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Exploring the influence and mechanism of different frying methods on the flavor quality of low-salt sour meat

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ABSTRACT

To obtain nutritious, healthy, and flavor-enriched sour meat products, the effects of different frying methods (microwave, air-frying, and traditional frying) on the flavor quality of low-salt sour meat were evaluated using metabolomics and other flavor analysis techniques. The pH value of the sour meat rose dramatically, while the TBARS value dropped significantly after frying. E-nose and E-tongue results showed that air-frying could reduce acidity and improve umami. The comprehensive analysis of all samples revealed the identification of 107 volatile flavor compounds, including 10 unique aroma compounds that were specifically detected in the AF group. Additionally, the air frying process notably increased the free amino acid and nucleotide concentrations in sour meat by 53.58% and 159.29%, respectively, while causing a significant reduction in both fatty acid and lactic acid content by 22.84% and 49.29%, respectively. All three frying methods altered the flavor of the samples, but air frying performed better in terms of flavor and texture.

1. Introduction

Sour meat is a traditional fermented meat product from ethnic minority regions in China, and the production method is mainly based on natural fermentation. Pork, rice flour, salt, and other seasonings (chili powder, zanthoxylum powder, pepper powder, etc.) are mixed thoroughly and fermented under sealed conditions for 1–2 months to obtain mature sour meat products, which are characterized by firm texture, sour taste, ester flavor, fat but not greasy, rich in probiotics and postbiotic elements, and high nutritional value (Lv et al., 2023). The proteins, carbohydrates, and fats in raw meat can be degraded to produce large amounts of free amino acids, active peptides, organic acids, and fatty acids through biochemical reactions such as the metabolism of microorganisms and nitrate reduction, which give sour meat distinct flavor and nutritious qualities (Perea-Sanz et al., 2019; Zhong et al., 2021). Meanwhile, some of the degradation products also serve to prolong the shelf life of sour meat products by inhibiting the growth of various disease- and spoilage-causing microorganisms (Zhong et al., 2022). In addition, it has been reported that the oxidative degradation of unsaturated fatty acids is the main source of most volatile flavor substances in sour meat (Mariutti & Bragagnolo, 2017). It is noteworthy that low-salt fermentation serves the dual purpose of decreasing sodium intake for consumers and enhancing the flavor characteristics of sour meat products. Consequently, leveraging the high protein and unsaturated fatty acid content of Tibetan pork in conjunction with the sodium reduction and flavor enhancement effect attributed to low-salt fermentation represents a productive strategy to boost the nutritional profile and flavor excellence of sour meat (Wang et al., 2021).

Fermented sour meat can be cooked in a variety of ways according to regional eating customs, such as steaming, frying, deep-frying, or boiling. Among these, deep-frying is one of the favored methods of cooked sour meat in some regions (Lv et al., 2019; Zhong et al., 2022).

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Deep-frying can not only remove the fishy flavor but also give the food a golden color, a crispy texture, and a distinctive flavor. However, traditional fried foods deemed to carry health risks, and immoderate consumption can readily result in chronic disorders like obesity, hyperlipidemia, cardiovascular disease, and diabetes, as well as heighten the probability of carcinogenicity induced by heterocyclic amines, which arise as metabolic byproducts of the frying procedure (Cahill et al., 2014). As a result, researchers have concentrated increasingly on new frying techniques to improve the flavor and safety of fried foods. Microwave frying, as a common household cooking technique, which primarily relies on the intermolecular friction within the food matrix during microwave heating, thus completing the conversion of high-frequency electromagnetic energy to thermal energy and realizing the heating process of food (Li et al., 2019). Furthermore, the monolinear oxygen generated during microwave heating can trigger protein and lipid oxidation, which actively contributes to flavor development (Abdel et al., 2021). Air-frying, an emerging frying method, is thought to be a superior way to improve food flavor while preserving food quality and safety (Song et al., 2020). Air-frying utilizes the circulating hot air as a heating medium, through the hot air in uniform contact with the food, to achieve the purpose of cooking food. Compared with traditional frying approaches, air frying uses a significantly lower amount of oil, and the unique hot air circulation heating technique in air frying can expedite the oxidation of food, enhancing its flavor attributes. Concurrently, this method facilitates rapid dehydration of the food surface, creating a firm outer layer to minimize moisture loss and preserve the succulent taste of the food (Cheng et al., 2023). Air-frying has been reported to greatly reduce the amount of oil in the chicken pieces compared to traditional frying, but it also reduces the overall amount of volatile flavor substances in the chicken pieces, resulting in a partial loss of flavor (Cao et al., 2020). Meanwhile, a comparative study of air-fried and traditional fried sturgeon steak revealed that air-fried sturgeon steak has a higher essential amino acid content, better flavor quality, and better digestibility (Liu et al., 2022).

To sum up, it is necessary to explore a healthy and convenient frying method without destroying the good flavor of low-salt fermented sour meat. Consequently, this research conducted a comparative analysis to assess the impact of air frying, microwave frying, and conventional frying on the flavor characteristics of low-salt fermented sour meat. Multi-omics technologies, including flavoromics and metabolomics, were subtly utilized to study the effect of three frying methods on the formation pathway of volatile flavor compounds (VOCs) in low-salt sour meat. Additionally, the electronic nose (*E*-nose) and electronic tongue (E-tongue) systems were used to analyze the differences between low-salt fermented sour meat products treated by different frying methods in terms of flavor profile and taste characteristics. This work may build a theoretical foundation for the healthization and industrial development of sour meat products.

2. Materials and methods

2.1. Sour meat preparation

Three independent baches of Tibetan pig hind leg (three legs in total) out of three 12-month-old Tibetan pigs (male, castrated, approximately 25–30 kg) were provided by the Abazhou Bowen Husbandry Technology Co., Ltd., Jiuzhaigou, China. Tibetan pork was cut into slices (about 3 cm × 5 cm × 0.5 cm) and assembled randomly. The rice (Thai Jasmine Rice) was purchased from the local supermarket (Fuzhou, Fujian Province, China). The rice (250 g) was continuously stir-fried on an 800 W (MC-CL22X1–102, Midea Co., Ltd., Guangdong, China) induction cooker for 40 min until the rice was golden brown at a time and ground into flour (sifted through 20 mesh). The pork was marinated with 3.0% (*w*/w) NaCl for 2 h. The rice flour was added to the pork at 10.0% (w/w) after draining the brine. Tibetan pork was mixed well with the ingredients and filled into the container to seal it. Then the containers were placed in a well-ventilated room (20–25 °C) in triplicate for fermentation. After 28 days of fermentation, the sour meat was vacuum packaged and kept at −80 °C for further experiments. All the samples were fermented from the same batch.

2.2. Cooking processing

Sour meat samples taken from the freezer at −80 °C were thawed at 4 °C and returned to room temperature. The moisture on the surface was then wiped off with kitchen paper and used for subsequent cooking. Raw meat group (Raw group): the sour meat samples without frying treatment. Microwave frying group (M group): with the sour meat samples being heated on a ceramic dish at 640 W for 50 s + 50 s in a microwave oven (EG823LC8-NS, Midea Co., Ltd., Guangdong, China), turning it over halfway to finally reach a core temperature above 71 °C. Deep-frying group (F group): with the samples being fried in soybean oil at 180 °C for 50 s in a deep fryer (XJ-6 K116, Xiangjiang Electric Appliance Co., Ltd., Hubei, China) to reach their final core temperature of >71 °C. Air-frying group (AF group): with the sour meat samples being heated at 180 °C for 5 min + 5 min in an air fryer (MF-KZ40Q4–403, Midea Group, Guangdong, China) that had already been preheating for 5 min, flipping it halfway through to bring its ultimate core temperature to 71 °C or higher.

2.3. pH

The pH value was measured as previously reported with some modifications (Dong et al., 2021). Two grams of minced sample were placed into a centrifuge tube with 18 mL of distilled water added, shaken for 30 min at room temperature (25 °C) in a shaker (SHA—C, Guohua Co., Ltd., Jiangsu, China), and then filtered. Finally, A pH meter (Mettler-Toledo Instrument Co., Ltd., Shanghai, China) was used to measure the pH of the filtrate.

2.4. TBARS

TBARS was conducted according to the method of Luo et al. (2021). Specifically, 0.3 g of minced sample was homogenized in a solution containing 3 mL of thiobarbituric acid (TBA) and 17 mL of trichloroacetic acid-hydrochloric acid (TCA-HCl). The resulting mixture was heated in boiling water for 30 min. Following the cooling of the mixture, 5 mL of chloroform was introduced, and subsequent to centrifugation (3000 ×*g*, 10 min) utilizing the H1650R centrifuge (Xiangyi Co., Ltd., Hunan, China), absorbance was measured at 532 nm. The quantification of TBARS levels was expressed in milligrams of malondialdehyde per kilogram of meat.

2.5. Electronic nose analysis

The *E*-nose analysis system (PEN3 Airsense, Schwerin, Germany) was used to examine the flavor characteristics of sour meat samples. Two grams of minced sour meat samples were placed in a 20 mL headspace bottle in a water bath at 60 °C for 10 min, and then brought to room temperature for determination. The headspace gas flow rate was 400 mL/min, and the measurement time and cleaning time were both 120 s. Ensure that the signal is restored to the baseline before each test. Each sample was measured three times in parallel, and the average value was taken.

2.6. Electronic tongue analysis

The taste characteristics of sour meat samples were analyzed using SA402B E-tongue analysis system (Insent Company, Kanagawa, Japan). The minced samples (15 g) were mixed with distilled water (150 mL), and the suspension was filtered with four layers of gauze, then centrifuged at 4000 ×*g* at 4 °C for 15 min. Finally, the upper oil layer was removed and then filtered using filter paper, and the filtrate was collected for the E-tongue analysis.

2.7. Identification and analysis of volatile compounds

Three grams of sour meat samples were allowed to equilibrate in a 20 mL extraction flask at 60 °C for five minutes. Next, a 50/30 μm fiber (Supelco, Bellefonte, PA, USA) with DVB/CAR/PDMS coating was inserted into the flask, followed by a water bath at 60 °C for 40 min to adsorb the volatiles. Then, the fiber was immediately inserted into the injection port of the gas chromatograph (SHIMADZU GC–MS-QP2010Ultra, Shimadzu, Kyoto, Japan) at 230 °C for 5 min in a non-split mode. Compounds were analyzed on a DB-WAX (30 m × 0.25 mm × 0.25 μm) capillary column. The rising temperature program was set as follows: initial column temperature, 40 °C, held for 5 min; then, the temperature ramped to 50 °C, held for 2 min (rate of 10 °C/min); then, increased to 120 °C, held for 2 min (rate of 4 °C/min); finally, risen to 230 °C, held for 5 min (rate of 12 °C/min) (Wang, et al., 2022).

The NIST11.lib mass spectrometry database was applied to retrieve the measured compounds and eventually select substances with a match higher than 80%. Peak area normalization method was utilized to calculate the relative content of each component.

2.8. Metabolite analysis and identification

Sour meat samples (100 ± 2 mg) were weighed into a 2 mL LEP tube with three steel beads and 1000 μL of tissue extract (75% 9:1 methanol: chloroform, 25% H_2O) were accurately added. Next, the mixture was put in a TISSUELYSER-II tissue grinder (Jingxin Pharmaceutical Machinery Co., Ltd., Shanghai, China) to grind for 60 s at 50 Hz and repeated twice, followed by sonication for 30 min and an ice bath for 30 min. Afterward, the mixture was centrifuged at 12,000 ×*g* for 10 min at 4 °C and the supernatant was collected, concentrated, and dried; then 200 μL of 2-chloro-*L*-phenylalanine solution was used to re-solubilize the sample (4 ppm, 4 °C). And the filtrate was detected by liquid chromatography-mass spectrometry (LC-MS) (Huang et al., 2022).

ACQUITY UPLC® HSS T3 column (2.1 × 150 mm, 1.8 μm) (Waters, Milford, MA, USA) was utilized for LC-MS analysis. The LC conditions were as follows: flow rate, 0.25 mL/min; column temperature, 40 °C; injection volume, 2 μL. The mobile phases were 0.1% (*v*/v) formic acid acetonitrile (C) and 0.1% (v/v) formic acid water (D), and acetonitrile (A) and 5 mmol/L ammonium formate water (B) in positive and negative ionization modes, respectively. The MS conditions were as follows: An electrospray ion source (ESI) was used with positive (3.5 kV) and negative (2.5 kV) ion modes. The sheath gas was 30 arb, the auxiliary gas was 10 arb, the capillary temperature was 325 °C, the full scan resolution was 70,000, and the scan range was 81–10,000. With a collision voltage of 30 eV, the HCD was employed for secondary cleavage, and dynamic exclusion was applied to eliminate unnecessary MS/MS data. (Liu, Wang, et al., 2022).

The metabolomics processing program ProgenesisQI (Waters Corporation, Milford, USA) was utilized to perform various tasks such as peak identification, peak integration, retention time normalization, and more, and finally obtained information containing retention time, quality-acquisition charge ratio, peak intensity, etc. Then, the software was utilized to identify the characteristic peaks, match MS and MS/MS information to metabolic databases, and identify metabolites based on mass spectral match scores. MoNA (http://mona.fiehnlab.ucda.vis.edu/), Metlin (http://metlin.scripps.edu/), and a company database (BioNovoGene, China) were the sources of all metabolites.

2.9. Multivariate statistical analysis and data processing

Each experiment was repeated at least three times. The results were expressed as mean ± standard deviation using one-way ANOVA ($p<0.05$). SIMCA-14.1 software (Umetrics AB, Umea, Vasterbotten, Sweden) was applied for the principal component analysis (PCA) and orthogonal-partial least squares discriminant analysis (OPLS-DA). GraphPad Prism 7.05 (San Diego, CA, USA) and TBtools 1.09 (GitHub, Inc., Guangzhou, China) and an online platform (https://www.bioinformatics.com.cn) were used to plot the date of flavoromics and metabolomics.

3. Results and discussion

3.1. pH and TBARS

The pH values of sour meat samples treated by different frying methods are shown in Fig. 1a. Since the pH of sour meat dropped greatly following fermentation processing, the pH of the fried sour meat was also at a lower level. Simultaneously, the pH of sour meat samples significantly rose ($p<0.05$), after frying treatment compared to the control group (pH = 4.46), with the greatest increase in pH in group AF to 4.65 and a lesser increase in groups M and F to 4.51. This may be related to the fact that sour meat products' proteins were denatured and broken down by heat during the cooking process, resulting in the unfolding of the protein structure and a reduction in the number of effective acidic groups (O'Neill et al., 2019). Additionally, the variations in fatty acids due to lipid oxidation during frying processing and the decrease in lactic acid content brought on by water loss may also lead to an elevation in pH (Joseph et al., 1997). Among the changes in pH were consistent with the trend of lactate content changes in our subsequent metabolomics analyses. Meanwhile, during air-frying, the intense treatment conditions increased the degradation of fatty acids into volatile flavor compounds, and the decrease in fatty acid content accelerated the hydrolysis of lipids, both of which were in a state of dynamic balance. However, the accumulation of lipid hydrolysis products in microwave and deep-frying treatments slowed the rate of lipid hydrolysis because fatty acids could not be converted to volatile flavor compounds abundantly. Therefore, the AF group showed the highest pH, and the M and F groups exhibited a lower pH.

The thiobarbituric acid reactive substances (TBARS) is widely used to evaluate the degree of lipid oxidation in meat products. It focuses mainly on assessing the level of oxidation by observing the color reaction between malondialdehyde, a byproduct of the oxidative breakdown of unsaturated fatty acids in fats and oils, and thiobarbituric acid. The results depicted in Fig. 1b exhibit a significant decrease in TBARS values for the sour meat samples following their preparation through different frying methods. Compared to the control group, the TBARS values of the M, F, and AF groups decreased by 67.2%, 55.5%, and 54.6%, respectively. This may be attributed to the fact that high levels of malondialdehyde generated during the fermentation process of sour meat, leading to a notable elevation in TBARS values. Subsequent heat treatment may facilitate the interaction between the accumulated malondialdehyde from fermentation and various constituents in sour meat, including nucleotides, nucleic acids, proteins, and free amino acids, thereby causing a marked reduction in TBARS values. ($p<0.05$) (Rao et al., 2022). Furthermore, the M group sour meat samples exhibited the lowest TBARS values. This could be explained by the fact that microwaves mainly relied on the electromagnetic effect to induce friction among the polar molecules in the sour meat, thereby achieving the heating effect. Nonetheless, the constant collision between molecules could improve the chance of malondialdehyde interacting with other components, consequently resulting in lower TBARS values in the M group (Motasemi & Afzal, 2013).

3.2. E-nose analysis

Electronic nose is a non-destructive testing technology that mimics human sensory organs to accurately and quickly distinguish the overall odor properties of a sample. It is frequently used in conjunction with gas chromatography–mass spectrometry (GC–MS) to make up for the

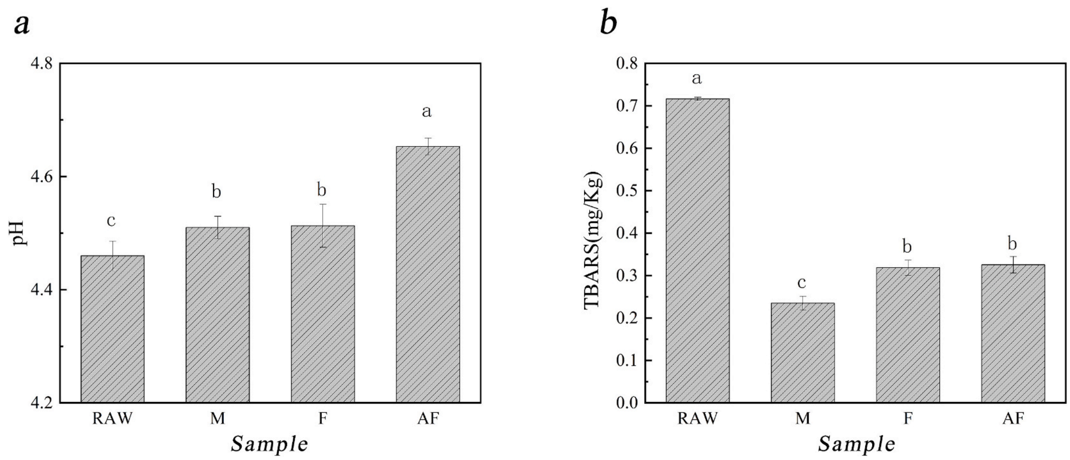

Fig. 1. Effect of different frying methods on the pH and TBARS in low-salt sour meat. pH (a); TBARS (b). (RAW, the sour meat without frying; M, Microwave frying; F, Deep-frying; AF, Air-frying).

limitations of GC–MS in flavor identification. The flavor profiles represented by the ten sensors of the electronic nose are shown in Fig. 2a. From the distribution in Fig. 2b, sour meat samples cooked by different frying methods revealed obvious dispersion in overall flavor attributes. The total contribution of PC1 and PC2 was 97.3%. This finding demonstrated that the electronic nose could be an effective means for flavor identification of sour meat cooked by various frying methods. Meanwhile, there was a partial overlap was observed between groups M and AF in the PCA plots, suggesting that the cooked sour meat samples from groups M and AF share some similarity in flavor. In addition, the samples of the M, AF, and RAW groups were distributed on the negative side of PC1, indicating that the microwave and air-frying were able to better preserve the original flavor of sour meat. This phenomenon was consistent with the results shown in the radar chart (Fig. 2c). The reason for the large flavor alteration in group F may be related to its direct contact with oil during processing. All the samples responded strongly to W1W (sulfides, pyrazines), W2W (organic sulfides and aromatic compounds, etc.), W1S (short-chain alkanes), and W5S (carbon-nitrogen oxides) (Fig. 2d). These findings aligned with the outcomes of GC–MS analysis, except for sulfur-containing compounds, which were not detected in GC–MS. This could be attributed to various factors, including rigorous extraction methods, sensitivity of the electronic nose sensor, and the limitations of GC–MS in detecting sulfides (Cheng et al., 2021).

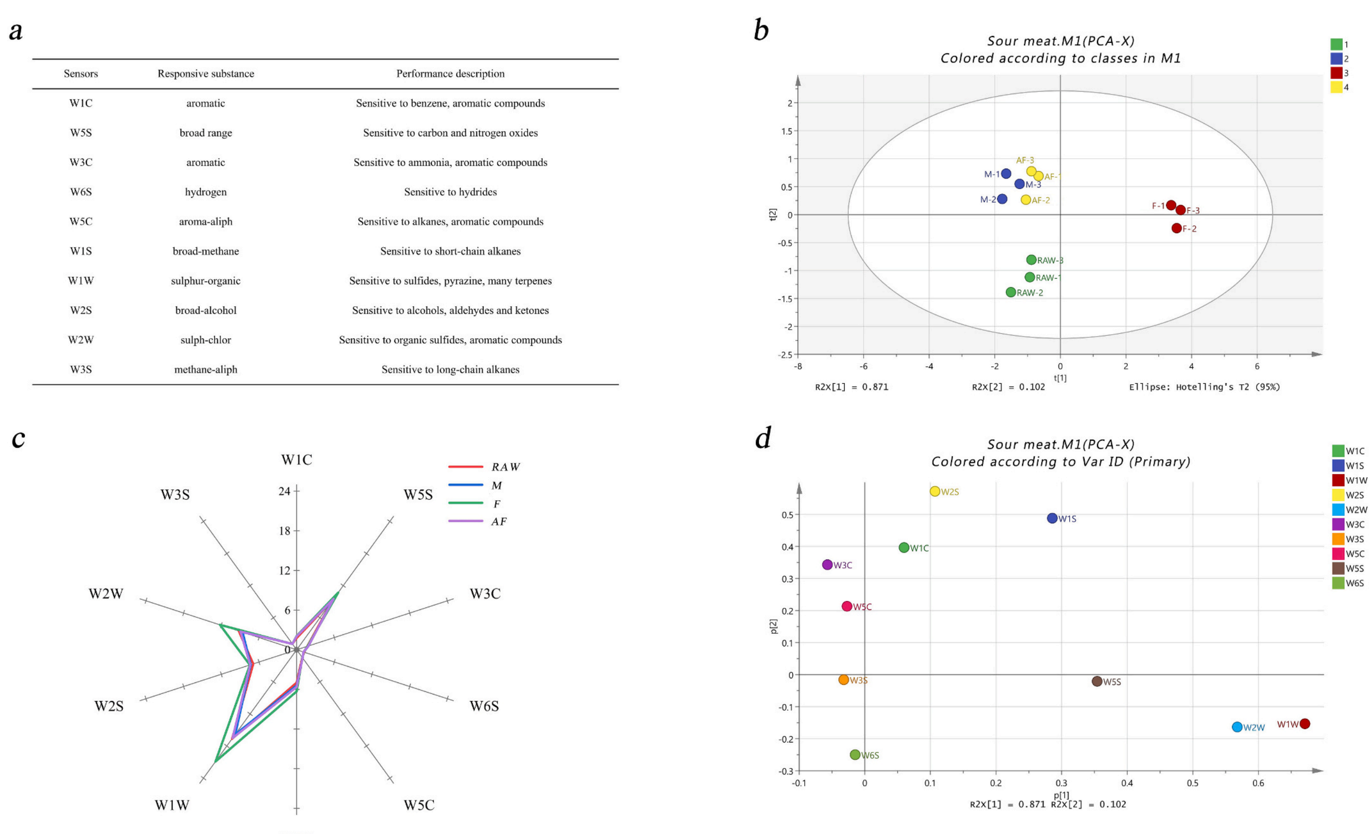

Fig. 2. Effect of different frying methods on the odor attribute of low-salt sour meat. *E*-nose sensor attribute (a); E-nose PCA score graph (b); E-nose Radar chart (c); E-nose loading graph (d). (RAW, the sour meat without frying; M, Microwave frying; F, Deep-frying; AF, Air-frying).

3.3. GC–MS analysis

The OPLS-DA prediction results are shown in Fig. 3a, with good reproducibility of the samples in each group and significant differences between groups. The cumulative variance contribution was 88.7%, while PC1 and PC2 explained 54.2% and 34.5% of the variables, respectively. As shown by the R^2Y of 0.995 and the Q^2 of 0.991, the present model represents a higher level of reliability. To further determine the reliability of the OPLS-DA model, a 200-permutation test was employed for evaluation. The intercept of Q^2 on the y-axis was below 0.5, and Q^2 was less than R^2 at a transverse coordinate was 1. Notably, R^2 was extremely near Q^2, confirming the reliability of the model without overfitting (Fig. 3b). Therefore, the original model provided a sufficient explanation for the variations between the samples (Liao et al., 2023).

A total of 107 volatile taste compounds were identified in sour meat samples cooked by various frying methods based on different mass spectrometry results and retention times. This comprehensive list

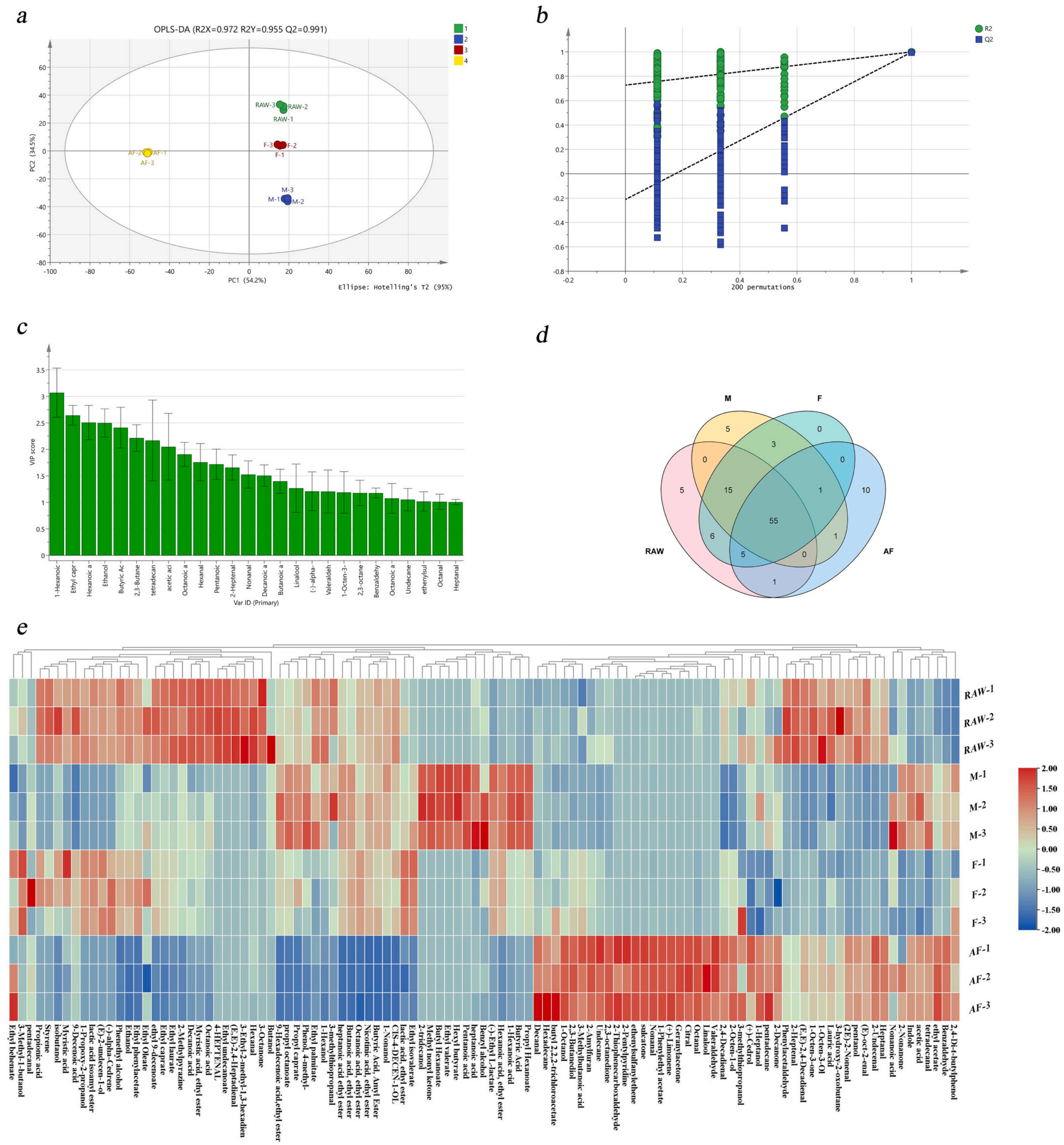

Fig. 3. Effect of different frying methods on the volatile components of low-salt sour meat. OPLS-DA score plot (a); Permutation test result (b); VIP value (c); Venn diagram (d); Clustering heat map (e). (RAW, the sour meat without frying; M, Microwave frying; F, Deep-frying; AF, Air-frying).

includes 19 aldehydes, 21 alcohols, 13 acids, 28 esters, 7 aliphatic hydrocarbons, 2 phenols, 9 ketones, and 8 others compounds (Table S1). Compared to the group of sour meat samples without frying treatment, microwave and air-frying treated sour meat samples showed 5 and 10 divergent flavor compounds, respectively. In contrast, the traditional frying method did not result in differential flavor compounds (Fig. 3d).

In order to more intuitively observe the variations in volatile flavor substances between sour meat samples prepared using different frying methods, the volatile flavor substances detected in the four groups of samples were subjected to clustered heat map analysis (Fig. 3e).

Esters are one of the most essential volatile flavor components in fermented meat products, imparting a fruity fragrance to the product. Their low flavor threshold makes them contribute significantly to the overall flavor of fermented meat products. Esters in sour meat may be created by the esterification reaction of alcohols and acids. However, studies have demonstrated that esters can generate a pungent smell at higher concentrations, which makes them less acceptable to consumers (Jiang et al., 2021). As depicted in Fig. 3e, the air-frying treatment resulted in a significant reduction or even non-detection of esters such as butanoic acid ethyl ester, octanoic acid ethyl ester, and heptanoic acid ethyl ester in the sour meat. This observation suggests that the air-frying procedure holds potential for enhancing the flavor profile of sour meat. The observed trend was in line with what Lu et al. (2022) found. The dropped in ester concentration in the sour meat could be attributed to the special air-heat-flow heating mode of the air fryer, which can promote esters to undergo alcoholysis, ammonolysis, ester exchange, and reduction reactions with other volatile constituents (Lu et al., 2022).

Aldehydes are low-threshold volatile compounds with pleasant floral and fruity aromas that are primarily produced by the degradation of unsaturated fatty acids. The Projection Variable Importance Score (VIP) analysis effectively distinguished all aldehydes, with eight specific aldehydes exhibiting a VIP value >1. These aldehydes comprise myristaldehyde, hexanal, 2-heptenal, nonanal, decanal, valeraldehyde, benzaldehyde, octanal, and heptanal. Among them, hexanal was usually considered as an indicator of the degree of lipid oxidation, and the trend of its content was consistent with our earlier TBARS findings. In addition, it has been reported that hexanal contributes to the grassy aroma of meat products, octanal and valeraldehyde have a citrus-like odor and a cheese-like aroma, respectively. These aldehydes are recognized for their significant contributions to flavor development in sour meat products (An et al., 2020). In this manuscript, air-frying treatment significantly increased the content of aldehydes in the sour meat, whereas microwave and traditional frying did not yield a significant impact on the content of aldehydes in the sour meat. This suggested that the air-frying maturation method can improve the overall aroma of sour meat products by increasing the content of aldehydes.

In this investigation, the main alcohols identified in the samples of sour meat following various frying treatments were 1-octen-3-ol, 1-pentanol, 1-octanol, 1-heptanol, 1-nonanol, linalool, 2-octen-1-ol, ethanol, and so forth. As shown in Fig. 3c, it was found that only ethanol, 1-octen-3-ol, and linalool were judged to be differentially volatile metabolites. Ethanol is mainly derived from the fermentation of carbohydrates, and the high odor threshold of ethanol limits its impact on meat flavor (Yang et al., 2018). 1-Octen-3-ol, a product of fat β-oxidation with mushroom aroma, is a common unsaturated alcohol in fermented meat products with a relatively low odor threshold, which significantly impacts the overall flavor of sour meat products (Wang, Li, et al., 2022). The contents of ethanol and 1-octen-3-ol in sour meat samples treated with different frying methods were significantly lower ($p < 0.05$), which may be due to the ability of heat treatment to promote the loss of volatile flavor compounds (Luo et al., 2022). Furthermore, linalool, identified exclusively in sour meat samples subjected to air-frying, exuded a delightful floral fragrance and contributed to refining the overall aroma of the sour meat products. This observation underscores the beneficial influence of air-frying treatment in augmenting the flavor characteristics of sour meat products (Muriel et al., 2004).

The majority of volatile acid compounds are generated by the degradation of fatty acids, which usually possess an irritating odor. In this study, the content of volatile acid compounds, such as hexanoic acid, octanoic acid, decanoic acid, etc., in the AF group was much lower than that of other groups. Additionally, compared to the other sour meat samples, the AF group also contained significantly higher levels of 2-pentylfuran, which is an oxidation product of linoleic acid with a pleasant buttery flavor and contributes to the flavor during processing (Qi et al., 2022). The results based on GC–MS instrumental analyses demonstrated that the sour meat products matured by air-frying possess more superior flavor qualities.

3.4. E-tongue analysis

As shown in Fig. 4a, the PC1 and PC2 were 78.9% and 16.0%, respectively, with a cumulative contribution of 94.9%, indicating that they can represent most of the information about the sample's taste. Meanwhile, the samples of both groups AF and F were distributed in the first quadrant, suggesting a certain degree of similarity in the taste attributes of the sour meat samples treated by traditional deep-frying and air-frying. Groups M and RAW were positioned on the negative axis of PC1, suggesting that the sour meat samples from group M closely resembled the original flavor profile of sour meat. This proximity could be attributed to the shorter microwave time and milder microwave conditions. The effects of different frying methods on the basic taste indicators of sour meat are shown in Fig. 4b. Different frying methods of maturation treatments drastically reduced the sour taste of the meat and boosted its bitter, salty, and umami tastes. The most notable alterations in taste attributes were observed in the sour meat that underwent air fried. Sourness was the most important taste profile in sour meat, with a trend of RAW > M > F > AF, which was consistent with the variation trend in pH in our previous study (Fig. 1a). In addition, the air-frying treatment considerably improved the salty and fresh flavors of the sour meat. The alteration in salty taste could be linked to the significant water reduction in the sour meat treated with the air fryer. Air-frying primarily utilizes circulating flow of hot air to achieve the effect of cooking. This airflow also contributes to moisture removal from the sour meat, elevating its osmotic pressure and consequently intensifying its salty flavor (Liu, Huang et al., 2022). Meanwhile, the change in umami may be related to the metabolites such as umami amino acids and nucleotides in sour meat and so forth, which was consistent with the results of the subsequent LC-MS analyses. In summary, the *E*-tongue results indicated that sour meat matured by air-frying can improve umami taste, lower the sour taste, which contributed to the development of a gentle flavor in sour meat products.

3.5. Metabolic variation analysis

Free amino acids play a pivotal role in shaping food flavor by not only providing their unique flavor but also engaging with other flavor-presenting substances like nucleotides, reducing sugars, etc. This interaction promotes the formation of the distinctive flavor of food (Zhang et al., 2019). The free amino acids in the samples of sour meat were affected differently by various frying methods. In comparison to untreated sour meat samples, the contents of umami amino acids (like glutamic acid and aspartic acid) in sour meat samples cooked by different frying methods were significantly reduced. Glutamic acid, aspartic acid, and their sodium salts have been reported to impart food a distinctive umami flavor. However, the trends of glutamic acid and aspartic acid following various frying treatments did not coincide with the trends of the umami taste presented by the E-tongue. This discrepancy might be due to the fact that the presentation of umami taste necessitates the synergistic effect of umami amino acids, umami nucleotides, and other substances. Meanwhile, the levels of bitter amino acids, including leucine, isoleucine, phenylalanine, among others, in the samples of sour meat exhibited varying degrees of elevation following

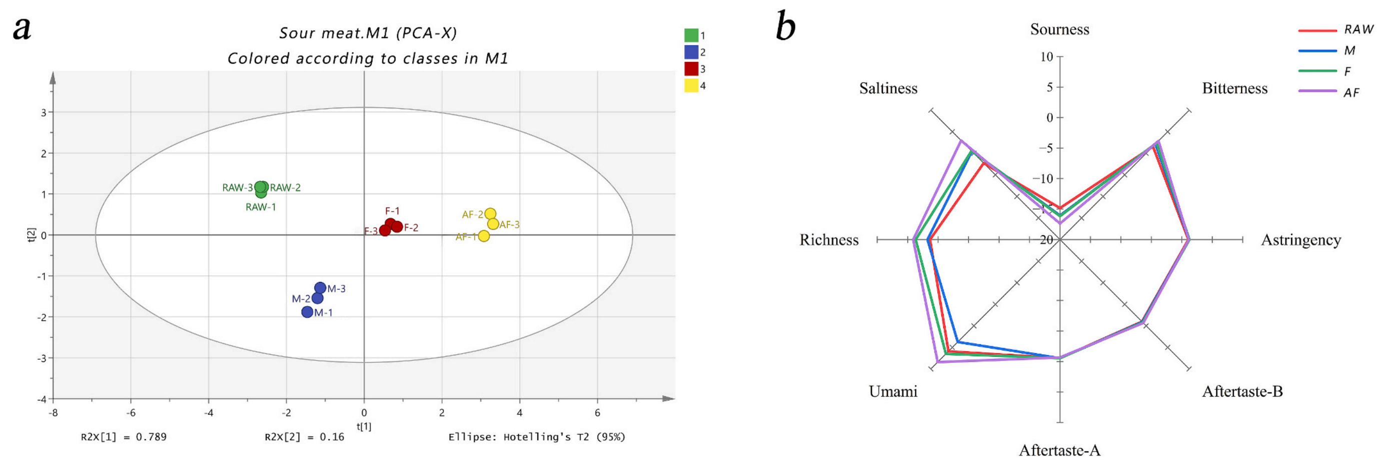

Fig. 4. Effect of different frying methods on the taste attribute of low-salt sour meat. E-tongue PCA score graph (a); E-tongue Radar chart (b). (RAW, the sour meat without frying; M, Microwave frying; F, Deep-frying; AF, Air-frying).

the application of distinct frying methods. The content of bitter amino acids was elevated slightly in group M as opposed to the control group, while a significant elevation was observed in groups F and AF, aligning with the pattern of bitter taste variation as indicated by our previous E-tongue investigations. Arginine is a multifunctional amino acid with the ability to produce a pleasant flavor, improve food quality, and exhibit certain antioxidant properties that may aid in repairing intestinal injuries and boost immune responses. (Ma et al., 2010; Sukhotnik et al., 2005). According to the findings of Li et al. (2021), the thermal processing treatment can reduce the amount of arginine in tilapia, and the air-frying treatment is favorable for the preservation of arginine in tilapia, which is similar to the results of our study. In this study, a notable reduction in arginine levels in sour meat samples following various frying maturation methods, with microwave and air-frying

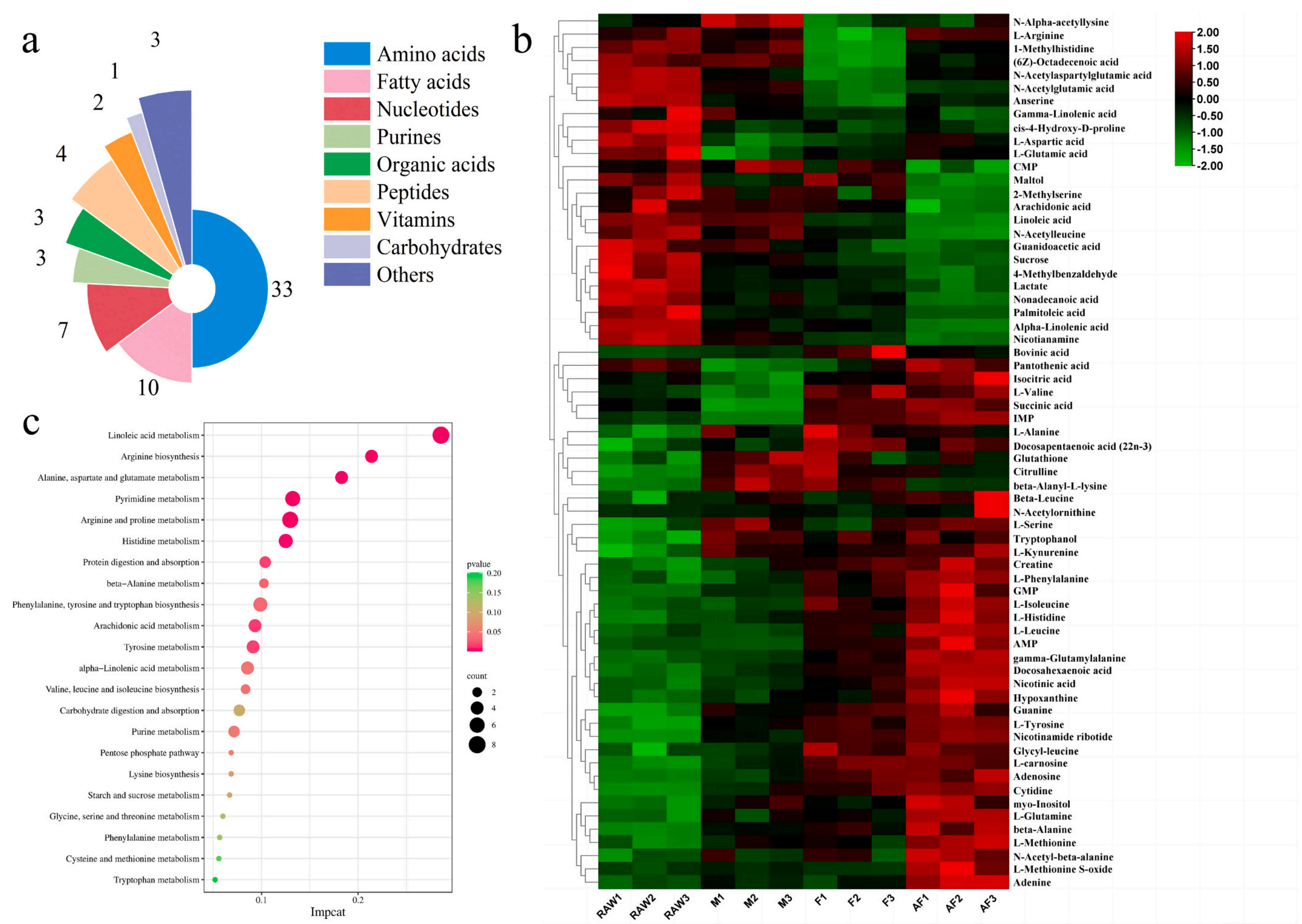

Fig. 5. Effect of different frying methods on the metabolites of low-salt sour meat. Classified pie chart (a); Clustering heat map (b); Metabolic pathways (c). (RAW, the sour meat without frying; M, Microwave frying; F, Deep-frying; AF, Air-frying).

treatments demonstrating a slight decline in arginine content compared with the traditional frying treatment. These findings suggest that air-frying and microwave treatments can be beneficial to the preservation of arginine in sour meat. In addition, it has been noted that large amounts of carnosine could be created in meat products during cooking and other processing, enhancing the umami taste of the products (Luo et al., 2022). Meanwhile, carnosine has also been noted for its significant role in inhibiting the scavenging of free radicals during heat treatment of meat products and promoting the formation of flavor. In this study, the content of carnosine was remarkably raised in all the sour meat samples after maturation treatment. It may be one of the factors that confer favorable taste and flavor to fried sour meat products.

ATP and its degradation products are important flavor presenting substances in fermented meat products. During the processing of fermented meat products, ATP could degrade into ADP, AMP, IMP, Hx, and HxR successively (Cheng et al., 2023). Among them, AMP and IMP are the main umami nucleotides, whereas Hx and HxR are the main contributors to bitter taste. As illustrated in Fig. 5b, all frying methods exerted a considerable influence on the content of nucleotides in the sour meat samples, with variations observed among the different frying methods. Compared with the RAW group, the contents of AMP increased by 84.42%, 170.25%, and IMP by 89.10% and 130.87% in the F and AF groups. Conversely, the M group exhibited a decrease of 6.37% and 47.15% in AMP and IMP levels, respectively, which was in line with the trend in the degree of response of the umami sensors in the findings of our *E*-tongue study. This may be due to the degradation of ATP is primarily regulated through the enzymes ATPase, ADPase, AMP dehydrogenase, phospholipase, and nucleosidase, and the effects of different heat treatments on enzyme activities is contingent upon several factors, encompassing temperature, time, medium, and enzyme type. Consequently, diverse frying techniques yield disparate effects on the content of IMP and AMP (Li et al., 2021). Meanwhile, the content of Hx in sour meat samples after different frying treatments was substantially raised, with the AF group exhibiting the most obvious upward trends compared to the RAW group, which was increased by 46.8%. Besides, the contents of nucleotide degradation products like adenosine, adenine, and nicotinamide nucleotides increased to different degrees, indicating that the macromolecular nucleotides in the sour meat were decomposed as a result of the heat treatment, resulting in an effect on the flavor of the sour meat products. All of these metabolites have been described to have certain functional properties and are beneficial to human health. Among them, nicotinamide nucleotides, also known as coenzyme I, serves as precursors for the synthesis of NAD (nicotinamide adenine dinucleotide), which possesses the function of regulating cellular senescence and maintaining energy balance in the body. Adenosine serves as a precursor in the biosynthesis of essential compounds like ATP and AMP and plays a crucial role in modulating various physiological processes. Its functions include promoting vasodilatation to prevent thrombosis, accelerating vasoconstriction in the hepatic vasculature to promote the breakdown of hepatic glycogen, and other relevant activities (Gyllenhammar et al., 2018).

Fat oxidation is the main source of volatile flavor compounds in fermented meat products, and the high proportion of unsaturated fatty acids in Tibetan pork amplifies the impact of heat processing on its flavor profile. As can be seen from Fig. 5b, the contents of polyunsaturated fatty acids such as linoleic acid, α-linolenic acid, etc. significantly declined in sour meat samples treated with different frying methods, with the contents of the AF group significantly lower than those of the M and F groups. This could be attributed to the relatively long heat treatment time required for air-frying maturation, thus accelerating the oxidative degradation of unsaturated fatty acids in sour meat. Consequently, this finding could explain the highest level of volatile aldehydes in the air-fried sour meat samples. Meanwhile, the amount of saturated fatty acids such as stearic acid, palmitic acid, and others was reduced after thermal processing treatment, especially stearic acid, whose content in the M, F, and AF groups decreased by 51.9%, 76.7%, and 40.3%, respectively. In addition, the level of docosahexaenoic acid in sour meat samples cooked by different frying methods was significantly increased, with the content of the AF group being dramatically higher than that of the M and F groups. Docosahexaenoic acid is recognized as an essential amino acid converted from linoleic acid and α-linolenic acid and is known for its cognitive benefits such as memory and thinking ability enhancement, intelligence improvement, and intellectual and visual development. (Carlson et al., 2013). Therefore, it is evident that air-frying has the potential to enhance the nutritional quality of sour meat products to a certain degree.

Organic acids are the main acidic components in fermented meat products, with their content and type exerting a significant effect on the sensory attributes of the products. Common organic acids found in fermented meat products include lactic acid, succinic acid, malic acid, citric acid, tartaric acid, etc. In this study, tartaric acid was not detected in all sample groups, while malic acid and citric acid were not identified as differential metabolites due to the lack of significant differences in their contents. Lactic acid was the most abundant organic acid in all the sample groups, with a mild acidic flavor that was a good acid agent (Wang, Li, et al., 2022). The lactic acid content of sour meat was significantly decreased by different heat treatments, and the lowest lactic acid content was found in the AF group. This finding could be related to the massive loss of lactic acid along with the moisture in sour meat caused by heat treatment. The reduction of lactic acid content may be one of the major reasons for the rising pH in the samples of sour meat after frying treatment. In accordance with the study of Yang et al. (2022), succinic acid serves as an ideal acidifier due to its ability to provide appropriate acidity levels and through its derivative succinate, lessens fat oxidation and improves the color of meat products. Furthermore, succinic acid and its sodium version have a strong promotion effect on food umami and can synergize with other umami components like amino acids and nucleotides to improve the umami taste of food. In our study, the change in succinic acid content was affected differently by various frying methods. Compared to the RAW group, the succinic acid content was significantly higher in the F and AF groups, while it was substantially lower in the M group, which was consistent with the trend of the intensity of the response of the umami sensor in the *E*-tongue analysis. Consequently, we surmised that succinic acid also plays an important part in the formation of the umami taste of sour meat products.

3.6. Differential metabolic pathway analysis

KEGG was used to analyze the metabolic pathways of 66 differential metabolites in order to understand the internal relationships of metabolites more intuitively (Fig. 5c). The significantly enriched pathways with Pathway Impact values >0.05 were selected for analysis, among which 22 significant metabolic pathways were screened out. Among them, the metabolism of linoleic acid had the greatest influence on the flavor of fried sour meat products. It is well known that linoleic acid is an important flavor precursor of volatile aldehydes, which is capable of generating stable aldehydes, such as hexanal and heptanal, under the action of heat-induced oxidation. Besides, the metabolism of arachidonic acid is considered to be one of the metabolic pathways that exerts a pronounced impact on the flavor profile of fried sour meat products. However, in our previous study, lower levels of arachidonic acid were detected, which might be able to serve as favorable proof that arachidonic acid is adequately metabolized during processing. Therefore, we hypothesized that thermal processing-induced oxidative degradation of fatty acids is one of the most important pathways contributing to the flavor formation of fried sour meat products.

In addition, the digestion and absorption of proteins also have a significant effect on the flavor of fried sour meat products (Fig. 5c). Among them, the metabolism of alanine, aspartic acid and glutamic acid had the most significant effect on the flavor of fried sour meat. As flavor amino acids, aspartic acid and glutamic acid have a crucial role in the flavor attributes of fried sour meat (Wang, Dong, et al., 2022). Nonetheless, in the process of deep-frying acidic meat, the heat-induced production of free radicals would attack the free amino acids, resulting in the formation of readily degradable amino acid radicals, which subsequently impact the flavor of sour meat through further oxidative degradation. Meanwhile, the synthesis and metabolism of arginine also had a high impact value, which justified the change of arginine content in the metabolic analysis. Therefore, we hypothesized that the high temperature during frying induced the oxidative degradation process of proteins and produced a series of volatile flavor substances, which contributed to the formation of the overall flavor of fried sour meat products.

3.7. Metabolic network prediction of flavor formation

The prediction of the pathways for the formation of flavor substances in fried sour meat is shown in Fig. 6. The formation of flavor in fried sour meat products originates from two main components, the oxidative decomposition of proteins and fats as well as the metabolism of certain carbohydrates during the fermentation process, and the heat-induced oxidation during the frying process. Fat oxidation is the most important pathway for flavor formation, and the high temperature treatment during frying induces the generation of many lipid radicals, which triggers a free radical chain reaction, thus promoting lipid oxidation and hydrolysis. In addition, it has been reported that fat may also participate in Strecker degradation and Maillard reaction to form a series of volatile flavor components that are important contributors to the overall flavor of the product (Shahidi & Oh, 2020). Fatty acids are the main products of oxidative degradation of fats and are also important flavor precursors. According to our study, aldehydes such as hexanal, heptanal, and octanal were mainly responsible for the formation of specific flavors in fried sour meat products. The enzymatic oxidation of linoleic acid during fermentation, coupled with the autoxidation of linoleic acid induced by high temperatures during frying, likely underlies the mechanism behind their formation.

In addition, α-keto acids are a key intermediate in the production of volatile flavor substances from free amino acids. During fermentation, as a result of transaminases acting on some branched-chain amino acids, α-keto acids are produced. α-keto acid decarboxylase subsequently reacts with the α-keto acids to produce aldehydes, alcohols, and ketones. The primary alcohol compound found in sour meat products, 1-octen-3-ol, is predominantly generated through the degradation of unsaturated fatty acids like linoleic acid and arachidonic acid, as well as the decomposition of the amino acid Strecker in the Ehrlich pathway. Additionally, its formation may also involve the oxidation of certain straight-chain aldehydes (Liu, Shen, et al., 2022; Y. Wang et al., 2020). Finally, carbohydrates are mainly derived from the primary accumulation in animal muscle tissues, which are pyrolyzed into easily absorbed monosaccharides during processing. They also play a partial role in the Maillard reaction as flavor precursors to generate volatile flavor substances.

4. Conclusion

To sum up, the frying methods influenced the odor and taste attributes of low-salt sour meat. The pH levels of the sour meat samples maturing by different frying methods were markedly elevated, while the

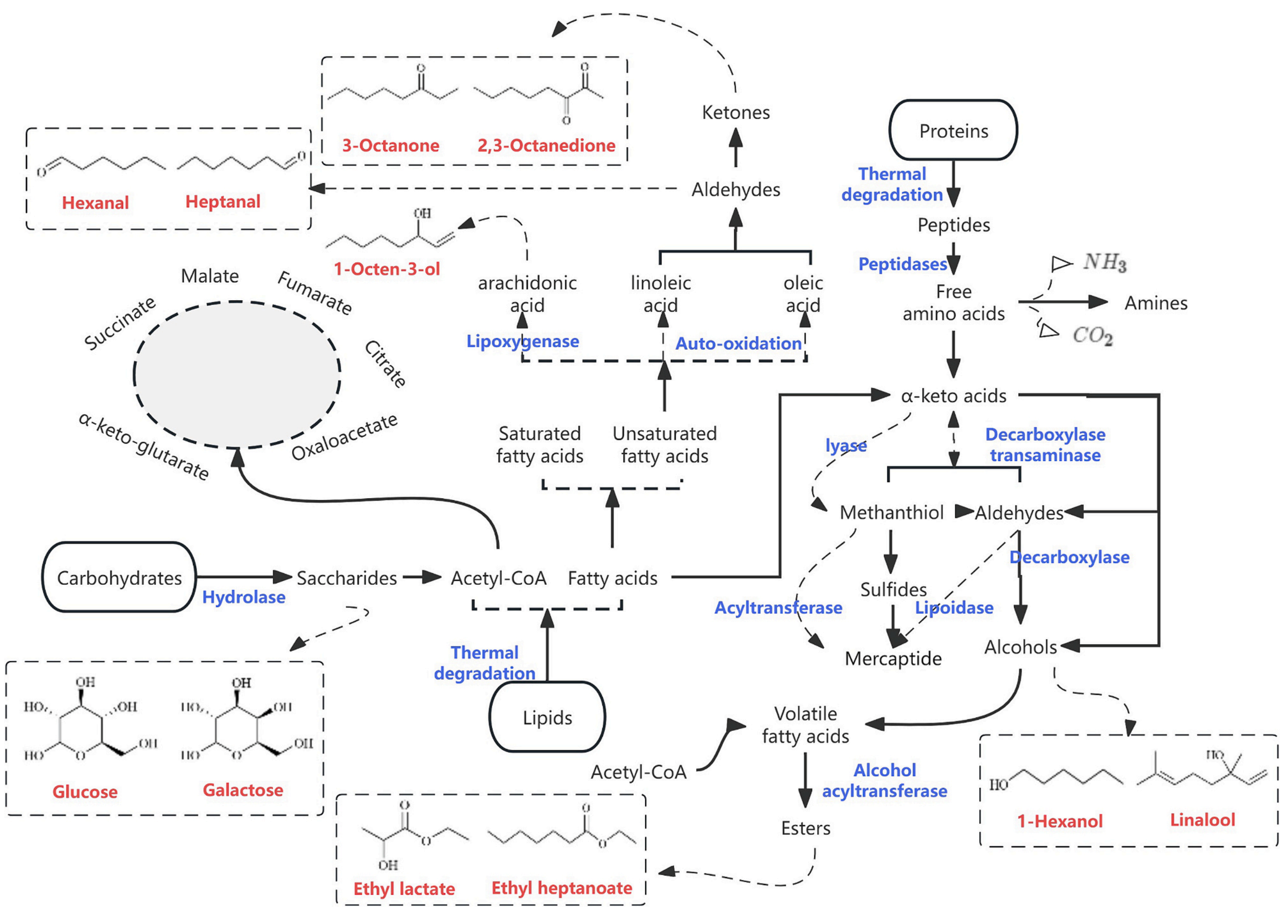

Fig. 6. Predicted metabolic network for flavor formation in low-salt sour meat during frying. Red represents the flavor substance, blue represents the reaction pathway, and black represents the flavor precursor substance or structural formula. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

TBARS values were notably reduced in comparison to the untreated low-salt sour meat samples. Combining the principal component analysis of E-nose and E-tongue with GC–MS and LC-MS proved effective in distinguishing low-salt sour meat samples cooked using various frying methods. The GC–MS results indicated that the unique hot-air circulation heating mode of air frying could facilitate heat-induced reactions within low-salt sour meat, thereby contributing to optimizing the composition ratio of volatile flavor compounds and the generation of distinctive flavor profiles. Amino acids, fatty acids, nucleotides, and organic acids emerge as the primary differential metabolites in sour meat prepared by various frying methods, serving as crucial precursors responsible for the flavor variations observed in low-salt sour meat. Among them, the metabolism pathways involving linoleic acid and alanine, aspartic acid, and glutamic acid were identified as the principal origins of discrepant metabolites, implying that lipid and protein oxidation are the main determinants of the flavor of fried low-salinity meat. In summary, all varied frying methods exhibited a favorable impact on the formation of low-salt sour meat flavor. In this study, air frying treatment could be a good choice for cooking low-salt sour meat because of its flavor optimization effect and nutritional enhancement effect. These experimental results expand the avenues for the deep processing of low-salt sour meat products and establish a theoretical foundation for their healthy and industrial development.

Ethical Guidelines

Ethics approval was not required for this research.

CRediT authorship contribution statement

Lujie Cheng: Writing – original draft, Visualization, Investigation, Formal analysis. **Qia Wang:** Writing – review & editing, Methodology, Formal analysis. **Xiefei Li:** Writing – review & editing, Validation. **Xinyuan Huang:** Methodology, Investigation, Formal analysis. **Fengping An:** Writing – review & editing. **Zhang Luo:** Writing – review & editing, Conceptualization. **Jingjing Wang:** Writing – review & editing. **Qiaohui Zeng:** Writing – review & editing. **Peng Shang:** Supervision, Funding acquisition. **Zhendong Liu:** Funding acquisition, Conceptualization. **Qun Huang:** Supervision, Project administration.

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.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2024.101591.

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