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Moroccan *Trichoderma* species: a distinctive source of volatile organic compounds

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Volatile organic compounds (VOCs) in fungi have been studied less than those produced by bacteria. In this study, gas chromatography-mass spectrometry (GC-MS) was used to identify the volatile compounds for the first time in four Moroccan *Trichoderma* strains. *T. orientale*, *T. asperellum* (1), *Trichoderma* sp. (3), and *T. asperellum* (2), which were isolated from saffron bulbs and compost, the observed fungal VOCs were identified using the National Institute of Standards and Technology (NIST) library. According on the NIST database, sixty-six (66) VOCs were identified, belonging to various chemical families: monoterpenes, sesquiterpenes, diterpenes, alcohols, aldehydes, ketones, carboxylic acids, furans, terpenic alcohols, naphthalenes and aromatic hydrocarbons. In our study we found that *T. asperellum* (2) produced the most VOCs with 45%, followed by *Trichoderma* sp. (3) 39%, *T. orientale* 30%, and *T. asperellum* (1) 29%. In addition to common VOCs produced like ethanol; 1-propanol, 2-methyl, and 1-butanol, 3-methyl, the four strains produced a variety of other VOCs. *T. asperellum* (2) produced eighteen, *T. orientale* twelve, *Trichoderma* sp. (3) ten, and *T. asperellum* (1) eight. Further comparisons with bibliographic databases revealed that 14 VOCs were not previously identified in any fungus and are novel to the Trichoderma species studied. Additionally, 05 VOCs were mentioned in previous studies without specifying their applications, and 11 VOCs were found in organisms other than *Trichoderma* spp. This research provides valuable insights into the VOCs production by different *Trichoderma* strains and highlights the diversity and novelty of compounds produced by these fungi.

Keywords: *Trichoderma* spp., volatile organic compounds, GC-MS, Morocco

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INTRODUCTION

Since 1938, several chemists have studied odorous compounds of different chemical classes in fungal species using efficient analytical methods. The aim was either to uncover new sources of flavor in industry or to look for taxonomic markers. Indeed, six families of VOCs have been identified: hydrocarbons and heterocycles, alcohols, phenols and their derivatives, aldehydes, acids and their derivatives, and sulfur compounds. In addition to their roles in fungus-animal interactions and growth processes, some of these secondary metabolites also play a crucial role in signaling mechanisms within fungal communities (Chiron & Michelot, 2005). Fungal VOCs play a significant role in plant growth and disease control. Over 400 different bacterial and fungal species were found to produce nearly 1000 microbial VOCs, with 300 VOCs produced only by fungi of them. Some fungal VOCs have been found to positively influence plant growth and are commonly used in mycofumigation practices to combat plant diseases (Roy & Banerjee, 2019). The sporophore is a solid structure composed of chemicals that give fungi their unique odor. These VOCs, responsible for the characteristic odors, belong to different chemical families and exhibit distinct levels of solubility, stability, and volatility (Chiron & Michelot, 2005). In 1953, Dobbs and Hinson demonstrated the existence of a fungistatic factor, which indicates the widespread presence of an antifungal organic compound deep in the soil, the origin of which was unknown. In 1973, Watson and Ford demonstrated in their research that highlighted two key theories influencing the prevention of fungal propagule germination and fungal hypha development. The first theory demonstrates that fungal inhibition is caused by the lack of nutrients. The second one suggests the presence of antifungal microorganisms in the soil. The confluence of these two hypotheses explains VOCs-induced fungistase (Kaddes et al., 2020). Fungal VOCs promote plant defense through many methods, including initiating host protection systems and resistance to pathogens (Werner et al., 2016).

The fundamental mechanism is a change in the equilibrium of K+ ion currents and a disturbance of the pH gradient. Which limits fungal mycelial development and spore germination (Kaddes et al., 2019). *Trichoderma* species are fungi that have attracted significant research interest and prompted extensive studies. They are well-known for producing many secondary metabolites, including volatile organic compounds (VOCs). In Siddiquee's review, he identified 479 VOCs emitted by various *Trichoderma* species. These VOCs include a diverse range of compounds such as hydrocarbons, heterocycles, aldehydes, ketones, alcohols, phenols, thioalcohols, thioesters, and their derivatives, including benzene derivatives and cyclohexanes. Various VOCs are widely recognized for their applications in agriculture and industry, as well as medicine due to their antibacterial and immunosuppressive properties (Siddiquee, 2014).

Emerging technologies for the detection of VOCs are potentially useful analytical tools for determining these volatiles in fungal cultures. Chromatographic methods can be used to identify and quantify individual volatile compounds. Static headspace (SHS), dynamic headspace and solid phase microextraction (SPME) techniques have gained significant importance in the analysis of VOCs. Volatile fungal metabolites are generally detected using gas chromatography (GC) techniques and have been detected in various genera, including *Aspergillus*, *Fusarium*, *Mucor*, *Penicillium* and *Trichoderma* (Stoppacher et al., 2010 and Shahiri Tabarestani et al., 2016). Mass spectrometry is a powerful technique for detecting individual volatile molecules within complex mixtures. The identification of these molecules and confirmation of their structure are typically achieved by comparing the mass spectra of the molecules detected with the reference spectra in the library (Stoppacher et al., 2010).

Gas chromatography-mass spectrometry is a powerful technique for the direct detection of VOCs, as the fungi are grown directly in headspace vials, and detection by GC-MS-HS is fully automated (Güler et al., 2015). Indeed, *Trichoderma* species have been widely used in agriculture as biostimulants and biofongicides, to increase the growth and protection of vegetables. The study of VOCs produced by *Trichoderma* species is an emerging field of research, due to their potential biotechnological applications in agriculture (Jiménez-Bremont et al., 2024). *Trichoderma* species with the most researched VOCs profiles are *T. harzianum*, *T. virens*, *T. viride*, *T. atroviride*, *T. Koningii*, *T. album*, and *T. pdeudokoningii* (Astudillo et al., 2000; Siddiquee, 2014 and Speckbacher et al., 2021). However, limited research has been conducted on other *Trichoderma* species, such as *T. asperelloides* (Ruangwong et al., 2021), *T. asperellum* (Hamrouni et al., 2020; Degani et al., 2021 and Mulatu et al., 2022), and *T. Longibrachiatum* (Mulatu et al., 2022). Moreover, the VOCs profile produced by these fungi varies across genera and species, with certain species generating a broad spectrum of VOCs (Nieto-Jacobo et al., 2017; González-Pérez et al., 2018 and González-Pérez et al., 2022). There is a growing literature on bacterial VOCs and their role in signaling within terrestrial environments (Schulz & Dickschat, 2007 and Junker & Tholl, 2013). However, the ecological role of fungal VOCs has received much less attention (Bennett et al., 2012 and Bitas et al., 2013).

Thus, this research aims to identify the various volatile organic compounds released by Moroccan *Trichoderma asperellum* and *T. orientale* strains, whose VOCs profiles have not been previously studied, and to compare these profiles with those of other organisms through a comprehensive bibliographic review to investigate their various biotechnological applications.

MATERIALS AND METHODS

Materials

Autoclave, incubator and Agilent GC-MS system (Figure 2).

Fungal material

T. orientale (OM980237), *T. asperellum* (1) (OP364043), and *Trichoderma* sp. (3) (TBS3) were isolated from saffron bulbs (*Crocus sativus*) brought back from Taliouine (Souss-Massa, South of Morocco), while *T. asperellum* (2) (KU987252) was isolated from compost returned from Missour (Eastern Morocco). The fungal species were subcultured on a sterile PSA medium (Potato sucrose agar: 200 g potato, 20 g sucrose, 15 g Agar-agar, 1000 mL distilled water) for 7 days at 25°C and in the dark. For GC-MS analysis and headspace measurements, a 5 mm diameter fragment from the actively growing culture tips was placed inside 10 mL headspace vials containing 5 mL of sterile PSA medium. The control vials contained only the sterile PSA medium. All vials were properly crimped with head-to-screw plugs incorporating gas-teflon/silicone septum and incubated for 5 days at 22°C (Shahiri Tabarestani et al., 2016). Three repetitions were performed for each fungal species using (three replicates) and three control samples. [Note: The 10 mL headspace vials were chosen for two reasons: 1st: to trap a high concentration of VOCs, as these are released in small quantities. In other words, to avoid the loss of certain VOCs when using vials of 20 mL or more. 2nd: the size of these vials (10 mL) is adapted to the GC-MS sample incubator (90°C for 10 min) before automatic injection by COMBI PAL (CTC ANALYTICS)].

Morphological identification of fungal strains

The *Trichoderma* strains, isolated from saffron and compost, are naturally occurring species that were morphologically identified in the Plant, Animal Production and Agro-Industry Laboratory. Macroscopic observations of *Trichoderma* strains revealed rapid colony growth on PSA medium after four days. *Trichoderma* strain colonies exhibited rapid growth, initially filamentous, and became green and granular by the fourth day (Figure 1A). Microscopic observations under a light microscope (×400) showed that the mycelium is septate, hyaline, and extremely branched. The conidiophores were well developed, bearing two or more phialides, with primary branching occurring at nearly 90 degrees to the main axis. The bowling-pin-shaped phialides are arranged in whorls (Figure 1B). The spores are round, dark green, smooth, and small, and they began to form rapidly in the center of colonies (Figure 1C). As the culture matures, the presence of globular chlamydospores becomes evident (Figure 1D).

Phylogenetic tree of fungal strains

Phylogeny of *Trichoderma*: Fungi; Ascomycota; Sordariomycetes; Hypocreomycetidae; Hypocreales; Hypocreaceae; Trichoderma. (https://speciesfungorum.org/Names/fundic.asp). The strain *T. orientale* (OM980237) submitted to NCBI on 18 march 2022 (https://www.ncbi.nlm.nih.gov/nuccore/2207397505/OM980237), *T. asperellum* (1) (OP364043) submitted on 11 september 2022 (https://www.ncbi.nlm.nih.gov/nuccore/OP364043), and *T. asperellum* (2) (KU987252) submitted on PLN 19 november 2016 (https://www.ncbi.nlm.nih.gov/nuccore/KU987252) (Khirallah et al., 2017). These *Trichoderma* strains were obtained from the mycotheque of Plant, Animal Productions and Agro-Industry Laboratory, Faculty of Sciences, Ibn Tofail University, Kenitra, Morocco.

Volatile extraction, chromatographic conditions, and GC-MS analysis

After 10 minutes of equilibrium at 90°C, the volatile metabolites were extracted from the headspace of the fungal cultures using the automatic processor COMBI PAL (CTC ANALYTICS) samples. The identification of VOCs was carried out using an Agilent GC coupled with an Agilent MS detector. Volatile compounds were separated using an apolar capillary column HP-5MS (5% phenyl, 95% methyl siloxane) 30 m long with 0.25 mm internal diameter and 0.25 µm film thickness of the stationary phase. The oven temperature program is steady at 40°C for 2 minutes before increasing at a rate of 10°C/min to 200°C, then at 15°C/min to 260°C, where it was held for 10 minutes. The injector temperature was adjusted to 270°C, with the splitless injection mode, and the carrier gas flow rate (helium He) was 1 mL.min^{-1}. The GC-MS analysis was carried out with 70ev ionization energy, detection in scan mode, and a scan zone of 25 to 550 m/z. It was carried out for three repetitions of *Trichoderma* species cultures and control. The observed fungal volatile compounds were identified using the library NIST-MS (version 2.4, construction 2020).

Bibliographic analysis

The Moroccan *Trichoderma* strains were compared to other *Trichoderma* species and living organisms that were referenced in VOCs papers, using databases: Lens.org (Lens Version 9.1.3): https://www.lens.org; Google Scholar: https://scholar.google.com; National Center for Biotechnology Information, PubChem Compound Summary: https://pubchem.ncbi.nlm.nih.gov/compound/ and Chem spider (Search and share chemistry): https://www.chemspider.com (All the databases were accessed on May between 03-20, 2024).

RESULTS AND DISCUSSION

By comparing the mass spectra of the volatile compounds produced by the fungi with entries in the NIST-MS library (version 2.4, construction 2020), GC-MS analysis allowed the identification of volatile organic compounds in the headspace of the solid cultures of the four Moroccan species of *Trichoderma* [*T. orientale*, *T. asperellum* (1), *T. asperellum* (2), and *Trichoderma* sp. (3)], which were isolated from saffron corms and compost. The chromatographic profiles of these species, along with the control, are presented in Figure 3 (3A, 3B, 3C, 3D, 3E). Sixty-six (66) VOCs were identified, predominantly belonging to chemical families of monoterpenes, sesquiterpenes, diterpenes, alcohols, aldehydes, ketones, carboxylic acids, furans, terpenic alcohols, naphthalenes and aromatic hydrocarbons (Table 1). Fungal VOCs are the result of fungal metabolism and material breakdown by enzymes and mold acids during growth (Moularat et al., 2008). Fast-growing molds are the main sources of fungal VOCs (Schuchardt & Kruse, 2009).

The comparison of volatile organic compounds (VOCs) emitted by *Trichoderma* species, revealed that the strain *T. asperellum* (2) exhibited the highest percentage production of VOCs with 45% (30 VOCs), followed by *Trichoderma sp.* (3) with 39% (26 VOCs), *T. orientale* 30% (20 VOCs), and *T. asperellum* (1) with 29% (19 VOCs) (Figure 4). The nature and quantity of VOCs produced by fungi vary depending on the substrate on which the mold grows, the moisture level, and the species involved (Thrasher & Crawley, 2009). Molds can generate hundreds of VOCs.

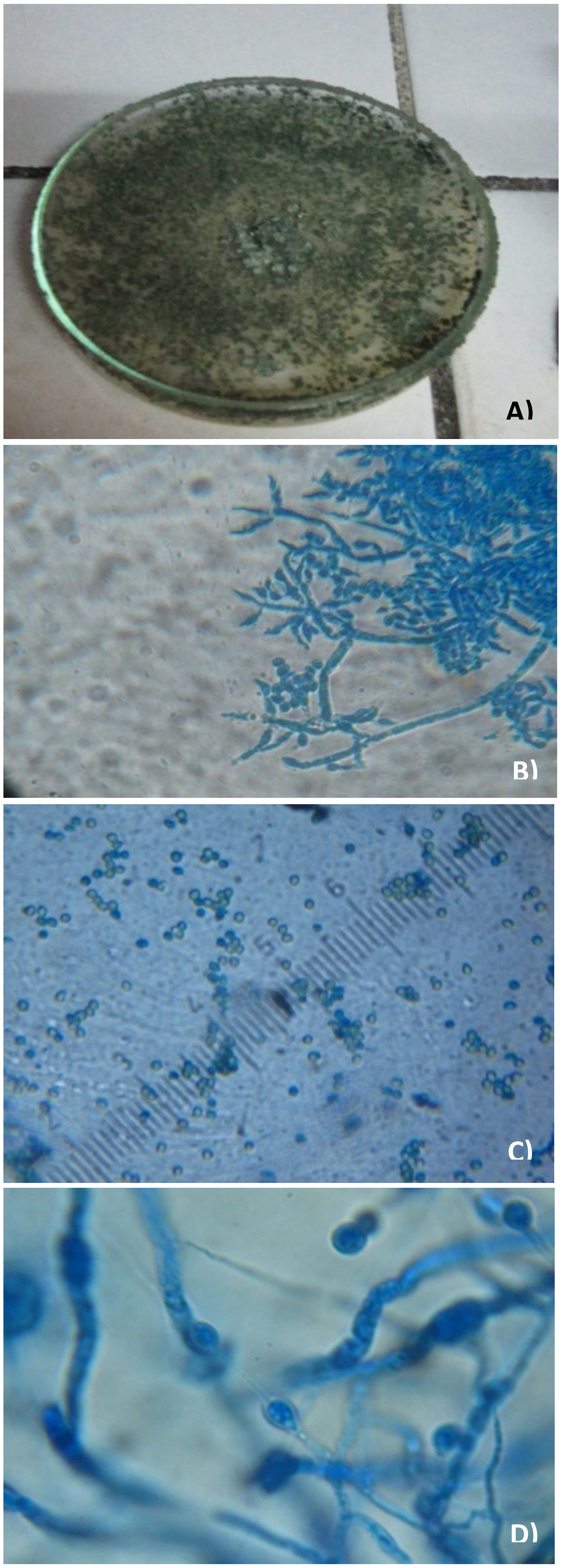

Figure1. Morphological description of *Trichoderma* strains. (A) 7-day-old culture on PSA medium, (B) conidiophores, (C) Conidia, (D) chlamydospores.

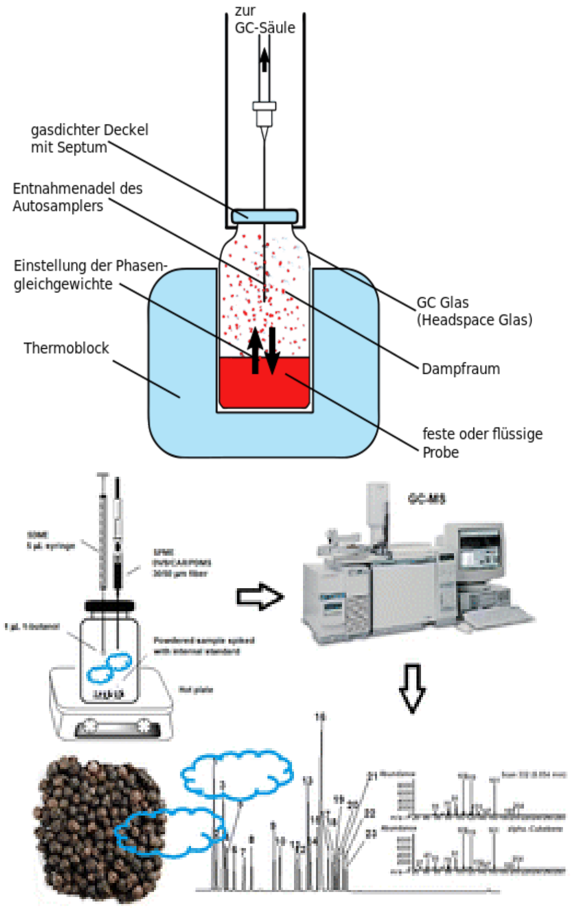

Figure 2. Gas chromatography-mass spectrometry-headspace (GC-MS-HS) used for the detection of VOCs.

Twenty-five (25) fungal VOCs were found for *Trichoderma atroviride* alone. When fungi are grown in the presence of mycotoxins, the concentration of fungal VOCs might fluctuate considerably. Indoor environments frequently contain many species able to produce mycotoxins (Stoppacher et al., 2010).

The four species produced several different volatile organic compounds. *T. asperellum* (2) produced eighteen (18) VOCs: Propanal, 2-methyl-; Butanal; 2-Butanone; Butanal, 2-methyl-; 1-Pentanol; 3-Heptanone, 5-methyl-; 2-Octanone; 2-Octanol; α -Phellandrene; α -Terpinene; β -Cymene; Octanoic acid, ethyl ester; Nonanal; 2-Octanol, acetate; Decanal; α -Curcumen; Epizonarene and Calamenene. The strain of *T. orientale* produced twelve (12): Acoradiene; Sativene; Cubebene; α -Caryophyllene; Benzenemethanol, α -methyl-; 1-Butanol, 3-methyl-, acetate; 1-Octen-3-ol; Butanone; Benzaldehyde; 3-Heptanone, 6-methyl-; Benzoic acid and pentanedioic acid. *Trichoderma sp.* (3) produced ten (10): Carnegine; Acetaldehyde, hydroxy; 2-Propanone, 1-hydroxy; Butyrolactone; 2-Propenoic acid; Furfural; 2-

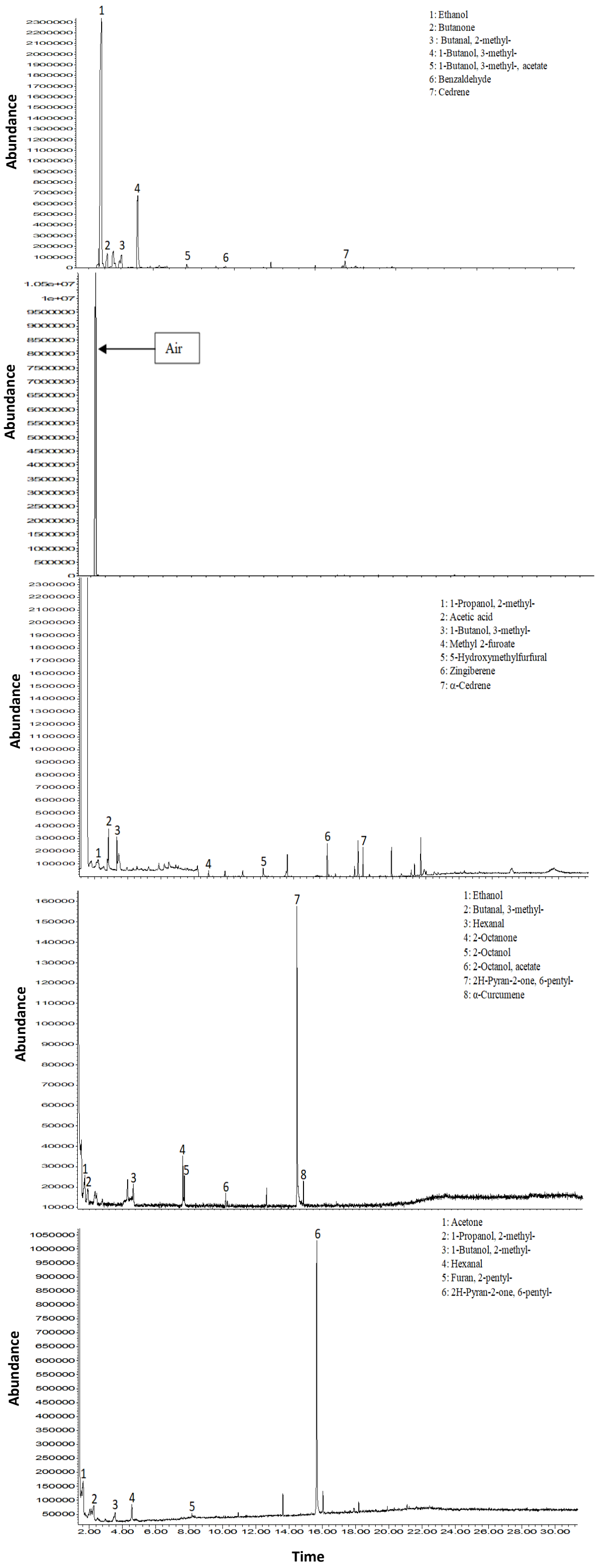

Figure 3. GC-MS chromatograms of the VOCs detected in the headspace of *Trichoderma* species isolated from saffron and compost culture after 5 days of growth, A: *T. orientale*, B: *T. asperellum* (1), C: *T. asperellum* (2), D: *Trichoderma* sp. (3), E: Control

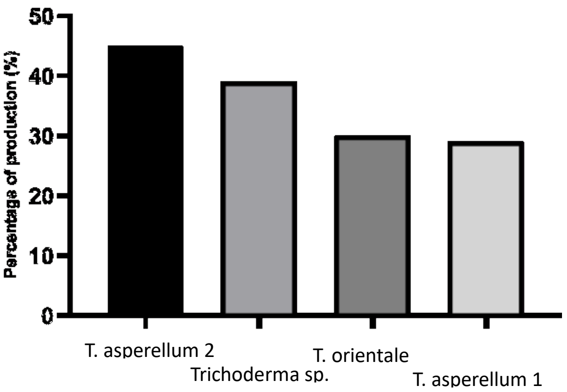

Figure 4. Percentage of volatile organic compounds production by Moroccan *Trichoderma* species isolated from saffron and compost.

Furanmethanol; Cubebol; 1-Bisabolone and β-Naphthyl myristate. *T. asperellum* (1) produced eight (08): β -Terpinen; β -Phellandrene; β -Curcumen; Zingiberene; β -Sesquiphellandrene; α -Cedrene; Cembrene and Zingiberenol. Conversely, these *Trichoderma* species emitted common volatile organic compounds. Ethanol; 1-Propanol, 2-methyl and 1-butanol, 3-methyl are the three (03) VOCs detected in the four species studied. While γ -Terpinene; 1-Propanol; 6-pentyl-2H-pyran-2-one and Furan, 2-pentyl are the four (04) VOCs produced by *T. asperellum* (2*)* and *Trichoderma* sp. (3). *T. asperellum* (1) and *Trichoderma* sp. (3) produced three (03) VOCs in common, acetic acid; 5-Hydroxymethylfurfural and Methyl 2-furoate. *T. orientale* and *Trichoderma* sp. (3) issued two (02): Cedrene and Alanine. *T. orientale* and *T. asperellum* (2) developed one (01): Butanal, 3-methyl-. *T. asperellum* (1)*, T. asperellum* (2*), and Trichoderma sp*. (3) produced three (03): Hexanal; Acetone and Toluene. *T. orientale*, *T. asperellum* (1) and *T. asperellum* (2) produced one (01): carbon dioxide. *T. orientale, T. asperellum* (1) and *Trichoderma* sp. (3) also emitted a common (01) VOC: 1-Butanol, 2-methyl (Table 1).

The four Moroccan *Trichoderma* species produced high levels of volatile organic compounds. The identified VOCs retention times ranged from 1,237 to 21,683 minutes. Cembrene has the longest retention time, while alanine has the lowest. The retention period of VOCs differed between four *Trichoderma* strains, with *T. orientale* having the smallest time (1,255 min) for carbon dioxide and the longest (20.06 min) for α-caryophyllene (α-humulene). The shortest duration for *T. asperellum* (1) is 1,273 minutes (carbon dioxide), while the longest is 21,683 minutes (cembrene). The shortest period for *T. asperellum* (2) is 1,279 minutes (carbon dioxide), while the longest is

Table 1: Comparative list of volatile organic compounds released by the four Moroccan *Trichoderma* species isolated from saffron and compost

Chemical families	VOCs detected	Chemical formula	*Trichoderma species*			
			T. orientale	*T. asperellum* 1	*T. asperellum* 2	*Trichoderma* sp. 3
Monoterpenes	Acoradiene	$C_{15}H_{24}$	+	-	-	-
	Sativene	$C_{15}H_{24}$	+	-	-	-
	Cubebene	$C_{15}H_{24}$	+	-	-	-
	γ -Terpinene	$C_{10}H_{16}$	-	-	+	+
	β -Terpinen	$C_{10}H_{16}$	-	+	-	-
	β -Phellandrene	$C_{10}H_{16}$	-	+	-	-
	Alanine	$C_3H_7NO_2$	+	-	-	+
	Carnegine	$C_{13}H_{19}NO_2$	-	-	-	+
	α -Phellandrene	$C_{10}H_{16}$	-	-	+	-
	α -Terpinene	$C_{10}H_{16}$	-	-	+	-
	β -Cymene	$C_{10}H_{14}$	-	-	+	-
Sesquiterpenes	Cedrene	$C_{15}H_{24}$	+	-	-	+
	α -caryophyllene	$C_{15}H_{24}$	+	-	-	-
	β -Curcumen	$C_{15}H_{22}$	-	+	-	-
	Zingiberene	$C_{15}H_{24}$	-	+	-	-
	β -Sesquiphellandrene	$C_{15}H_{24}$	-	+	-	-
	α -Cedrene	$C_{15}H_{24}$	-	+	-	-
	α -Curcumen	$C_{15}H_{22}$	-	-	+	-
	Epizonarene	$C_{15}H_{24}$	-	-	+	-
	Calamenene	$C_{15}H_{22}$	-	-	+	-
Diterpenes	Cembrene	$C_{20}H_{32}$	-	+	-	-
Alcohols	Ethanol;	C_2H_6O	+	+	+	+
	1-Propanol, 2-methyl	$C_4H_{10}O$	+	+	+	+
	1-Butanol, 3-methyl	$C_5H_{12}O$	+	+	+	+
	1-Butanol, 2-methyl	$C_5H_{10}O$	+	+	-	+
	Benzenemethanol, α -methyl-	$C_8H_{10}O$	+	-	-	-
	1-Butanol, 3-methyl-, acetate	$C_7H_{14}O_2$	+	-	-	-
	1-Octen-3-ol	$C_8H_{16}O$	+	-	-	-
	1-Propanol	C_3H_8O	-	-	+	+
	1-Pentanol	$C_5H_{12}O$	-	-	+	-
	2-Octanol	$C_8H_{18}O$	-	-	+	-
	2-Octanol, acetate	$C_{10}H_{20}$	-	-	+	-
Aldehydes	Butanal, 3-methyl-	$C_5H_{10}O$	+	-	+	-
	Benzaldehyde	C_7H_6O	+	-	-	-
	Acetaldehyde, hydroxy	$C_2H_4O_2$	-	-	-	+
	Hexanal;	$C_6H_{12}O$	-	+	+	+
	Butanal;	C_4H_8O	-	-	+	-
	Propanal, 2-methyl-	C_4H_8O	-	-	+	-
	Butanal, 2-methyl-	$C_5H_{10}O$	-	-	+	-
	Nonanal	$C_9H_{18}O$	-	-	+	-
	Decanal	$C_{10}H_{20}O$	-	-	+	-
ketones	Acetone	C_3H_6O	-	+	+	+
	Butanone	C_4H_8O	+	-	-	-
	3-Heptanone, 6-methyl-	$C_8H_{16}O$	+	-	-	-
	2-Propanone, 1-hydroxy	$C_3H_6O_2$	-	-	-	+
	Butyrolactone	$C_4H_6O_2$	-	-	-	+
	6-pentyl-2H-pyran-2-one	$C_{10}H_{14}O_2$	-	-	+	+
	2-Butanone	C_4H_8O	-	-	+	-
	3-Heptanone, 5-methyl-	$C_8H_{16}O$	-	-	+	-
	2-Octanone	$C_8H_{16}O$	-	-	+	-
Carboxylic acids	Acetic acid	$C_2H_4O_2$	-	+	-	+
	Benzoic acid	$C_7H_6O_2$	+	-	-	-
	Pentanedioic acid	$C_5H_8O_4$	+	-	-	-
	2-Propenoic acid	$C_3H_4O_2$	-	-	-	+
	Octanoic acid, ethyl ester	$C_8H_{16}O_2$	-	-	+	-
Furans	5-Hydroxymethylfurfural	$C_6H_6O_3$	-	+	-	+
	Furfural	$C_5H_4O_2$	-	-	-	+
	2-Furanmethanol	$C_5H_6O_2$	-	-	-	+
	Furan, 2-pentyl	$C_9H_{14}O$	-	-	+	+
	Methyl 2-furoate	$C_6H_6O_3$	-	+	-	+
Terpenic alcohols	Zingiberenol	$C_{15}H_{26}O$	-	+	-	-
	Cubebol	$C_{15}H_{26}O$	-	-	-	+
	1-Bisabolone	$C_{15}H_{24}O$	-	-	-	+
Naphthalenes	β -Naphthyl myristate	$C_{24}H_{34}O_2$	-	-	-	+
Aromatic hydrocarbons	Toluene	C_7H_8	-	+	+	+
	Carbon dioxide	CO_2	+	+	+	-
Total number	**66**		**20**	**19**	**30**	**26**

+: Present; **-:** Absent. The chemical formulas of the VOCs were retrieved from the PubChem database.

15,466 minutes (calamenene). Regarding *Trichoderma* sp. (3), the smallest time is alanine (1,237 min), and the longest is cedrene (17,898 min) (Table 2). Most microbiological VOCs have distinctive odors, with each fungal species capable of producing a unique set of volatile compounds. However, research has demonstrated that different fungal species often produce similar VOCs (Inamdar et al., 2020). Even within the same family or among isolates of the same species, significant variations in the compounds produced can occur. This indicates that secondary metabolic compounds exhibit chemical uniqueness at the species level. However, it has been demonstrated that different species can produce the same class of secondary metabolites, and even identical secondary metabolites (Tabarestani et al., 2016). *T. atroviride* and *T. viride* produce ethanol, beta-bisabolene, alpha-farnesene, beta-himachalene, dl-limonene, beta-sesquiphellandrene, caryophyllene, and zingiberene, as reported by studies (Stoppacher et al., 2010; Polizzi et al., 2011; Polizzi et al., 2012 and Hung et al., 2013).

Table 3 indicated fourteen (14) VOCs newly detected in the Moroccan strains of *Trichoderma* investigated and not previously reported in other fungi: Benzenemethanol, α-methyl-; Pentanedioic acid; 2-Propenoique acid; β-Terpinen; Zingiberenol; 2-Octanol; 2-Octanol, acetate; Acetaldehyde, hydroxy; 2-Propanone, 1-hydroxy; β-Naphthyl myristate; 5-Hydroxymethylfurfural; Cubebol; 1-Bisabolone and Carnegine. Five VOCs (Benzenemethanol, α-methyl; β-Terpinen; 2-Octanol, acetate; Acetaldehyde, hydroxy and 2-Propanone, 1-hydroxy) have been cited in the literature without identifying their biotechnological uses or potential function in other domains. In addition, eleven (11) VOCs were detected in living species other than *Trichoderma* spp., particularly Butanone; Benzaldehyde; 3-Heptanone, 6-methyl-; Benzoic acid; Sativene; α-caryophyllene (or α-humulene); Propanal, 2-methyl-; 1-Pentanol; 3-Heptanone, 5-methyl-; Decanal, and Furfural. The bibliographic data revealed the inhibitory potential of VOCs generated by *Trichoderma* species against pathogenic fungi (Table 3). Indeed, ethanol; 1-propanol, 2-methyl and 1-butanol, 3-methyl are chemical compounds that play the role of inhibiting mycelial growth and attracting fungivores. Fungal VOCs have been investigated for their possible use as a fuel source called "mycodiesel" (Morath et al., 2012 and Lemfack et al., 2014). The bibliographic study of the newly discovered VOCs in Moroccan *Trichoderma* strains revealed the role of cubebol in inhibiting the growth of algae *Heterosigma akashiwo*, repulsive activities against crop pest snails (*Acusta despecta*), and lethal activity levels against the larvae of mosquitoes (Aedes). Cubebol is also a popular long-term cooling and cooling agent in the food business (Chen et al., 2023).

Furthermore, carnegine has been proven to have significant antibacterial activity (Bouaziz et al., 2016). Bisabolone is active against *Candida albicans* and *Saccharomyces cerevisiae* (Sarg et al., 1994). Benzoic acid is an antimicrobial food preservative, Acetic acid is antibiotic that treats infections caused by bacteria or fungus, Decanal and 1-Butanol, 3-methyl are antifungal agents (PubChem database). Ethanol synthesized by the four species examined was produced by *T. viride* and *T. reesei* (Kumar et al., 2014). *T. atroviride* CCM F536 generated 1-propanol, 2-methyl and 1-butanol, 3-methyl (Siddiquee, 2014). *T. longibrachiatum* EF5 contains 1-butanol, 2-methyl, which was developed by *T. orientale*, *T. asperellum* (1), and *Trichoderma* sp. (3). Furthermore, Acoradiene, Benzaldehyde, and 1-octen-3-ol found in *T. orientale* were formed by *T. guizhouense*, *Photorhabdus temperata*, and *T. atroviride*, respectively (Ullah et al., 2015; Li et al., 2021 and Speckbacher et al., 2021). *T. orientale*, *T. asperellum* (1), and *T. asperellum* (2) produced carbon dioxide, as did *T. hamatum*, *T. koningii*, and *T. viride* (Dal Bello et al., 1997; Chahal et al., 2014 and JayaMadhuri et al., 2020). Alcohols (hexanol; 2-ethyl-1hexanol; 1-butanol; 3-methyl-1-butanol; 2-methyl-1-propanol; 2-terpineol), terpenes (limonene), sesquiterpenes (thujocopsene, cedrene, farnesene), and ketones (pentanyldimethanone, pentanonone, acetonone) are among the most frequent fungus VOCs. Molds emit low molecular weight VOCs, such as alcohols, aldehydes, and ketones, which are generally non-specific during their primary growth phase. In contrast, high molecular weight VOCs (sesquiterpenes, aromatic hydrocarbons) are more specific to the growth stationary phase (Reboux et al., 2011). The bioactivity of *Trichoderma* depends on specialized metabolites called secondary metabolites; these include VOCs. For example, VOCs can enhance plant growth, modify root structure, and activate plant defenses against both biotic and abiotic stresses (Jiménez-Bremont et al., 2024). VOCs play an essential signaling role in the natural environments of fungi. Additionally, they ensure numerous ecological interactions between fungi, plants and bacteria (Morath et al., 2012).

Table 2. The retention time of VOCs identified in the four Moroccan *Trichoderma* species isolated from saffron and compost

T. asperellum (2)		*Trichoderma* sp. (3)		*T. orientale*		*T. asperellum* (1)	
Compound name	RT* (min)	Compound name	RT* (min)	Compound name	RT* (min)	Compound name	RT* (min)
Carbon dioxyde	1.279	Alanine	1.237	Carbon dioxyde	1.255	Carbon dioxyde	1.273
Ethanol	1.423	Ethanol	1.531	Alanine	1.291	Ethanol	1.483
Acetone	1.525	Acetone	1.633	Ethanol	1.808	Acetone	1.633
Butanal	1.657	1-Propanol	1.812	Butanone	2.149	1-Propanol, 2-methyl	2.262
1-Propanol	1.698	1-Propanol, 2-methyl	2.225	1-Propanol, 2-methyl	2.208	Acetic acid	2.944
Propanal, 2-methyl-	1.729	Acetaldehyde, hydroxy	2.776	Butanal, 3-methyl-	2.929	1-Butanol, 3-methyl	3.489
2-Butanone	1.914	Acetic acid	2.848	1-Butanol, 3-methyl	3.050	1-Butanol, 2-methyl	3.543
1-Propanol, 2-methyl	2.076	2-Propanone, 1-hydroxy	3.351	1-Butanol, 2-methyl	4.049	Toluene	3.980
Butanal, 3-methyl-	2.321	1-Butanol, 3-methyl	3.447	1-Butanol, 3-methyl-, acetate	7.064	Hexanal	4.555
Butanal, 2-methyl-	2.435	1-Butanol, 2-methyl	3.531	Benzaldehyde	8.877	β-Terpinen	8.885
1-Pentanol	3.309	Toluene	3.956	3-Heptanone, 6-methyl-	9.339	Methyl 2-furoate	9.867
1-Butanol, 3-methyl	3.315	2-Propenoic acid	4.333	Benzenemethanol, α-methyl-	11.805	β-Phellandrene	8.885
Toluene	4.053	Hexanal	4.555	1-octen-3-ol	11.817	5-Hydroxymethylfurfural	12.167
Hexanal	4.322	Furfural	5.256	Benzoic acid	14.412	β-Curcumene	16.131
3-Heptanone, 5-methyl-	7.597	2-Furanmethanol	5.885	Pentanedioic acid	16.287	Zingiberene	16.137
2-octanone	7.669	Butyrolactone	6.903	Acoradien	16.663	β-Sesquiphellandrene	16.538
Furan, 2-pentyl	7.681	Furan, 2-pentyl	8.172	Cedrene	16.853	Zingiberenol	17.688
2-Octanol	7.819	γ-Terpinene	8.885	Sativene	17.257	α-Cedrene	18.851
α-Phellandrene	7.921	Methyl 2-furoate	9.873	Cubebene	19.764	Cembrene	21.683
α-Terpinene	8.130	β-Naphthyl myristate	10.849	α-caryophyllène or α-humulene	20.060		
β-Cymene	8.268	5-Hydroxymethylfurfural	12.173				
γ-Terpinene	8.837	Cubebol	17.688				
Octanoic acid, ethyl ester	9.573	1-Bisabolone	19.257				
Nonanal	9.916	Carnegine	17.682				
2-Octanol, acetate	10.065	6-pentyl-2H-pyran-2-one	15.670				
Decanal	10.400	Cedrene	17.898				
6-pentyl-2H-pyran-2-one	14.622						
α-Curcumene	14.909						
Epizonarene	15.149						
Calamenene	15.466						

*RT: Retention time

In both intra- and interspecific associations, VOCs released by plants and microorganisms are essential for signaling, competition, and antagonism (Midzi et al., 2022 and Mukherjee et al., 2022). *Trichoderma* species are the subject of extensive research. They are known for their abundant production of numerous specialized metabolites, such as VOCs, which are used in both industry and agriculture (Singh et al., 2020). The volatile 6-pentyl-2H-pyran-2-one (6-PP), known for its characteristic coconut odor, has been used as an additive in the food industry (Lee et al., 2016). Furthermore, studies have indicated that this VOC can influence plant growth (Garnica-Vergara et al., 2016). Tomato plants sprayed with purified 6-PP metabolite exhibited increased biomass, a well-developed root system that enhances nutrient uptake, and improved resistance to pathogens (Vinale et al., 2008). Until now, most studies on fungal VOCs have focused on their food and aromatic properties. However, these compounds also have potential applications as semiochemicals for insects or as indirect indicators of fungal growth in agriculture. In addition, research on fungal volatiles has also been carried out for various purposes, including monitoring deterioration, chemotaxonomy biofilters, and biodiesel. Similarly, to detect plant and animal diseases, for mycofumigation and as far as plant health (Hung et al., 2015).

Table 3. Search for potential biotechnological applications of volatile organic compounds identified in Moroccan strains of *Trichoderma* in comparison with other living organisms based on different databases.

Compound name	VOCs producers	Potential applications or functions	Bibliographical references	Natural products found in Plantae and other living organisms and their role with data available in compound summary description and Taxonomy (PubChem database)
Acoradiene	*Fusarium oxysporum*	Nematicide Major component of the male pheromone of *Gnatocerus cornutus*	(Tashiro et al., 2004 and Freire et al., 2012)	*Thujopsis dolabrata*, *Cistus monspeliensis*, and other organisms. (32 items)
Sativene	Bipolaris victoriae S27 *Cladosporium cladosporioides CL-1* *Helminthosporium & Fomitopsis pinicola Bipolaris eleusines*	Plant-growth regulator Improving seedlings growth	(Kramer & Abraham, 2012; Li et al., 2018; Sridharan et al., 2020 and Wang et al., 2020)	*Metacalypogeia alternifolia*, *Solanum lycopersicum*, and other organisms. (8 items)
Cubebene	*F. solani*	Chemical communication	(Ana et al., 2020)	*Piper cubeba*, *Cinnamomum aromaticum*, and other organisms. (77 items)
γ-Terpinene	*T. atroviride ATCC 74058* Cumin *Escherichia coli*	Pharmaceutical and cosmetics industrie Alternative biofuel	(Chiron & Michelot, 2005; Tahri et al., 2016 and Qi et al., 2018)	*Camellia sinensis*, *Artemisia thuscula,* and other organisms. - Antioxidant activity. - Plant metabolite (931 items)
β-Terpinene	Satureja hortensis L. (Lamiaceae family)		(Rezaei-Chiyaneh et al., 2023)	*Perilla frutescens*, *Artemisia sericea*, and other organisms. (49 items)
β-Phellandrene	*T. atroviride ATCC 74058* *Synechocystis* sp. PCC 6803	Photosynthetic generation of hydrocarbons	(Bentley et al., 2013 and Siddiquee, 2014)	*Helichrysum taenari*, *Pinus densiflora*, and other organisms. - Plant metabolite. (385 items)
Alanine	*Trichoderma* spp. *Trichoderma harzianum*	Source of nitrogen in the culture media	(Calistru et al., 1997; Tashpulatov et al., 1998 and Siddiquee, 2014)	Metabolite found in or produced by Escherichia coli (strain K12, MG1655). (2,811 items)
Carnegine	Hammada scoparia	Bactericidal activity Molluscicidal activity	(Mezghani-Jarraya et al., 2009 and Bouaziz et al., 2016)	
α-Phellandrene	*T. atroviride* ATCC 74058 *Curcuma longa* L. leaves	*In vitro* anti-inflammatory activity	(Siddiquee, 2014 and Mossmann et al., 2024)	*Artemisia thuscula*, *Espeletia weddellii*, and other organisms. - Volatile oil component, - Plant metabolite, - Antimicrobial agent. (463 items)
α-Terpinene	*T. atroviride ATCC 74058*		(Siddiquee, 2014)	*Camellia sinensis, Artemisia thuscula*, and other organisms. - Volatile oil component - Plant metabolite. (621 items)
β-Cymene	*Trichoderma* spp. *Marasmius oreades*	Anti-inflammatory activity	(Chiron & Michelot, 2005; Lee et al., 2016 and Marques et al., 2019)	Magnolia officinalis, Cymbopogon martinii, and other organisms. (68 items)
Cedrene	*Trichoderma longibrachiatum EF5* *T. longibrachiatum AU158* *Trichoderma guizhouense* *Rhododendron* species	Antifungal activity Arabidopsis lateral growth stimulation Repellent activities	(Bai et al., 2019; Sridharan et al., 2020; Li et al., 2021 and Mulatu et al., 2022)	*Aristolochiaceae*, *Lonicera japonica*, and *other organisms.* (25 items)
α-caryophyllène	*Gyrinops walla* *Lactarius mitissimus*	Antifungal activity	(Kramer & Abraham, 2012; Bitas et al., 2013 and Munasinghe et al., 2021)	*Trichogonia grazielae*, *Callilepis laureola*, and *other organisms.* (978 items)
β-Curcumene	*Trichoderma guizhouense* Curcuma amada	Antibacterial; larvicidal insecticidal properties	(Li et al., 2021 and Narayanankutty et al., 2021)	*Curcuma xanthorrhiza*, *Curcuma kwangsiensis*, and *other organisms.* (17 items)
Zingiberene	*T. atroviride* ATCC 74058 Ginger essential oil	Antibacterial activity against *Staphylococcus*	(Stoppacher et al.,2010 and Wang et al., 2020)	*Humulus lupulus*, *Zanthoxylum simulans*, and other organisms. (55 items)

		aureus and *Escherichia coli*		
β-Sesquiphellandrene	*Trichoderma* spp.		(Lee et al., 2016)	*Curcuma kwangsiensis, Curcuma phaeocaulis*, and other organisms. (67items)
α-Cedrene	*Trichoderma* spp. *Alternaria alternata* *Muscodor albus*		(Kramer & Abraham, 2012; Lee et al., 2016 and Weikl et al., 2016)	*Camellia sinensis*, *Magnolia officinalis*, and other organisms. - Human urinary metabolite - Volatile oil component (136 items)
α-Curcumene	*T. atroviride (ATCC 74058* Essential oil of *Saussurea lappa* roots Essential oil of *Vetiveria zizanioides*	Larvicidal activity Antimycobacterial activity	(Gupta et al., 2012; Liu et al., 2012 and Siddiquee, 2014)	*Solanum tuberosum,* and other organisms. (128 items)
Epizonarene	*T. longibrachiatum* *T. harzianum* *T. viride*		(Citron et al., 2011)	*Araucaria columnaris*, *Teucrium leucocladum*, and other organisms. (21 items)
Calamenene	*T. longibrachiatum* *T. harzianum* *T. viride*		(Citron et al., 2011)	*Camellia sinensis*, *Calypogeia muelleriana*, and other organisms. 138 items
Cembrene	Endophytic fungus isolated from *Taxus yunnanensis*	Antibiotic: Stronger inhibition to *Staphylococcus aureus*, *Bacillus subtilis* and *Candida albicans*	(Chen et al., 2009)	*Boswellia sacra*, *Pinus nigra*, and other organisms. (6 items)
Ethanol	*Ceratocystis fimbriata* *Trichoderma viride* & *Saccharomyces cerevisiae* *Trichoderma reesei*	Mycelial growth inhibition	(Kumar et al., 2014 and Kaddes et al., 2020)	*Humulus lupulus*, *Tuber melanosporum*, and other organisms. - Antiseptic drug, - Polar solvent, - Neurotoxin, a central nervous system depressant, a teratogenic agent, - Disinfectant, - Human metabolite, - Saccharomyces cerevisiae metabolite, - Escherichia coli metabolite and a mouse metabolite. (679 items)
1-Propanol, 2-methyl	*Phomopsis sp.* *T. atroviride CCM F536*	Fungivore attractant	(Morath et al., 2012 and Siddiquee, 2014)	*Angelica gigas*, *Tuber melanosporum,* and other organisms. - *Saccharomyces cerevisiae* metabolite. (526 item)
1-Butanol, 3-methyl	*Muscodor albus* *Saccharomyces cerevisiae* (souche CR1) *T. atroviride* (CCM F536)	Mycelial growth inhibition Rice seeds germination and seedling growth inhibition	(Siddiquee, 2014; Kaddes et al., 2020 and Nguyen et al., 2024)	*Ambrosiozyma monospora*, *Humulus lupulus*, and other organisms. - *Saccharomyces cerevisiae* metabolite - Antifungal agent. (604 items)
1-Butanol, 2-methyl	*Paenibacillus jamilae HS-26* *T. longibrachiatum EF5* *Saccharomyces cerevisiae* (souche CR1)	Mycelial growth inhibition Herbicide: Rice seeds germination and seedling growth inhibition	(Wang et al., 2019; Kaddes et al., 2020; Sridharan et al., 2020 and Nguyen et al., 2024)	*Francisella tularensis*, *Camellia sinensis*, and other organisms. - Saccharomyces cerevisiae metabolite. (485 items)
Benzenemethanol, α-methyl-	*Bunium persicum*	Larvicidal activity essentiel oil	(Sanei-Dehkordi et al., 2016)	
1-Butanol, 3-methyl-, acetate	*Muscodor albus* *T. atroviride*	Antifungal activity	(Morath et al., 2012 and Speckbacher et al., 2021)	*Humulus lupulus*, *Zingiber mioga*, and other organisms. - Metabolite - Saccharomyces cerevisiae metabolite. (40 items)
1-octen-3-ol	*Agaricus bisporus* *Penicilum paneum* *T. atroviride* *Saccharomyces cerevisiae*	Conidia germination inhibition Pollinating insects attractant	(Bitas et al., 2013; Kaddes et al., 2020 and Speckbacher et al., 2021)	*Camellia sinensis*, *Tetradenia riparia*, and other organisms. - Insect attractant, - Volatile oil component, - Fungal metabolite and - Antimicrobial agent. (192 items)
1-Propanol	*T. atroviride*		(Speckbacher et al., 2021)	*Tuber melanosporum*, *Zea mays*, and other organisms. (478 items)
1-Pentanol	*Tuber magnatum Pico* *Ischnoderma benzoinum* *Armillaria mellea*		(Chiron & Michelot, 2005)	*Camellia sinensis*, *Angelica gigas*, and other organisms. - Plant metabolite - Human metabolite.

				(146 items)
2-Octanol	Green Gram Plant	Moth insect Attractant	(Mobarak et al., 2022)	*Daphne odora, Zea mays*, and other organisms. - Volatile oil component - Plant metabolite (32 items)
2-Octanol, acetate	*Zingiber officinale* Fresh Rhizomes		(Nishimura, 1995)	
Butanal, 3-methyl-	*Bacillus safensis, B. pumilus & B. subtilis* *Tuber mesentericum* *Fusarium oxysporum* *Trichoderma* spp.	Antifungal activity	(Mauriello et al., 2004; Lee et al., 2016; Erjaee et al., 2019 and Speckbacher et al., 2021)	*Francisella tularensis*, *Eucalyptus pulverulenta,* and other organisms. - Flavouring agent, - Plant metabolite, - Volatile oil component, - Saccharomyces cerevisiae metabolite. (566 items)
Benzaldehyde	*Tuber mesentericum* *Tuber excavatum* *Tuber aestivum* *Tuber panniferum* *Photorhabdus temperata*	Biomarqueurs Fongistatique Post harvest antifungal activity Insecticidal Activity	(Mari & Guizzardi, 1998; Mauriello et al., 2004; Bitas et al., 2013; Ullah et al., 2015 and Sridharan et al., 2020)	*Camellia sinensis*, *Humulus lupulus*, and other organisms. - Flavouring agent, - Fragrance, - Odorant receptor agonist, - Plant metabolite. (874 items)
Acetaldehyde, hydroxy				*Arabidopsis thaliana* and *Homo sapiens* - Fundamental metabolite - Human metabolite (481 items)
Hexanal	*Bacillus subtilis—ESR 24* *Trichoderma atroviride* TRS25 *Boletus edulis* & *Pleurotus ostreatus*	Antifungal activity Signalisation	(Chiron & Michelot, 2005; Nawrocka et al., 2018 and Jayakumar et al., 2021)	*Camellia sinensis*, *Humulus lupulus*, and other organisms. - Human urinary metabolite. - Volatile compound associated with undesirable flavours. (792 items)
Butanal	*Trichoderma* spp.		(Lee et al., 2016)	*Gossypium hirsutum*, *Ligusticum striatum*, and other organisms. - Metabolite found in or produced by *Escherichia coli* (strain K12, MG1655). - Biomarker, - Escherichia coli metabolite and a mouse metabolite. (509 items)
Propanal, 2-methyl-	*Tuber melanosporum Vitt* *Tuber aestivum (Chatin) Vitt*		(Chiron & Michelot, 2005)	*Angelica gigas*, *Tuber melanosporum*, and other organisms. - Saccharomyces cerevisiae metabolite. (473 items)
Butanal, 2-methyl-	*Trichoderma* spp. *Tuber magnatum Pico* *Tuber borchii Vitt* *Saccharomyces cerevisiae*		(Chiron & Michelot, 2005 and Lee et al., 2016)	*Tuber melanosporum*, *Zea mays*, and other organisms. - Volatile oil component, - Plant metabolite - Saccharomyces cerevisiae metabolite. (484 items)
Nonanal	*Pseudomonas spp*. & bacteria isolated from canola and soya plants *Trichoderma* spp. *T. harzianum FA1132*	Antifungal activity	(Hewavitharana et al., 2014; Siddiquee, 2014 and Lee et al., 2016)	*Camellia sinensis, Humulus lupulus*, and other organisms. - Human metabolite and a plant metabolite. (451 items)
Decanal	*Pseudomonas spp.* & bacteria isolated from canola and soya plants *Tuber aestivum (Chatin) Vitt*	Antifungal activity	(Chiron & Michelot, 2005 and Hewavitharana et al., 2014)	*Camellia sinensis*, *Gymnodinium nagasakiense,* and other organisms. - Antifungal agent, - Fragrance and a plant metabolite. (368 items)
Acetone	*T. harzianum FA1132* *T. atroviride*		(Siddiquee, 2014 and Speckbacher et al., 2021)	*Humulus lupulus*, *Angelica dahurica* var. *formosana*, and other organisms. (543 items)
Butanone	Bacillus & Pseudomonas spp. *Tuber aestivum*. *Tuber borchii* *Tuber melanosporum*	Promoting plant growth	(Chiron & Michelot, 2005; Sharifi & Ryu, 2016 and Tyagi et al., 2020)	*Alpinia chinensis*, *Tuber melanosporum,* and other organisms. - Polar aprotic solvent - Bacterial metabolite. (468 items)
3-Heptanone, 6-methyl-	*Marasmius oreades*		(Chiron & Michelot, 2005)	*Origanum hypericifolium* (1 item)

2-Propanone, 1-hydroxy	Honeybee (Citrus Royal jelly)		(Montaser et al., 2023)	*Durio zibethinus*, *Capsicum annuum*, and other organisms. - Human metabolite, - Escherichia coli metabolite and a mouse metabolite. (493 items)
Butyrolactone	*Aspergillus versicolor F62*	Anti-inflammatory activity	(Gong et al., 2014)	*Aethus indicus*, *Streptomyces*, and other organisms. - Neurotoxin - Metabolite - Pharmacological agent - Solvent. (72 items)
6-pentyl-2H-pyran-2-one	*Trichoderma sp. YMF 1.00416* *T. viride* *T. harzianum* *T. koningii* *Myrothecium sp.* *Trichoderma harzianum*	Nematicidal activity Anti-fungal activity	(Scarselletti & Faull, 1994 and Siddiquee, 2014)	*Myrothecium*, *Trichoderma aureoviride*, and other organisms. - Metabolite. (12 items)
2-Butanone	*T. atroviride CCM F536*		(Siddiquee, 2014)	*Alpinia chinensis*, *Tuber melanosporum*, and other organisms. (468 items)
3-Heptanone, 5-methyl-	*Marasmius oreades* Platynereis dumerilii and Nereis succinea		(Chiron & Michelot, 2005 and Fletcher et al., 2022)	*Emblica officinalis* *Thymus vulgaris* (garden thyme) *Thymus quinquecostatus* (3 items)
2-Octanone	Cocultures of *F. oxysporum* and *T. atroviride*		(Speckbacher et al., 2021)	*Heracleum dissectum*, *Eryngium foetidum*, and other organisms. - Metabolite. (50 items)
Acetic acid	*T. longibrachiatum*	Nematicidal activity	(Siddiquee, 2014)	*Camellia sinensis*, *Microchloropsis*, and other organisms. Antibiotic that treats infections caused by bacteria or fungus. (111 items)
Benzoic acid	*Agaricus blazei Murrill* *Lactarius helvus* *Gyrophragmium dunalii* *Hydnum repandum*	Antifungal activity	(Calderón et al., 1993; Chiron & Michelot, 2005 and Wu et al., 2009)	*Desmos chinensis*, *Paeonia emodi*, and other organisms. - Antimicrobial food preservative, - Plant metabolite, - Human xenobiotic metabolite, - Algal metabolite - Drug allergen. (446 items)
Pentanedioic acid	*Corynebacterium glutamicum*	C5 plasticizer synthesis	(Sohn et al., 2022)	*Glycine max, Drosophila melanogaster,* and other organisms. - Human metabolite - Daphnia magna metabolite. (90 items)
2-propenoic acid		An excellent source of carboxylic rich (COOH) coatings	(Kováčik et al., 2024)	*Cocos nucifera*, *Gynerium sagittatum*, and other organisms - Metabolite. (20 items)
Octanoic, ethyl ester acid	*Boletus edulis*		(Chiron & Michelot, 2005)	*Gardenia jasminoides*, *Mandragora autumnalis*, and other organisms. - Metabolite - Metabolite found in or produced by Saccharomyces cerevisiae. (62 items)
5-Hydroxymethylfurfural	*Alpinia oxyphylla*	Neuroprotective effects against Alzheimer's disease	(Johnson et al., 2024)	Tropicoporus linteus, Peperomia leptostachya, and other organisms. - An indicator and a Maillard reaction product. (148 items)
Furfural	*Agaricus bisporus* *Lentinula edodes*		(Chiron & Michelot, 2005)	*Francisella tularensis*, *Angelica gigas*, and other organisms. - A Maillard reaction product and a metabolite. (163 items)
2-Furanmethanol	*Oxyporus latem arginatus* Grape berries infected with *Aspergillus carbonarius -strain OTA5010-*	Antifungal activity	(Giannoukos et al., 2020)	*Perilla frutescens*, *Zea mays*, and other organisms. It has a role as a Maillard reaction product. (72 items)

Furan, 2-pentyl	*Bacillus megaterium* XTBG34 *T. atroviride CCM F536* *T. atroviride ATCC 74058*	Plant growth promotion	(Paul & Park, 2013 and Siddiquee, 2014)	Magnolia officinalis, Daphne odora, and other organisms. - Aspergillus metabolite - Human urinary metabolite, - Volatile oil component, - Insect repellent, - Flavouring agent, - Plant growth stimulator - Bacterial metabolite. (149 items)
Methyl 2-furoate	*T. atroviride CCM F536*		(Siddiquee, 2014)	*Actinidia chinensis*, *Carica papaya,* and *Mangifera indica* (12 items)
Zingiberenol	*Mormidea v-luteum* Ginger (*Zingiber officinale*) Rhizomes	Different functions in chemical communication Aggregation pheromone	(Khrimian et al., 2015 and Moliterno et al., 2021)	*Zingiber officinale* (5 items)
Cubebol	Sugi (*Cryptomeria japonica*) bark	Algal inhibitory activities Repellent activities Larval lethal activity Lasting cooling and refreshing agent in the food industry	(Saijo et al., 2013 and Chen et al., 2023)	*Pinus densiflora, Picea glehnii,* and other organisms. (45 items)
1-Bisabolone	*Apium graveolens*	Antimicrobial activity Antifeedant action	(Sarg et al., 1994)	*Stevia eupatoria* and *Bethencourtia palmensis* (6 items)
β-Naphthyl myristate		Used as the substrate to improve the detection sensitivity and specificity significantly lipase inhibitors	(Tang et al., 2016)	
Carbon dioxide	*Trichoderma hamatum* *Trichoderma koningii* & *Penicillium janthinellum* *Trichoderma viride*	Anti-fungal activity	(Dal Bello et al., 1997; Chahal et al., 2014 and JayaMadhuri et al., 2020)	Produced during respiration by all animals, fungi, and microorganisms that depend directly or indirectly on living or decaying plants for food. - solvent, - Vasodilator agent, - Anaesthetic, - Antagonist, - Member of greenhouse gas, - Human metabolite, - Member of food packaging gas, - Food propellant, - Refrigerant, - Saccharomyces cerevisiae metabolite, an Escherichia coli metabolite, and a mouse metabolite. (481 items)
Toluene	*T. harzianum FA1132* *T. atroviride CCM F536* *B. axarquiensis-ESR 7,* *B. subtilis-ESR 24* & *B. licheniformis-ESR 26*		(Siddiquee, 2014 and Jayakumar et al., 2021)	*Basella alba, Zingiber mioga*, and other organisms. Used in veterinary medicine to treat various parasites in dogs and cats. (105 items)

Most of the published research on fungal VOCs has been driven by economic interests. For instance, food and flavor chemists have analyzed VOCs in fungi for their taste properties. Agronomists utilize these compounds as indicators of mold damage to crops. Building scientists have utilized fungal VOCs as indicators of hidden mold growth in water-damaged buildings. Entomologists study them as chemical cues that either attract or repel certain insect species. Mycologists, on the other hand, describe them as spore inhibitors and signals of fungal development. Phytopathologists consider them to be stress metabolites (Note: For each of these topics, specific bibliographical references are provided in the following sections). Olfaction and aroma: Fungal volatiles contribute to the desirable flavor properties of certain cheeses, sausages, beverages, Asian food products, etc. Consequently, odor analysis has been used to control the quality of these fermented foods (Kinderlerer, 1989 and Bruna et al., 2001). Similarly, the VOCs profiles of gourmet macrofungi (chanterelles and truffles) were analyzed (Fraatz & Zorn, 2011). Many fungal VOCs are chemically identical to desirable plant products and are classified as "bioidentical" natural flavor ingredients, offering a wide range of possibilities in the food industry (Lomascolo et al., 1999). A famous example is the production of 6-pentyl-α-pyrone, a lactone with a characteristic coconut odor, by certain *Trichoderma* species (Prapulla et al., 1992). Malodors as indicators of spoilage: Microbial metabolism is primary cause for undesirable flavors and odors in feeds and foodstuffs, which can serve as indirect markers of contamination (Schnürer et al., 1999). For instance, in a jam factory, VOCs have been used to detect spoilage (Nieminen et al., 2008) and on bakery products (Keshri et al., 2002). In addition, VOCs sampling has been used to monitor fungal contamination of stored cereals (Jeleń & Wasowicz, 1998). Indoor molds also produce VOCs, and their detecting provides a nondestructive method of locating mold within buildings (Matysik et al., 2008). Fingerprinting and chemotaxonomy: Fungal VOCs have the potential to be used in chemotaxonomy for distinguishing between fungal spaces based on their odors. The VOCs signatures of the virulent and avirulent entomopathogenic species *Metarrhizium anisopliae* and *Beauveria bassiana* showed consistent profiles (Hussain et al., 2010). Fungi belonging to distinct functional groups (ectomycorrhizal, pathogenic, and saprophytic) were found to have distinct odorant profiles, especially in the pattern of sesquiterpenes. These profiles could be used to predict members of various ecological groups (Muller et al., 2013). Biofilters and biodiesel: Fungi can produce a wide range of volatile compounds and can also metabolize them. As a result, they have been effectively integrated into biofilters to degrade volatile contaminants, offering an efficient method for air purification and environmental management (VergaraFernandez et al., 2011). Since fungi are known to use plant biomass, it has been suggested that they could be used to produce compounds that resemble diesel, which are referred to as "biodiesel" or "mycodiesel" (Grigoriev et al., 2011). For instance, a variety of *Ascocoryne* species produced VOCs mixtures, some of which resemble the target molecules for biofuel. These mixtures included alkanes, alkenes, alcohols, ester, ketones, acids, benzene derivatives, and terpenes (Mallette et al., 2014). Disease detection: Disease can be detected by odorants (Casalinuovo et al., 2006) and have been used by plant pathologists and medical mycologists. The powdery mildew fungus *Uncinula necator*, which causes severe infection in vineyards, is a case in point in the plant world. From diseased grapes, Darriet et al. (2002) identified several unique odorants, such as 1-octen-3-one (mushroom-like), (Z)-1,5 octadien-3-one (geranium-like), and an unspecified fishy odor. The common mold *Aspergillus fumigatus*, which can result in invasive pulmonary aspergillosis, a condition linked to high mortality rate in immunocompromised patients, is an example from the realm of human disease. *Aspergillus fumigatus* produces farnesene when cultured in vitro, and terpene volatiles have been proposed as a potential tool for the early detection of invasive aspergillosis (Heddergott et al., 2014). Patients with *Aspergillus fumigatus* infections had the compound 2-pentylfuran found in their breath (Chambers et al., 2009). Research on fungal VOCs has been conducted across several academic disciplines. Scientists from diverse fields, including analytical chemistry, developmental mycology, food and flavor research, entomology, olfaction, perfumery, and toxicology, have performed valuable studies. Collaboration among experts from these areas will enable a comprehensive understanding and characterization of the crucial role that gas-phase biogenic molecules play in various fields.

CONCLUSION

In this study, the volatile profiles of four *Trichoderma* species [*T. orientale*, *T. asperellum* (1), *Trichoderma* sp. (3), and *T. asperellum* (2)] isolated from saffron bulbs and compost, were analyzed using GC-MS. Our finding revealed significant variations in the volatile mixtures generated by each *Trichoderma* species, with 66 VOCs belonging to various chemical families. The existence of specific volatile profiles emitted by *Trichoderma* spp., contributing to their unique VOCs signatures. Notably, some of these VOCs were detected for the first time in Moroccan *Trichoderma* species and have not previously been identified in any fungi. Previous studies have mentioned VOCs without specifying their applications, and some have identified these compounds in organisms other than *Trichoderma* spp. A comparison of bibliographic data with other living organisms revealed several potential biotechnological applications, hence the necessity to conduct further research to investigate their potential use in agriculture and biotechnology. As a result, this research has provided valuable information on the production of VOCs by different Moroccan strains of *Trichoderma* and highlights the diversity and novelty of the compounds produced by these fungi. It would be valuable to further investigate the roles and potential applications of Benzenemethanol, α-methyl; β-Terpinen; 2-Octanol, acetate; Acetaldehyde, hydroxy; and 2-Propanone, 1-hydroxy. Additional functional studies on these new VOCs, particularly in biocontrol, could yield promising results. A better understanding of the capabilities of these VOCs, can unlock new opportunities for sustainable agriculture and promote a healthier, more diverse ecosystem.

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