC-MS conditions were injection pre-dwell time was 0.1 min (hot needle injection), injector temperature was 250°C and 1 µL sample was injected into GC port using spitless mode. The initial temperature was 50°C, which increased to 280°C at 6°C/min. The MS was operated at 70 eV, trap current emission of 100 µA, source temperature of 250°C, transfer line temperature of 280°C and MS quad temperature of 150°C. Scanning was performed from m/z 50 to 550 amu. Data were acquired and analyzed using ChemStation software (G1701EA GC/MSD, Agilent). For non-target analysis, compounds were identified by analyzing their mass spectra and comparing them with the NIST14 database (Gaithersburg, MD, USA) and Wiely12 (John Wiley & Sons, Inc., Hoboken, New Jersey, USA). The components were identified by comparing their mass spectra with those of Wiley09 and the National Institute of Standard Technology (NIST 14) mass spectral database. The strategy of accepting the identification of compounds based on the reverse match for untargeted analysis was adopted from Alsufyani et al. (2017).

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Data pre-processing and statistical analysis

The chromatograms of all technical replicates (n ≥ 6) were pre-processed, and the means of replicates were used for the canonical analysis of principal coordinates using CAP12.exe (version 12, University of Auckland, Auckland, New Zealand). http://www.stat.auckland.ac.nz/~mja. Scatter plots to visualize the distribution of the samples were drawn using SigmaPlot (version 11.0, Systat Software, Bayshore, USA) to collect biomarkers for discriminating treatments.

Results

Molecular identification of insects

Molecular identification was attempted by amplifying and sequencing the mitochondrial COI isolated from the collected termites and black ants. A phylogenetic tree was constructed to confirm the identity of the used insects (*Fig. 2*). Assembled COI sequences were deposited in GenBank with the following accession numbers: termites, *A. ochraceus* (OR920052) and black ants, *Messor cf. wasmanni* (OR920053). As shown in the phylogenetic tree, there are two major clades, one for *A. ochraceus* and the other for *M. wasmanni*. *A. ochraceus* collected from Taif city has a COI sequence that differs from other species collected from different regions in Saudi Arabia. Similar observation was recorded for the black ants (*M. wasmanni*) as shown in *Figure 2*.

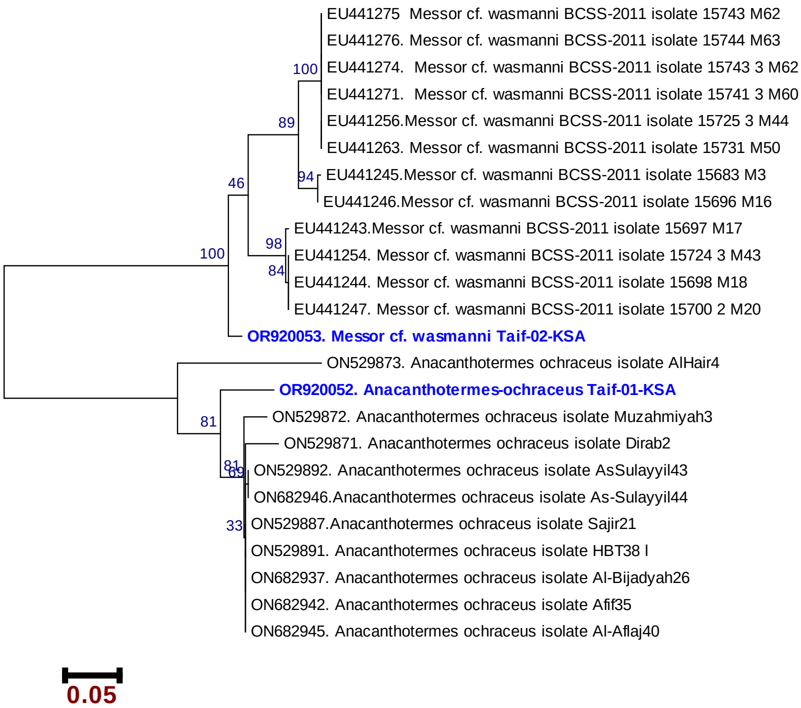

Figure 2. *The maximum likelihood method phylogenetic tree is based on the Tamura-Nei model (Tamura and Nei, 1993). The tree was constructed based on the partial sequence of COI of both termites (591 nts) and black ants (480 nts) collected from the Taif region. International and local sequences from the Genbank database were used to confirm the identity of our collected insects and their genetic relatedness with other sequences of the same species. Insect species used in the current study are shown in blue fonts and bootstrapping values are shown above the branches*

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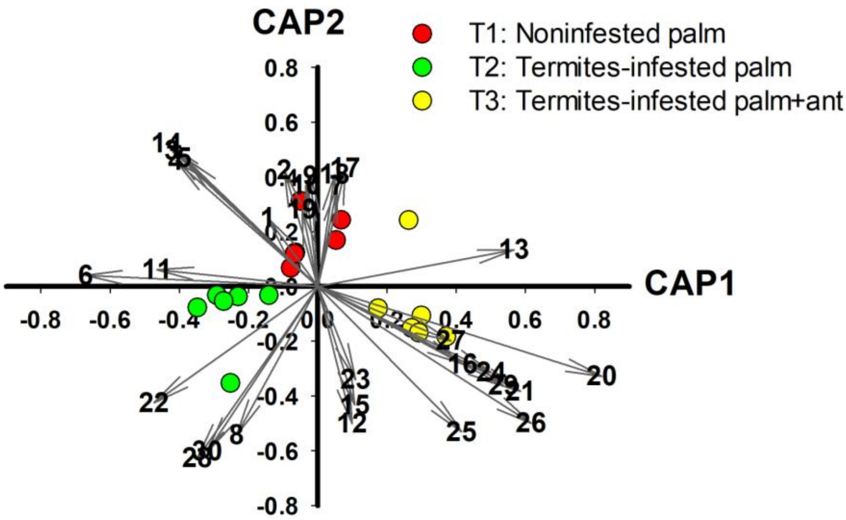
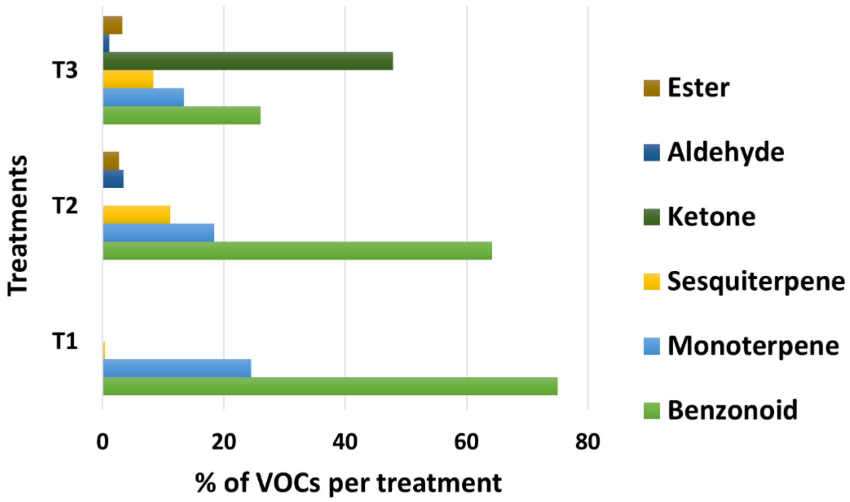
Overview of the compounds detected in the headspace

The CLS extraction method revealed thirty compound concentrations in the chemosphere of the various date palm *P. dactylifera* treatments, as shown in *Figure 1* and *Table 1,* these compounds have been tentatively identified for non-target analysis, with ketone, aldehydes, esters, benzene derivatives, and terpenes being the main components (*Fig. 3B*).



(A)



(B)

Figure 3. *(A) CAP analysis of the headspace compounds (n = 30) released by non-infested date palm trees T1(red), termite-infested date palm trees T2(green) and termite-infested palm after adding ant T3(yellow) analyzed by GC/MS (n = 18 samples), two canonical axes of the canonical analysis of principle coordinates (CAP) analysis are plotted. CAP analysis demonstrates the separation of the samples based on the presence and absence of headspace compounds. Scaled vectors of the headspace compounds (ID numbers) were significant (|r| > 0.2) for the separation of the groups, miss-classification error = 9.52%. The numbers refer to the headspace compounds in Table 1. (B) A histogram displays the emissions percentage of VOCs released in the chemosphere of three date palm treatments (T1, T2 and T3)*

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The chemosphere constituents of the noninfested date palm were benzene derivatives (75%) and monoterpenes (24.5%). After infestation by *A. ochraceus* the chemosphere of the termite-infested palm released consisted of terpenes: sesquiterpene (11.2%), monoterpenes (18.4%), aldehyde (3.5%), ester (2.7%) and benzene derivatives (64%) but no ketone was detected. After the black ants were added to the infested palm, the chemosphere profile of the tripartite community consisted of ketone (47.8%), benzene derivatives (25.9%), monoterpenes (13.3%), sesquiterpene (8.4%), aldehyde (1.1%) and esters (3.3%) (*Fig. 3B*). Sesquiterpenes were identified only after infestation as seen in *Table 1*. Thus, this specific sesquiterpene blend can be used as an indicator for the infestation status of date palm trees by *A. ochraceus* (T2), which was characterized by Valencene (bio # 26), Guaiene (bio # 28), (+)-Sativen (bio #29) and (-)-Aristolene (bio #30), which continued in (T3). The presence of *M. wasmanni* in the infested date palm trees (T3) was indicated with two sesquiterpenes, namely Longifolene (bio #24) and Aromadendrene (bio #25), in addition to the previous sesquiterpenes in (T2). The collected headspace compounds were analyzed by GC-MS and subjected to multivariate analysis, i.e., canonical analysis of principle coordinates (CAP) (*Fig. 3A*), to identify biomarkers in the headspace characteristic for each treatment. CAP separated GC datasets of aphid species based on the absence and existence of compounds or abundance of compounds among headspace profiles. Each compound represents a vector: the higher the vector, the more significant the vector is in treatment separation.

Significant biomarkers within the chemosphere for group separation

Figure 3A’s statistical analysis of PCA revealed significant differences between VOCs in the chemosphere, either through quantitative or qualitative estimation (*Figs. 3A* and *4*).

The major identifiable compounds presented in *Table 1* and *Figure 3A*, with an accuracy of more than 90%, can differentiate between three communities. In *Figure 3A*, only distinguishing volatile compounds are shown and the overlapping compounds are excluded. Many other peaks were also observed, with their matching degrees to the NIST database being very low or their amounts much lower than others.

Non-infested date palm: in noninfested palm trees, VOCS represented by α-pinene (bio # 3), α-phellandrene (bio # 5), tricyclene (bio # 14),1,2-diethyl benzene (bio # 17) and 1,4-diethyl benzene (bio # 18) were detected in the chemosphere (*Table 1; Fig. 3A*).

Termite-infested date palm: This system describes the interaction between the date palm and *A. ochraceus*. Date palms are responding to termites by emitting VOCs into the chemosphere. VOCS is represented by 3-carene (bio # 6), camphene (bio # 8), (+)-4-carene (bio # 22), guaiene (bio # 28) and (-)-aristolene (bio # 30). Adding biotic variables caused changes in this community, which affected the chemosphere profile (*Table 1; Fig. 3A*).

Termite infested palm + ant: The addition of black ant *M. wasmanni* allowed for the identification of the following compounds γ-terpinene (boi #13), m-methyl acetophenone (boi #16), 7,8-dihydro-β-ionone (boi #27), aromadendrenem (boi #25), (+)-sativen (bio # 29), isobornyl acetate (bio # 20) and bornyl acetate (bio # 21) (*Table 1; Fig. 3A*).

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Table 1. *VOCs (n = 30) were released into the chemosphere before and after date palm infestation by A. ochraceus in the presence or absence of M. wasmanni (18 samples). Experiments were conducted under laboratory conditions, and CLS extracted metabolites and analyzed by GC-MS*

Substances classes	Biomarkers	Compounds	Palm treatments		
			Noninfested palm (T1) (n = 6)	**Termites-infested date palm (T2) (n = 6)**	**Termites-infested date palm + ant (T3) (n = 6)**
Benzenoid	Bio1	1,3-dimethyl- Benzene^	+	+	+
	Bio 2	Ethylbenzene^	+	+	+
	Bio 9	(1-methylethyl)- Benzene^	+	+	+
	Bio 10	1,3,5-trimethyl- Benzene^	+	+	+
	Bio 11	1-ethyl-3-methyl- Benzene^	+	+	+
	Bio 12	1-methyl-4-(1-methylethyl)- Benzene^	+	+	+
	Bio 15	1,3-diethyl- Benzene^	+	+	+
	Bio 17	1,2-diethyl- Benzene^	+	+	+
	Bio 18	1,4-diethyl- Benzene^	+	+	+
Total of benzenoid			**9**	**9**	**9**
Monoterpene	Bio 3	alpha-Pinene^	+	+	+
	Bio 4	unknown	+	+	**0**
	Bio 5	alpha-Phellandrene	+	+	+
	Bio 6	3-Carene	**0**	**0**	+
	Bio 7	Terpinolene	+	+	+
	Bio 8	Camphene	+	+	+
	Bio 13	γ-Terpinene	+	+	+
	Bio 14	Tricyclene^	+	**0**	**0**
	Bio 22	(+)-4-Carene	+	**0**	**0**
Total of monoterpene			**8**	**6**	**6**
Sesquiterpene	Bio 23	Copaene^	+	**0**	**0**
	Bio 24	Longifolene	**0**	**0**	+
	Bio 25	Aromadendrene	**0**	**0**	+
	Bio 26	Valencene	**0**	+	+
	Bio 28	Guaiene	**0**	+	+
	Bio 29	(+)-Sativen	**0**	+	+
	Bio 30	(-)-Aristolene	**0**	+	+
Total of sesquiterpene			**1**	**4**	**6**
Ketone	Bio 16	m-methyl acetophenone	**0**	**0**	+
	Bio 27	7,8-Dihydro-β-ionone	**0**	**0**	+
Total of			**0**	**0**	**2**
Aldehyde	Bio 19	3-Isopropylbenzaldehyde*	**0**	+	+
Total of aldehyde			**0**	**1**	**1**
Ester	Bio 20	Isobornyl acetate^	**0**	**0**	+
	Bio 21	Bornyl acetate^	**0**	+	+
Total of ester			**0**	**1**	**2**

The compounds were identified by the NIST library and RT (tentative identification). * Several key compounds were verified by comparison with authentic standards. The metabolites marked with a "^" had a reverse match between 990-850 (0): not detected, (+): detected

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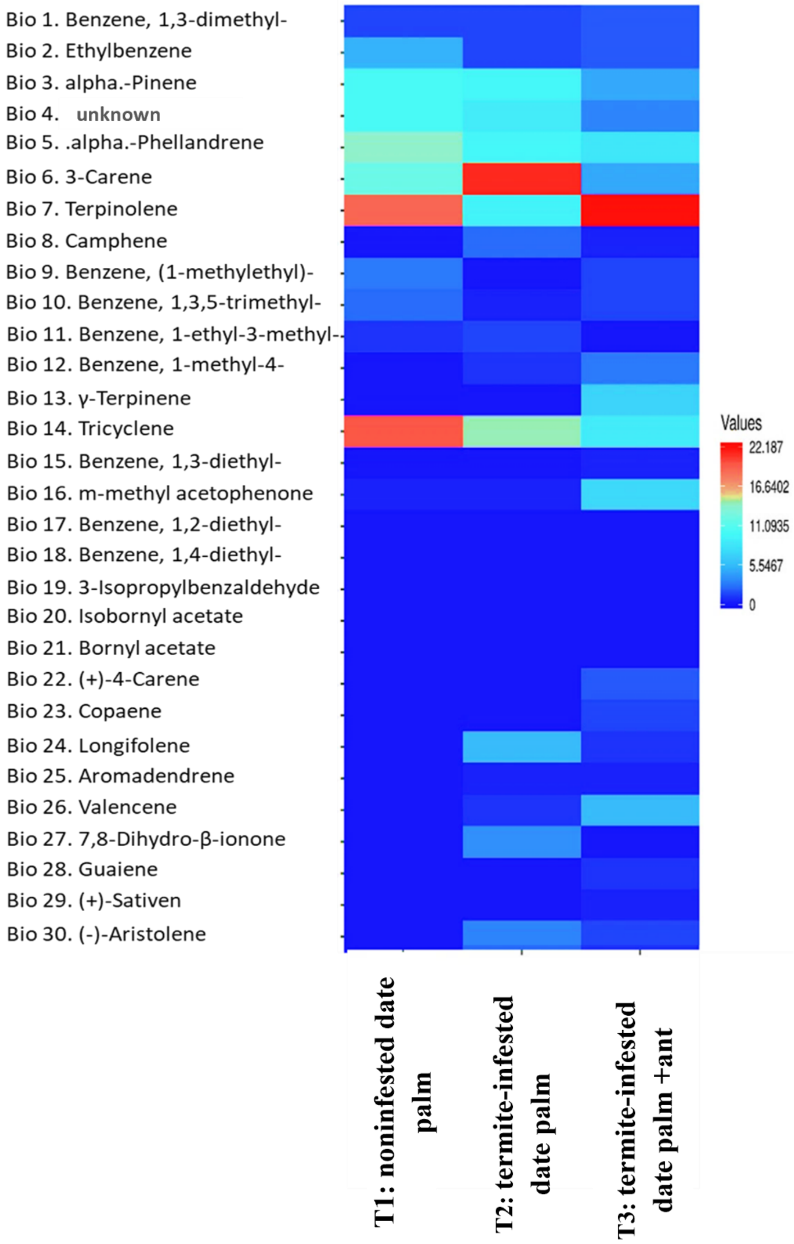

Figure 4. *Heat map of the mean percentage of volatile chemical compounds released into the headspace of three treatments (T_1, T_2 and T_3) based on the GC/MS analysis. The average percentages ranged from 0 to 22%. Smaller values were shown as deep blue and higher as red cells in the horizontal direction*

Discussion

Insect infestation of date palms can be controlled via several combined pest management approaches, including chemical insecticides (diazinon, imidacloprid, phosmet, and phosphine) (Yasin et al., 2019), host plant resistance and male sterilization practices (Faleiro, 2006). Due to the limitations of chemical insecticides and their hazardous side effects, researchers focus on finding eco-friendly, more sustainable agriculture and human health-compatible alternatives to control palm pests (Bianchi et al., 2006). Recently, biological control approaches have attracted the

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attention of researchers worldwide. This approach uses biocontrol agents such as parasitoids, predators, parasites, and microorganisms to destroy crop-harmful pests. The interaction between termites, palms and ants involves the release of several semiochemicals with various functions. Because of their importance in regulating behavior of pests, semiochemicals play a major role in biological management and are being considered in integrated pest management approaches (Smart, 2014). Therefore, the current study was designed to resolve intricate interplay and chemical signaling in the ecological dynamics of the tripartite community of termites (significant date pests palm), ants (predators of termites) and date palm trees. In such a community, several semiochemicals are released, orchestrating the interaction between the three species.

GC-MS analysis of volatile organic compounds revealed a rich tapestry of relationships among these organisms. According to our results, some identified compounds were distinguished for each group with profound ecological implications. Chemical compounds detected from the healthy palm trees were monoterpenes and benzene. Of these compounds, α-pinene is a monoterpene that acts as a repellent to red palm weevil at low concentrations (Ji et al., 2021). The scarcity of studies concerning the volatile compounds of palm trees hampered the interpretation of the importance of some chemical compounds. Five compounds, 3-carene, camphene, (+)-4-carene, guaiene and (-)-aristolene were released by termites while feeding on palm trees. The semiochemicals camphene, (+)-4-carene, and 3-carene are hydrogenated monoterpenes that have been reported to be among the defense mechanisms of termites and act as alarming pheromones (Mitaka and Akino, 2021). Because of their completely biodegradable, safe, and effective nature, monoterpenes can be employed as ideal lead compounds for the generation of new insecticides as well as potentially alternative pest control agents. Several studies reported monoterpenes' insecticidal activities (Abdelgaleil et al., 2009; Santos et al., 2011; Xie et al., 2014). 3-carene along with α-pinene was also released by some plant species, such as *Cupressus sempervirens*, with potent pesticide activity against the maize weevil (*Sitophilus zeamais*) (Langsi et al., 2020). Thus, the secretion of these monoterpenes plays a role in alarming individuals of termite communities and acting as repellent agents to their enemies.

Sesquiterpenes are another group of volatile compounds that have insecticidal activity against many insects. Several insects and plants release sesquiterpenes and have been tested against *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae) (Lima Janaína, 2013; Zhu et al., 2003). Two sesquiterpenes, guaiene and (-)-aristolene, were identified in the headspace of palm trees infested with termites. Several species of Reticulitermes, subfamily Heterotermitinae, consistently release significant amounts of sesquiterpenes (Clement et al., 2015). Sesquiterpenic compounds were reported in the frontal gland secretions of soldiers from different subfamilies of Termitidae (Scheffrahn, 1988; Valterová, 1988). Sesquiterpenes of termite origin may act as defensive molecules with repellent activities against ants, toxins or antihealants (Everaerts et al., 1993; Scheffrahn, 1988).

Several semiochemicals were detected from black ants upon addition to the bitrophic community including monoterpenes, ketone, sesquiterpene, and ester. Several studies γ-terpinene is a monoterpene released by ants in the headspace that has a strong insecticidal activity against different insects, including *Odontotermes obesus*, *Heterotermes sulcatus* Mathews (Blattodea: Rhinotermitidae). In these studies, γ-terpinene was extracted along with other compounds from various plants such as *Origanum vulgare* and *Pittosporum undulatum* and *Coleus amboinicus* (Gong and Ren,

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2020; Salem et al., 2020; Singh et al., 2002). Aromadendrene is another sesquiterpene with insecticidal activity against the larval stage of *Aedes aegypti* (de Morais, 2022). Two ester compounds were detected in the biosphere of the tripartite community: the isobornyl acetate and bornyl acetate. These two compounds are known for their insecticidal solid activities (Song, 2022).

Thus, the GC-MS-identified compounds in our study seem to play multiple roles during the interaction of the organisms in the tripartite community (date palm trees, termites and ants). They may act as repellent, alarming or killing semiochemicals. These compounds can be incorporated into integrated pest management systems to effectively control termites affecting the date palm trees. Semiochemicals are widely utilized in monitoring, mass trapping, mating disruption, attract-and-kill, and push-pull methods to control pest insects in agriculture, stored goods, and forests (Sharma et al., 2019). In addition, semiochemicals can be employed indirectly to spread the entomopathogenic microbes and to attract entomopathogenic microbes for insect management (Murali-Baskaran et al., 2018). Pathogen dispersal is made more efficient by auto-dissemination systems, which use both appealing, species-specific semiochemicals and entomopathogens. This method, often known as “lure and kill” or “attract and kill,” works well to manage a variety of insect pests (Vega, 2007).

Conclusions

This study discovered that volatile compound profiles changed significantly due to termites and related organisms during the date palm infestation. The distinct biomarkers determine the infestation status of date palms by *A. ochraceus*. The chemosphere often differs between the three statuses of date palms. Thus, the study results can determine the status of the date palm, whether it is intact, infested by *A. ochraceus,* or infested in the presence of ants. According to the literature, the identified compounds from the three organisms have multiple functions such as repelling, alarming, or killing activities. In recent years, the integration of semiochemicals with natural enemies has gained importance, and biological control agents and semiochemicals have become indispensable components of the integrated pest management of insect pests. Therefore, VOCs active against termites can be integrated into pest management approaches to control termites effectively. In addition, alarming or defense VOCs of termites can be counteracted to suppress their defense mechanisms.

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