



## GC-MS and GC×GC-ToF-MS analysis of roasted / broth flavors produced by Maillard reaction system of cysteine-xylose-glutamate

An-Ran Zheng<sup>a,b</sup>, Chao-Kun Wei<sup>a,b,c,\*\*</sup>, Dun-Hua Liu<sup>a</sup>, Kiran Thakur<sup>b,c</sup>, Jian-Guo Zhang<sup>b,c</sup>, Zhao-Jun Wei<sup>b,c,\*</sup>

<sup>a</sup> School of Food and Wine & School of Agriculture, Ningxia University, Yinchuan, 750021, PR China

<sup>b</sup> School of Biological Science and Engineering, Collaborative Innovation Center for Food Production and Safety, North Minzu University, Yinchuan, 750021, PR China

<sup>c</sup> School of Food and Biological Engineering, Hefei University of Technology, Hefei, 230009, PR China

### ARTICLE INFO

Handling Editor: Professor Aiqian Ye

#### Keywords:

Maillard reaction  
Cysteine  
Glutamate  
Flavor  
Volatile components

### ABSTRACT

Maillard reaction products (MRPs) with roasted/broth flavors were prepared and analyzed for the resulting flavor differences. The identification of volatile compounds in MRPs was carried out by GC-MS and GC × GC-ToF-MS. A total of 88 compounds were identified by GC-MS; 130 compounds were identified by GC × GC-ToF-MS, especially acids and ketones were identified. Principal component analysis (PCA) was used to visualize the volatile compounds, and the roasted/broth flavors were differentiated. The contents and types of pyrazines were more in roasted flavors; thiol sulfides and thiophenes were more in broth flavors. All in all, the differences in volatile compounds producing roasted/broth flavors were studied through the cysteine-xylose-glutamate Maillard reaction system, which provided a theoretical basis for the future use of Maillard reaction to simulate meat flavor.

### 1. Introduction

Maillard reaction (MR) is also defined as a non-enzymatic browning reaction, which usually occurs in the reaction between amino groups (such as amino acids, peptides, or proteins) and carbonyl groups (such as reducing sugars) during food thermal processing or food storage (Ma et al., 2022). The meat flavor prepared by MR has been widely used in meat processing, flavor improvement, and aroma enhancement. In MR, the substrate has a significant influence on the final flavor. For example, cysteine, which is an important precursor for the formation of sulfur-containing compounds in meat flavor, has been shown to have an irreplaceable effect on the production of meat flavor (Cerny, 2007). *L*-cysteine is degraded by Strecker to produce ammonia, acetaldehyde, and hydrogen sulfide. These components will trigger a series of reactions in the subsequent MR to form various meaty compounds such as cysteine-ribose (Cerny, 2007) and cysteine-xylose (Li and Liu, 2022) reaction system. Ribose can also participate in the MR to produce meat flavor, the thermal stability is the lowest among common reducing sugars and is

prone to pyrolysis (Rose et al., 2015). The products were mainly umami and thick flavor when glutamate participates in the reaction, while glycine produces caramel flavor (Wong et al., 2010). However, the cysteine-reducing sugar MR system produces a flavor that is closer to that of roasted meat (Xue et al., 2019).

Glutamate is a common food flavoring agent widely distributed in nature. At present, glutamic acid-based MR is mainly used to improve the flavor and umami of meat products (Charve et al., 2018). Zhou et al. (2016) analyzed the effects of reactant ratio, temperature, and time on color and flavor of Maillard reaction products (MRPs) of the *L*-ascorbic acid/*L*-glutamic acid system, and the results showed that excessive glutamic acid can produce a strong roasting aroma and nut aroma. Yu et al. (2013) used isotope labeling method to study the MR of glutamic acid and ascorbic acid to produce pyrazines, and evaluated the aroma of the pyrazines produced. It was found that the reaction product can produce roasted burnt aroma and nutty aroma.

However, the flavor produced by cysteine-xylose MR system was closer to that of roasted meat. After glutamic acid participates in the

\* Corresponding author. School of Biological Science and Engineering, Collaborative Innovation Center for Food Production and Safety, North Minzu University, Yinchuan, 750021, PR China.

\*\* Corresponding author. School of Food and Wine & School of Agriculture, Ningxia University, Yinchuan, 750021, PR China.

E-mail addresses: [zhenganran17@163.com](mailto:zhenganran17@163.com) (A.-R. Zheng), [weichaokun@nxu.edu.cn](mailto:weichaokun@nxu.edu.cn) (C.-K. Wei), [dunhualiu@126.com](mailto:dunhualiu@126.com) (D.-H. Liu), [kumarikiran@hfut.edu.cn](mailto:kumarikiran@hfut.edu.cn) (K. Thakur), [zhangjianguo@hfut.edu.cn](mailto:zhangjianguo@hfut.edu.cn) (J.-G. Zhang), [zjwei@hfut.edu.cn](mailto:zjwei@hfut.edu.cn) (Z.-J. Wei).

<https://doi.org/10.1016/j.crf.2023.100445>

Received 2 November 2022; Received in revised form 31 December 2022; Accepted 12 January 2023

Available online 14 January 2023

2665-9271/© 2023 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

cysteine-xylose MR system, the flavor of the product tends to change from grilled meat flavor to broth flavor (Wei et al., 2022). Glycine was involved in the cysteine-xylose MR system which also had a similar trend (Cao et al., 2017). In addition, Wei et al. (2019) studied the effect of protease hydrolysates on cysteine-xylose MR system and found that protease hydrolysates were beneficial to enhance the umami and kokumi taste of MRPs. Therefore, by regulating the cysteine-xylose-other amino acids MR system, it is possible to simulate the flavors of roasted/broth. Wu et al. (2021) analyzed the flavor substances of 6 Chinese dry-cured Spanish mackerel by electronic nose, gas chromatography olfaction and GC × GC - ToF - MS, and the results showed that the drying time was beneficial to the formation of key flavor substances. Zeng et al. (2020) analyzed the flavor substances of *Cordyceps militaris* chicken soup by GC-MS, GC × GC - ToF - MS, and GC-IMS, proving that the application of enzymatic hydrolysis technology is conducive to the dissolution of protein in the soup and the formation of flavor. Therefore, the combined analysis of GC-MS and GCGC is conducive to the comparison of subtle differences in flavor components.

The purpose was to use the cysteine-xylose-glutamate MR system to generate roasted/broth flavors and to evaluate the resulting volatility by GC-MS and GC × GC-ToF-MS. This study could reveal the differences in the flavors of roasted/broth produced by the Maillard system.

## 2. Materials and methods

### 2.1. Materials and chemicals

L-cysteine (99%), L-glutamic acid (98%), D-xylose (98%) were purchased from Shanghai Macleans Biochemical Technology Co., Ltd. Normal alkanes (C<sub>7</sub>–C<sub>30</sub>), methanol, and 1,2-dichlorobenzene were purchased from Beijing Chemical Reagent Company. Sodium dihydrogen phosphate (NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O), disodium hydrogen phosphate (Na<sub>2</sub>HPO<sub>4</sub>·12H<sub>2</sub>O), and other chemicals were purchased from Shanghai Macleans Biochemical Technology Co., Ltd., which were of analytical grade.

### 2.2. Preparation of Maillard reaction products

Preparation of roasted flavor MRPs.

3.0 g of xylose, 0.2 g of cysteine, 1.0 g of glutamic acid were added into 30.0 mL of distilled water and the pH of the reaction solution was adjusted to 7 with phosphate buffer solution, and transferred into an ampoule bottle and stored at 90 °C. The reaction was carried out for 1.5 h.

Preparation of broth flavor MRPs.

3.0 g of xylose, 0.2 g of cysteine, 3.0 g of glutamic acid were added into 30.0 mL of distilled water, and the pH of the reaction solution was adjusted to 6 with phosphate buffer solution, and transferred into an ampoule bottle and stored at 100 °C, and allowed to react for 2 h.

### 2.3. E-nose analysis

The analysis of roasted/broth flavors by E-nose (PEN 3 Airsense Analytics GmbH, Schwerin, Germany) was performed based on the method of Yang et al. (2016) with appropriate modifications. 0.1 g of the roasted/broth flavors sample was placed in 20 mL headspace vial containing 5 mL of distilled water and allowed to equilibrate at 25 °C for 30 min. The sampling time was set as 1 s/group, the sensor self-cleaning time was set as 300 s, the sensor zeroing time was set as 10 s, the sample preparation time was set as 5 s, the injection flow rate was set as 300 mL/min, and the analysis sampling time was set as 120 s. The samples were detected by headspace aspirating method at room temperature of 20 °C, and each sample was detected in parallel three times (Zhu et al., 2016).

### 2.4. GC-MS analysis

For SPME processing, 0.5 g of sample was added to a 20 mL headspace vial. The CAR/PDMS fiber head was aged in advance according to the instructions, and then SPME extraction was performed on each sample. The samples were equilibrated at 50 °C for 10 min, adsorbed in headspace for 20 min, and analyzed.

Gas chromatography conditions: Chromatographic column was HP-5 MSUI (30 m × 0.25 mm, 0.25 μm) capillary column; injection volume 1 μL; Carrier gas: helium (99.999%), flow rate: 1 mL/min; Injection port temperature: 250 °C; The column temperature program was maintained at 40 °C for 2 min, 40–100 °C at 2 °C/min, 100–150 °C at 4 °C/min, held for 2 min; raised to 280 °C at 20 °C/min, held for 10 min. Mass spectrometry conditions: EI source, ionization voltage 70 eV; ion source temperature 230 °C; mass scanning range m/z 40–450.

RI: C<sub>7</sub>–C<sub>30</sub> normal alkanes were injected under the same GC-MS conditions.

$$m = \rho \times V \times A/A_0 \times 1000$$

where m is the absolute content of the identified compound/ng; ρ is the concentration of 1,2-dichlorobenzene (50 μg/μL); V is the volume of 1,2-dichlorobenzene (2 μL), A is the peak area of the identified compound; A<sub>0</sub> is the area of 1,2-dichlorobenzene. The quantification of volatile flavor substances is achieved by calculating m value with internal standard method, while the qualitative analysis is achieved through MS and RI.

### 2.5. GC × GC-ToF-MS analysis

SPME sample processing and internal standard quantification were consistent with GC-MS. The parameters of GC × GC-ToF-MS were carried out according to the method of Li et al. (2021) with some modifications: DB-WAX (30 m × 250 μm × 0.25 μm) and DB-17MS (2 m × 100 μm × 0.10 μm), respectively were used as first and second dimension columns. Helium (99.999%) was used as the carrier gas at a flow rate of 1.0 ml/min and an injection volume of 1 μL. The initial temperature was kept at 40 °C for 3 min; the temperature was raised to 250 °C at 5 °C/min and held for 5 min, the inlet temperature was 250 °C; the column temperature of the second dimension was 255 °C, and the desorption temperature was always lower than that of the first dimension. The ion source temperature was set to 250 °C, and the ionization potential of the mass spectrometer was 70 eV. Spectra were collected in the mass range of m/z 33–500 at a collection rate of 50/s.

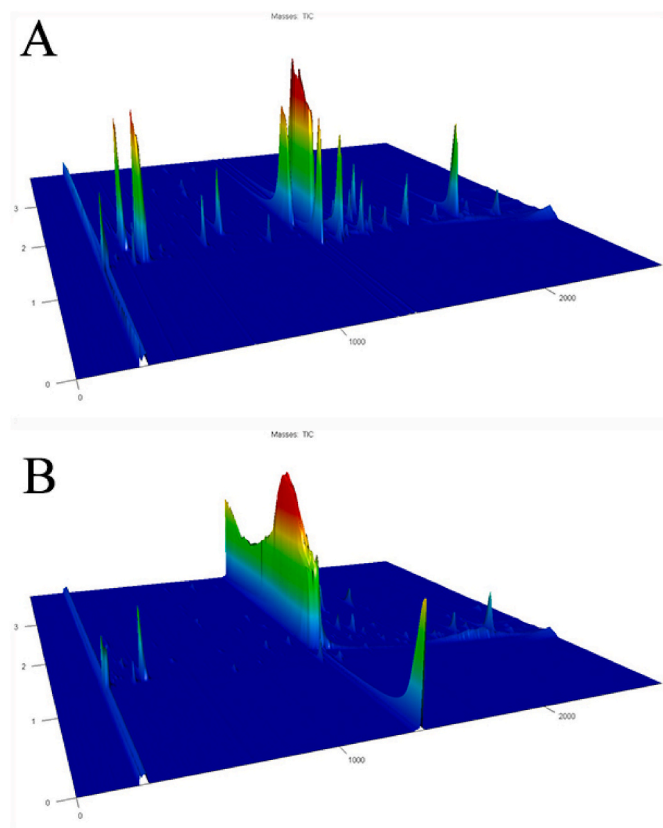
### 2.6. Statistical analysis

All the experiments were repeated three times and data were analyzed by ANOVA with SPSS Statistics 20.0 and expressed as mean ± standard deviation (n = 3), (P < 0.05). Linear retention indices (RI) and mass spectra of GC × GC-ToF-MS and GC-MS data were identified by comparison with the NIST database. The Omicshare Cloud platform (<http://www.omicshare.com/tools/>), the Microbiology Letter platform (<http://www.bioinformatics.com.cn/>), and the Metware Cloud platform (<https://cloud.metware.cn/>) were used for differential substance related data analysis.

## 3. Results and discussion

### 3.1. GC × GC-ToF-MS 3D plots of roasted/broth flavors MRPs

GC × GC-ToF-MS was used for the three-dimensional chromatographic analysis of the volatile components of the roasted/broth flavors prepared by MR system (Fig. 1). Column I represented the retention time of the compound, and Column II represented the chemical polarity. As shown in Fig. 1, the highest number of base peaks in the roasted flavor



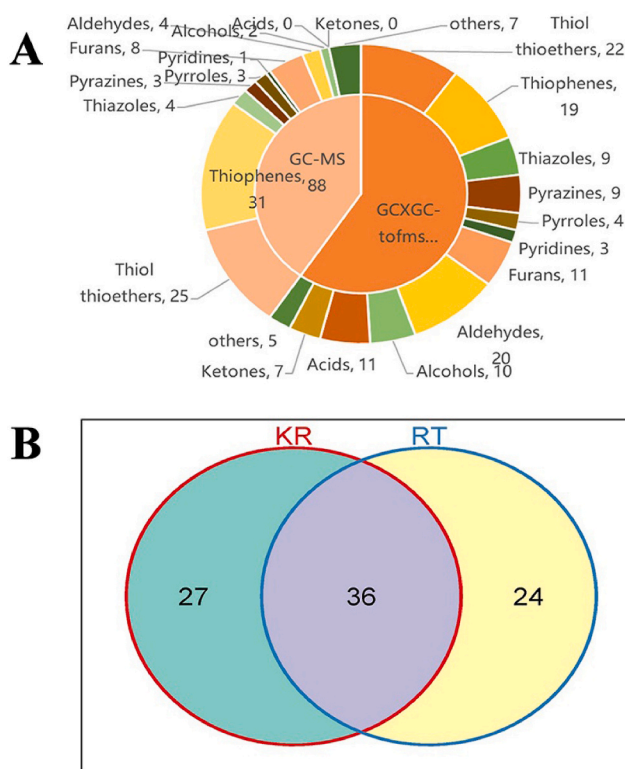
**Fig. 1.** 3D-chromatograms of volatile compounds (vocs) in roasted/broth flavors were analyzed by GC × GC-ToF-MS. Column I indicates the retention time of the compound and Column II indicates the chemical polarity of the compound. KR (roasted flavor), RT (broth flavor).

indicated that more volatile components were produced, while the higher number of base peaks in the broth flavor indicated that some volatile components were present in broth flavor at higher levels than those in the roasted flavor (Table S1). The results indicated some differences in both types and contents of volatile components in the roasted/broth flavors.

### 3.2. GC × GC-ToF-MS and GC-MS analysis of volatile compounds

Roasted/broth flavors products were prepared in cysteine-xylose-glutamate MR system. The volatile compounds were identified by GC × GC-ToF-MS and GC-MS, and the results were shown in Table S1 and Table S2. It can be seen from Fig. 2 that a total of 130 volatile compounds have been identified in the roasted/broth flavors, including 12 categories: 22 thiol thioethers, 19 thiophenes, 9 thiazoles, 9 pyrazines, 4 pyrroles, 3 pyridines, 11 furans, 20 aldehydes, 10 alcohols, 11 acids, 7 ketones, and 5 others. A total of 88 volatile compounds were identified by GC-MS, including 10 classes: 25 thiol sulfides, 31 thiophenes, 4 thiazoles, 3 pyrazines, 3 pyrroles, 1 pyridines, 8 furans, 4 aldehydes, 2 alcohols, and 7 others.

According to the data measured by GC × GC-ToF-MS, acid compounds were small part of volatile compounds found in roasted/broth flavors, and the main acid compounds identified in roasted/broth flavors were decanoic acid, furoic acid, and isovaleric acid, of which decanoic acid had a fatty flavor. However, they were not identified in GC-MS. They were mostly oxidized by aldehydes, which had a high flavor threshold and usually had a slight sensory impact on the overall odor of the roasted/broth flavor (Zhao et al., 2019). Among the aldehydes, the most abundant aldehyde in the broth flavor was nonanal, which has the aroma of rose and citrus, responsible for a fruity aroma of broth, and the



**Fig. 2.** Types of volatile components in roasted/broth flavors under two identification methods (a) and the difference in volatile species identified by (Table S2).

lowest content was methoxyacetaldehyde (Tian et al., 2019). The most abundant aldehyde in the roasted flavors was *E,E*-2,4-decadienal, *E,E*-2,4-decadienal, as an unsaturated aldehyde had the smell of fat and seaweed. The lowest content was observed for pyruvaldehyde. It was also found in GC-MS that nonanal was the most abundant aldehyde in broth flavor. Benzaldehyde is produced by Strecker degradation of amino acid and has a nutty and fruity aroma (Loch et al., 2016). The content of benzaldehyde and phenylacetaldehyde in grilled meat flavor was higher than that of broth flavor, which is due to the fact that grilled meat flavor reacts at higher reaction temperature and lower water activity during the preparation process, which leads to the easy Strecker degradation of amino acids (Hidalgo and Zamora, 2019). Aldehydes are mainly the main degradation products of lipid oxidation, but MR-induced degradation of amino acids also produced some aldehydes. Its strong volatility and low odor threshold exerted a strong synergistic effect with many other substances. Altogether, they give the food a burnt, meaty, and delicate fragrance. Phenylacetaldehyde is also used as one of Strecker aldehydes, contributing to the smell of honey. It was identified in all samples, but its content was higher in broth flavor. GC-MS could not identify the presence of phenylacetaldehyde. Ketones mainly come from amino acid degradation, MR, and fat oxidation (Zhang et al., 2016). The results showed that the substances had less effect on the flavor, contributing to the meat flavor and sweetness of the roast meat/broth flavor. A total of seven ketones were identified by GC × GC-ToF-MS, while the broth flavor (13.51 µg/L) contained more than the roast flavor (15.41 µg/L), and 2-methyl-3-hydroxy-4-pyrone was only detected in the roast flavor. Some ketones can also be used as intermediates for certain heterocyclic compounds. The transformation of these intermediates with increasing reaction time was responsible for the decrease of ketones in the broth flavor. However, using GC-MS, no ketones were identified, probably because GC × GC-ToF-MS has higher sensitivity.

Nitrogen-containing compounds are mainly composed of pyrazines,

pyrroles, pyridines, and thiazoles. Pyrazine compounds are very important volatile flavor substances formed by the condensation of Strecker aldehydes and ketones, with a low flavor threshold, and generally have flavors such as nuts, roasted aromas, and burnt aromas (An and Kimpe, 2009). The  $\alpha$ -aminoketone pathway is one of the most important pathways for the formation of pyrazine compounds (Yaylayan, 2003). Dicarbonyl compounds and amino compounds are degraded and condensed by Strecker to form  $\alpha$ -amino ketones (Yaylayan, 2003).  $\alpha$ -Amino ketones are further condensed or condensed with other amino ketones to form a dihydropyridazine and finally oxidized to pyrazine (Adab et al., 2021). In the identification of GC  $\times$  GC-ToF-MS, a total of 9 kinds of pyrazine compounds were identified, and 2-methylpyrazine, 2,5-dimethylpyrazine, 2,6-dimethylpyrazine and 2,6-dimethylpyrazine were found in high content. At GC  $\times$  GC ToF-MS, 9 pyrazine compounds were identified, including 2-methylpyrazine, 2,5-dimethylpyrazine, 2,6-dimethylpyrazine and 2-ethyl-5-methylpyrazine; 3 pyrazine compounds were identified by GC-MS, namely pyrazine, 2-methylpyrazine and 2,5-dimethylpyrazine. Both identification methods found the existence of 2-methylpyrazine and 2,5-dimethylpyrazine in grilled meat flavor, indicating that they are the main volatile substances in roasted flavor. With the increase of pH, the type and content of pyrazine compounds were increased (Shen et al., 2019). Geib et al. (2018) reported the influence of pH changes on flavor substances in cysteine - furanone system, and observed that pyrazines were only detected at pH 7.1, and not at pH 5.1 and pH 2.2. It was detected that the ammonia produced by the degradation of cysteine had a higher activity at pH 7.1, so pyrazines were detected. The identification of higher species and content of pyrazines in roasted flavor than in broth flavor is the main reason for the lower species and content of pyrazines in broth flavors because broth flavors are prepared under weakly acidic and low-temperature conditions when the amino groups of cysteine and glutamic acid are protonated in a partially acidic environment and are unable to undergo nucleophilic attack (Cui et al., 2021).

Furan compounds are a class of oxygen-containing heterocyclic compounds, one of the characteristic volatile compounds formed in MR and caramelization reaction. They are mainly formed by reducing sugar-amino acids through a series of steps of dehydration, decomposition, and cyclization (Shen et al., 2021). Furans has unique sensory properties and can significantly improve the flavor of food (Zhang et al., 2015). Yahya et al. (2014) found that furan and its derivatives are easy to produce the characteristic aroma of barbecue, cooked meat, coffee, and marine food. In Table S1, furfural was the most abundant furan compound with a baked potato flavor and a sweet taste. The content in the broth flavor (271.35  $\mu\text{g/L}$ ) was higher than in the roast flavor (207.92  $\mu\text{g/L}$ ), and the difference between them was mainly since the broth flavor was prepared under weakly acidic conditions, where, in addition to the occurrence of the MR, the three water molecules in the xylose molecule were removed under acidic conditions to form the five-member heterocyclic compound furfural (D. Y. Li et al., 2020).

The main thiazole compounds were 2-acetyl thiazole, 4-ethyl-2,5-dimethyl thiazole, and 2-methyl thiazole. The content of 2-acetylthiazole in the roasted flavor was higher than that in the broth flavor, possibly because the pH of the roasted flavor was higher than that of the broth flavor. Li and Liu (2021) reported that the content of 2-acetylthiazole increased with the increase of pH, and effect of pH on MR - derived aroma volatiles from cooked meat system causanda nutty and popcorn smell.

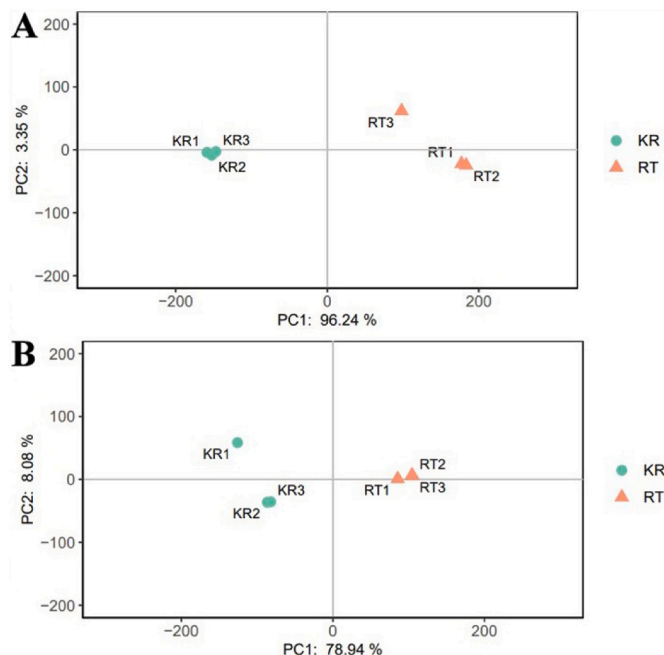
The sulfur compounds have a very low odor threshold, and generally have meat, onion, garlic, and sulfide flavors, which is the basic component of various meat flavors. In the roasted/broth flavors, 22 volatile compounds were identified by GC  $\times$  GC-ToF-MS, which were basically the same as those identified by GC-MS. However, 2-mercaptoethanol and ethylene glycol ethyl ether were not identified by GC-MS, which is probably due to the better peak resolution and higher sensitivity of GC  $\times$  GC-ToF-MS. The content of thioether compounds in the two groups was as high as 5.99%, 2-thiophene mercaptan, bis (2-methyl-

3-furyl) disulfide, 2-furfuryl mercaptan, 2-thiophenyl mercaptan, bis (2-thiophenyl) disulfide and 3-mercapto-2-butan-2-one were the main detected components. Among them, 2-methyl-3-furanthiol and 2-furanthiol had a lower odor threshold with a roast, coffee, and meat aroma. Bis (2-methyl-3-furyl) disulfides and difuryl disulfides are dimers of 2-methyl-3-furanethiol and 2-furanethiol, respectively, and are also important compounds in broth flavor (Mottram et al., 1995). 2-Methyl-3-furanethiol and furfuranethiol were found in cooked beef, pork, and chicken (Jayasena et al., 2013), as well as in the reaction products of cysteine and ribose and the thermal degradation products of thiamine (Farmer and Mottram, 2010). Cerny and Guntz-Dubini (2013) found the model reaction of cysteine and xylose and found that the carbon backbone of furfuryl mercaptan comes from xylose. Cyclization produces furfural, which reacts with  $\text{H}_2\text{S}$  formed by the degradation of cysteine to obtain furfuryl mercaptan. In addition to the above, the carbon skeleton of 2-methyl-3-furan mercaptan can also be composed of fragments from xylose and cysteine. Tan and Ai (2012) reported the effect of reaction temperature and time on the formation of fragrance compounds in the ascorbic acid-cysteine mode reaction and found that when the reaction temperature reaches 160  $^\circ\text{C}$ , 2-methyl-3-furanthiol, furfurylthiol, and 2-thiophene contents were reduced to an undetectable level. It can be seen that the formation of these substances is not only strongly dependent on pH but also greatly dependent on temperature. It may be that the MR increases the reactivity of the fragrance substances at higher temperatures, and the sulfhydryl groups attached to the molecular structure are easily removed at high temperatures and converted into the corresponding furan and thiophene derivatives or they are intermediate with other activities. The body polymerizes into macromolecular compounds.

Temperature is a prerequisite for the production of thiophenes at the beginning of MR, and thiophenes play an important role in the overall meat flavor (Chen et al., 2018). A total of 19 thiophene compounds were identified by GC  $\times$  GC-ToF-MS in the roasted/broth flavors, and thiophene compounds usually have onion, burnt, coffee flavor, etc. The thiophenes can be obtained by the reaction of furan and  $\text{H}_2\text{S}$ , and the furan compounds can be obtained by the formation of the corresponding Amadori rearrangement products from amino acids and reducing sugars, followed by 1,2-enolization and cyclization. Among them, 2-methyl, thieno[2,3-b]and thiophene, and 3,4-dimethylthiophene are the compounds with high content in roast/broth flavor. The content of the thiophene compounds identified by GC  $\times$  GC-ToF-MS was higher for the roasted meat flavor (238.67  $\mu\text{g/L}$ ) than for the broth flavor (188.12  $\mu\text{g/L}$ ).

### 3.3. PCA of volatile compounds identified by GC $\times$ GC-ToF-MS and GC-MS

Principal component analysis (PCA) is a statistical analysis method for multivariate analysis (Y. Li et al., 2020). The data base for GC  $\times$  GC-ToF-MS principal component analysis is based on the relative contents of the compounds in Table S1. The data for principal component analysis by GC-MS are based on the absolute contents of the compounds as shown in Table S2. The PCA score graphs was shown in Fig. 3. In Fig. 3 A, the variance contribution of the first principal component was 96.24%, and the variance contribution of the second principal component was 3.35%, and the cumulative variance contribution was 99.59%. In Fig. 3 B, the variance contribution of the first principal component was 78.94%, the variance contribution of the second principal component was 8.08%, and the cumulative variance contribution was 87.02% (Table S2). Although only 88 volatile components were detected by GC-MS, their contribution to variance was 99.59%, which could represent the whole information of the samples. As it can be seen from Fig. 3, the roasted/broth flavors were located farther apart, indicating that the volatiles produced by roasted/broth flavors were significantly different.



**Fig. 3.** Principal component analysis (PCA) of roasted/broth flavors was performed based on the volatile compounds identified by GC × GC-ToF-MS (a) and GC-MS (b). Score plot of peak area by GC × GC-ToF-MS and absolute content intensity by GC-MS for roasted/broth flavors. KR (roasted flavor), RT (broth flavor).

**3.4. Electronic nose analysis**

As shown in Fig. 4 A, the sensor response radar chart of the electronic nose to the roasted/broth flavors was prepared by MR system of cysteine-xylose-glutamate. In the roasted/broth flavors W1W, the sensor response value was the highest, followed by W1S, W2S, and W5S, indicating that most flavor components produced by MR were sulfides, alcohols, and nitrogen oxides. These results are similar to the results of Scaman et al. (2006) in the MR of cysteine in a heat-responsive meat flavor production system, which pointed out that sulfide plays an important role in meat flavor. In fact, some sulfur-containing compounds are important sources of essential meat flavor, such as H<sub>2</sub>S, methyl mercaptan, 2,2-dimethylpropyl mercaptan, and 3-methyl-2-butane mercaptan. Compared with the roasted meat flavor, the response value of the broth flavor sensor was higher, which may be due to the fact that the broth flavor was prepared at a lower pH and reaction

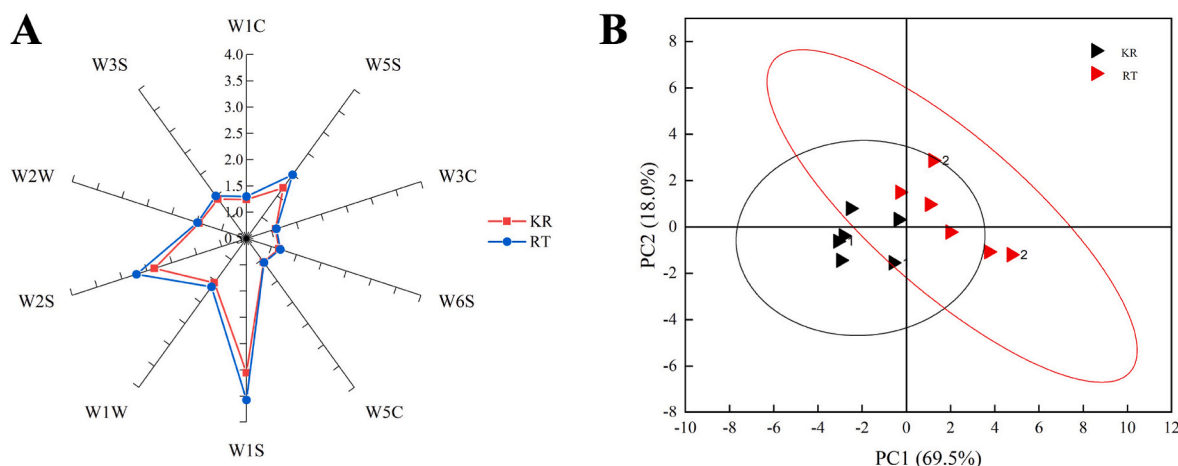
temperature. PCA is a linear transformation of the original data vector through dimensionality reduction, and the whole process does not contain sample information (Gabriela et al., 2021). This method obtains the most important factors and the highest contribution rate to find differences between samples. The results of the electronic nose data for the roasted/broth flavor are shown in Fig. 4 A. It can be seen from Fig. 4 B that the variance contribution rates of PC1 and PC2 were 69.5% and 18.0% respectively, and the total variance contribution rate was 87.5%, which was greater than 85%, indicating that PC1 and PC2 can represent the main information of the sample and can better explain the sample information. The broth flavor was mainly distributed in the first and fourth quadrants, and the flavor of roasted meat was mainly distributed in the second and third quadrants, but some of the flavors of roasted/broth were distributed in the second quadrant, indicating that the overall flavor of the two was somewhat different, but there were also similarities. The combination of electronic nose technology and PCA analysis can qualitatively analyze the aroma in the flavor of roasted/broth, and the analysis results were related to the type and content of volatile substances.

**3.5. OPLS-DA analysis of roasted/broth flavors**

Compared with PLS-DA (partial least squares discriminant analysis), OPLS-DA (orthogonal partial least squares discriminant analysis) can effectively eliminate influences that have cannot influence the screening of different substances (Huang et al., 2021). To accurately obtain the difference of flavor substances between the two groups and within the group, this study used the OPLS-DA supervised discriminate method to analyze the volatile substances of roasted/broth flavors. Fig. 4 showed the OPLS-DA score and the results were in good agreement with those of PCA. It can be seen that there is an obvious separation between the roasted meat flavor and the broth flavor. The parameter R<sub>2</sub>X = 0.821 in the model indicates that the explanatory rate of the independent variable X was 99.6%; R<sub>2</sub>Y = 1 and the explanatory rate of the categorical variable Y was 100%; Q<sub>2</sub>Y = 0.992, indicating that the model predicts the sample variables at the degree of 99.9%, and the P < 0.05 of Q<sup>2</sup> and R<sub>2</sub>Y (Li et al., 2021). The results shown that the OPLS-DA model was stable and has a high predictive power.

**3.6. Screening and analysis of flavors difference substances in roasted/broth**

From the 130 volatile compounds (Table S1), based on P value < 0.05, FC > 2, and FC < 0.5, 78 substances were selected (Ai et al., 2021). As shown in Fig. S1, with the variable importance in projection VIP (Variable important in projection) > 1 as the condition of the OPLS-DA



**Fig. 4.** Comparison of characteristic response values of electronic nose sensors for roasted/broth flavors (a); PCA score map and load double map (b).

model, 48 different metabolites were screened out, as shown in Table 1.

To observe the variation trend of volatile matter concentration in roasted/broth flavors more directly, heat maps were prepared according to the relative content of each difference, and the differences between the data were visualized by different color gradients. As shown in Fig. 5, the color of the heat map represented the relative expression level of the different substances in roasted/broth flavors. The color changes from green to yellow and then to red indicated that the abundance of the different substances was gradually increased. The high and low expressions interacted together, red was the high-expression component and green was the low-expression component. According to the clustering of the two sets of samples, Fig. 5 can be divided into two regions, the barbecue sample was the first region, and the broth sample was the second region.

The results of the heat map analysis of differential substances showed these compounds: 2-methyl-3-furanthiol, 2-methyl-3-methylthiofuran, 1-butanethiol, 2-acetylpyrrole. The content of lutidine and 2-acetylthiophene were significantly higher than the barbecue flavor; the roasted flavors contained 2-thiophenethiol, furfuryl alcohol, 3-furaldehyde, 2,5-dimethylpyrazine, 4-methylthiazole, and furan propylene. The aldehyde

content was significantly higher than the broth flavor.

#### 4. Conclusion

GC × GC-ToF-MS and GC-MS were used to identify the flavor volatiles of roasted/broth prepared by cysteine-xylose-glutamate MR system. A total of 130 volatile substances were identified by GC × GC-ToF-MS, while 88 volatile substances were identified by GC-MS, of which 36 were common substances. The contents of thiophenes, thiazoles, and pyrazines were higher in the roasted meat flavor than in the broth flavor; the contents of thiols and thioethers, furans, aldehydes and acids were higher in the broth flavor than in the roasted meat flavor, which were consistent with the results of the data measured by GC-MS. Differences in volatiles between roasted/broth flavors were clearly distinguished by electronic nose analysis, with higher response values for nitrogen oxides, methyl groups, sulfides, and aldehydes and ketones in roasted flavor than in broth flavor. The differential analysis of roasted/broth flavors provides a basis for simulating the preparation of meat flavors using the MR.

**Table 1**  
Flavor Differential Substances of roasted/broth flavors.

No.	Compounds	VIP	P value	fdr	Log2FC	Type
1	2-Methyl-3-thienyl (2-methyl-3-furyl) disulfide	1.252253	0.003401	0.028897	-1.580008234	down
2	Furan-2-ylmethanethiol	1.21538	0.022474	0.060781	-1.358654419	down
3	2-Thiophenethiol	1.217529	0.019128	0.058784	-3.719946495	down
4	2-Methyl-3-furanthiol	1.154528	0.043302	0.09743	1.039497734	up
5	2-Methyl-3-(methylthio)furan	1.24423	7.84E-04	0.01686	3.749571263	up
6	2-Thienyl disulfide	1.232116	0.004922	0.028897	-1.530046706	down
7	1-Butanethiol	1.224867	0.012561	0.042908	1.521685585	up
8	3-Mercapto-2-butanone	1.259457	0.001338	0.01686	2.375855012	up
9	2-Ethyl-5-methylthiophene	1.238888	0.001256	0.01686	2.812408744	up
10	2-Cyclopentene-1,4-dione	1.258642	0.004405	0.028897	-1.508620456	down
11	Thieno(2,3-b)thiophene	1.214903	0.020537	0.060781	4.210356664	up
12	2-Acetylthiophene	1.267803	9.92E-04	0.01686	2.074785464	up
13	3-Thienylacetic acid	1.244861	0.011371	0.042141	Inf	up
14	3-Methylthiophene-2-aldehyde	1.248316	0.007774	0.033775	3.357550625	up
15	2-Propanoylthiophene	1.268863	0.001313	0.01686	Inf	up
16	2-Formyl-2,3-dihydrothiophene	1.201954	0.027495	0.067964	Inf	up
17	2,5-dimethyl-4-ethyl thiazole	1.254585	7.45E-04	0.01686	1.766555493	up
18	4-Methylthiazole	1.205808	0.004439	0.028897	-2.175646005	down
19	2-Methyl thiazole	1.214805	0.022593	0.060781	-2.269910652	down
20	methylpyrazine	1.236405	0.002097	0.024017	-2.771150968	down
21	2,6-Dimethylpyrazine	1.271569	2.52E-05	0.003179	-Inf	down
22	2,5-Dimethylpyrazine	1.204116	0.027509	0.067964	-Inf	down
23	2-Ethyl-5-methylpyrazine	1.220781	0.008603	0.033873	-2.685356547	down
24	2-Acetyl-1H-pyrrole	1.191752	0.005913	0.028897	1.497447974	up
25	2-Methylfuran	1.247034	0.010022	0.038267	Inf	up
26	2-Amylfuran	1.218511	0.004537	0.028897	3.456238679	up
27	2-Ethylfuran	1.260574	0.001218	0.01686	2.044286873	up
28	1-(Furan-2-yl)ethanone	1.240483	0.012421	0.042908	-Inf	down
29	3-Furaldehyde	1.25915	0.004924	0.028897	-Inf	down
30	Furylacrolein	1.230659	0.005983	0.028897	-1.766512389	down
31	(2E,4E)-Deca-2,4-dienal	1.211979	0.005641	0.028897	-1.716526394	down
32	Acetaldehyde	1.212281	0.013546	0.044915	-1.447354531	down
33	Methoxyacetaldehyde	1.239827	0.0126	0.042908	-Inf	down
34	Lauryl aldehyde	1.191119	0.00777	0.033775	2.492840039	up
35	Nonanal	1.266598	1.55E-04	0.009794	3.339620289	up
36	Phenylacetaldehyde	1.19373	0.014057	0.045416	1.35907213	up
37	Isobutyraldehyde	1.26564	0.002641	0.027735	Inf	up
38	Butyraldehyde	1.237312	0.005351	0.028897	-2.523354725	down
39	N-butanol	1.238287	0.005866	0.028897	-1.442574217	down
40	Propylene Glycol	1.2505	0.008418	0.033873	-Inf	down
41	Furfuryl Alcohol	1.200793	0.022672	0.060781	-2.202954462	down
42	Glycolic acid	1.15833	0.048848	0.107979	Inf	up
43	Butyric Acid	1.217273	0.00359	0.028897	-2.154881507	down
44	5,6-Dihydro-2H-pyran-2-one	1.211712	0.017256	0.054355	1.136717974	up
45	3-Hydroxy-2-methyl-4H-pyran-4-one	1.256004	0.006192	0.028897	-Inf	down
46	3,5-Dimethylpyridine	1.096123	0.031036	0.075202	1.703529639	up
47	2,4-Di-tert-butylphenol	1.146422	0.022343	0.060781	-1.037793545	down
48	Ether	1.220433	0.020791	0.060781	-Inf	down

VIP: variable importance in projection; FC: fold change. P-value: projection value; Log2FC: fold change log transform.

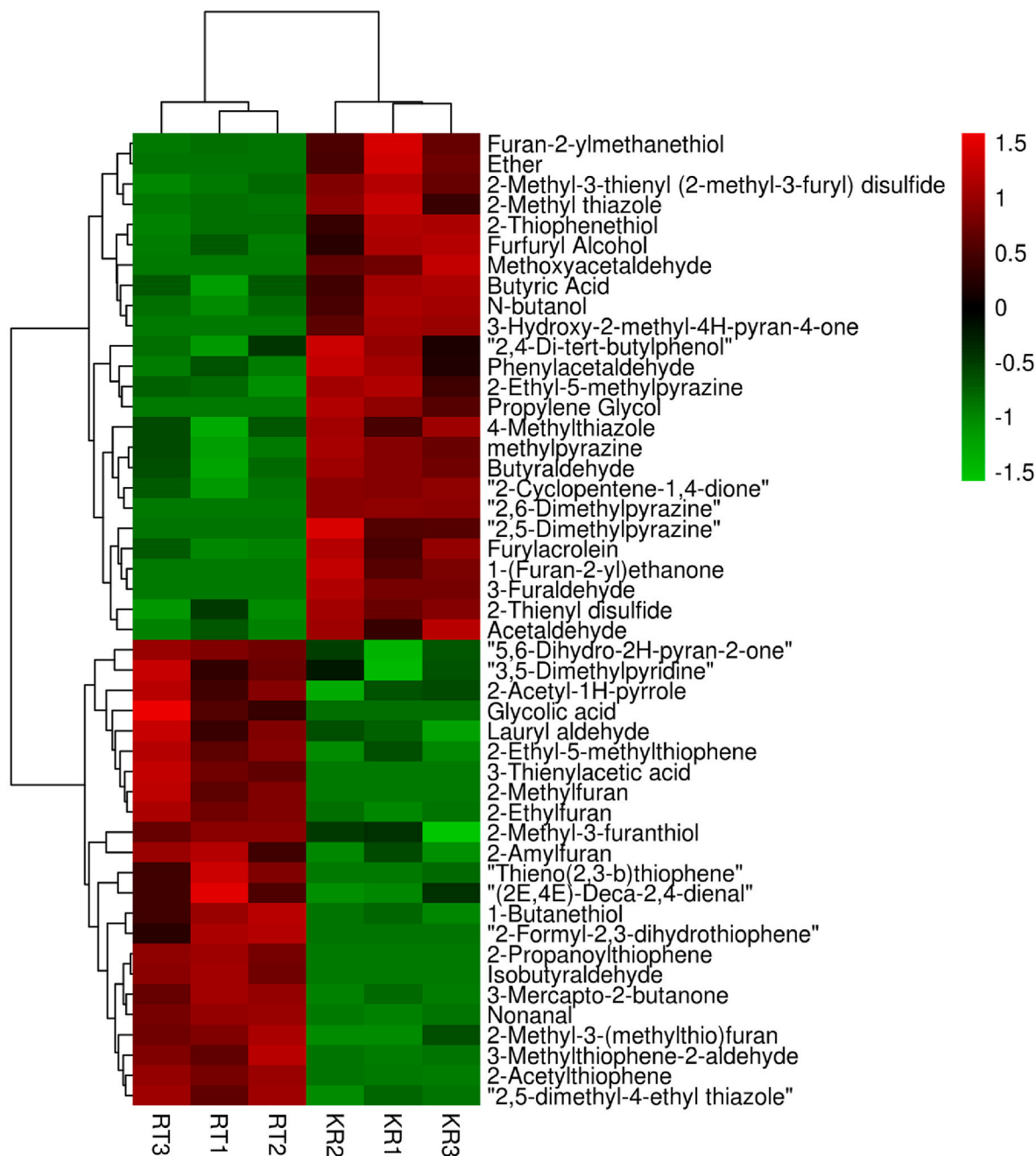


Fig. 5. Differential matter volcano map (a), cluster heat map (b).

#### CRediT authorship contribution statement

**An-Ran Zheng:** Investigation, Formal analysis, Conceptualization. **Chao-Kun Wei:** Investigation, Supervision, Formal analysis, Methodology, Validation, Writing – review & editing. **Dun-Hua Liu:** Formal analysis, Data curation. **Kiran Thakur:** Writing – original draft, Writing – review & editing. **Jian-Guo Zhang:** Methodology, Validation. **Zhao-Jun Wei:** Project administration, Funding acquisition.

#### 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

No data was used for the research described in the article.

#### Acknowledgements

This work was supported by the National Natural Science Foundation of Ningxia Province (2021AAC02019, 2021AAC03050), the Youth talent cultivation project of North Minzu University (2021KYQD27, FWNX14), the Major Projects of Science and Technology in Anhui Province (202203a06020009), Key research and development projects in Ningxia province (2021BEF02013, 2021BEB04049).

## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.crfs.2023.100445>.

## References

- Adab, C., Fz, A., Bs, A., Tr, A., Feng, C.D., 2021. Tracking volatile flavor changes during two years of aging of Chinese vinegar by hs-spmc-gc-ms and gc-o. *J. Food Compos. Anal.* 106, 104295 <https://doi.org/10.1016/j.jfca.2021.104295>.
- Ai, J., Wu, Q., Battino, M., Bai, W.B., Tian, L.M., 2021. Using untargeted metabolomics to profile the changes in roselle (*Hibiscus sabdariffa* L.) anthocyanins during wine fermentation. *Food Chem.* 364, 130425 <https://doi.org/10.1016/j.foodchem.2021.130425>.
- An, A., Kimpe, N.D., 2009. Formation of pyrazines from ascorbic acid and amino acids under dry-roasting conditions. *Food Chem.* 115 (4), 1417–1423. <https://doi.org/10.1016/j.foodchem.2009.01.071>.
- Cao, C., Xie, J., Hou, L., Zhao, J., Chen, F., Xiao, Q., 2017. Effect of glycine on reaction of cysteine-xylose: insights on initial maillard stage intermediates to develop meat flavor. *Food Res. Int.* 99 (1), 444–453. <https://doi.org/10.1016/j.foodres.2017.06.012>.
- Cerny, C., 2007. Origin of carbons in sulfur-containing aroma compounds from the maillard reaction of xylose, cysteine and thiamine. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 40 (8), 1309–1315. <https://doi.org/10.1016/j.lwt.2006.09.008>.
- Cerny, C., Guntz-Dubini, R., 2013. Formation of cysteine-s-conjugates in the maillard reaction of cysteine and xylose. *Food Chem.* 141 (2), 1078–1086. <https://doi.org/10.1016/j.foodchem.2013.04.043>.
- Charve, J., Manganiello, S., Glabasnja, A., 2018. Analysis of umami taste compounds in a fermented corn sauce by means of sensory-guided fractionation. *J. Agric. Food Chem.* 66 (8), 1863–1871. <https://doi.org/10.1021/acs.jafc.7>.
- Chen, X., Yu, J., Cui, H., Xia, S., Zhang, X., Yang, B., 2018. Effect of temperature on flavor compounds and sensory characteristics of maillard reaction products derived from mushroom hydrolysate. *Molecules* 23 (2), 247. <https://doi.org/10.3390/molecules23020247>.
- Cui, H., Yu, J., Zhai, Y., Feng, L., Ho, C.T., 2021. Formation and fate of amadori rearrangement products in maillard reaction. *Trends Food Sci. Technol.* 115 (1), 391–408. <https://doi.org/10.1016/j.tifs.2021.06.055>.
- Farmer, L.J., Mottram, D.S., 2010. Effect of cysteine and ribose on the volatile thermal degradation products of a triglyceride and three phospholipids. *J. Sci. Food Agric.* 60 (4), 489–497. <https://doi.org/10.1002/jsfa.2740600414>.
- Gabriela, M.R.N.A., Dresch, D., Melchert, W.R., 2021. Use of non-volatile compounds for the classification of specialty and traditional brazilian coffees using principal component analysis. *Food Chem.* 360 (30), 130088 <https://doi.org/10.1016/j.foodchem.2021.130088>.
- Geib, E., Baldeweg, F., Doerfer, M., Nett, M., Brock, M., 2018. Cross-chemistry leads to product diversity from atromentin synthetases in aspergilli from section nigri. *Cell Chem. Biol.* 26 (2), 223–234. <https://doi.org/10.1016/j.chembiol.2018.10.021>.
- Hidalgo, F.J., Zamora, R., 2019. Formation of phenylacetic acid and benzaldehyde by degradation of phenylalanine in the presence of lipid hydroperoxides: new routes in the amino acid degradation pathways initiated by lipid oxidation products. *Food Chem. X* 2, 100037. <https://doi.org/10.1016/j.fochx.2019.100037>.
- Huang, Q., Dong, K., Wang, Q., Huang, X., Wang, G.Z., An, F., Luo, P., 2021. Changes in volatile flavor of yak meat during oxidation based on multi-omics. *Food Chem.* 371, 131103 <https://doi.org/10.1016/j.foodchem.2021.131103>.
- Jayasena, D.D., Dong, U.A., Nam, K.C., Jo, C., 2013. Flavour chemistry of chicken meat: a review. *Asian-Australas. J. Anim. Sci.* 26 (5), 732–742. <https://doi.org/10.5713/ajas.2012.12619>.
- Li, X., Liu, S.Q., 2021. Impacts of thermal treatment, xylose and cysteine addition on aroma compounds profile in lactic acid bacterium fermented pork hydrolysates. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 152 (3), 112368. <https://doi.org/10.1016/j.lwt.2021.112368>.
- Li, X.Z., Liu, S.Q., 2022. Effect of heat treatment on amino acids and volatile compounds of enzymatic pork trimmings hydrolysate supplemented with xylose and cysteine. *Food Biosci.* 46, 101538 <https://doi.org/10.1016/j.fbio.2021.101538>.
- Li, D.Y., Yuan, Z., Liu, Z.Q., Yu, M.M., Guo, Y., Liu, X.Y., Zhang, M., Liu, H.L., Zhou, D.Y., 2020. Effect of oxidation and maillard reaction on color deterioration of ready-to-eat shrimps during storage. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 131, 109696. <https://doi.org/10.1016/j.lwt.2020.109696>.
- Li, Y., Wu, Y., Quan, W., Jia, X.D., He, Z.Y., Wang, Z.J., 2020. Quantitation of furosine, furfurals, and advanced glycation end products in milk treated with pasteurization and sterilization methods applicable in China. *Food Res. Int.* 140 (10), 110088 <https://doi.org/10.1016/j.foodres.2020.110088>.
- Li, J., Wu, H., Wang, L., Huang, Y.J., Wang, L.B., 2021. Key taste components in two wild edible Boletus mushrooms using widely targeted metabolomics. *Biochem. Systemat. Ecol.* 96, 104268 <https://doi.org/10.1016/j.bse.2021.104268>.
- Loch, C., Reusch, H., Ruge, I., Godelmann, R., Pflaum, T., Kuballa, T., Schumacher, S., Lachenmeier, D., 2016. Benzaldehyde in cherry flavour as a precursor of benzene formation in beverages. *Food Chem.* 206, 74–77. <https://doi.org/10.1016/j.foodchem.2016.03.034>.
- Ma, H.C., Chen, Y.T., He, Z.Z., Wei, Y.H., 2022. Facile fluorescent glucose detection based on the maillard reaction. *Colloids Surf. A Physicochem. Eng. Asp.* 18, 128339 <https://doi.org/10.1016/j.colsurfa.2022.128339>.
- Mottram, D.S., Madruga, M.S., Whitfield, F.B., 1995. Some novel meatlike aroma compounds from the reactions of alkanediones with hydrogen sulfide and furanthiols. *J. Agric. Food Chem.* 43 (1), 189–193. <https://doi.org/10.1021/jf00049a035>.
- Rose, M., Holland, J., Dowding, A., Petch, S., White, S., Fernandes, A., Mortimer, D., 2015. Investigation into the formation of pahn in foods prepared in the home to determine the effects of frying, grilling, barbecuing, toasting and roasting. *Food Chem. Toxicol.* 78, 1–9. <https://doi.org/10.1016/j.fct.2014.12.018>.
- Scaman, C., Nakai, S., Aminari, M., 2006. Effect of pH temperature and sodium bisulfite or cysteine on the level of maillard-based conjugation of lysozyme with dextran, galactomannan and mannan. *Food Chem.* 99 (2), 368–380. <https://doi.org/10.1016/j.foodchem.2005.08.003>.
- Shen, H., Zhao, M., Sun, W.Z., 2019. Effect of pH on the interaction of porcine myofibrillar proteins with pyrazine compounds. *Food Chem.* 287, 93–99. <https://doi.org/10.1016/j.foodchem.2019.02.060>.
- Shen, M., Liu, L., Zhang, F., Jiang, Y., Xie, M., 2021. Effects of processing parameters on furan formation in canned strawberry jam. *Food Chem.* 358 (1), 129819 <https://doi.org/10.1016/j.foodchem.2021.129819>.
- Tan, Z., Ai, Y.U., 2012. Effect of reaction temperature and time on the formation of volatiles from the maillard reaction of ascorbic acid with arginine. *Sci. Technol. Food Indus.* 33 (6), 143–152. <https://doi.org/10.13386/j.issn1002-0306.2012.06.053>.
- Tian, H.X., Xu, X.L., Chen, C., Yu, H.Y., 2019. Flavoromics approach to identifying the key aroma compounds in traditional Chinese milk fan. *J. Dairy Sci.* 102 (11), 9639–9650. <https://doi.org/10.3168/jds.2019-16796>.
- Wei, C.K., Ni, Z.J., Thakur, K., Liao, A.M., Huang, J.H., Wei, Z.J., 2019. Aromatic effects of immobilized enzymatic oxidation of chicken fat on flaxseed (*Linum usitatissimum* L.) derived maillard reaction products. *Food Chem.* 306 <https://doi.org/10.1016/j.foodchem.2019.125560>, 125560-125560.
- Wei, D., Wei, C., Zhang, H., 2022. Grey correlation degree for analysis of the correlation between electronic nose responses to meat flavors and their sensory scores. *Meat Res.* 36 (5), 49–53. <https://doi.org/10.7506/rlyj1001-8123-20220120-004> (in Chinese).
- Wong, K.H., Aziz, S.A., Mohamed, S., 2010. Sensory aroma from maillard reaction of individual and combinations of amino acids with glucose in acidic conditions. *Int. J. Food Sci. Technol.* 43 (9), 1512–1519. <https://doi.org/10.1111/j.1365-2621.2006.01445.x>.
- Wu, S., Yang, J., Dong, H., Liu, Q., Li, X., Zeng, X., Bai, W., 2021. Key aroma compounds of Chinese dry-cured Spanish mackerel (*Scomberomorus niphonius*) and their potential metabolic mechanisms. *Food Chem.* 342, 128381 <https://doi.org/10.1016/j.foodchem.2020.128381>.
- Xue, P.M., Ping, Z., Hong, L.T., Zhi, S.W., Peng, W., 2019. Effects of different enzymatic hydrolyses of mutton tallow on the aroma characteristics of the maillard reaction of xylose–cysteine based on gc/ms, ek/mose, and statistical analysis. *Eur. J. Lipid Sci. Technol.* 122 (3), 1900212 <https://doi.org/10.1002/ejlt.201900212>.
- Yahya, H., Linforth, R.S.T., Cook, D.J., 2014. Flavour generation during commercial barley and malt roasting operations: a time course study. *Food Chem.* 145 (15), 378–387. <https://doi.org/10.1016/j.foodchem.2013.08.046>.
- Yang, W.J., Yu, J., Pei, F., Mariga, A.M., Ma, N., Fang, Y., Hu, Q.H., 2016. Effect of hot air drying on volatile compounds of flammulina velutipes detected by hs-spmc-gc-ms and electronic nose. *Food Chem.* 196 (1), 860–866. <https://doi.org/10.1016/j.foodchem.2015.09.097>.
- Yaylayan, V.A., 2003. Recent advances in the chemistry of strecker degradation and amadori rearrangement: implications to aroma and color formation. *Food Sci. Technol. Res.* 9 (1), 1–6. <https://doi.org/10.3136/fstr.9.1>.
- Yu, A.N., Tan, Z.W., Wang, F.S., 2013. Mechanistic studies on the formation of pyrazines by maillard reaction between l-ascorbic acid and l-glutamic acid. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 50 (1), 64–71. <https://doi.org/10.1016/j.lwt.2012.07.001>.
- Zeng, X., Liu, J., Dong, H., Bai, W., Yu, L., Li, X., 2020. Variations of volatile flavor compounds in Cordyceps Militarlis chicken soup after enzymolysis pretreatment by SPME combined with GC-MS, GC×GC-TOF MS and GC-IMS. *Int. J. Food Sci. Technol.* 55, 509–516. <https://doi.org/10.1111/ijfs.14294>.
- Zhang, G.L., Liang, Y., Zhu, J.Y., Jia, Q., Gan, W.Q., Sun, L.M., Hou, H.M., 2015. Oxidative stress-mediated antiproliferative effects of furan-containing sulfur flavors in human leukemia jurkat cells. *Food Chem.* 180 (1), 1–8. <https://doi.org/10.1016/j.foodchem.2015.01.122>.
- Zhang, K., Wu, N., Wang, X., Wu, X., Wang, S., Long, X., 2016. Effects of 3 feeding modes on the volatile and nonvolatile compounds in the edible tissues of female Chinese mitten crab (*Eriocheir sinensis*). *J. Food Sci.* 81 (4–6), S968–S981. <https://doi.org/10.1111/1750-3841.13229>.
- Zhao, J., Wang, T., Xie, J., Xiao, Q., Cheng, J., Chen, F., Wang, S., Sun, B., 2019. Formation mechanism of aroma compounds in a glutathione-glucose reaction with fat or oxidized fat. *Food Chem.* 270, 436–444. <https://doi.org/10.1016/j.foodchem.2018.07.106>.
- Zhou, Y.Y., Li, Y., Yu, A.N., 2016. The effects of reactants ratios, reaction temperatures and times on maillard reaction products of the l-ascorbic acid/l-glutamic acid system. *Food Sci. Technol.* 36 (2), 268–274. <https://doi.org/10.1590/1678-457X.02415>.
- Zhu, J.C., Feng, C., Wang, L.Y., Niu, Y.W., Xiao, Z.B., 2016. Evaluation of the synergism among volatile compounds in oolong tea infusion by odour threshold with sensory analysis and e-nose. *Food Chem.* 221, 1484–1490. <https://doi.org/10.1016/j.foodchem.2016.11.002>.