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Gas chromatography—ion mobility spectrometry-based fingerprint analysis of volatile flavor compounds in ginger cultivated under different conditions

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ABSTRACT

Ginger is widely acclaimed for its pungent aroma, nutritional benefits, and unique pharmacological properties, making it essential in culinary and medicinal applications. This study investigates volatile flavor profile differences in ginger resulting from various cultivation practices. Gas chromatography–ion mobility spectrometry (GC–IMS) was utilized to isolate and identify volatile compounds. Subsequent analyses, including relative odor activity values (ROAV) and multivariate statistical analysis, precisely identified key flavor compounds differentiating organically cultivated ginger from conventional field-grown varieties. A total of fifty-six volatile compounds were identified, comprising 17 esters, 4 alcohols, 7 ketones, 18 terpenoids, 6 aldehydes, and 4 miscellaneous compounds, with esters and terpenoids constituting over 50 % of total volatiles. Compounds such as α -phellandrene, β -citronellal, butyl 2-propenoate, 2-heptanone-D, and 3-octanone predominantly contributed lemon, banana, and citronella notes in organically cultivated ginger. In contrast, citral dominated in conventional ginger. This research significantly advances our understanding of ginger's aroma under varied cultivation conditions and demonstrates GC–IMS's utility in effectively profiling ginger flavor, thereby guiding improved cultivation and management.

1. Introduction

Ginger (Zingiber officinale Roscoe), a member of the Zingiberaceae family, has been cultivated globally for over 2000 years for its medicinal and culinary properties (Nicoll and Henein, 2009). Key ginger-producing regions include China, India, Nigeria, Australia, Jamaica, and Haiti (Bartley and Jacobs, 2000). Notably, China dominates as the primary producer and exporter, contributing more than 50 % to the global export market, with significant production from provinces like Shandong, Hebei, Liaoning, and Fujian (Han et al., 2022). Ginger is rich in proteins, carbohydrates, dietary fiber, and bioactive components such as gingerol and terpenes, traditionally employed for therapeutic purposes, including treatment of colds, nausea, and headaches (Kiyama, 2020; Mao et al., 2019; Shahrajabian et al., 2019). Its distinctive pungent aroma and flavor profoundly impact consumer preferences and its diverse use in the food and beverage industry (Ding et al., 2012; Li et al., 2021).

Traditional soil-based cultivation methods for ginger encounters challenges such as nematodes and pathogens, adversely affecting crop yield and quality (Wang et al., 2022; Yanagawa et al., 2022). Organic

production systems provide effective coping strategies to meet this challenge (Verma et al., 2020). This systems use materials of organic origin (such as plants, animals, and fertilizers) and promote the application of compost, fertilizers, organic waste, crop rotation, pest control, etc., through biological measures. It has been shown that improved soil structure because of better aggregation in organically amended soil will help to open up soil pores and channels in fine-textured soil, and consequently it helps in better growth of the crops (Řezáčová et al., 2021). Organic fertilizers help in keeping balance in carbon and nitrogen (C/N) ratio of the soil and also enhance the soil fertility and productivity. Due to more biological activities in soil, the nutrients that are in the lower depths are made available to the plants (Rasul et al., 2015). Organic fertilizers have nearly all the vital plant nutrients required for plant growth and produce other non-nutrient benefits also by providing foods for soil microbes, different growth-promoting organic acids, improving soil structure, water holding capacity, etc (Verma et al., 2020).

Organically cultivated ginger is characterized by higher productivity, enhanced ginger flavor, and a richer nutritional composition compared to conventionally grown ginger. Research indicates that

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Fig. 1. Organic farming practices (A) and field farming practices (B).

organic cultivation significantly increases ginger yield, dry matter content, and oleoresin concentration (Verma et al., 2019). The characteristic volatile flavor compounds in ginger are critical sensory indicators that substantially influence consumer purchasing decisions (Vedashree et al., 2020). and play a vital role in ginger's deep processing and comprehensive utilization (Okonkwo et al., 2024). Consequently, developing rapid, precise analytical methods for evaluating the volatile flavor quality of organic ginger and identifying characteristic flavor compounds is crucial for quality assessment.

Among analytical methods employed to detect volatile aroma compounds in food, electronic nose technology, gas chromatography-mass spectrometry (GC-MS), and gas chromatography-olfactometry-mass spectrometry (GC-O-MS) are commonly used. Although GC-MS is robust and widely adopted, it typically requires extensive sample pretreatment, which can be time-consuming and complex due to the inherent complexity of food matrices. (Gu et al., 2021). Gas chromatography-ion mobility spectrometry (GC-IMS) has recently emerged as an innovative analytical technique, combining the powerful separation capabilities of GC with the rapid detection afforded by ion mobility spectrometry (IMS) (Wang et al., 2020). Equipped with an automatic headspace injector and integrated databases for automated data acquisition, GC-IMS facilitates rapid, sensitive analysis of volatile compounds in liquid and solid samples without extensive preparation (Wang et al., 2020). GC-IMS has been successfully applied to flavor analysis and quality evaluation in diverse food products, including honey (Wang et al., 2021), rice (Gu et al., 2020), wheat grains (Jin et al., 2023), and onion (Jin et al., 2024). However, its application in profiling volatile flavor compounds of ginger under different conditions remains insufficiently unexplored.

In this study, we employed GC–IMS along with advanced multivariate statistical methods (PCA and OPLS-DA) to delineate the difference in volatile flavor profiles and aroma composition between organically and conventionally cultivated ginger. Fresh ginger samples were utilized to compare the flavor changes induced by these cultivation practices. Advanced statistical tools, including principal component analysis (PCA) and partial least squares discriminant analysis (OPLS-DA), were applied to identify potential differential flavor components in ginger samples. Additionally, a GC–IMS library was developed to facilitate a comprehensive understanding of characteristic flavor profiles in ginger cultivated under varied cultivation conditions, thereby providing a theoretical framework for quality evaluation, optimization of cultivation methods, and targeted ginger breeding strategies.

2. Materials and methods

2.1. Materials

The experimental material was 'Jinchang Ginger' (*Zingiber officinale* Roscoe), a Shandong Mianjiang variety characterized by fewer rhizome branches, larger rhizomes, fewer and sparser nodes, and superior commercial quality. All accessions were cultivated in the greenhouse located

in Weifang, Shandong, China (119°18′E, 36°16′N).

Two cultivation methods, field planting (PT) and organic planting (YJ), were employed (Fig. 1). For organic planting (YJ), a nutrient substrate was prepared by mixing pine tower powder from Changbai Mountain with red pine wood powder in a 1:3 ratio, followed by autoclaving and fermentation. The substrate mixture was packed into individual planting bags (27 cm \times 27 cm \times 37 cm; wall thickness of 2 mm) and arranged on waterproof platforms within standardized large spring and fall shed. Each planting bag contained two ginger plants, resulting a planting density equivalent to 2700 substrate bags per hectare or 5400 plants per hectare. In contrast, field planting (PT) utilized fertile sandy soil within a 6-m arched trellis system. Plants were spaced 20 cm apart within rows, with 75 cm spacing between rows, resulting in a planting density of 4500 plants per mu (~67,500 plants per hectare). The planting period spanned annually from April to October in both 2022 and 2023. Healthy ginger free from any deterioration, decay or pest infestation were harvested each October. Sampling involved randomly selecting three plots of 7.5 m² per treatment, from which three plants per plot were collected. To mitigate metabolic differences between different rhizome sections, samples were consistently taken from the middle portion of the ginger rhizomes. Following harvest, samples from each treatment were promptly sealed in plastic bags, transported to the laboratory, washed thoroughly with deionized water, and air-dried at room temperature.

2.2. Sample preparation

Fresh ginger samples were sliced into small pieces, approximately 2 \times 2 mm in size. Subsequently, 1 g of ginger slices was weighed into a 20 mL sealed headspace vial. Each experiment were conducted in triplicate. The samples were then incubated at 50 $^{\circ}\text{C}$ for 15 min with shaking at 500 rpm before injection.

2.3. Ion mobility spectrometry

Volatile compounds in ginger samples from PT and YJ treatments were analyzed using a GC-IMS system according to the method described by Li et al. (2019), with minor modifications. The analysis was conducted using a FlavourSpec® GC-IMS system (G.A.S., Dortmund, Germany) equipped with a PAL RSI autosampler unit (CTC Analytics AG, Zwingen, Switzerland). Following incubation, 500 µL of headspace gas was automatically injected into the system injector (operating at 85 °C in splitless mode) using a heated syringe. Volatile compounds were initially separated using a Restek MXT-WAX column (0.53 mm × 15 m, $1~\mu\text{m})$ at a constant temperature of 60 °C, followed by secondary separation within the ion mobility spectrometry drift tube. High-purity nitrogen (99.999 %) served as both the carrier gas and drift gas, maintained at a drift tube temperature of 45 $^{\circ}\text{C}$ and a drift gas flow rate of 75 mL/min. The total analytical duration was 55 min, adhering to a specific carrier gas program: 2 mL/min (0-2 min), ramped from 2 to 10 mL/min (2-10 min), from 10 to 100 mL/min (10-30 min), and

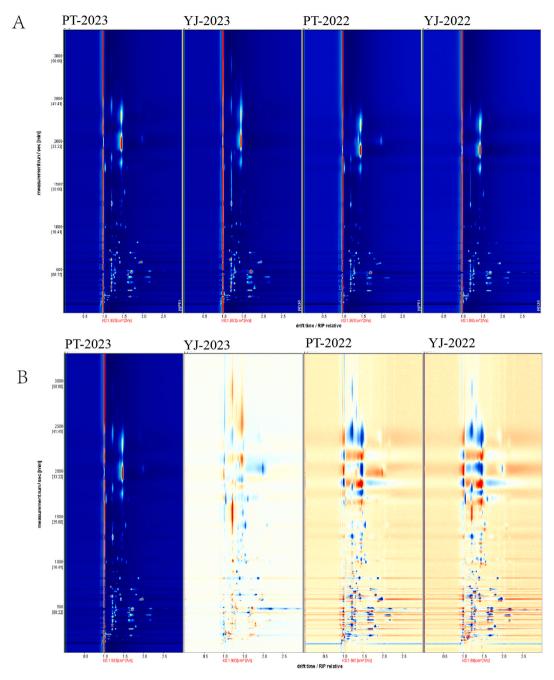


Fig. 2. (A) Two-dimensional (2D) chromatogram of the ginger samples in HS-GC-IMS. (B) Discrepancy spectrum of the ginger samples in HS-GC-IMS.

maintained at 100 mL/min (30-55 min).

N-Ketones C4-C9 (Shanghai Aladdin Biochemical Technology Co., Ltd., China) were utilized as external references to calculate the retention index (RI) of the volatile compounds. The compounds were identified by comparing their RI and drift time against a pre-established GC–IMS reference library. Quantification of volatile compounds was achieved using standard curves, with the relative content expressed as the peak volume.

2.4. Relative odor average value

The relative odor average value (ROAV) for each compound was determined based on the relative content and sensory threshold according to the methods outlined by Zhu et al. (2020). ROAV indicates the relative contribution of each volatile compound to the overall aroma contributors. Compounds with ROAV $\geq \! 1$ are considered significant

aroma contributors (Zhang et al., 2022). The compound with the highest contribution in the sample was assigned an ROAV of 100, and ROAVs of other compounds were calculated using the following equation:

$$ROVA = \frac{C_i}{C_{max}} \times \frac{T_{max}}{T_i} \times 100$$

Where C_i and T_i represent the relative mass fraction (%) and sensory threshold of each compound, respectively, and C_{max} and T_{max} denote the relative mass fraction (%) and sensory threshold of the most impactful ingredient, respectively.

2.5. Statistical analysis of data

Data were presented as mean \pm standard deviation. Statistical analysis was carried out using a T-test in Microsoft Excel (2016), and

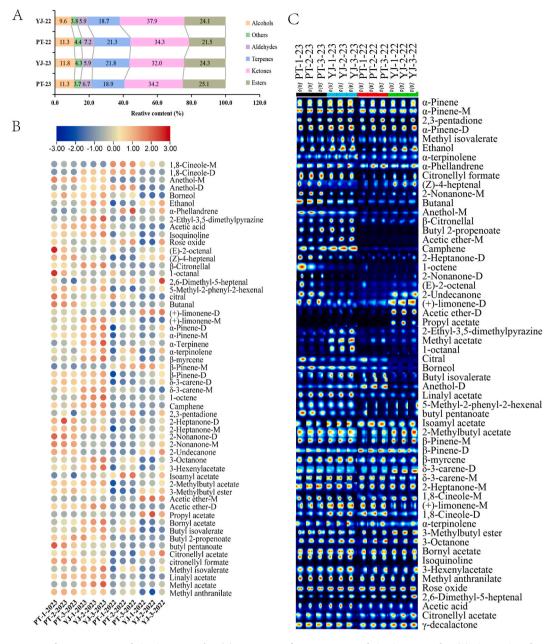


Fig. 3. (A) The percentage of aroma compounds in ginger samples. (B) Heat maps of aroma compounds in ginger samples. (C) Fingerprint of aroma compounds in ginger samples.

differences were considered statistically significant at P < 0.05.

3. Results

3.1. Comparative analysis of volatile compounds using GC-IMS

The volatile aroma components in four ginger samples (PT-22, PT-23, YJ-22, and YJ-23) were analyzed using headspace gas chromatography–ion mobility spectrometry (HS-GC–IMS). Fig. 2A illustrates a two-dimensional vertical spectrum, where the drift time is displayed along the x-axis and retention time along the y-axis, with Ko values ranging between 1.980 and 1.981 $\rm cm^2/V_S$. The normalization of drift times was achieved relative to the reactant ion peak (RIP), which served as a standard reference. Each point appearing to the right of the RIP represents a detected volatile organic compound. Color intensity, varying from white (indicating lower concentration) to red (indicating higher concentration), visualizes differences in volatile compound abundance.

The analysis indicated a comparable distribution of volatile composition in both organic (YJ) and conventional (PT) ginger samples, predominantly clustering at retention times between 200 to 750 s and drift times ranging from 1.0 to 2.3 s. This similarity suggests relatively consistent volatile composition profile across different cultivation methods.

To better elucidate differences attributable to cultivation conditions, the PT-23 spectrum served as a reference (Fig. 2B). Subsequently, difference spectra were generated by subtracting the reference spectrum, highlighting specific alterations in volatile compound concentrations. The color gradient of difference spectra, ranging from blue (reduced concentration) to red (increased concentration), clearly illustrates distinct fluctuations in the concentrations and distribution patterns of volatile substances between organic and conventional ginger samples. These visual differences underscore notable variations in the volatile composition influenced by distinct cultivation conditions, as evidenced by alterations in the positions, numbers, and intensities of detected peaks.

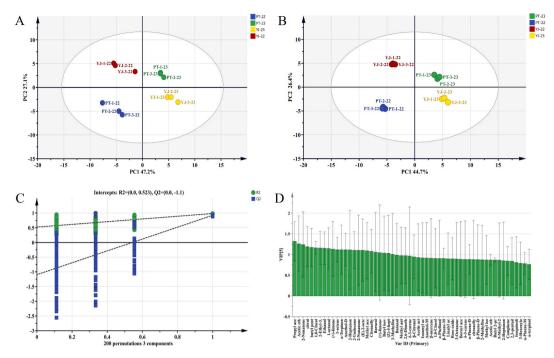


Fig. 4. Principal component analysis (A), orthogonal partial least squares-discriminant analysis (B), permutation (C) and VIP Plot of Potential Volatile Compounds (D) in gingers.

3.2. Volatile compounds identified by HS-GC-IMS

Fig. 3A illustrates the relative percentage content of volatile aroma compounds in ginger, highlighting esters as primary contributors to ginger's fruity, floral, and sweet flavors. Notably, the ester content was higher in organic (YJ) ginger compared to conventional (PT) ginger in 2022, while this pattern reversed in 2023. The heatmap (Fig. 3B) details the distinctive ester profiles of ginger samples, revealing that YJ ginger predominantly contained esters such as 2-methylbutyl acetate, acetic ether-M, acetic ether-D, propyl acetate, and butyl 2-propenoate, contributing significantly to fruity and floral notes. In contrast, PT ginger samples primarily comprised methyl anthranilate and butyl pentanoate. The inconsistent patterns observed among other esters over the two-year study suggest a complex interaction of volatile compounds, potentially influenced by environmental variable like climatic conditions, soil conditions, and microbial composition (Das et al., 2020; Rasul et al., 2015)

Alcohol compound concentrations were relatively stable across all samples, varying slightly between 14.1 % and 16.2 %, as illustrated in Fig. 3A. While PT ginger showed minor year-to-year variability, YJ ginger exhibited an increase in alcohol content. The alcohols identified, including anethol-M, anethol-D, borneol, and ethanol, play a substantial role in the aromatic profile of ginger.

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potentially influenced by environmental variable like climatic conditions (Soundar Raju et al., 2015). Moreover, citral, 1,8-cineole, β -linalool, and bornyl acetate have previously been reported as crucial odorants in fresh ginger (Zhang et al., 2021).

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Aldehyde content remained consistent across PT samples during the two years but exhibited a decreasing trend in YJ ginger. The primary aldehydes identified include (E)-2-octenal, (Z)-4-heptenal, 1-octanal, 2,6-dimethyl-5-heptenal, butanal, and 5-methyl-2-phenyl-2-hexenal. These compounds contribute buttery, lemon, citronella, and cocoa aromas and a pungent odor to ginger.

Fingerprints analysis (Fig. 3C) visually summarizes the variation of volatile organic compounds under different cultivation conditions. Notably, in the red area I, the YJ-22 samples demonstrated higher concentrations of volatile compounds such as 2-undecanone, (+)-limonene-D, acetic ether-D, propyl acetate, methyl acetate, 1-octanal, and 2-ethyl-3,5-dimethylpyrazine relative to the PT samples. These findings highlight significant influences of cultivation practices and cultivation years on the accumulation and composition of aroma-active substances in ginger.

3.4. Relative odor activity values (ROAV) of volatile compounds

The overall aroma contribution of individual volatile compounds in ginger is influenced by their concentration as well as their odor thresholds, quantified through relative odor activity values (ROAV). Compounds exhibiting high ROAV values are considered significant contributors to ginger aroma profile (Eduardo et al., 2010). The ROAV measures a compound's contribution to the food aroma system, emphasizing the role of both concentration and sensory threshold in defining its overall impact. In this study, out of the 56 identified volatile compounds (Table 2), three exhibited an ROAV ≥ 1 , indicating substantial influence on ginger's aromatic characteristics. These prominent

 Table 1

 Identification of volatile compounds in the ginger samples (PT and YJ) based on GC-IMS.

Number	Compounds	CAS#	RI	DT	Relative content				VIP	Flavor Description
					PT-23	YJ-23	PT-22	YJ-22		
Alcohols										
1	Anethol-M	C104461	1814.5	1.22466	$22037.9 \pm 1979.27a$	$17622.48 \pm 204.89 \ b$	$7542.55 \pm 453.39c$	$10592.46 \pm 1003.08 \ d$	1.115	\
2	Anethol-D	C104461	1266.7	1.21537	$2609.36 \pm 74.94a$	$3966.71 \pm 223.91 \text{ b}$	$5060.84 \pm 273.87c$	$1550.68 \pm 166.33 \ d$	0.917	\
3	Borneol	C507700	1704	1.22198	$11,491 \pm 1102.86a$	$11769.42 \pm 773.24a$	$9579.64 \pm 698.32 \text{ b}$	$5749.64 \pm 307.5c$	1.060	camphor
4 Others	Ethanol	C64175	933.9	1.14354	$6913.62 \pm 364.77 \ b$	$11022.01 \pm 1062.53 a$	$4110.36 \pm 1468.67c$	$9874.53 \pm 1092.96a$	1.154	sweet
5	2-Ethyl-3,5-dimethylpyrazine	C13925070	1476.6	1.22955	$1661.02 \pm 151.13~b$	$3843.18 \pm 490.07a$	$1035.62 \pm 229.16c$	$1625.95 \pm 21.9 \text{ b}$	1.160	\
5	Acetic acid	C64197	1472.4	1.0498	$3124.94 \pm 80.24a$	$3189.12 \pm 12.19a$	$2896.73 \pm 63.25 \text{ b}$	$2759.5 \pm 21.99c$	0.931	sour
7	Isoquinoline	C119653	1291.4	1.18388	$8087.85 \pm 432.98 \text{ ab}$	$9079.27 \pm 549.69a$	$6765.08 \pm 1093.09 bc$	$5988 \pm 949.15c$	0.810	herbal, benzaldehy
8	Rose oxide	C16409431	1336.3	1.35225	$4456 \pm 77.32a$	$4448.31 \pm 44.58a$	$4571.47 \pm 121.69a$	$4336.76 \pm 263.91a$	0.899	powerful geranium
Aldehydes										-
9	(E)-2-octenal	C2548870	1430	1.3328	$786.03 \pm 355.58a$	$325.43 \pm 8.73 \text{ b}$	$383.52 \pm 182.07 \text{ b}$	$491.97 \pm 37.94 \text{ ab}$	1.104	Fatty
10	(Z)-4-heptenal	C6728310	1220	1.6298	$3292.16 \pm 116.02a$	$2472.98 \pm 100.81c$	$1536.33 \pm 145.05 d$	$2903.41 \pm 196.9 \mathrm{b}$	1.032	buttery flavor
11	1-octanal	C124130	1299.5	1.82	$1540.9 \pm 812.92a$	$442.32 \pm 25.62 \text{ ab}$	$604.84 \pm 571.51 \text{ b}$	$381.53 \pm 156.48 \text{ b}$	1.147	citrus, honey
12	2,6-Dimethyl-5-heptenal	C106729	1347.4	1.17635	$5521.98 \pm 1873.15a$	$7765.75 \pm 1441.29a$	$8499.95 \pm 1643.01a$	$9311.54 \pm 4005.1a$	0.974	melon
13	5-Methyl-2-phenyl-2-hexenal	C21834924	1483.6	1.45244	$3009.24 \pm 171.31a$	$3058.03 \pm 161.71a$	$1827.67 \pm 162.65c$	$2474.88 \pm 292.57 \text{ b}$	0.867	cocoa aroma
14	Butanal	C123728	836.1	1.11944	$3710.88 \pm 264.21a$	$2477.01 \pm 45.44 \text{ b}$	$1329.51 \pm 181.54c$	$1637.03 \pm 303.3c$	0.985	pungent odor
Terpenes										1. 0.
15	1,8-Cineole-M	C470826	1194.8	1.29865	$4311.49 \pm 96.09c$	$4137.72 \pm 43.42c$	$5492.87 \pm 15.03a$	$4779.58 \pm 179.84 \text{ b}$	1.162	mint, sweet
16	1,8-Cineole-D	C470826	1217.9	1.2984	$4900.01 \pm 174.19c$	7424.22 ± 85.48 b	$7998.7 \pm 46.04a$	4909.61 ± 111.68c	0.910	mint, sweet
17	β-Citronellal	C106230	1489.9	1.3466	$3928.76 \pm 284.38 \mathrm{b}$	$5001.89 \pm 151.85a$	$3011 \pm 67.91c$	$3039.99 \pm 349.44c$	0.947	lemon-, citronella
18	citral	C5392405	1679.4	1.05574	$9095.98 \pm 3320.06a$	$6423.14 \pm 421.25 \text{ ab}$	$8223.49 \pm 2142.3a$	2956.46 ± 88.62 b	1.191	lemon
19	(+)-limonene-D	C138863	1218.2	1.21537	2580.21 ± 80.44 b	$2135.89 \pm 48.55c$	$1926.78 \pm 221.63c$	$3664.2 \pm 113.86a$	1.044	lemon, orange
20	(+)-limonene-M	C138863	1205.3	1.71883	17666.62 ± 847.96 b	$23750.97 \pm 469.48a$	18509.52 ± 205.82 b	$11217.59 \pm 215.2c$	1.132	lemon, orange
20 21	α-Phellandrene	C99832	1182.7	1.21652	2726.72 ± 48.64 b	$2929.51 \pm 34.96 \text{ ab}$	$3250.83 \pm 398.29a$	2974.76 ± 250.23 ab	0.908	fresh, peppery
22	α-Pinene-D	C80568	1026.1	1.66509	$7642.26 \pm 5.34 \text{ ab}$	$8882.06 \pm 415.43a$	6762.41 ± 963.06 b	$7392.83 \pm 816.62 \text{ b}$	0.884	pine
23	α-Pinene-M	C80568	1020.1	1.29791	$5817 \pm 19.51a$	$6200.01 \pm 97.49a$	5122.9 ± 445.64 b	5225.47 ± 391.9 b	0.783	pine
23 24	α-Terpinene	C99865	1198.8	1.21579	1646.69 ± 148.96 b	$2061.88 \pm 132.25a$	1505.24 ± 56.34 b	1651.15 ± 67.96 b	1.117	woody, lemon odo
2 4 25	α-terpinelee α-terpinolene	C586629	1251.4	1.21274	$3404.43 \pm 153.02a$	$3831.57 \pm 212.47a$	$3489.3 \pm 637.89a$	$3269.73 \pm 534.76a$	0.763	lilac
25 26	*		1151.8	1.212/4		$7247.07 \pm 259.73a$		$5600.67 \pm 328.69c$	0.763	
	β-myrcene	C123353			6181.59 ± 99.51 b		6134.87 ± 392.84bc			sweet, balsamic
27	β-Pinene-M	C127913	1122.5	1.21842	$8068.73 \pm 217.5a$	$8237.57 \pm 80.08a$	9334.9 ± 400.37 b	9528.71 ± 358.71 b	0.878	woody or resinous
28	β-Pinene-D	C127913	1123.5	1.63042	$6416.43 \pm 128.45 \text{ ab}$	$6904.28 \pm 293.73a$	$3778.49 \pm 805.14c$	5444.33 ± 584.89 b	0.908	woody or resinous
29	δ-3-carene-D	C13466789	1164.9	1.67615	$13001.6 \pm 220.71a$	$13157.75 \pm 436.31a$	9015.52 ± 644.85 b	$11480.22 \pm 920.26c$	0.886	lemon, resin
30	δ-3-carene-M	C13466789	1163.8	1.28736	4366.4 ± 253.96 b	$5055.38 \pm 45.83a$	$4112.91 \pm 90.72 \text{ b}$	$3488.56 \pm 76.15c$	0.966	lemon, resin
31	1-octene	C111660	836.9	1.15652	$317.17 \pm 6.12 \mathrm{b}$	$1107.27 \pm 139.43a$	$114.26 \pm 19.83 \text{ b}$	$271.89 \pm 67.31c$	1.120	gasoline
32	Camphene	C79925	1089.1	1.21188	$7757.02 \pm 2270.92 \text{ b}$	$11874.59 \pm 689.05a$	$1745.87 \pm 472.65c$	$1696.87 \pm 333.63c$	0.844	camphor
Ketones										
33	2,3-pentadione	C600146	1062.8	1.20934	$5996 \pm 664.06a$	$7001.39 \pm 450.12a$	$6806.95 \pm 929.21a$	$6113.07 \pm 870.38a$	1.114	sweet odor
34	2-Heptanone-D	C110430	1185.6	1.62291	$5955.82 \pm 934.09a$	$3544.73 \pm 245.07 \text{ b}$	$2420.87 \pm 302.06 \text{ b}$	$3305.74 \pm 610.49 \text{ b}$	0.854	fruity, banana
35	2-Heptanone-M	C110430	1183.9	1.25993	$2700.93 \pm 39.65a$	$2680.38 \pm 16.12a$	$2215.9 \pm 121.02c$	$2473.68 \pm 73.38 \ b$	1.243	fruity, banana
36	2-Nonanone-D	C821556	1397.9	1.88039	$3514.19 \pm 556.43a$	$1040.27 \pm 21.19 \ b$	$830.19 \pm 71.89 \text{ b}$	$1436.47 \pm 485.15 \ b$	1.105	hot milk, green
37	2-Nonanone-M	C821556	1397.2	1.40412	$5206.65 \pm 248.13a$	$2833.92 \pm 93.21 \ b$	$1958.33 \pm 256.42c$	$3434.77 \pm 722.83 \ b$	1.107	hot milk, green
38	2-Undecanone	C112129	1604.7	1.54641	$5352.5 \pm 357.12a$	$2195.63 \pm 157.7 \; b$	$1294.63 \pm 51.4c$	$5142.45 \pm 827.02a$	0.894	peach
39 Esters	3-Octanone	C106683	1289.5	1.30669	$2515.12 \pm 32.66 \ b$	$2941.84 \pm 156.59a$	2504.98 \pm 219.12 b	2188.13 \pm 232.97 b		herb, butter, resin
40	3-Hexenylacetate	C3681718	1323.7	1.30867	$2413 \pm 88.65 \text{ ab}$	$2805.06 \pm 194.68a$	$2160.12 \pm 323.09 \ b$	$2116.29 \pm 309.77 \; b$	0.789	green, fruity
41	Isoamyl acetate	C123922	1106	1.30561	$1842.8 \pm 325.75 \ b$	$2214.1 \pm 68.96 \text{ b}$	$2940.18 \pm 378.57a$	$2336.56 \pm 337.95 \text{ b}$	0.923	banana
42	2-Methylbutyl acetate	C624419	1122.5	1.7198	$3886.06 \pm 167.05a$	$4228.95 \pm 100.56a$	$2083.48 \pm 464.04c$	$3167.35 \pm 366.76 \text{ b}$	0.876	apple, banana
43	3-Methylbutyl ester	C27625350	1275.9	1.42949	$6477.71 \pm 160.53a$	6037.87 ± 75.53 b	$4413.47 \pm 218.56c$	$6272.93 \pm 218.95 \text{ ab}$	1.006	has a fruity odor
44	Acetic ether-M	C141786	889.3	1.09719	1020.92 ± 77.78 bc	1442.01 ± 179.46 b	$776.32 \pm 204.01c$	$5709.45 \pm 514.28a$	1.262	pineapple
45	Acetic ether-D	C141786	894	1.32525	805.34 ± 19.04 b	$1127.94 \pm 164.06a$	$99.33 \pm 6.67c$	$111.35 \pm 16.58c$	0.870	pineapple
46	Propyl acetate	C109604	976.3	1.16209	245.9 ± 28.9 b	492.06 ± 86.41 b	135.04 ± 11.63 b	$2462.84 \pm 565.08a$	1.324	fruity (pear–raspbe
	pji uccuic	0107007	270.3	1.10207	2 10.7 ± 20.7 D	172.00 ± 00.71 D	100.0 1 ± 11.00 D	2 102.01 ± 303.00d	1.027	Tarry (Pear-183PDE

Table 1 (continued)	ontinued)									
Number	Jumber Compounds	CAS#	RI	DT	Relative content				VIP	Flavor Description
					PT-23	YJ-23	PT-22	YJ-22		
47	Bornyl acetate	C76493	1289	1.21557	7439.55 ± 264.81 b	8334.12 \pm 245.55a	7202.88 ± 348.1 bc	$6665.54 \pm 445.71c$	0.888	balsamic
48	Butyl isovalerate	C109193	1270.2	1.90883	$3007.32 \pm 168.08 \ \mathrm{b}$	$4090.69 \pm 271.38a$	$3555.13 \pm 699 \text{ ab}$	$1648.67 \pm 181.05c$	1.037	apple
49	Butyl 2-propenoate	C141322	917.6	1.66456	$7650.79 \pm 1638.02a$	$8156.48 \pm 2021.29a$	$617.04 \pm 81.24 \mathrm{b}$	$720.05 \pm 30.16\mathrm{b}$	0.869	/
20	butyl pentanoate	C591684	1301.6	1.40969	$2717.41 \pm 592.72a$	$1460.9\pm55.57\mathrm{b}$	$925.48 \pm 458.03 \mathrm{b}$	$811.84 \pm 189.74\mathrm{b}$	1.175	apple, pineapple, green
51	Citronellyl acetate	C150845	1690.7	1.46866	$33919.17 \pm 1099.16 \text{ b}$	$31215.21 \pm 488.84c$	$25659.1 \pm 1570.11 d$	$36958.62 \pm 2030.01a$	1.080	rose, dust
52	citronellyl formate	C105851	1273.8	1.9742	15935.52 \pm 356.6a	$12407.29 \pm 168.53 \ b$	$4049.38\pm834.2c$	$7412.95 \pm 1642.65 d$	0.880	rose, dust
53	Methyl isovalerate	C556241	1020.8	1.21256	$5314.24 \pm 71.53 \text{ ab}$	$6123.58 \pm 242.3a$	$5356.12 \pm 631.2 \text{ ab}$	$5079.93 \pm 473.64c$	0.873	
54	Linalyl acetate	C115957	1567.6	1.21662	$18741.32 \pm 1375.14a$	$20414.64 \pm 768.05a$	$11343.51 \pm 1079.11 \; b$	$9164.34 \pm 514.7c$	0.900	sweet, fruit
55	Methyl acetate	C79209	831.5	1.20102	$267.13\pm19.08~\mathrm{b}$	$748.87 \pm 116.57a$	$290.78 \pm 104.52\mathrm{b}$	$258.52 \pm 71.47 \text{ b}$	1.095	pungent, apple
26	Methyl anthranilate	C134203	1335.2	1.69491	$4321.63 \pm 44.3a$	$4098.85 \pm 88.77 \text{ ab}$	$3660.2\pm237.03\mathrm{bc}$	$3147.47 \pm 506.85c$	0.974	honey, flower
Total					$335319.023 \pm 7349.3 a$	$345481.54 \pm 10427.22a$	$248573.41 \pm 15211.98 \ b$	$266409.52 \pm 21963.81 \; b$		

Notes: All values are expressed as the mean $(n = 3) \pm standard$ deviation. M: monomer; D: dimer; RI, retention index; DT, drift time; PT-22, field planting of gingers in 2022; YJ-22, organic planting of gingers in 2023; YJ-23, organic planting of gingers in 2023.

compounds included α -phellandrene (fresh, peppery scent), β -citronellal (lemon-like and citronella notes), and citral (lemon scent). Additional compounds with notable contributions (ROAV \geq 0.1) encompassed 1, 8-cineole-D and 1,8-cineole-M (mint, sweet scent), anethol-M, (Z)-4-heptenal (buttery note), 2-heptanone-D and 2-heptanone-M (fruity, banana-like), 3-octanone (butter, resinous), and butyl 2-propenoate. Collectively, these compounds significantly enhance ginger's distinctive aroma profile, providing characteristic fresh, peppery, lemon-like, citronella, minty, sweet, fruity, banana-like, and buttery aromatic notes.

Terpenes, characterized by low odor thresholds and potent aroma properties, demonstrated notably high ROAV values (Li et al., 2019). Particularly, α -Phellandrene and β -Citronellal showed higher ROAV values in YJ ginger, contributing predominant lemon-like and citronella notes. Conversely, citral exhibited a higher ROAV value in conventional (PT) ginger, contributing a strong lemon aroma. Other terpenoids were present in relatively lower concentrations and therefore had lower ROAV values, consistent with previous findings indicating aldehydes typically exhibit low threshold values but strong aromatic impact (Ying et al., 2009).

Esters, typically generated through the esterification of fatty acids with alcohols, mainly exhibit fruity and floral odors (Cao et al., 2021). Notably, butyl 2-propenoate had higher ROAV values in YJ ginger samples, contributing pungent, apple, and sweet fruit notes. Additionally, ketones such as 2-heptanone-D and 3-octanone, identified at higher concentration in organic ginger, further enhanced banana and buttery notes within ginger's complex aroma profile.

3.5. Multivariate statistical analysis

Principal Component Analysis (PCA) was utilized to differentiate ginger samples based on their volatile compound profiles, particularly to detect subtle distinctions among samples (Segelke et al., 2020). The PCA score plot presented in Fig. 4A illustrates that principal component 1 (PC1) and principal component 2 (PC2) explained 46.7 % and 25.9 % of the total variance, respectively, cumulatively accounting for 72.6 % to the variance, demonstrating sufficient explanatory power for the data set. PCA clearly categorized the ginger samples into distinct groups aligned with their respective cultivation methods and years. This distribution suggests a strong model fit and predictability for identifying ginger vintages.

Furthermore, orthogonal partial least squares discriminant analysis (OPLS-DA), a supervised discriminant analysis technique, was employed to model the relationship between volatile aromatic substances and sample categories to enhance prediction accuracy. As shown in Fig. 4B, the OPLS-DA model effectively differentiated the four groups of ginger samples, corroborating the PCA results. The fit indices were compelling, with R²X (the fit index of the independent variable) at 0.982, R²Y (the fit index of the dependent variable) at 0.989, and the predictive index of the model (Q^2) at 0.962, all suggesting that the model's results were robust and reliable. \mathbb{R}^2 and \mathbb{Q}^2 values greater than 0.5 generally indicate a good fit (Triba et al., 2015). Following 200 permutation tests (Fig. 4C), the negative y-axis intercept of the Q2 regression line statistically validated model robustness, effectively excluding overfitting risks. This statistical confirmation demonstrates the model's reliability for comparative evaluation of ginger cultivation methodologies. The OPLS-DA score plots (Fig. 4B) demonstrated classification efficacy comparable to PCA, achieving clear segregation of ginger samples into four distinct clusters. This orthogonal separation underscores the method's precision in discriminating cultivation-condition-specific metabolic signatures.

The impact of cultivation methods on ginger's aroma was measured using VIP scores from data analysis. Fig. 4D shows that 24 compounds had VIP scores above 1, meaning they greatly affected the ginger aroma. These compounds include 3 alcohols, 7 esters, 4 ketones, 3 aldehydes, 6 terpenes, and 1 other compound. The compounds that had significant

Table 2Relative thresholds for volatile compounds.

Number	Compound	CAS#	Thresholds (mg/kg)	PT-23	YJ-23	PT-22	YJ-22
Alcohols							
1	Anethol-M	C104461	0.016	0.1912	0.1612	0.0773	0.126
2	Anethol-D	C104461	0.016	0.0226	0.0363	0.0519	0.018
3	Borneol	C507700	0.52	0.0031	0.0033	0.0030	0.002
4	Ethanol	C64175	0.62	0.0015	0.0026	0.0011	0.003
Others							
5	2-Ethyl-3,5-dimethylpyrazine	C13925070	0.00004	0.0001	0.0001	0.0002	0.000
6	Acetic acid	C64197	0.013	0.0334	0.0359	0.0365	0.040
7 8	Isoquinoline Rose oxide	C119653 C16409431	0.1	0.0062	0.0065	0.0075	0.008
	rose oxide	C10409431	0.1	0.0002	0.0003	0.0073	0.006
Aldehydes 9	(E)-2-octenal	C2548870	0.003	0.0364	0.0159	0.0210	0.031
10	(Z)-4-heptenal	C6728310	0.0034	0.1344	0.1065	0.0210	0.031
11	1-octanal	C124130	0.17	0.0013	0.0004	0.0006	0.000
12	2,6-Dimethyl-5-heptenal	C106729	0.016	0.0479	0.0710	0.0871	0.110
13	5-Methyl-2-phenyl-2-hexenal	C21834924	/	0.0 17 5	\	\	\
14	Butanal	C123728	0.1	0.0052	0.0036	0.0022	0.003
Terpenes							
15	1,8-Cineole-M	C470826	0.00267	0.2242	0.2269	0.3372	0.341
16	1,8-Cineole-D	C470826	0.00267	0.2548	0.4070	0.4911	0.350
17	α-Phellandrene	C99832	2.9	5.7653	14.0644	4.2441	7.748
18	β-Citronellal	C106230	0.0004	1.3636	1.8305	1.2339	1.448
19	citral	C5392405	0.00015	8.4190	6.2682	8.9869	3.757
20	(+)-limonene-D	C138863	0.0539	0.0066	0.0058	0.0059	0.013
21	(+)-limonene-M	C138863	0.0539	0.0455	0.0645	0.0563	0.039
22	α-Pinene-D	C80568	0.1	0.0044	0.0054	0.0043	0.005
23	α-Pinene-M	C80568	0.1	0.0106	0.0130	0.0111	0.014
24	α-Terpinene	C99865	7.9	0.0037	0.0037	0.0041	0.004
25 26	α-terpinolene	C586629 C123353	0.08 0.1125	0.0000 0.0059	0.0000 0.0070	0.0000 0.0071	0.000 0.007
27	β-myrcene β-Pinene-M	C123333 C127913	0.1125	0.0039	0.0070	0.0071	0.007
28	β-Pinene-D	C127913	0.18	0.0062	0.0067	0.0085	0.009
29	δ-3-carene-D	C13466789	9.3	0.0049	0.0056	0.0034	0.010
30	δ-3-carene-M	C13466789	9.3	0.0002	0.0002	0.0002	0.000
31	1-octene	C111660	0.0046	0.0001	0.0001	0.0001	0.000
32	Camphene	C79925	1.98	0.0096	0.0352	0.0041	0.011
Ketones							
33	2,3-pentadione	C600146	0.015	0.0555	0.0683	0.0744	0.077
34	2-Heptanone-D	C110430	0.0035	0.2363	0.1483	0.1134	0.180
35	2-Heptanone-M	C110430	0.0035	0.1071	0.1121	0.1038	0.134
36	2-Nonanone-D	C821556	0.032	0.0152	0.0048	0.0043	0.008
37	2-Nonanone-M	C821556	0.032	0.0226	0.0130	0.0100	0.020
38	2-Undecanone	C112129	3	0.0002	0.0001	0.0001	0.000
39	3-Octanone	C106683	0.0013	0.2686	0.3313	0.3159	0.320
Esters	0.11	00001710	0.000	0.0070	0.0456	0.0000	0.044
40	3-Hexenylacetate	C3681718 C123922	0.009	0.0372	0.0456	0.0393	0.044
41	Isoamyl acetate		0.4685	0.0005	0.0007	0.0010	0.001
42 43	2-Methylbutyl acetate 3-Methylbutyl ester	C624419 C27625350	0.14	0.0039	0.0044	0.0024	0.004
44	Acetic ether-M	C141786	0.88	0.0002	0.0002	0.0001	0.001
45	Acetic ether-D	C141786	0.88	0.0002	0.0002	0.0001	0.001
46	Propyl acetate	C109604	1.00	0.0000	0.0001	0.0000	0.000
47	Bornyl acetate	C76493	0.44	0.0023	0.0028	0.0027	0.002
48	Butyl isovalerate	C109193	/		\	\	\
49	Butyl 2-propenoate	C141322	0.0029	0.3663	0.4117	0.0349	0.047
50	butyl pentanoate	C591684					
51	Citronellyl acetate	C150845	1.05	0.0045	0.0044	0.0040	0.006
52	citronellyl formate	C105851		\		\	\
53	Methyl isovalerate	C556241	0.011	0.0671	0.0815	0.0798	0.088
54	Linalyl acetate	C115957	0.04	0.0650	0.0747	0.0465	0.043
55	Methyl acetate	C79209	5.1	0.0000	0.0000	0.0000	0.000
56	Methyl anthranilate	C134203	0.000006	100.00	100.00	100.00	100.0

differences (P < 0.05) among the four ginger groups were mostly 14 kinds of compounds (Table 1). The distinct profiles of these compounds, particularly in YJ ginger where they are more concentrated, suggest specific volatile markers for ginger from different cultivation practices. This indicates that organic cultivation may enhance the accumulation of these key compounds. A comparative analysis was conducted on the volatile compounds in PT samples over two consecutive years. The compounds with VIP values greater than 1 included 14 compounds. In

the PT-2023 samples, only the content of 1,8-Cineole decreased, while the contents of the other compounds showed significant increases. A similar trend was observed in the YJ samples over two consecutive years.

PCA and OPLS-DA analyses showed distinct aroma profiles between YJ and PT ginger samples, meaning there are big differences between commonly and organically cultivated gingers. The OPLS-DA model helped us understand these flavor differences over the two years of the

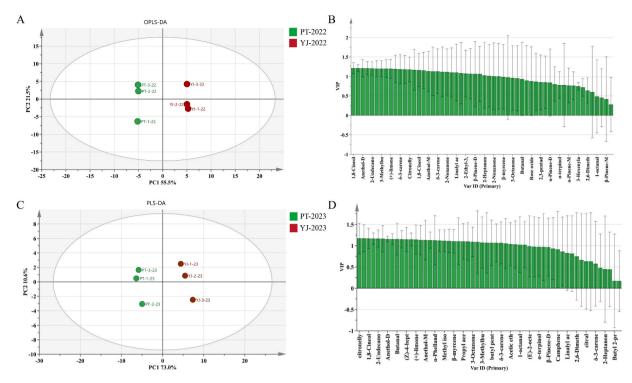


Fig. 5. (A) OPLS-DA plots for PT-22 and YJ-22 samples. (B)VIP charts of PT-22 and YJ-22 samples. (C) OPLS-DA plots for PT-23 and YJ-23 samples (D)VIP charts of PT-23 and YJ-23 samples.

study (Fig. 5). For the 2022 samples (Fig. 5A), compounds like 1,8-cineole-D, (+)-limonene-D, anethol-D, propyl acetate, 2-undecanone, 3-methylbutyl ester, (Z)-4-heptenal, and acetic ether-M were key in telling the samples apart. They mainly give sweet, lemon, peach, and pineapple flavors. Notably, compounds such as (+)-limonene-D, 2-undecanone, 3-methylbutyl ester, (Z)-4-heptenal, and acetic ether-M were found more in organic ginger, giving it a unique flavor. In the 2023 samples, the main compounds contributing to the aroma were citronellyl formate, 2-nonanone-M, 1,8-cineole-D, 1-octene, 2-undecanone, 2-nonanone-D, and anethol-D. These provided rose, green, sweet, citrus, and peach flavors. Interestingly, among the top 10 flavor compounds with VIP scores over 1.0, only methyl acetate, anethol-D, and 1-octene were higher in organic ginger. This shows volatile compound profiles can change with different cultivation conditions and years.

4. Discussion

Volatile compounds in plants are a class of lipophilic substances with low molecular weights and high vapor pressures, which can freely cross cellular membranes and be released into the surrounding environment (Pichersky et al., 2006). In this study, the volatile compounds of ginger under different cultivation methods (organic and field planting) were identified and analyzed by GC-IMS for two consecutive years. The results show that esters and terpenoids are the major volatile compounds in ginger, and organic planting can increase the relative content of these compounds. The quality, yield, and accumulation of bioactive compounds in ginger are influenced by soil, cultivation method, and other factors, and this study aligns with other studies (Das et al., 2020; R and L, 2020). These findings support the viability of organic treatments for improving ginger growth and highlight the potential for integrating sustainable practices in ginger cultivation.

In this study, the volatile compound fingerprints of ginger measured under the two cultivation methods were found to differ. A total of 56 volatile compounds were identified across all ginger samples, including 17 esters, 4 alcohols, 7 ketones, 18 terpenes, 6 aldehydes, and 4 other compounds. The ROAV values for 11 volatile compounds were greater

than 0.1. In YJ ginger, the main compounds were α -phellandrene, β-citronellal, butyl 2-propenoate, 2-heptanone-D, and 3-octanone. In PT ginger, citral was the predominant volatile compound. Some studies have shown that the biosynthesis of volatile compounds depends on the availability of carbon, nitrogen, and sulfur, as well as energy provided by primary metabolism (Dudareva et al., 2013). In this study, the differences in ginger volatile compounds were attributed to organic and field cultivation methods and their respective soil types. Furthermore, soil microbial communities play an important role in nutrient mobilization and uptake for plants (Liu et al., 2017). Ginger is a long-duration crop (7-8 months) that requires a balanced and judicious supply of different nutrients for high rhizome productivity and superior quality. Organic manures and amendments are excellent soil conditioners that supply nutrients slowly but steadily to crops over an extended period (Das et al., 2020; R and L, 2020). This slow and steady release of nutrients by organic manure over an extended duration is particularly significant for long-duration crops like ginger and tomato (Das et al., 2017). It was also observed that the volatile compounds of ginger from the same cultivation pattern varied from year to year. Various factors such as experimental methods, environmental conditions, genetic influence, ontogeny, variety, and the high-temperature climatic patterns used can cause variations in volatile compounds (Gong et al., 2023; Praveen Kumar et al., 2025; Vedashree et al., 2020).

In the present study, terpenoids were the most diverse, in line with Chen et al. (2023). Terpenes are characterized by the isoprene unit (C5) and form a large group of phytochemicals, serving as the sources of pleasant smell, spicy taste, and numerous pharmacological activities (Wink, 2010). It is worth noting that the concentration and composition of terpenoids vary among different ginger varieties, growing conditions, and processing methods (Edo et al., 2025). Therefore, the quality and sourcing of ginger influence the terpenoid content and, consequently, its therapeutic potential. Ginger has been used as a food enhancer in food processing to improve food aroma and flavor (Laelago Ersedo et al., 2023). This, combined with its aroma and flavor potential, has led to its widespread use in various food, beverage, and confectionery industries (Gaur et al., n.d.; Iara Gomes De Oliveira et al., 2024; Kumar et al.,

2018). Thus, the quality of ginger requires improvement to meet the different needs of the food and pharmaceutical industries. The food value enhancement and the bioactivity of different ginger cultivars are influenced by agroclimatic conditions and cultivar types (Praveen Kumar et al., 2025). Ginger production requires improvement through better cultivation. This study has investigated the effect of cultivation practices on the flavor quality of ginger and has provided valuable insights into the organic cultivation of ginger.

5. Conclusion

This study explored the HS-GC–IMS technology to distinguish between different ginger cultivation and management practices. Using this approach, we detected 56 volatile compounds in ginger samples. Esters and terpenes were the predominant groups, accounting for more than 50 % of the total detected volatiles. These findings were further analyzed using ROAV, PCA, and OPLS to effectively discriminate between the YJ and PT ginger cultivation practices. The principal volatile components identified in YJ and PT ginger included α -phellandrene, β -citronellal, citral, butyl 2-propenoate, 2-heptanone-D, and 3-octanone.

Thus, rapid GC–IMS technology, advanced chemometric techniques, and pattern recognition algorithms demonstrated superior capability in identifying and differentiating ginger based on cultivation methods. This approach provides a valuable strategy for confirming ginger cultivation practices, offers a comprehensive aroma profile, and furnishes crucial flavor data that can aid in managing ginger cultivation and processing ginger products. However, this study focused solely on how cultivation methods affect the accumulation of volatile compounds in ginger, without considering the impact of environmental factors such as climate conditions. This aspect will be addressed in future research.

CRediT authorship contribution statement

Qian-qian Jia: Conceptualization, Methodology, Data curation, Writing – original draft, Writing – review & editing. Jia-xing Li: Data curation, Formal analysis, Investigation, Visualization. Sen Yang: Validation, Resources, Investigation. Ding-ding Su: Supervision, Writing – review & editing, Funding acquisition, Final approval of the version to be published. All authors have read and approved the final manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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