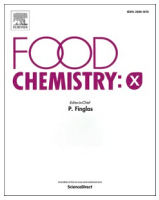
Food Chemistry: X 23 (2024) 101581

Contents lists available at ScienceDirect

Food Chemistry: X

journal homepage: www.sciencedirect.com/journal/food-chemistry-x



Characterization of key aroma-active compounds in fermented chili pepper (*Capsicum frutescens* L.) using instrumental and sensory techniques

Yue Xiao [a,b,c], Shiyao Zhang [a,b,c], Xinyu Wang [a,b,c], Xinyi Zhao [a,b,c], Zhijia Liu [a,b,c], Chuanqi Chu [a,b,c], Yanfei Wang [a,b,c], Xiaosong Hu [a,d], Junjie Yi [a,b,c,*]

[a] *Faculty of Food Science and Engineering, Kunming University of Science and Technology, Kunming 650500, China*
[b] *Yunnan Key Laboratory for Food Advanced Manufacturing, Kunming 650500, China*
[c] *International Green Food Processing Research and Development Center of Kunming City, Kunming 650500, China*
[d] *College of Food Science and Nutritional Engineering, China Agricultural University, Beijing 100083, China*

ARTICLE INFO

Keywords:
Fermented chili pepper
Aroma-active compounds
Odor activity value
Perceptual interactions

ABSTRACT

The aroma profile of fermented chili pepper was analyzed using gas chromatography–mass spectrometry (GC–MS) coupled with chromatography-olfactometry (GC-O). A total of 19 aroma-active compounds were detected, exhibiting aroma intensities spanning from 1.8 to 4.2. And 12 aroma-active compounds were determined as pivotal odorants through odor activity value (OAV) calculation. Concentrations of these aroma-active compounds were quantified and subsequently employed in reconstructing the aroma profile of fermented chili pepper. Quantitative descriptive sensory analysis and electronic nose analysis proved that the aroma profile of fermented chili pepper was basically reconstituted. Omission experiments confirmed that methyl salicylate, linalool, 2-methoxy-3-isobutylpyrazine, and phenylethyl alcohol were the key aroma-active compounds of fermented chili pepper. Moreover, the perceptual interactions between the key aroma-active compounds were investigated. It was found that methyl salicylate masked the floral aroma, while phenylethyl alcohol had an additive effect on the aroma of linalool and 2-methoxy-3-isobutylpyrazine.

1. Introduction

The aroma of fermented chili pepper (*Capsicum frutescens* L.) is a crucial quality attribute that directly influences consumer preference (Ye et al., 2020). The specific aroma profile of fermented chili pepper is influenced by the production methods, raw materials, and strains of microorganisms used (Liu et al., 2023). In recent years, our research group have conducted a comprehensive study on the aroma profile of fermented chili pepper, exploring various factors such as different ripening levels and cultivars of the peppers, fermentation vessels and brines, as well as bacterial and yeast strains (Ye et al., 2022; Ye et al., 2022; Xiao et al., 2023; Zhang et al., 2022). The analysis identified hundreds of volatile compounds in fermented chili pepper, with terpenes (such as β-ocimene and linalool), esters (like methyl salicylate and ethyl hexanoate), volatile acids (like acetic acid and hexanoic acid), and alcohols (including hexanol and phenylethyl alcohol) being the most prevalent (Liu et al., 2023). However, a comprehensive comparative study on olfactometric and sensory analysis of the aroma profile of fermented chili pepper is still needed.

Our previous works have conducted a systematic analysis of the aroma of fermented chili peppers at different fermentation time using headspace solid-phase microextraction-gas chromatography–mass spectrometry (HS-SPME-GC–MS) and multivariate data analysis (Ye et al., 2022; Ye et al., 2022; Zhang et al., 2022). Although HS-SPME-GC–MS can be used for qualitative and quantitative analysis of the aroma profile, individual differences in aroma perception thresholds mean that only a small subset of the numerous volatile compounds significantly contribute to sensory perception, with varying aroma impacts (Ni et al., 2022). To address this gap, gas chromatography-olfactometry (GC-O) leverages the human olfactory system as a detection tool to effectively identify aroma active compounds (Zhao et al., 2021). Additionally, odor activity value (OAV) analysis, as a crucial method for determining the individual aroma compounds' contributions to the overall aroma profile, can be combined with HS-SPME-GC–MS and GC-O to estimate the odor contribution of quantitative aroma compounds (Tan et al., 2022). Despite this, the chromatographic process

* Corresponding author at: Faculty of Food Science and Engineering, Kunming University of Science and Technology, Kunming 650500, China
E-mail addresses: xyz1620@163.com (Y. Xiao), zs1zhangshiyao@163.com (S. Zhang), wangxy2255@163.com (X. Wang), z18280182766@163.com (X. Zhao), liuzhijia@kust.edu.cn (Z. Liu), 7180112069@stu.jiangnan.edu.cn (C. Chu), wangyfkust@163.com (Y. Wang), huxiaos@263.net (X. Hu), junjieyi@kust.edu.cn (J. Yi).

https://doi.org/10.1016/j.fochx.2024.101581
Received 30 April 2024; Received in revised form 7 June 2024; Accepted 17 June 2024
Available online 19 June 2024

separates the olfactory stimulus from the food, leading to a lack of sensory interactions, including synergistic and inhibitory effects (Baldovini & Chaintreau, 2020). Therefore, it is essential to incorporate sensory techniques, such as aroma recombination and omission experiments, to validate the true contribution of aromatic compounds (Ma et al., 2022). Furthermore, Feller's additive model can be utilized to assess the interaction effects of certain mixtures, enabling a more precise determination of the perceptual interactions of key aroma-active compounds (Li et al., 2023). To date, combined instrumental and sensory techniques have been employed to systematically conduct flavor analysis and characterize key aromatic compounds in various foods, such as apple juice (Niu et al., 2019), fermented tea (Shen et al., 2023), and wine (Qian et al., 2024). However, no studies have utilized instrumental and sensory techniques to identify key aroma-active compounds of fermented chili peppers. In addition, the perceptual interaction of these aromatic compounds remains unknown, presenting significant challenges to scientifically elucidating the aroma quality of fermented chili peppers.

To address this knowledge gap, this study aims to achieve the following objectives: (1) accurately identify and quantify the aromatic compounds of fermented chili pepper using HS-SPME-GC–MS and GC-O; (2) evaluate the aroma contribution of the aromatic compounds through OAV calculations; (3) validate the key aroma-active compounds using recombination aroma model and omission experiments; and (4) explore perceptual interactions of the key aroma-active compounds based on Feller's additive model.

2. Materials and methods

2.1. Chemicals

The authentic standards were purchased for volatile qualitative and quantitative analysis. Acetic acid (≥ 99.8%), 4-methylvaleric acid (99.8%), myristic acid (≥ 99.5%), palmitic acid (≥ 99%), methyl butyrate (> 99.5%), isobutyl isovalerate (> 98%), isoamyl isovalerate (98%), methyl salicylate (≥ 99.5%), hexyl hexanoate (> 98%), methyl palmitate (99%), ethyl palmitate (≥ 99%), 1-hexanol (> 99.5%), benzyl alcohol (≥ 99.5%), phenylethyl alcohol (> 99.5%), *β*-myrcene (≥ 90%), linalool (98%), geraniol (≥ 99%), ethylbenzene (≥ 99.7%), o-xylene (≥ 99%), 2-heptanone (≥ 99.8%), phenylacetaldehyde (95%) and 2-methoxy-3-isobutylpyrazine (99%) were supplied by Sigma-Aldrich Chemical Co., Ltd. (Shanghai, China). 3-Methyl-1-butanol (98%) and (*Z*)-linalool oxide (98%) were supplied by Macklin Biochemical Co., Ltd. (Shanghai, China). (*E*)-*β*-Farnesene (98%), *β*-ocimene (98%) and hexyl 2-methylbutyrate (98%) were supplied by Yien Chemical Technology Co., Ltd. (Shanghai, China).

2.2. Fermented chili pepper samples preparation

Based on our previous studies, it has been established that green chili peppers exhibit a wider variety and higher content of aroma compounds (Ye et al., 2022). When fermented in jars with aged brine for 30 days, they demonstrate superior and more characteristic flavor profiles (Zhang et al., 2022). Therefore, in this study, green chili peppers that were fermented in jars with aged brine for 30 days were selected as the samples. These fermented chili pepper samples were sourced from Hongbin Green Food Group Co. Ltd. (Jianshui, Yunnan, China). Approximately 100 g of fermented chili peppers with brine were collected from three sampling points from the top to the bottom of the jar and from four sampling points from the side to the center of the jar. The samples from each sampling point were thoroughly mixed to ensure the final samples were representative. It was also verified that the fermented chili peppers exhibited no visible blemishes, diseases, or physical damage. Subsequently, the samples were promptly transported to the laboratory under refrigeration, sealed within sterile plastic bags. All samples were preserved at −40 °C until further analysis.

2.3. Headspace solid-phase microextraction

Following our previous study, the aroma extraction procedure was conducted (Ye et al., 2020). The HS-SPME holder coupled with a 50/30 μm divinylbenzene/carboxen/polydimethylsiloxane (DVB/CAR/PDMS) fiber (Zhenzheng, Qingdao, China) was used. The 3 g sample was added to a 10 mL headspace bottle and sealed using a screw cap equipped with a PTFE/silicone septum. Following this, the sample bottle was allowed to equilibrate at 40 °C for 15 min, after which the pre-aged fiber was adsorbed for 40 min. The fiber was inserted into the GC injector for desorption, where it was held at 250 °C for a duration of 5 min.

2.4. Gas chromatography–mass spectrometry analysis

The volatile compounds were examined employing a method similar to our prior research, with certain adjustments (Zhang et al., 2022). The GC–MS system (QP2010, Shimadzu, Japan) equipped with a DB-5MS capillary column (30 m × 0.25 mm, 0.25 μm) was used for volatiles analysis. The carrier gas (helium, > 99.99%) flow rate was 2 mL/min. The oven temperature program commenced at 45 °C and was maintained for 5 min, then ramped up to 250 °C at a rate of 5 °C/min, and held for 2 min. The mass spectrometer operated in electron-impact mode at 70 eV, scanning a mass range of 35–500 amu (*m/z*).

The volatile compounds of fermented chili pepper were qualitatively analyzed through MS characterization and comparison of retention indices (RIs). The MS data were scrutinized against the NIST 2014 library, with qualitative analysis performed based on the correlation and structural data derived from MS. Compounds were deemed identified when the matching degree exceeded 80%. The RIs were determined under identical GC–MS conditions using n-alkanes (C5-C25) standards (Anpel, Shanghai, China). Additionally, the obtained RIs were compared with standard RIs corresponding to the target compounds. Quantification of the compounds extracted via SPME was carried out using external standards. Standard curves for volatile compounds were generated by analyzing a series of standard compounds with known concentrations.

2.5. Gas chromatography-olfactometry analysis

The fermented chili pepper samples were evaluated using an olfactory detector port (ODP4, Gerstel, Germany) connected to the GC instrument (Niu et al., 2019). The analytical parameters were consistent with those of GC–MS. To maintain olfactory sensitivity, humidified air was introduced into the sniffing cone at a constant flow rate of 50 mL/min, preventing dehydration of the nasal mucosa. Prior to GC-O analysis, 15 panelists received training with a minimum of 35 aroma reference compounds. They were instructed to familiarize themselves with the sensory descriptors and aroma intensities (AIs) for each compound. During GC-O analysis, panelists positioned their noses near the sniffing port. Upon detecting a scent, they activated the "olfactometer button," verbalized a description, and recorded their observations. A six-point scale ranging from 0 to 5 was utilized: 0 indicated absence, 1 represented very faint, 2 indicated faint, 3 signified moderate, 4 suggested strong, and 5 represented very strong. The AI score for aromas sharing identical retention times and similar descriptions was determined by averaging ratings from the 15 panelists. Each panelist performed GC-O/AI analysis three times for every sample to ensure reliability.

2.6. Sensory analysis

Based on sensory evaluation, odor threshold, aroma recombination, omission experiments, and aroma perception of aromatic compounds were determined. The research protocol had been submitted to the Medical Ethics Committee of Kunming University of Science and Technology (Reference No. KMUST-MEC-077) for ethical approval. All sensory evaluation participants participated voluntarily and consented to

the use of their information.

2.6.1. Odor threshold detection and odor activity value calculation

The odor threshold measurement was conducted following the ASTM-E1432 method (Cometto-Muñiz & Abraham, 2008). Prior to the test, the 15 panelists were briefed on the aroma characteristics of each compound and provided with a standard solution to familiarize themselves with its aroma. Each session comprised 10 forced-choice examinations, each consisting of three brown bottles containing a solution. Among them, one bottle contained a solution with aroma compounds at decreasing concentrations (achieved through successive 2-fold dilution), while the remaining two bottles contained blank model solutions. The concentration/response function was a psychometric function and followed a S-curve ($P = 1/[1 + \exp.(-(x - C)/D)]$). Here, x represented the concentration of the aromatic compound [log (μg/kg)], C indicated the olfactory threshold of the aromatic compound [log (μg/kg)], D served as a parameter defining the gradient of the function for each odorant, and P denoted the probability of detection corrected by chance factor [$P = (3p - 1)/2$], with p representing the proportion of correct responses for each concentration. The experimental threshold was the concentration at which the probability of correct detection reached 50%. To assess aroma contributions of aromatic compounds, the OAV was calculated, which was to determine the concentration of the target odorant divided by the corresponding threshold (Pang et al., 2012).

2.6.2. Aroma recombination

The aroma recombination process entailed combining different aromatic compounds with OAV $\geq$ 1, which were incorporated into the odor-blank matrix at their respective detected concentrations and thoroughly mixed to achieve an overall aroma profile (Chen et al., 2022). To account for the potential influence of non-volatile compounds on aroma emission, an odor-blank matrix was prepared following a method outlined in a previous study with slight adaptations (Liu et al., 2022). The procedure involved weighing 50 g of fermented chili pepper and 50 mL of ultrapure water into a rotary evaporation flask. Subsequently, volatiles were evaporated in a 70 °C water bath, with this step reiterated multiple times (no <5) until the sensory impact almost disappeared. Following this, the fermented chili pepper samples were dried using an oven at 70 °C.

The aroma profiles of both the initial fermented chili pepper and the reconstructed model were assessed by 15 panelists following the approach outlined by Ni et al. (2022) with certain adjustments. Before the analysis, training sessions were conducted to enable panelists to accurately perceive and describe the odor characteristics of fermented chili pepper. Following this, the panelists underwent training with standardized aroma samples to establish a consensus on the chosen aroma descriptors (fruity, green, floral, citrus, herbal, and spicy notes) and their intensities. Fruity evoked the scent of ripe fruits; green denoted an aroma reminiscent of grass; floral was the light aroma of fresh flowers; citrus evoked the fragrance of citrus fruits; herbal was characterized by the scent of various herbs; and spicy exhibited pungent characteristics. The precise descriptions were delineated using aqueous solutions containing reference odorants as follows: isoamyl isovalerate for "fruity" attribute, phenethyl alcohol for "floral" attribute, linalool for "citrus" attribute, β-ocimene for "herbal", β-myrcene for "spicy" attribute and 2-methoxy-3-isobutylpyrazine for "green". Each sample (5 g) was deposited into a scent-free cup and assigned a random three-digit numerical identifier. Subsequently, each panelist received a sample randomly and assessed it quantitatively using a 10-point interval scale, ranging from 0 (no discernible odor) to 9 (extremely potent odor). Conducted in a sensory laboratory with a consistent room temperature of approximately 25 °C, the evaluation yielded the ultimate score for each characteristic, determined by averaging the ratings assigned by all participating panelists.

2.6.3. Omission experiments

The individual contribution of each aroma-active compound was evaluated using omission experiments (Chen et al., 2022). The single aroma-active compounds were removed from the comprehensive aroma recombination model. Subsequently, panelists compared each omission model with two complete recombination models through a triangle test to evaluate differences. To ensure unbiased assessment, all recombination samples were randomly assigned three-digit codes, and panelists were instructed to select the different sample among them during sensory analysis.

2.6.4. Perceptual interaction analysis

Based on threshold detection method, perceptual interactions of key aroma-active compounds were investigated. Perceptual interactions were evaluated using Feller's additive model (Miyazawa et al., 2008). The detection probability $P(AB)$ was defined as follows: $P(AB) = P(A) + P(B) - P(A)P(B)$, where $P(A)$ and $P(B)$ indicated the probabilities of two compounds in the mixture were detected, respectively. The threshold values obtained from the actual experimental model and the basic additive theoretical model were compared. When the experimental threshold of the mixture surpassed the theoretical value, it indicated a degree of mask effect. When the ratio of the experimental threshold to the theoretical threshold was below 0.5, it indicated a synergistic effect. Alternatively, if the ratio of the experimental threshold to the theoretical threshold fell between 0.5 and 1.0, it signified an additive effect (Li et al., 2023).

2.7. Electronic nose analysis

Electronic nose analysis (cNose, Shanghai Bosin Industrial Development Co., Ltd., China) equipped with 18 metal oxide sensors was utilized to further monitor and distinguished the aroma profiles of fermented chili pepper, recombination model and omission models. The electronic nose analysis was conducted based on the method described by Zhang et al. (2023) with minor modifications. The 5 g homogenized sample was placed in a 20 mL headspace vial, sealed with a Teflon/Silicon spacer, and balanced at 40 °C in a water bath for a duration of 30 min. Throughout 120 s measurement duration, the volatile gases at a rate of 1 L/min induced fluctuations in the sensor's conductance. The sufficient duration of sampling time ensured that the sensor signals reached a stable value.

2.8. Statistical analysis

The experiments were conducted in triplicate, and the results were expressed as mean ± standard deviation. The results of triangle sensory test were analyzed using the one-tailed test to determine significant differences ($p < 0.05$). Data visualization and statistical analysis were performed using Origin 2021 software (Origin Lab, Northampton, USA).

3. Results and discussion

3.1. Aroma compounds identification and quantification

The volatile compounds in fermented chili pepper samples were determined using HS-SPME-GC–MS, and the representative total ion chromatography was shown in Fig. 1A. A total of 28 volatile compounds were identified in the fermented chili pepper samples, categorized based on their chemical structure into acids (4), esters (8), alcohols (5), terpenes (6), and others (5) (Table 1). Among these compounds, acids were the most abundant, followed by alcohols and terpenes, collectively representing approximately 79.18% of the total volatile contents in fermented chili pepper. The distribution of volatiles closely resembled to the findings of previous studies (Zhang et al., 2022).

Sour and pungent in character, acids were the most abundant class in fermented chili pepper. The concentration of acetic acid was 1301.92 ±

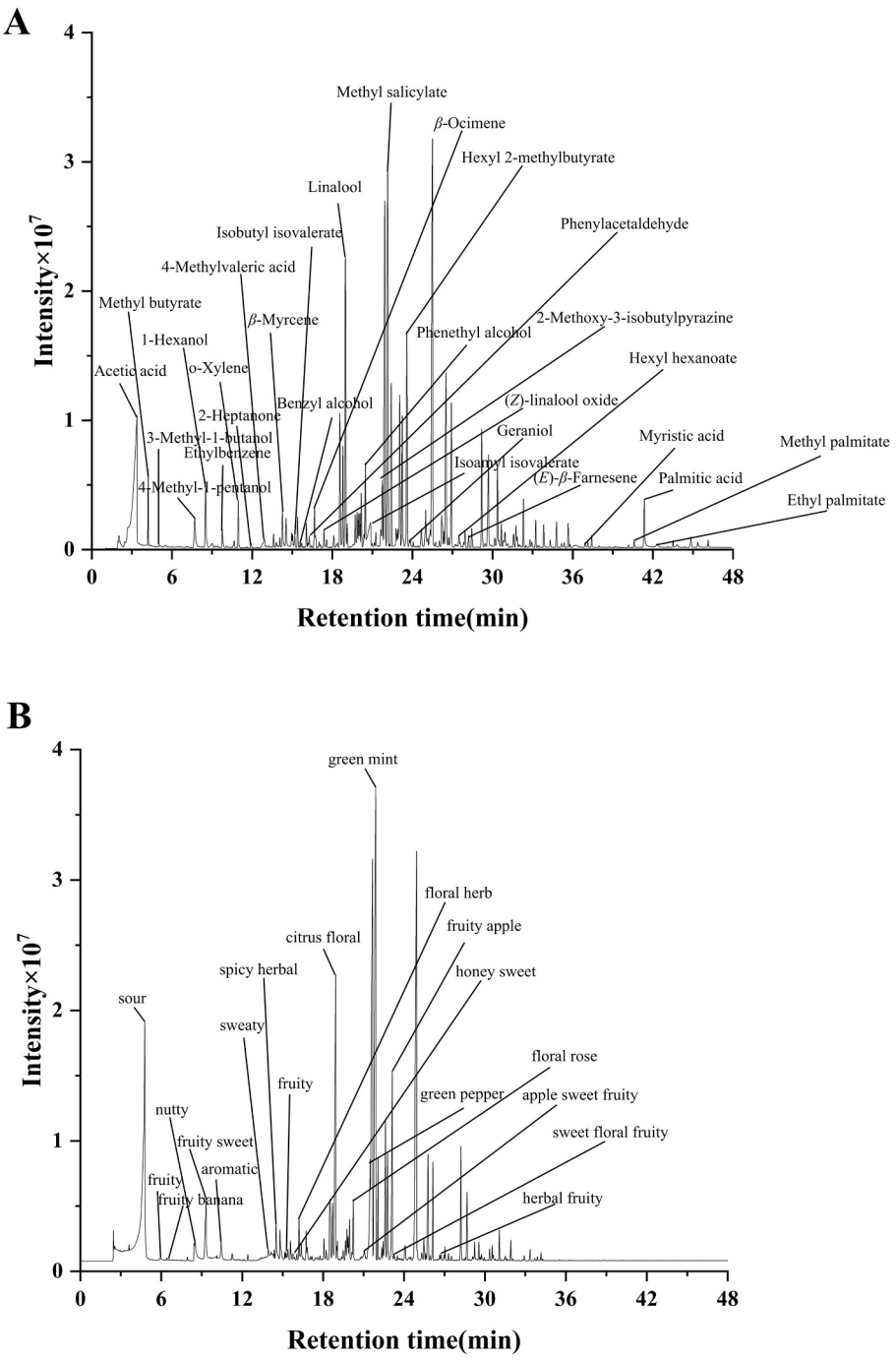


Fig. 1. Total ion chromatography of volatile compounds in fermented chili pepper by GC–MS (A) and GC-O (B).

80.65 μg/kg, contributing a delightful acidic note to the food. It was generated through two pathways: the metabolic breakdown of threonine, producing acetaldehyde subsequently oxidized into acetic acid, and the oxidative phosphorylation and tricarboxylic acid cycle within the cells (Kun et al., 2022). Furthermore, fermented chili pepper contained significant quantities of fatty acids, synthesized through the fatty acid synthesis pathway from acetyl-CoA (El-Shamy & Farag, 2022).

Another crucial group influencing aroma, alcohols were generated through sugar metabolism, as well as the decarboxylation and dehydrogenation of amino acids. Five alcohols were identified, with 4-methyl-1-pentanol (428.57 ± 42.25 μg/kg) emerging as the predominant one in fermented chili pepper, followed by phenylethyl alcohol, 1-hexanol, 3-methyl-1-butanol and benzyl alcohol. The 4-methyl-1-pentanol was closely associated with microbial metabolism and had previously been detected in Chinese pickled pepper inoculated with *Lactiplantibacillus plantarum* (Tian et al., 2023).

Terpenes were synthesized as secondary metabolites by the activity of glycosylases from microorganisms, which capable of cleaving the bond between terpenes and sugars or initiating de novo production (Yang et al., 2020). Among these compounds, linalool (325.70 ± 32.31 μg/kg) appeared with a higher concentration in fermented chili pepper, which could be synthesized by yeasts through the mevalonate pathway, originating from geraniol diphosphate (Holt et al., 2019).

Esters were primarily formed through reaction catalyzed by esterase between alcohols and acids, sourced from glucoses and amino acids during microbial metabolism (Al-Dalali et al., 2020). In addition, esters

Table 1
Volatile compounds detected in fermented chili pepper by GC–MS and GC-O.

Compounds	RI_1[a]	RI_2[b]	Standard curve	R^2	Content(μg/kg)	Odor threshold (μg/kg)	OAV	Odor description[c]	Aroma intensity
Acids									
Acetic acid	650	646	y = 0.0104× + 2.3444	0.9869	1301.92 ± 80.65	387.66	3.36	sour	3.6
4-Methylvaleric acid	960	955	y = 0.0015× + 0.0904	0.9883	33.65 ± 4.39	33.93	0.99	sweaty	2.4
Myristic acid	1754	1756	y = 0.0034× + 0.127	0.9722	47.96 ± 1.66	–	–	–	–
Palmitic acid	1959	1960	y = 0.0108× + 4.2491	0.9876	194.05 ± 34.31	–	–	–	–
Esters									
Methyl butyrate	717	721	y = 0.045× + 0.4822	0.9703	9.00 ± 0.06	15.06	0.69	fruity	2.3
Isobutyl isovalerate	1007	1005	y = 0.0021× + 0.0633	0.9773	25.11 ± 3.09	26.44	0.95	fruity	3.5
Isoamyl isovalerate	1106	1105	y = 0.0028× + 0.4662	0.9842	64.81 ± 8.18	16.32	3.97	apple sweet fruity	3.9
Methyl salicylate	1195	1193	y = 0.0173× + 0.6642	0.9751	102.18 ± 13.63	3.32	30.78	green mint	4.1
Hexyl 2-methylbutyrate	1237	1237	y = 0.0085× + 1.2395	0.9639	72.50 ± 6.19	42.97	1.69	fruity apple	3.2
Hexyl hexanoate	1385	1384	y = 0.0062× + 0.1357	0.9640	8.51 ± 1.40	334.58	0.03	herbal fruity	2.1
Methyl palmitate	1922	1927	y = 0.0052× + 0.0432	0.9925	46.39 ± 2.56	–	–	–	–
Ethyl palmitate	1991	1991	y = 0.0049× + 0.0995	0.9636	12.97 ± 1.39	–	–	–	–
Alcohols									
3-Methyl-1-butanol	727	726	y = 0.0014× + 0.0131	0.9883	133.85 ± 8.59	140.58	0.95	fruity banana	3.5
4-Methyl-1-pentanol	840	837	y = 0.0012× + 0.0645	0.9795	428.57 ± 42.25	83.65	5.12	nutty	3.1
1-Hexanol	868	865	y = 0.0019× + 0.0833	0.9685	164.26 ± 17.59	41.82	3.93	fruity sweet	3.8
Benzyl alcohol	1031	1031	y = 0.0023× + 0.0343	0.9711	17.64 ± 1.63	–	–	–	–
Phenylethyl alcohol	1106	1107	y = 0.0036× + 0.822	0.9868	233.24 ± 28.23	40.13	5.81	floral rose	4.1
Terpenes									
β-Myrcene	990	991	y = 0.0013× + 0.3975	0.9933	78.20 ± 7.08	8.76	8.93	spicy herbal	3.6
β-Ocimene	1049	1043	y = 0.0008× + 0.2004	0.9826	286.48 ± 34.01	83.65	3.42	floral herb	3.8
(*Z*)-Linalool oxide	1074	1071	y = 0.005× + 0.0991	0.9726	13.59 ± 1.92	–	–	–	–
Linalool	1100	1101	y = 0.0085× + 0.0812	0.9926	325.70 ± 32.31	8.32	39.15	citrus floral	4.2
Geraniol	1251	1253	y = 0.0084× + 0.3876	0.9766	11.43 ± 1.26	14.52	0.78	sweet floral fruity	3.1
(*E*)-*β*-Farnesene	1455	1456	y = 0.0022× + 0.0422	0.968	26.16 ± 4.13	–	–	–	–
Others									
Ethylbenzene	864	869	y = 0.0166× + 4.2112	0.9908	58.33 ± 6.53	66.12	0.88	aromatic	3.7
o-Xylene	872	878	y = 0.0173× + 6.3576	0.9865	385.41 ± 37.43	–	–	–	–
2-Heptanone	890	889	y = 0.0193× + 0.0102	0.9971	15.24 ± 1.20	–	–	–	–
Phenylacetaldehyde	1047	1044	y = 0.0033× + 0.0749	0.9799	42.96 ± 6.16	2.16	19.89	honey sweet	3.4
2-Methoxy-3-isobutylpyrazine	1177	1178	y = 0.0156× + 0.01	0.9939	23.62 ± 1.58	5.75	4.11	green pepper	3.7

[a] RI_1: the retention index obtained by GC–MS.
[b] RI_2: the retention index obtained by GC-O.
[c] Odor description: The odor characteristic of each volatile compound sensed on sniffer port during GC-O experiments. The "-" mark indicated that the volatile compound was not sniffed.

could be formed through non-enzymatic esterification reactions involving alcohols and organic acids during fermentation and aging processes (Li et al., 2023). Methyl salicylate (102.18 ± 13.63 μg/kg), also known as sweet birch oil, imparted a mint smell and exhibited the highest content among esters.

Furthermore, ethylbenzene, o-xylene, 2-heptanone, phenylacetaldehyde, and 2-methoxy-3-isobutylpyrazine were identified in fermented chili pepper. Ethylbenzene and o-xylene were aromatic hydrocarbons, while the presence of 2-heptanone (a ketone) suggested the involvement of a lipid metabolic pathway that could influence flavor characteristics. Phenylacetaldehyde, as an aldehyde in fermented chili pepper, likely originated primarily from the Strecker degradation of amino acids (Liu et al., 2023). Additionally, 2-methoxy-3-isobutylpyrazine, a pyrazine derivative derived from the food matrix, was abundant before chili pepper fermentation (Xiao et al., 2023).

3.2. Aroma-active compounds identification

Of the 28 volatile compounds, 19 aroma-active compounds were identified through GC-O/AI analysis (Fig. 1B). The AIs of these aroma-active compounds ranged from 1.8 to 4.2 in fermented chili pepper samples (Table 1). Discrepancies in AIs might be primarily attributed to concentration variations among these compounds, as well as potential differences in mass transfer coefficients (Weterings et al., 2020). Aroma active compounds with higher intensity exhibited higher AIs, thereby making a more important contribution to the distinctive aroma profile of fermented chili pepper.

Linalool (AI = 4.2), characterized by citrus and floral scent, exhibited the highest intensity, followed by methyl salicylate (AI = 4.1) and phenylethyl alcohol (AI = 4.1), contributing green, mint, and rose-floral notes, respectively. The sour aroma in fermented chili pepper was attributed to acetic acid (AI = 3.6). Furthermore, 4-methylvaleric acid (AI = 2.4) produced a sweaty note, which was produced in the presence of 2-isobutylmalate synthase via the degradation of leucine (Wang et al., 2022). Esters such as methyl butyrate (AI = 2.3), isobutyl isovalerate (AI = 3.5), isoamyl isovalerate (AI = 3.9), hexyl 2-methylbutyrate (AI = 3.2), and hexyl hexanoate (AI = 2.1) predominantly imparted fruity note, playing a pivotal role in defining the characteristic aroma of fermented chili pepper. Alcohols contributed importantly to aroma profile, each exhibiting unique characteristics: 3-methyl-1-butanol (AI = 3.5) provided a fruity and banana odor, 4-methyl-1-pentanol (AI = 3.1) offered a nutty scent, 1-hexanol (AI = 3.8) imparted a fruity and sweet odor, and phenylethyl alcohol (AI = 4.1) contributed a floral and rose fragrance. Terpenes, such linalool (AI = 4.2) and *β*-ocimene (AI = 3.8), primarily conveyed herbal and floral notes. Geraniol (AI = 3.1), derived from the same precursor, geranyl pyrophosphate, exhibited a unique sweet characteristic, setting it apart from linalool (Wang et al., 2020). Meanwhile, the robust spicy scent was derived from *β*-myrcene (AI = 3.6). The presence of ethylbenzene (AI = 3.7) introduced subtle aromatic note. Phenylacetaldehyde (AI = 3.4) enriched the olfactory experience with its green, sweet, and floral scent, while 2-methoxy-3-isobutylpyrazine (AI = 3.7) introduced a green and peppery undertone. These complex interactions and contributions of individual compounds contributed to the intricate and diverse aroma characteristics of fermented chili pepper.

3.3. Aroma-active compounds contributions

After determining the quantitative and odor thresholds for aroma-active compounds, the odor activity value (OAV) was calculated to further screen for compounds with greater contributions (Table 1). The S-curves following the threshold measurements are shown in Supplementary Fig. 1. Among the 19 aroma-active compounds, 12 compounds with OAVs ≥1 were identified for their higher aroma activity (Fig. 2).

Among these compounds, linalool had the highest OAV (OAV = 39.15), which was consistent with the findings from GC-O/AI analysis. It exhibited the characteristics of citrus and floral, which might enhance the citrus characteristics of fermented chili pepper (Ni et al., 2022). The *β*-myrcene (OAV = 8.93), with its herbal and spicy notes, played a pivotal role in the aroma profile of fermented pepper paste (Li et al., 2023). Linalool and *β*-myrcene had also been identified as important odorant compounds in pepper oil (Sun et al., 2020). In addition, *β*-ocimene (OAV = 3.42) was identified as an essential aroma compound in fermented chili pepper, contributing to herb and floral notes. Methyl salicylate (OAV = 30.78), widely used in perfumery and approved as a flavoring agent in foodstuffs, imparted a distinctive wintergreen aroma, enhancing the overall pleasant aroma of fermented chili pepper. Isoamyl isovalerate (OAV = 3.97) and hexyl 2-methylbutyrate (OAV = 1.69) were also important esters and contributed fruity note to fermented chili pepper. The primary alcohols identified were phenylethyl alcohol (OAV = 5.81), 4-methyl-1-pentanol (OAV = 5.12) and 1-hexanol (OAV = 3.93), imparting fruity and floral notes, respectively. Specifically, phenylethyl alcohol, primarily derived from *L*-phenylalanine metabolism, imparted a floral and rose fragrance (Sieiro-Sampedro et al., 2019). Meanwhile, *L*-phenylalanine served as the precursor of phenylacetaldehyde (OAV = 19.89), a major contributor to sweet and honey notes at a low concentration. The green and pepper aromas were contributed by 2-methoxy-3-isobutylpyrazine. Despite its relatively low concentration, 2-methoxy-3-isobutylpyrazine (OAV = 4.11) played a crucial role as an important contributor to the olfactory profile of fermented chili pepper. It had been reported that 2-methoxy-3-isobutylpyrazine was also the important aroma-active compound of asparagus (Pegiou et al., 2023) and fermented pepper paste (Kang & Baek, 2014).

The findings revealed that there was no direct correlation between the concentrations and OAVs of the aroma-active compounds. For example, the concentration of phenylacetaldehyde (42.96 ± 6.16 μg/kg) was not high, but its strong aroma contribution (OAV = 19.89) resulted from its extremely low threshold (2.16 μg/kg). On the other hand, despite having higher concentration in fermented chili pepper, 3-methyl-1-butanol (133.85 ± 8.59 μg/kg) had a low aroma contribution (OAV = 0.95) due to its remarkably high threshold (140.58 μg/kg). Therefore, quantitative analysis combined with OAV calculation was essential for a more accurate description of aroma profile (Zhao et al., 2021).

Our combined analysis of AI and OAV demonstrated a strong correlation between aroma-active compounds with high AIs and those exhibiting high OAVs, such as linalool and methyl salicylate. This suggested a high level of agreement between these two methodologies. Comparable findings were also noted by Tan et al. (2022) and Niu et al. (2019) when examining important aromatic compounds of peach and apple juices using a combination of AI and OAV analysis, highlighting a positive association between these two analytical approaches.

3.4. Aroma profile simulation reproduction

The aroma profile of fermented chili pepper and its recombination

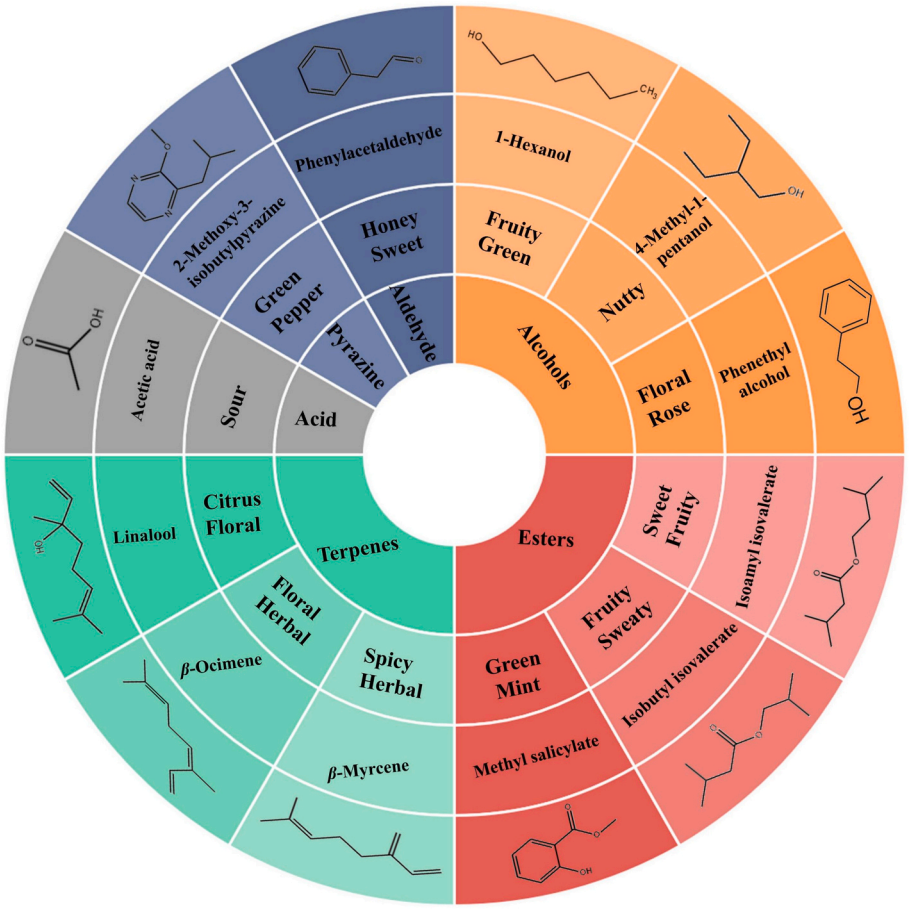


Fig. 2. The aroma wheel of the aroma-active compounds in fermented chili pepper identified by the calculation of OAVs.

model were determined through quantitative descriptive sensory analysis. Six attributes, fruity, green, floral, citrus, herbal and spicy notes, were selected for description. As shown in Fig. 3A, the recombination model exhibited a high similarity to the original fermented chili pepper aroma profile, with slight differences in some notes. Both fermented chili pepper and recombination model were characterized the most strongly by spicy note with a score of 8.4–7.8, followed by floral note (7.5–7.3). In the recombination model, a weaker presence of fruity, citrus and spicy odors was observed, while green, floral and herbal odors were perceived slightly higher.

The electronic nose profile showed that the aroma profile of the original fermented chili pepper and recombination model were similar for each sensor, although the response values showed slight differences (Fig. 3B). This aligns with the results of quantitative descriptive sensory analysis. The response values of the recombination model on sensor S5 and sensor S11 was higher than those of fermented chili pepper. Sensor S5 and S11 were sensitive to nitrogenous compounds and alkanes, respectively. This could be attributed to the interaction between 2-methoxy-3-isobutylpyrazine (nitrogenous compounds) and the extended control of oven temperature during drying, leading to the conversion of non-volatile compounds during the preparation of odor-blank matrix (Wang et al., 2020). The slight variance between the recombination model and the actual fermented chili pepper aroma might be influenced by several factors. On one hand, the detected odor thresholds might not precisely align with those in a real food matrix. On the other hand, accurately measuring the intricate and diverse interactions between odorants and the matrix could be challenging. Despite these challenges, the results still successfully simulated the typical aroma profile of fermented chili pepper, laying a foundation for further omission experiments.

3.5. Key aroma-active compounds verification

Based on the response signals of 18 electronic nose sensors of each omission model, principal component analysis (PCA) was conducted to validate the flavor variances (Fig. 3C). The two principal components (PC1 and PC2) explained 84.20% of the overall variance (75.90% and 8.30%, respectively). Based on the PCA results, the omission model could be separated on the PCA plot. The omitted acetic acid model, the omitted alcohols models (4-methyl-1-pentanol, 1-hexanol, and phenylethyl alcohol) and the omitted esters models (isobutyl isovalerate and isoamyl isovalerate) fell in the positive side of PC1, while the omitted terpenes models (linalool, β-myrcene, and β-ocimene) and omitted 2-methoxy-3-isobutylpyrazine model clustered in the negative side of PC1. However, unlike omitted isobutyl isovalerate model and omitted isoamyl isovalerate model, the aroma attributes observed in the omitting methyl salicylate model were more similar to those of omitting terpenes. This might be due to the similarity of methyl salicylate with terpenes in aroma profiles. At the same time, the aroma profile of phenylacetaldehyde exhibited resemblances to those of methyl salicylate and β-myrcene.

Using the recombination model as the reference, 12 omission models were evaluated using triangle test, and the results are presented in Table 2. Four essential odorants were collectively identified from the 12 aroma-active compounds, showing a significant distinction ($p < 0.05$) when omitted from the recombination models. In particular, the omission of methyl salicylate and linalool resulted in a highly significant difference ($p < 0.001$), emphasizing the crucial role of these aroma-active compounds in shaping the overall aroma profile of fermented chili pepper. Additionally, 2-Methoxy-3-isobutylpyrazine exhibited a highly significant difference ($p < 0.01$), and phenylethyl alcohol showed a significant difference ($p < 0.05$). According to our previous studies, methyl salicylate, 2-methoxy-3-isobutylpyrazine, and linalool were present in fresh chili pepper (Xiao et al., 2023; Ye et al., 2022). Among them, methyl salicylate and 2-methoxy-3-isobutylpyrazine were consumed as fermentation substrates. Linalool increased the content through the action of carbohydrate active enzymes, and yeasts produced phenylethyl alcohol through amino acid metabolism (Xiao et al., 2023). In general, methyl salicylate, linalool, 2-methoxy-3-isobutylpyrazine, and phenylethyl alcohol were identified as the primary key aroma-active compounds crucial for shaping the aroma profile of fermented chili pepper.

Through partial least squares regression analysis, methyl salicylate, linalool, and phenylethyl alcohol were identified as key aroma compounds in fermented chili pepper (Ye et al., 2022). Similarly, Li et al. (2023) concurred, based on detection frequency analysis and relative odor activity value assessments, that these compounds played pivotal roles in defining the aroma of fermented chili pepper. These findings are consistent with the results obtained from the combined instrumental and

Table 2
Omission experiments from the recombination model.

Odorants omitted from the recombination model	Right of selection[a]	Significance[b]
Acetic acid	6	
Isobutyl isovalerate	6	
Isoamyl isovalerate	3	
Methyl salicylate	12	***
4-Methyl-1-pentanol	7	
1-Hexanol	5	
Phenylethyl alcohol	9	*
β-Myrcene	6	
β-Ocimene	6	
Linalool	13	***
Phenylacetaldehyde	8	
2-Methoxy-3-isobutylpyrazine	10	**

[a] Number of correct judgments from fifteen panelists.
[b] * significant ($p \leq 0.05$); ** highly significant ($p \leq 0.01$); *** very highly significant ($p \leq 0.001$).

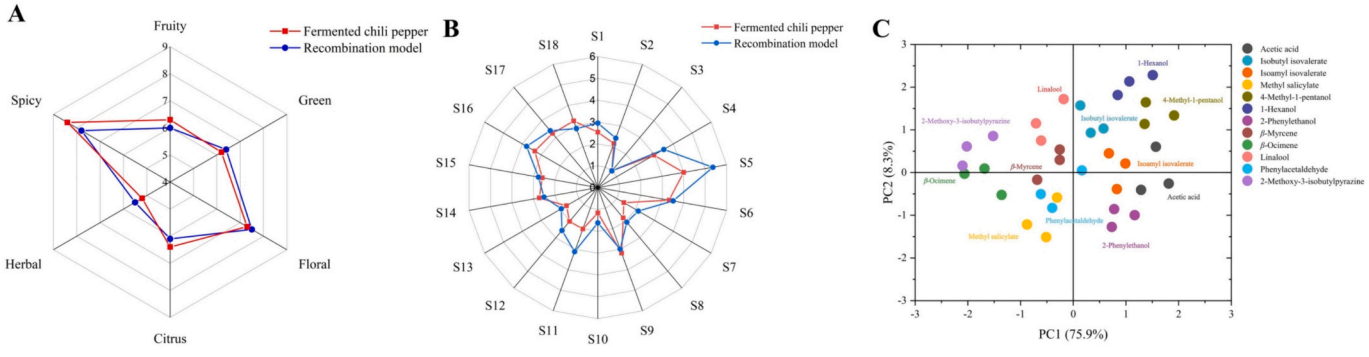


Fig. 3. Comparative aroma profile analysis of fermented chili pepper and aroma recombination model based on analysis of quantitative descriptive sensory (A) and electronic nose (B). PCA scores plot for omission model analysis based on electronic nose (C).

sensory techniques in this study. It is noteworthy that while 2-methoxy-3-isobutylpyrazine was infrequently reported as a crucial aroma component in fermented chili pepper, our study uncovered its significant contribution to the overall aroma profile.

3.6. Perceptual interaction between key aroma-active compounds

The threshold effects of the binary model involving four key aroma active compounds intermixed with each other were studied (Fig. 4). Among the six solution groups resulting from binary mixing, four groups exhibited masking effects: methyl salicylate with phenylethyl alcohol, methyl salicylate with linalool, methyl salicylate with 2-methoxy-3-isobutylpyrazine, and linalool with 2-methoxy-3-isobutylpyrazine. The ratios of actual odor threshold to theoretical odor threshold of the four groups were 1.98, 1.50, 1.12, and 2.16, respectively. Notably, in mixtures composed of compounds with large structural differences, masking effects were predominant (Zhu et al., 2017). Methyl salicylate exerted a negative influence on the on the overall aroma contribution post-mixing, likely attributable to its robust minty scent. This finding aligned with prior research indicating that esters could mask the aroma of other compounds, with methyl salicylate particularly masking floral aromas such as linalool and phenylethyl alcohol (Saison et al., 2009).

The ratios of actual odor threshold to theoretical odor threshold for mixtures of phenylethyl alcohol with linalool and phenylethyl alcohol with 2-methoxy-3-isobutylpyrazine were 0.60 and 0.62, respectively, falling between 0.5 and 1, indicating the presence of additive effects between the two groups. Compounds with similar structures or aromas were more likely to exhibit synergistic or additive effects (Zhu et al., 2017). Linalool, a chain terpene alcohol possessing the same hydroxyl group as phenylethyl alcohol, exhibited similar floral characteristics, likely accounting for the additive effect observed between phenylethyl alcohol and linalool. Additionally, research suggests that the degree of interaction among odorants depended on their intensity and pleasantness; when these factors were comparable, synergistic or additive effects were more pronounced (Li et al., 2023). Consequently, 2-methoxy-3-isobutylpyrazine and phenylethyl alcohol might have similar aroma intensity and pleasantness, facilitating additive effects between them.

Various factors influenced the processing of olfactory information in the olfactory system, including chemical similarity of odorants, odor intensity and odor pleasantness (Ma et al., 2021). In this study, the pairwise perceptual interaction between the four key aroma-active compounds often resulted in masking effects, particularly evident in the strong masking effect of methyl salicylate on floral fragrances. Through the combined action of phenylethyl alcohol with linalool or 2-methoxy-3-isobutylpyrazine, the floral or green aroma characteristics could be enhanced, respectively.

4. Conclusion

In this study, key aroma-active compounds and their characteristics of fermented chili pepper were systematically investigated. A total of 19 aroma-active compounds were detected, with 12 identified as important odorants through OAV analysis. Based in the results of aroma recombination and omission experiments, the key aroma-active compounds of fermented chili pepper were identified as methyl salicylate, linalool, 2-methoxy-3-isobutylpyrazine, and phenylethyl alcohol. It was observed that the aroma perception of the key aroma-active compounds often displayed a masking effect, with methyl salicylate exerting a strong masking effect on the floral aroma. Conversely, phenylethyl alcohol enhanced the floral and green flavor characteristics of fermented chili pepper through interactions with linalool and 2-methoxy-3-isobutylpyrazine, respectively.

The use of both instrumental and sensory techniques provided a comprehensive understanding of the contribution level of aroma-active compounds and enabled analysis of perceptual interactions. To further elucidate the underlying biochemical mechanisms and optimize fermentation processes for enhanced aroma quality, future research will focus on a more detailed examination of the key aroma-active

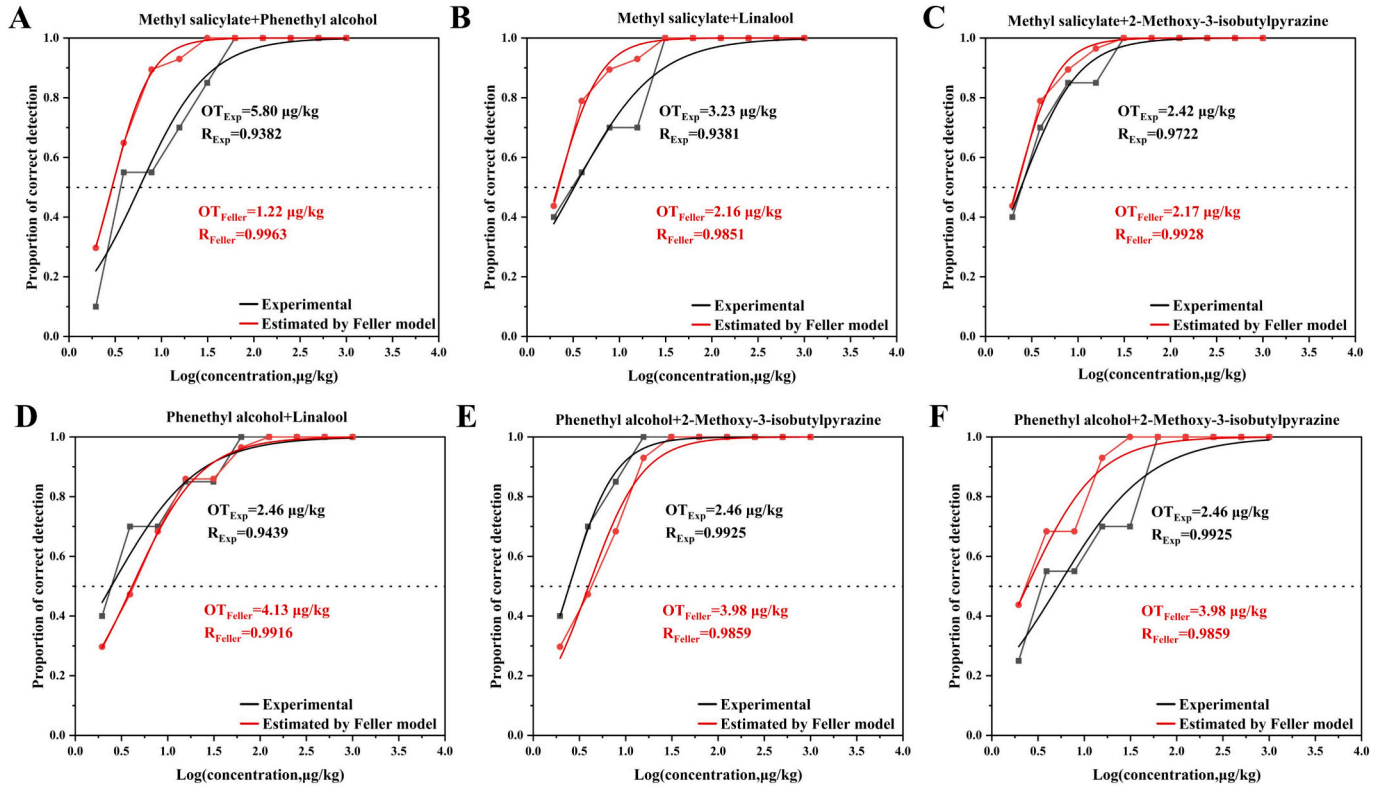


Fig. 4. Detection of perceptual interactions of key aroma-active compounds in fermented chili pepper.

compounds evolution at various fermentation stages.

CRediT authorship contribution statement

Yue Xiao: Writing – review & editing, Writing – original draft, Methodology, Investigation. **Shiyao Zhang:** Methodology, Investigation. **Xinyu Wang:** Writing – review & editing. **Xinyi Zhao:** Writing – review & editing. **Zhijia Liu:** Writing – review & editing, Supervision. **Chuanqi Chu:** Writing – review & editing. **Yanfei Wang:** Writing – review & editing. **Xiaosong Hu:** Supervision. **Junjie Yi:** Writing – review & editing, Supervision, Project administration, Conceptualization.

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

The data that has been used is confidential.

Acknowledgments

This work was supported by the Yunnan Provincial Natural Science Foundation (202301AW070015 and 202101BE070001-054), Major Science and Technology Projects in Yunnan Province (202102AE090050 and 202302AH360003), and Excellent Youth Funding of Yunnan Province (YNQR-QNRC-2018-109).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2024.101581.

References

Al-Dalali, S., Zheng, F., Sun, B., Zhou, C., Li, M., & Chen, F. (2020). Effects of different brewing processes on the volatile flavor profiles of Chinese vinegar determined by HS-SPME-AEDA with GC-MS and GC-O. *LWT - Food Science and Technology, 133*, Article 109969. https://doi.org/10.1016/j.lwt.2020.109969

Baldovini, N., & Chaintreau, A. (2020). Identification of key odorants in complex mixtures occurring in nature. *Natural Product Reports, 37*(12), 1589–1626. https://doi.org/10.1039/D0NP00020E

Chen, C., Liu, Z., Yu, H., Xu, Z., & Tian, H. (2022). Flavoromic determination of lactones in cheddar cheese by GC–MS–olfactometry, aroma extract dilution analysis, aroma recombination and omission analysis. *Food Chemistry, 368*, Article 130736. https://doi.org/10.1016/j.foodchem.2021.130736

Cometto-Muñiz, J. E., & Abraham, M. H. (2008). Human olfactory detection of homologous n-alcohols measured via concentration–response functions. *Pharmacology Biochemistry and Behavior, 89*(3), 279–291. https://doi.org/10.1016/j.pbb.2007.12.023

El-Shamy, S., & Farag, M. A. (2022). Volatiles profiling in heated cheese as analyzed using headspace solid-phase microextraction coupled to gas chromatography coupled to mass spectrometry. *EFood, 3*(1–2), Article e2. https://doi.org/10.1002/efd2.2

Holt, S., Miks, M. H., de Carvalho, B. T., Foulquié-Moreno, M. R., & Thevelein, J. M. (2019). The molecular biology of fruity and floral aromas in beer and other alcoholic beverages. *FEMS Microbiology Reviews, 43*(3), 193–222. https://doi.org/10.1093/femsre/fuy041

Kang, K.-M., & Baek, H.-H. (2014). Aroma quality assessment of Korean fermented red pepper paste (*gochujang*) by aroma extract dilution analysis and headspace solid-phase microextraction–gas chromatography–olfactometry. *Food Chemistry, 145*, 488–495. https://doi.org/10.1016/j.foodchem.2013.08.087

Kun, J., Meng, Q., Wei, C.-C., Xie, G., Yan, J., Ho, C.-T., & Tong, H. (2022). Characterization of the key compounds responsible for the fermented soybean-like cup aroma of raw Pu-erh tea using instrumental and sensory methods. *LWT - Food Science and Technology, 162*, Article 113458. https://doi.org/10.1016/j.lwt.2022.113458

Li, M., Xu, X., Bi, S., Pan, X., Lao, F., & Wu, J. (2023). Identification and validation of core microbes associated with key aroma formation in fermented pepper paste (*Capsicum annuum* L.). *Food Research International, 163*, Article 112194. https://doi.org/10.1016/j.foodres.2022.112194

Li, M., Zhan, P., Wang, P., Tian, H., Geng, J., & Wang, L. (2023). Characterization of aroma-active compounds changes of Xiecun Huangjius with different aging years based on odor activity values and multivariate analysis. *Food Chemistry, 405*, Article 134809. https://doi.org/10.1016/j.foodchem.2022.134809

Li, N., Li, G., Li, A., & Tao, Y. (2023). Synergy effect between fruity esters and potential odorants on the aroma of Hutai-8 rose wine revealed by threshold, *S* -curve, and σ–τ plot methods. *Journal of Agricultural and Food Chemistry, 71*(37), 13869–13879. https://doi.org/10.1021/acs.jafc.3c03733

Liu, C., Wang, C., Zheng, T., Zhao, M., Gong, W., Wang, Q., Yan, L., & Zhang, W. (2022). Characterization of key odor-active compounds in Sun-dried black tea by sensory and instrumental-directed flavor analysis. *Foods, 11*(12), 1740. https://doi.org/10.3390/foods11121740

Liu, J., Yu, Y., Ye, X., Wang, K., Zhu, Y., Wang, Y., Yu, Z., & Liu, P. (2023). Exploring the ecological interactions of Bacillus and their contribution to characteristic aroma components in Zhenjiang aromatic vinegar. *Food Bioscience, 54*, Article 102900. https://doi.org/10.1016/j.fbio.2023.102900

Liu, Z., Cai, S., Zhang, S., Xiao, Y., Devahastin, S., Guo, C., Wang, Y., Wang, T., & Yi, J. (2023). A systematic review on fermented chili pepper products: Sensorial quality, health benefits, fermentation microbiomes, and metabolic pathways. *Trends in Food Science & Technology, 141*, Article 104189. https://doi.org/10.1016/j.tifs.2023.104189

Ma, L., Gao, M., Zhang, L., Qiao, Y., Li, J., Du, L., Zhang, H., & Wang, H. (2022). Characterization of the key aroma-active compounds in high-grade Dianhong tea using GC-MS and GC-O combined with sensory-directed flavor analysis. *Food Chemistry, 378*, Article 132058. https://doi.org/10.1016/j.foodchem.2022.132058

Ma, Y., Tang, K., Xu, Y., & Thomas-Danguin, T. (2021). Perceptual interactions among food odors: Major influences on odor intensity evidenced with a set of 222 binary mixtures of key odorants. *Food Chemistry, 353*, Article 129483. https://doi.org/10.1016/j.foodchem.2021.129483

Miyazawa, T., Gallagher, M., Preti, G., & Wise, P. M. (2008). Synergistic mixture interactions in detection of Perithreshold odors by humans. *Chemical Senses, 33*(4), 363–369. https://doi.org/10.1093/chemse/bjn004

Ni, R., Yan, H., Tian, H., Zhan, P., & Zhang, Y. (2022). Characterization of key odorants in fried red and green huajiao (*Zanthoxylum bungeanum* maxim. And *Zanthoxylum schinifolium* sieb. Et Zucc.) oils. *Food Chemistry, 377*, Article 131984. https://doi.org/10.1016/j.foodchem.2021.131984

Niu, Y., Wang, R., Xiao, Z., Zhu, J., Sun, X., & Wang, P. (2019). Characterization of ester odorants of apple juice by gas chromatography-olfactometry, quantitative measurements, odour threshold, aroma intensity and electronic nose. *Food Research International, 120*, 92–101. https://doi.org/10.1016/j.foodres.2019.01.064

Pang, X., Guo, X., Qin, Z., Yao, Y., Hu, X., & Wu, J. (2012). Identification of aroma-active compounds in Jiashi muskmelon juice by GC-O-MS and OAV calculation. *Journal of Agricultural and Food Chemistry, 60*(17), 4179–4185. https://doi.org/10.1021/jf300149m

Pegiou, E., Siccama, J. W., Mumm, R., Zhang, L., Jacobs, D. M., Lauteslager, X. Y., … Hall, R. D. (2023). Metabolomics and sensory evaluation of white asparagus ingredients in instant soups unveil important (off-)flavours. *Food Chemistry, 406*, Article 134986. https://doi.org/10.1016/j.foodchem.2022.134986

Qian, X., Ling, M., Sun, Y., Han, F., Shi, Y., Duan, C., & Lan, Y. (2024). Decoding the aroma characteristics of icewine by partial least-squares regression, aroma reconstitution, and omission studies. *Food Chemistry, 440*, Article 138226. https://doi.org/10.1016/j.foodchem.2023.138226

Saison, D., De Schutter, D. P., Uyttenhove, B., Delvaux, F., & Delvaux, F. R. (2009). Contribution of staling compounds to the aged flavour of lager beer by studying their flavour thresholds. *Food Chemistry, 114*(4), 1206–1215. https://doi.org/10.1016/j.foodchem.2008.10.078

Shen, S., Zhang, J., Sun, H., Zu, Z., Fu, J., Fan, R., Chen, Q., Wang, Y., Yue, P., Ning, J., Zhang, L., & Gao, X. (2023). Sensomics-assisted characterization of fungal-flowery aroma components in fermented tea using *Eurotium cristatum*. *Journal of Agricultural and Food Chemistry, 71*(48), 18963–18972. https://doi.org/10.1021/acs.jafc.3c05273

Sieiro-Sampedro, T., Figueiredo-González, M., González-Barreiro, C., Simal-Gandara, J., Cancho-Grande, B., & Rial-Otero, R. (2019). Impact of mepanipyrim and tetraconazole in Mencía wines on the biosynthesis of volatile compounds during the winemaking process. *Food Chemistry, 300*, Article 125223. https://doi.org/10.1016/j.foodchem.2019.125223

Sun, J., Sun, B., Ren, F., Chen, H., Zhang, N., & Zhang, Y. (2020). Characterization of key odorants in Hanyuan and Hancheng fried pepper (*Zanthoxylum bungeanum*) oil. *Journal of Agricultural and Food Chemistry, 68*(23), 6403–6411. https://doi.org/10.1021/acs.jafc.0c02026

Tan, F., Wang, P., Zhan, P., & Tian, H. (2022). Characterization of key aroma compounds in flat peach juice based on gas chromatography-mass spectrometry-olfactometry (GC-MS-O), odor activity value (OAV), aroma recombination, and omission experiments. *Food Chemistry, 366*, Article 130604. https://doi.org/10.1016/j.foodchem.2021.130604

Tian, Y., Mu, Y., Su, W., & Qi, Q. (2023). Correlation between microbiota and volatile flavor compounds during inoculated fermentation of Chinese pickled pepper (*Paojiao*). *LWT - Food Science and Technology, 178*, Article 114642. https://doi.org/10.1016/j.lwt.2023.114642

Wang, C., Wang, Y., Chen, J., Liu, L., Yang, M., Li, Z., Wang, C., Pichersky, E., & Xu, H. (2022). Synthesis of 4-methylvaleric acid, a precursor of pogostone, involves a 2-isobutylmalate synthase related to 2-isopropylmalate synthase of leucine biosynthesis. *New Phytologist, 235*(3), 1129–1145. https://doi.org/10.1111/nph.18186

Wang, M.-Q., Ma, W.-J., Shi, J., Zhu, Y., Lin, Z., & Lv, H.-P. (2020). Characterization of the key aroma compounds in Longjing tea using stir bar sorptive extraction (SBSE) combined with gas chromatography-mass spectrometry (GC–MS), gas chromatography-olfactometry (GC-O), odor activity value (OAV), and aroma

recombination. *Food Research International, 130*, Article 108908. https://doi.org/10.1016/j.foodres.2019.108908

Weterings, M., Bodnár, I., Boom, R. M., & Beyrer, M. (2020). A classification scheme for interfacial mass transfer and the kinetics of aroma release. *Trends in Food Science & Technology, 105*, 433–448. https://doi.org/10.1016/j.tifs.2019.04.012

Xiao, Y., Zhang, S., Liu, Z., Wang, T., Cai, S., Chu, C., Hu, X., & Yi, J. (2023). Effect of inoculating *Pichia* spp. starters on flavor formation of fermented chili pepper: Metabolomics and genomics approaches. *Food Research International, 173*, Article 113397. https://doi.org/10.1016/j.foodres.2023.113397

Yang, X., Hu, W., Xiu, Z., Jiang, A., Yang, X., Sarengaowa, J., & Y., Guan, Y., & Feng, K.. (2020). Comparison of northeast sauerkraut fermentation between single lactic acid bacteria strains and traditional fermentation. *Food Research International, 137*, Article 109553. https://doi.org/10.1016/j.foodres.2020.109553

Ye, Z., Shang, Z., Li, M., Qu, Y., Long, H., & Yi, J. (2020). Evaluation of the physiochemical and aromatic qualities of pickled Chinese pepper (*Paojiao*) and their influence on consumer acceptability by using targeted and untargeted multivariate approaches. *Food Research International, 137*, Article 109535. https://doi.org/10.1016/j.foodres.2020.109535

Ye, Z., Shang, Z., Li, M., Zhang, X., Ren, H., Hu, X., & Yi, J. (2022). Effect of ripening and variety on the physiochemical quality and flavor of fermented Chinese chili pepper (*Paojiao*). *Food Chemistry, 368*, Article 130797. https://doi.org/10.1016/j.foodchem.2021.130797

Ye, Z., Shang, Z., Zhang, S., Li, M., Zhang, X., Ren, H., Hu, X., & Yi, J. (2022). Dynamic analysis of flavor properties and microbial communities in Chinese pickled chili pepper (*Capsicum frutescens* L.): A typical industrial-scale natural fermentation process. *Food Research International, 153*, Article 110952. https://doi.org/10.1016/j.foodres.2022.110952

Zhang, M., Yin, Y., Li, Y., Jiang, Y., Hu, X., & Yi, J. (2023). Chemometric classification of apple cultivars based on physicochemical properties: Raw material selection for processing applications. *Foods, 12*(16), 3095. https://doi.org/10.3390/foods12163095

Zhang, S., Xiao, Y., Jiang, Y., Wang, T., Cai, S., Hu, X., & Yi, J. (2022). Effects of brines and containers on flavor production of Chinese pickled chili pepper (*Capsicum frutescens* L.) during natural fermentation. *Foods, 12*(1), 101. https://doi.org/10.3390/foods12010101

Zhao, Y., Wei, W., Tang, L., Wang, D., Wang, Y., Wu, Z., & Zhang, W. (2021). Characterization of aroma and bacteria profiles of Sichuan industrial paocai by HS-SPME-GC-O-MS and 16S rRNA amplicon sequencing. *Food Research International, 149*, Article 110667. https://doi.org/10.1016/j.foodres.2021.110667

Zhu, J., Chen, F., Wang, L., Niu, Y., & Xiao, Z. (2017). Evaluation of the synergism among volatile compounds in oolong tea infusion by odour threshold with sensory analysis and E-nose. *Food Chemistry, 221*, 1484–1490. https://doi.org/10.1016/j.foodchem.2016.11.002