

# Study on the Effects of Marinating and Drying Processes on the Flavor of Yellow Beef and Yak Beef

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## ABSTRACT

To clarify the regulatory effect of marinating and drying process on beef flavor and the flavor differences of processed products from different beef varieties, this study took Shandong yellow beef and Qinghai yak beef as research objects. Using headspace solid phase microextraction-gas chromatography-mass spectrometry (GC-MS) and gas chromatography-ion mobility spectrometry (GC-IMS) technologies, combined with relative odor activity value (ROAV), principal component analysis (PCA), partial least squares discriminant analysis (PLS-DA) and other methods, the evolution characteristics of volatile flavor substances in beef before and after processing were analyzed, and the flavor profile differences of the finished beef jerky were compared. The results showed that aldehydes were the core flavor substances of both beef varieties in the fresh meat stage, with trans-2-nonenal and n-nonanal as the key contributors respectively. After marinating and drying, the proportion of aldehydes decreased significantly, terpenes became the dominant flavor substances of beef jerky, and new Maillard reaction products such as pyrazines and esters were generated. There were significant flavor differences between the two finished beef jerky, with 28 and 26 volatile substances detected respectively, and D-limonene, linalool and 2-ethyl-1,3,5-trimethylpyrazine were the key differential markers. This study provides a theoretical basis and data support for the differentiated development of characteristic beef jerky.

## KEYWORDS

Beef jerky; Gas chromatography-mass spectrometer (GC-MS); Gas chromatography-ion mobility spectrometry (GC-IMS); Volatile flavor compounds

## 1. INTRODUCTION

### 1.1. Overview of Beef

Beef is a high-quality ingredient known for its high protein and low fat content, rich in essential amino acids, vitamins, and minerals. Its processed products are favored by consumers for their nutritional value and convenience. [2] Beef ranks second in meat production in China. Beef jerky, produced from beef using traditional processing techniques, offers unique flavor, high nutrition, long shelf life, and is easy to carry for travel, making it a favorite convenient meat product among various ethnic groups in China. In recent years, consumers have demanded higher quality and flavor from beef jerky,

driving the industry towards specialization and differentiation. The raw material variety and processing technology are core factors determining the quality of finished beef jerky.

## **1.2. Current Research Status of Processing of the Two Beef Types**

Shandong yellow cattle, a high-quality local beef breed in North China, have moderate muscle fiber and even fat distribution, making them well-suited for jerky processing. Qinghai yak, an ancient and primitive livestock breed on the Qinghai-Tibet Plateau, has undergone long-term "natural selection + artificial breeding" to develop excellent characteristics adapted to the harsh, cold environment. Its meat has the advantages of high protein content, low fat content, and rich amino acid diversity, with excellent slaughter and meat production performance. [3] Significant differences exist in muscle fiber, fat content, and fascia distribution among different beef cuts, which directly affect the composition and release efficiency of flavor substances in processed products. [4] Currently, research on these two beef types mostly focuses on nutritional quality analysis of a single breed or optimization of processing techniques. Few studies systematically compare the dynamic evolution of flavor substances from fresh meat to finished product under the same processing technology, nor have they clearly defined the volatile flavor substance characteristics of the two finished jerky products. Existing research cannot explain the causes of flavor differentiation in jerky from different beef varieties under the same process, nor can it support flavor-directed regulation and differentiated product development for these two specialty raw materials.

## **1.3. Research Content and Objectives**

Existing domestic research on beef jerky [5] mostly focuses on optimizing general processing techniques for a single variety, rarely combining specialty raw materials [6] to study the dynamic changes in flavor and the flavor differences of finished jerky, thus failing to meet the practical needs of developing and promoting specialty snack foods. Although recent studies have examined flavor differences in beef jerky from different regions, confirming that origin, raw material variety, and processing technology jointly lead to significant differentiation of volatile components such as aldehydes, alcohols, and esters, [7] systematic research on the flavor formation mechanism, key marker differences, and raw material suitability of Shandong yellow beef and Qinghai yak beef under a uniform marinating and drying process remains scarce. This study aims to investigate the dynamic changes in volatile flavor substances from raw meat to processed jerky and to clarify the differences in the composition and content of volatile flavor substances between Shandong yellow beef jerky and Qinghai yak beef jerky. [8] HS-SPME-GC-MS can simultaneously determine multiple compounds with minimal interference, high quantitative accuracy, and reliable qualitative analysis. Additionally, headspace solid-phase microextraction coupled with gas chromatography-mass spectrometry can effectively separate and identify volatile components in complex mixtures, offering advantages such as high sensitivity, good reproducibility, low cost, simple operation, and convenience, making it widely used in food flavor analysis. [9] This study will analyze the causes of flavor differences and the influence of variety on the two types of beef jerky under this process. The core of this research is to systematically analyze the changes in volatile flavor substances from fresh beef to marinated and dried jerky, clarify the regulatory mechanism of the marinating and drying process on beef flavor, comprehensively compare the flavor profile differences of the two finished jerky products, and screen for key flavor markers that distin

## 2. MATERIALS AND METHODS

### 2.1. Materials and Equipment

#### 2.1.1. Main Materials

The main raw materials used in this study include Shandong yellow beef, Qinghai yak beef, green Sichuan pepper, etc. Details are shown in Table 1.

**Table 1.** Experimental Materials

Experimental Material Source	Experimental Material Source
Shandong Beef	Liangshan Loyalty and Ambition Hongtu Agricultural Technology Development Co., Ltd.
Qinghai Yak	Haidong Tibetan Autonomous Prefecture, Qinghai Province
Green Sichuan Pepper	Zibo Haiyue Longgong Logistics Port
Iodized Edible Salt	China Salt (Shanghai) Salt Industry Co., Ltd.
Chicken Bouillon Powder	Shanghai Totole Food Co., Ltd.
Mixed Spices	Zibo Haiyue Longgong Logistics Port

#### 2.1.2. Experimental Equipment

**Table 2.** Experimental Equipment

Experimental Equipment Model Manufacturer	Experimental Equipment Model Manufacturer	Experimental Equipment Model Manufacturer
Gas Chromatography-Mass Spectrometer7890AAgilent Technologies, Inc.	Gas Chromatography-Mass Spectrometer7890AAgilent Technologies, Inc.	Gas Chromatography-Mass Spectrometer7890AAgilent Technologies, Inc.
Gas Chromatography - Ion Mobility Spectrometer (FlavourSpec®) by G.A.S. GmbH, Germany	Gas Chromatography - Ion Mobility Spectrometer (FlavourSpec®) by G.A.S. GmbH, Germany	Gas Chromatography - Ion Mobility Spectrometer (FlavourSpec®) by G.A.S. GmbH, Germany
Electronic Analytical Balance AL104, Mettler Toledo Instrument Co., Ltd.	Electronic Analytical Balance AL104, Mettler Toledo Instrument Co., Ltd.	Electronic Analytical Balance AL104, Mettler Toledo Instrument Co., Ltd.
Metal Bath Solid Phase Microextraction Probe Holder KL01 Beijing Kanglin Technology Co., Ltd.	Metal Bath Solid Phase Microextraction Probe Holder KL01 Beijing Kanglin Technology Co., Ltd.	Metal Bath Solid Phase Microextraction Probe Holder KL01 Beijing Kanglin Technology Co., Ltd.
Solid Phase Extraction Syringe	Solid Phase Extraction Syringe	Solid Phase Extraction Syringe
Low-temperature Freezer	Low-temperature Freezer	Low-temperature Freezer
Electric Thermostatic Drying Oven	Electric Thermostatic Drying Oven	Electric Thermostatic Drying Oven

### 2.2. Experimental Methods

#### 2.2.1. Processing Technology of Beef Jerky

Fresh beef → Pretreatment → Cutting into strips → Marinating → Drying → Packaging

(1) Pretreatment: Select fresh Shandong yellow beef and Qinghai yak beef, remove fat, membrane, and external fat, cut into pieces weighing about 100g along the muscle fiber direction, and rinse with clean water to remove dirt and blood [10].

(2) Cutting into strips: Cut along the beef fiber direction into strips with a cross-section of 2 cm × 2 cm and a thickness of approximately 1.0–1.2 cm.

(3) Marinating: Put 100g of cut beef strips into a clean vacuum packaging bag. Add 1.27g of Sichuan pepper, 2.86g of salt, 3.02g of monosodium glutamate, 39.68g of water, and 9.63g of mixed spices. Mix thoroughly so that each beef strip is evenly coated with the marinade. Seal and marinate at a constant temperature of 4°C in a refrigerator for 24 hours.

(4) Drying: After marinating, remove the beef strips from the marinade. Tie a knot tightly at one end of each beef strip using heat-resistant sterile cotton thread. Mark according to the experimental groups and hang sequentially on a dedicated drying rack inside an oven, ensuring a spacing of ≥2 cm between strips without contact or overlap. Place the rack with beef strips into a constant temperature drying oven set at 60°C. Start the drying program and dry continuously for 5 hours. Do not open the oven door during drying to avoid temperature fluctuations affecting the moisture content and flavor substance retention of the beef jerky.

(5) Packaging: After drying, turn off the oven power. Allow the oven to cool naturally to room temperature before removing the beef jerky. Remove the cotton thread, weigh the dried beef jerky using an electronic balance, place it in sealed bags, and store at a constant temperature of 4°C in a refrigerator.

### 2.2.2. ROAV

Referring to the method of Liu Dengyong [11] for determining key flavor compounds in food, ROAV was calculated according to the following formula :

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

Where  $ROAV_i$  represents the ROAV of a volatile flavor substance;  $C_i$  represents the relative content of a volatile flavor substance;  $C_{max}$  represents the relative content of the flavor substance with the greatest contribution;  $T_{max}$  represents the threshold of the flavor substance with the greatest contribution;  $T_i$  represents the threshold of a certain flavor compound.

### 2.2.3. Detection of Volatile Substances by Gas Chromatography-Mass Spectrometry

Weigh 2g of sample into a 20 mL headspace vial and seal immediately. Preheat the vial in a metal bath at 60°C for 5 minutes for equilibration, then extract using an SPME fiber (activated) for 30 minutes. Insert the fiber into the GC injector for desorption at 250°C for 5 minutes.

Chromatographic conditions: Modified according to the experimental conditions of Yang Shengnan et al. [12] An HP-5MS column was used. High-purity helium was the carrier gas. The injector temperature was 250°C, flow rate 1 mL/min, split ratio 4:1. Temperature program: initial temperature 40°C held for 1 min, then increased at 5°C/min to 250°C and held for 5 minutes.

Mass spectrometric conditions: Electron ionization (EI) source energy 70 eV, ion source temperature 230°C, transfer line temperature 250°C, scan range m/z 35–500.

### 2.2.4. Detection of Volatile Substances by Ion Mobility Spectrometry

Referencing the method of Yao Wensheng et al., [8] with slight modifications. Weigh 2g of sample into a 20 mL headspace vial. Headspace conditions: incubate at 50°C for 20 min, syringe temperature 80°C, incubation rotation speed 500 r/min. FlavourSpec® instrument conditions: column type MXT-5 (15 m × 0.53 mm ID × 1 μm), analysis time 20 min, carrier/drift gas high-purity nitrogen.

### 2.2.5. Data Processing

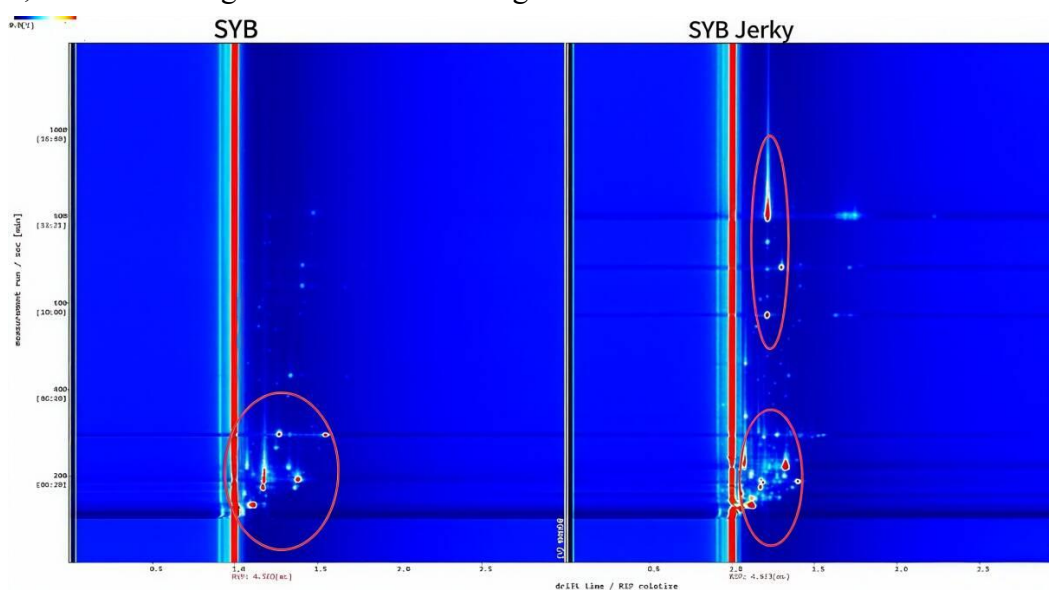
Microsoft Excel 2016 was used for initial data integration and calculation. Origin 2024 software, Simca 14.1 software, and Gallery Plot software were used for graphing.

## 3. RESULTS AND DISCUSSION

### 3.1. GC-IMS Analysis

#### 3.1.1. GC-IMS Analysis of Shandong Yellow Beef and Shandong Yellow Beef Jerky

The GC-IMS two-dimensional spectra of fresh Shandong yellow beef (SYB) and Shandong yellow beef jerky (SYB Jerky) are shown in Figure 1. The red vertical line at 1.0 on the horizontal axis represents the reactant ion peak (RIP). Each signal point on either side of the RIP represents a volatile organic compound. The color and brightness of the signal points represent the concentration of the substance; darker and brighter colors indicate higher concentrations.

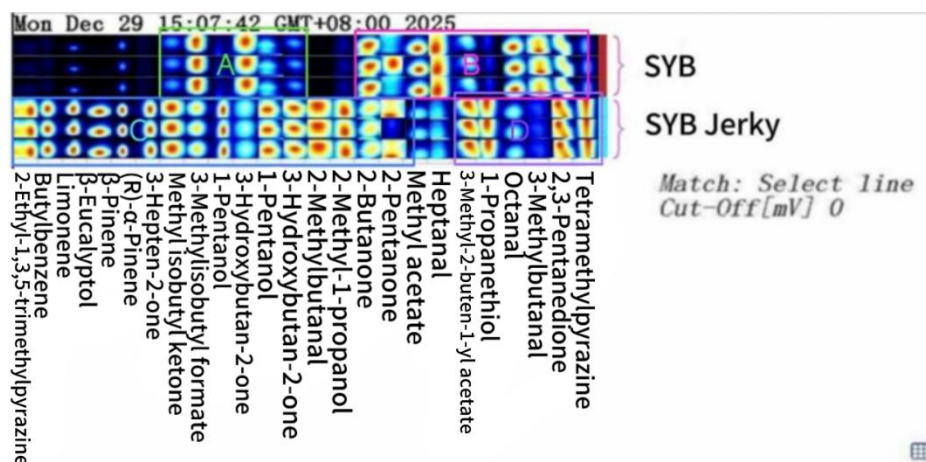


**Figure 1.** 2D spectra of SYB and SYB Jerky (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky)

Qualitative analysis of SYB and SYB Jerky via GC-IMS identified multiple characteristic peaks, covering aldehydes, ketones, esters, alcohols, nitrogen-containing heterocycles and other types of flavor substances (see Appendix Table 2 for details).

The volatile flavor substance fingerprint spectra of SYB and SYB Jerky were constructed using the Gallery Plot plug-in (Figure 2), which clearly divided 4 characteristic flavor regions, realizing the visualization of flavor differences before and after processing. Region A: Exclusive characteristic region of SYB. The core substances are (R)- $\alpha$ -pinene, 1,8-cineole and other terpenoids, which endow the raw meat with a faint plant-like fragrance. These substances are completely degraded and disappeared after processing, and can be used as marker substances to distinguish raw materials and finished products of Shandong Yellow Beef. Region B: Shared flavor region of both samples. The core substances are 3-methylbutanal, tetramethylpyrazine, heptanal, etc., which form the basic flavor skeleton of Shandong Yellow Beef. Among them, 3-methylbutanal and heptanal contribute fatty and green grassy notes, and are the core flavor substances in the raw meat stage. Region C: Exclusive processing characteristic region of SYB Jerky. The core substances are alkylpyrazines such as 2-ethyl-1,3,5-trimethylpyrazine, which are characteristic products of the Maillard reaction. They endow the beef jerky with rich roasted and nutty aromas, and are the newly added core flavor substances after processing. Region D: Flavor enhancement region of SYB Jerky. The core substances are

limonene, 3-methylthiopropanol, methyl acetate, etc. Among them, terpenoids such as limonene are derived from the spices used in curing, endowing the beef jerky with fresh citrus and herbaceous aromas, and are the core flavor-enhancing components after processing.

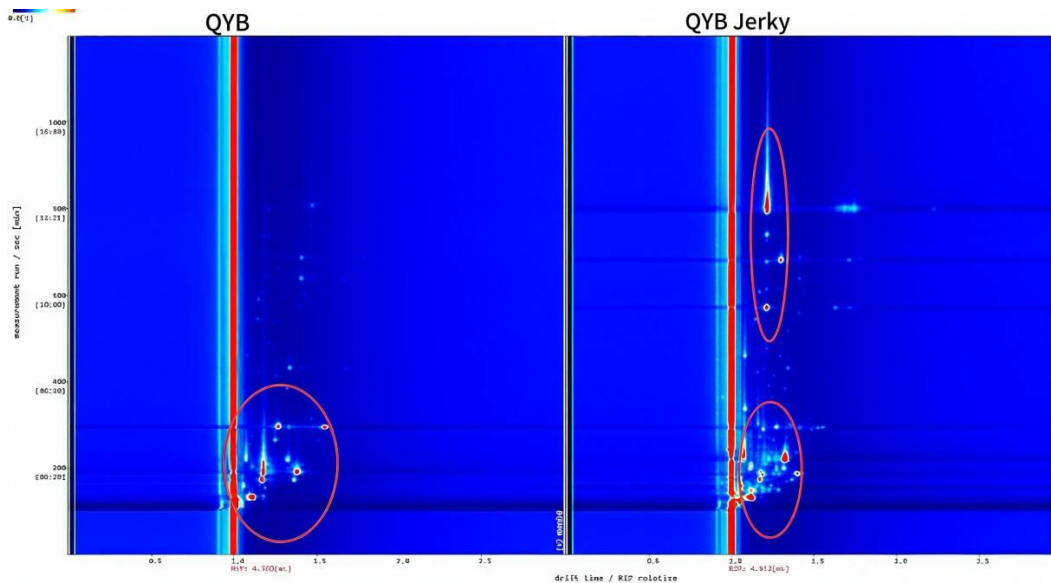


**Figure 2.** VOCs fingerprint profiles of SYB and SYB Jerky (SYB: Shandong Yellow Beef SYB Jerky:Shandong Yellow Beef Jerky)

Based on the qualitative results from the GC-IMS database, more than 20 volatile monomers were detected in SYB and SYB Jerky, covering aldehydes, alcohols, ketones, terpenes, esters, pyrazines and other categories. Among them, SYB is dominated by aldehydes, which are the core flavor category in the raw meat stage. The high proportion of aldehydes in raw meat originates from fatty acid oxidation. After processing, the content of aldehydes decreases, which is attributed to the volatilization of small-molecule aldehydes caused by high temperature, and the conversion of aldehydes into pyrazines via the Maillard reaction with amino acids. In SYB Jerky, the proportions of terpenes and esters increase significantly, and the types and contents of alcohols, ketones and pyrazines also rise remarkably, becoming important flavor components of the beef jerky. This confirms that the curing and drying process completely changes the composition of volatile flavor substances in Shandong Yellow Beef.

### 3.1.2. GC-IMS Analysis of Qinghai Yak Meat and Qinghai Yak Jerky

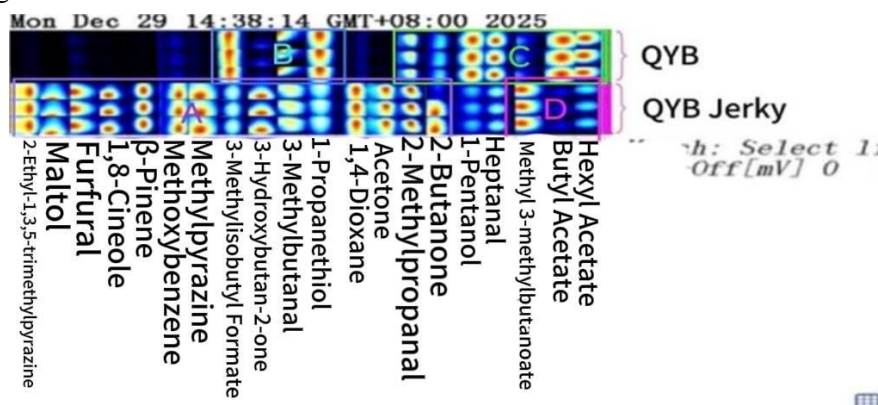
GC-IMS was used to analyze the volatile organic compounds in QYB and QYB Jerky. The two-dimensional spectra showed significant differences in the signal peak characteristics between QYB and QYB Jerky. The signal peaks of QYB were mainly concentrated in the retention time range of 200–600 s, with a small number and low intensity. In contrast, the number of signal peaks in QYB Jerky increased substantially. Not only did the signal peak intensity in the 200–600 s range improve significantly, but a large number of new signal peaks also appeared in the retention time range of 900–1500 s, indicating that curing and drying processing also significantly increased the variety and content of volatile flavor substances in Qinghai yak meat.



**Figure 3.** 2D spectra of QYB and QYB Jerky (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

Qualitative analysis of Qinghai yak meat and Qinghai yak jerky by GC-IMS (see Appendix Table 3 for details) identified multiple characteristic peaks. Most of them were qualitatively identified, covering various flavor substances such as esters, aldehydes, ketones, alcohols, terpenes, and nitrogen-containing heterocycles.

Fingerprint spectra plotted by the Gallery Plot plug-in were used to analyze the volatile aroma compounds of raw QYB and QYB Jerky (see Figure 4). Four characteristic flavor regions, A, B, C, and D, could be classified. Region A was the exclusive processing characteristic region of QYB Jerky, with core substances including 2-ethyl-1,3,5-trimethylpyrazine, maltol, etc., which are characteristic products of the Maillard reaction. They endow the jerky with strong roasted and caramel-like aromas, serving as newly formed key flavor substances after processing. Region B was the common flavor region of both samples, with aldehydes such as heptanal, 3-methylbutanal, and 2-methylpropanal as the core components, forming the basic flavor skeleton of Qinghai yak meat and acting as the main flavor contributors in the fresh meat stage. Region C was the exclusive characteristic region of QYB, dominated by terpenoids including  $\beta$ -pinene and 1,8-cineole, which provide fresh meat with a unique cool herbal aroma. The content of these substances decreased significantly after processing, with only a small amount retained in the final product. Region D was the flavor enhancement region of QYB Jerky, with core substances including ethyl acetate, hexyl acetate, furfural, etc. Among them, esters contribute a fresh fruity aroma to the jerky, acting as important modifying components of the flavor after processing.

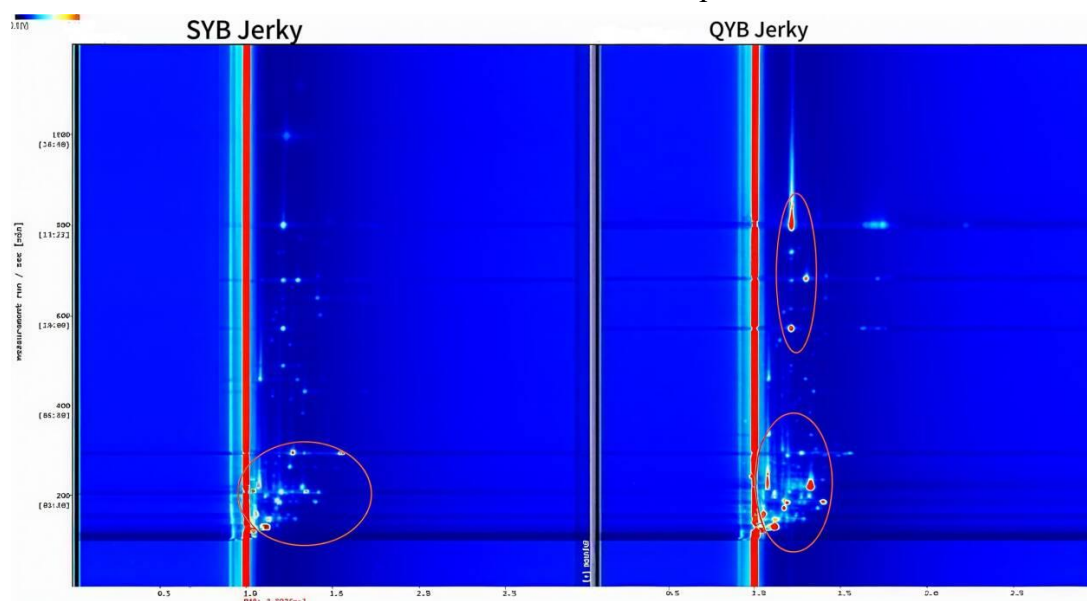


**Figure 4.** VOCs fingerprints of QYB and QYB Jerky (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

Combined with the gas chromatography retention time, ion migration time of volatile substances in Appendix Table 3 and GC-IMS database matching, qualitative analysis was performed on QYB and QYB Jerky. More than 20 volatile monomers were detected in QYB and QYB Jerky, including aldehydes, alcohols, ketones, terpenes, esters, pyrazines and others. Among them, QYB was dominated by aldehydes, which were the core flavor category in the fresh meat stage. In QYB Jerky, the proportion of terpenes increased significantly, and pyrazines and esters became important newly added flavor components. The remarkable increase in terpenes in the jerky mainly came from the composite spices added during curing. Such substances have low threshold and strong aroma activity, and are the core source of the fresh herbal aroma of the jerky. It confirms that the curing and drying process significantly changed the composition of volatile flavor substances in Qinghai yak meat, and terpenes became the dominant flavor substances in the finished jerky.

### 3.1.3. GC-IMS Analysis of Shandong Yellow Beef Jerky and Qinghai Yak Beef Jerky

GC-IMS was used to analyze the volatile organic compounds in Shandong Yellow Beef Jerky and Qinghai Yak Beef Jerky, and the results are shown in Figure 5. There were significant differences in the intensity and distribution of characteristic peaks between the two spectra, and the overall signal peaks of Qinghai Yak Beef Jerky were stronger. It can be seen from the spectra that the signal peak intensity and distribution differed obviously between the two types of jerky, and the overall signal peak brightness of QYB Jerky was higher, indicating a higher overall concentration of volatile substances. The signal peaks of both samples were mainly concentrated in the retention time range of 200–600 s, with more signal peaks in this range for SYB Jerky. In contrast, QYB Jerky showed denser signal peaks in the retention time range of 600–1500 s, suggesting significant differences in the polar distribution of volatile substances between the two products.

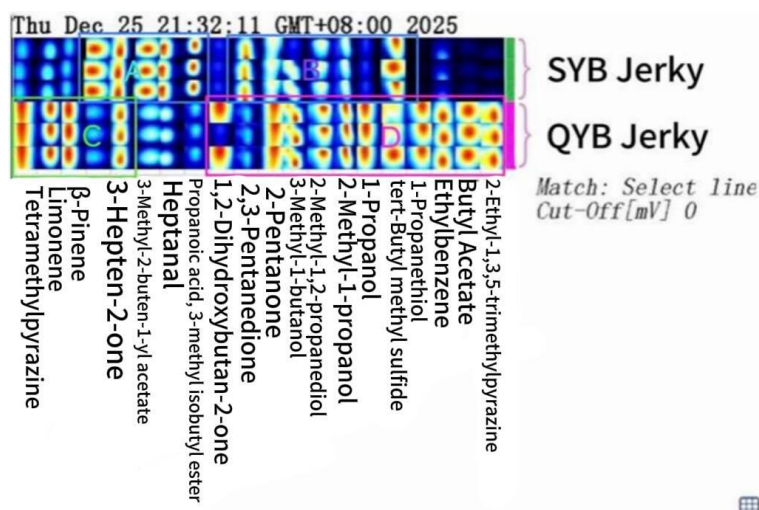


**Figure 5.** 2D spectra of SYB Jerky and QYB Jerky (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

Qualitative analysis of Shandong Yellow Beef Jerky (SYB Jerky) and Qinghai Yak Beef Jerky (QYB Jerky) was conducted using GC-IMS. Most of the characteristic peaks were identified, and the components included esters, aldehydes, ketones, nitrogen-containing heterocycles and other flavor substances (see Appendix Table 4 for details).

Visual analysis of volatile aroma compounds in SYB Jerky and QYB Jerky was performed using fingerprint spectra plotted by the Gallery Plot plug-in (see Figure 6). Four characteristic flavor regions, A, B, C and D, were classified. Region A was the exclusive characteristic aroma region of SYB Jerky, with 3-methyl-2-buten-1-ol acetate and other esters as the core substances, which endowed it with unique fruity and fatty aromas and served as characteristic markers distinguishing it from QYB Jerky.

Region B was the common flavor region of both beef jerky samples, with tetramethylpyrazine, 2,3-pentanedione and others as the core substances, forming the basic roasted and milky aroma base of beef jerky as the shared key flavor components. Region C was the exclusive characteristic aroma region of QYB Jerky, with limonene,  $\beta$ -pinene and other terpenoids as the core substances, retaining part of the herbal aroma of the raw material as its key characteristic flavor components. Region D was the characteristic aroma enhancement region of QYB Jerky, with 2-ethyl-1,3,5-trimethylpyrazine, ethyl butyrate and others as the core substances. Among them, the content of pyrazines was significantly higher than that in SYB Jerky, making its roasted and nutty aroma more prominent, while esters further enhanced its fruity aroma layers.



**Figure 6.** Fingerprints of SYB Jerky and QYB Jerky (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

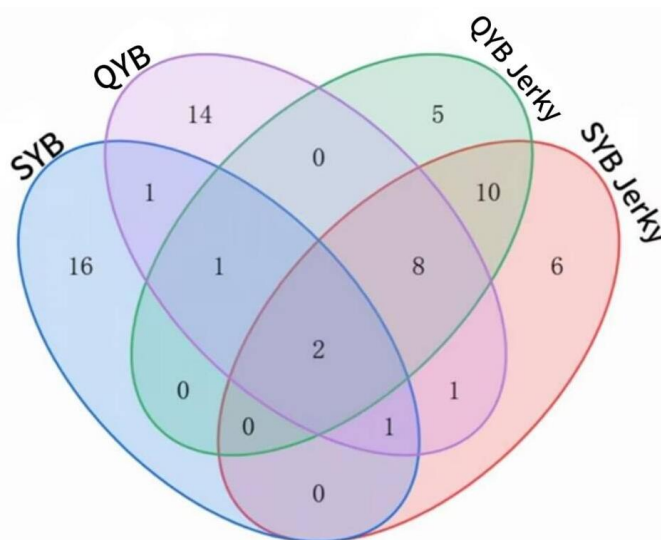
Qualitative analysis of volatile components in Shandong Yellow Beef Jerky and Qinghai Yak Beef Jerky was carried out based on the gas chromatography retention time and ion migration time of volatile substances in Appendix Table 4, combined with GC-IMS database matching. The results showed that more than 20 volatile monomers were detected from the two samples, including aldehydes, alcohols, ketones, terpenes, esters, pyrazines and other categories. Terpenes (such as limonene) were the main components in Shandong Yellow Beef Jerky, while the proportions of pyrazines (such as tetramethylpyrazine) and esters (such as ethyl butyrate) increased in Qinghai Yak Beef Jerky, and aromatic compounds also became important flavor components. Pyrazines exhibit nutty and roasted aromas produced during meat processing, and are typical products of the Maillard reaction between sugars and amino acids in meat processing. The differences in the categories and contents of these aroma components jointly constitute the characteristic flavors of Shandong Yellow Beef Jerky and Qinghai Yak Beef Jerky.

### 3.2. HS-SPME-GC-MS Analysis

Four beef samples (Shandong Yellow Beef, SYB; Shandong Yellow Beef Jerky, SYB Jerky; Qinghai Yak Beef, QYB; Qinghai Yak Beef Jerky, QYB Jerky) were analyzed using HS-SPME-GC-MS. The extraction efficiency of volatile flavor compounds in dried meat products is easily affected by factors such as sample amount, extraction temperature and time, and reasonable parameter settings can significantly improve the number and integrity of detected compounds [14]. Combined with the optimization experience of similar meat product detection, the extraction and detection parameters determined in this study could effectively cover major flavor substances such as aldehydes, terpenes and alcohols. Various volatile components were identified, and significant differences existed in the types and compositions of volatile flavor compounds among different samples (see Appendix Table 1 for details). Among them, hexanal and heptanal were the core volatile components common to meat products [15], and their detection further verified the reliability of the detection method in this study.

The types of volatile flavor compounds in SYB Jerky and QYB were more abundant than those in SYB. A total of 20 VOCs were identified in SYB, 28 in QYB, 28 in SYB Jerky and 26 in QYB Jerky, which could be classified into 8 categories.

In terms of category distribution, terpenoids accounted for a prominent proportion in jerky samples (11 in SYB Jerky, 12 in QYB Jerky), serving as the main source of the fresh herbal aroma in processed jerky. As the dominant flavor category in fresh meat, aldehydes were detected in 10 species in both SYB and QYB, and their content and number decreased significantly after processing. In addition, the differences in the number and distribution of ketones (7 in SYB Jerky, 3 in QYB), alcohols (6 in SYB, 3 in QYB), aromatic compounds (3 in SYB Jerky, 2 in QYB), hydrocarbons (4 in QYB, 3 in SYB Jerky), esters (1 in QYB Jerky) and ethers (1 in QYB) jointly formed the unique flavor profiles of different samples.



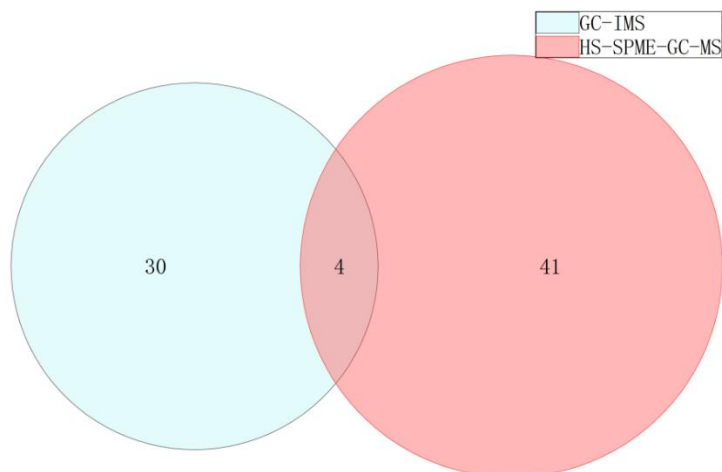
**Figure 7.** Venn diagram of volatile flavor compounds detected by GC-MS in four samples (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

### 3.3. Comprehensive Analysis Using GC-IMS and HS-SPME-GC-MS

Venn diagrams were plotted for the volatile flavor compounds detected by GC-IMS and HS-SPME-GC-MS, as shown in Figure 8. The study indicated that four flavor compounds were detected by both techniques, namely heptanal, eucalyptol, isovaleraldehyde, and 1-pentanol, suggesting a certain degree of specificity between the two analytical methods. The combined application of GC-IMS and HS-SPME-GC-MS, through technical complementarity, significantly improved the coverage of volatile flavor compound analysis in complex sample systems. This combined technique possesses both the high-throughput and rapid screening capacity of GC-IMS and the high-resolution and accurate qualification advantages of HS-SPME-GC-MS, enabling a more comprehensive characterization of the volatile components in the samples.

The results from GC-IMS and HS-SPME-GC-MS were both dominated by aldehydes, alcohols, terpenes, and esters. Aldehydes mostly originated from amino acid degradation and fatty acid oxidation, usually with low thresholds and strong odors. For example, the common components heptanal and isovaleraldehyde contributed characteristic flavors such as fatty and nutty aromas to the samples. Alcohols were mainly derived from fatty acid oxidation and carbohydrate metabolism, with relatively high thresholds, mostly presenting fruity and mellow notes, such as the common components eucalyptol and 1-pentanol. Terpenes were mostly characteristic components of the samples themselves, endowing products with fresh herbal and pine aromas. Esters were mainly produced by esterification between alcohols and acids, with low thresholds, mostly providing pleasant

fruity and ester-like fragrances. The synergistic effect of these components jointly constructed the flavor profiles of the samples.

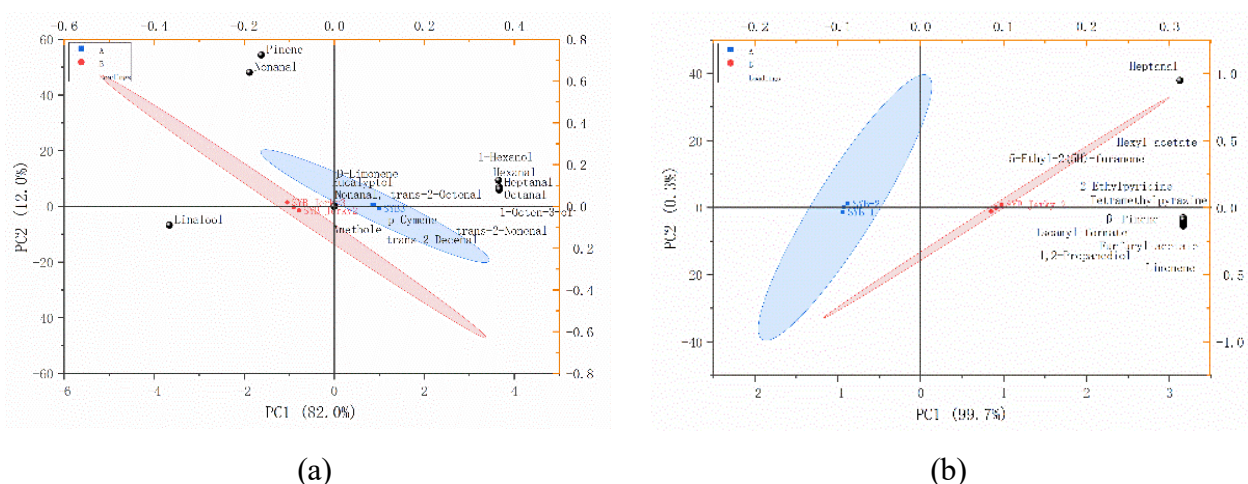


**Figure 8.** Venn diagram of VOCs detected by two analytical techniques

### 3.4. Differential Volatile Compound Screening

#### 3.4.1. Screening of Volatile Compounds Differences between Shandong Yellow Beef and Shandong Yellow Beef Jerky.

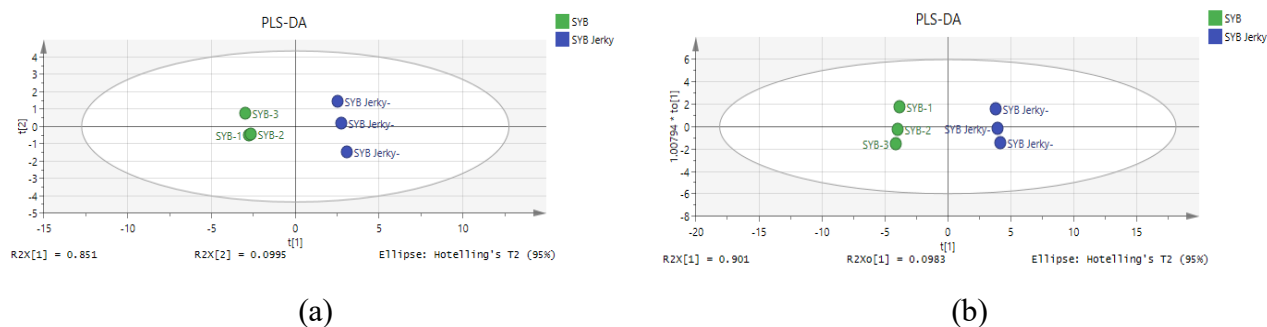
PCA is an unsupervised analytical method capable of linearly reducing the dimensionality of multivariate data. To reveal the differences in volatile compounds between SYB and SYB Jerky, dimensionality reduction was performed separately on the data obtained from GC-IMS and HS-SPME-GC-MS analyses, with the results shown in Figure 9. The cumulative contribution rate of the first two principal components in the GC-IMS model reached 100%, while that of the GC-MS model reached 94.0%, both sufficiently representing the overall flavor profiles of the samples. In the PCA plots of both techniques, SYB and SYB Jerky were completely separated, indicating significant differences in the composition and content of their volatile components. The consistent discriminatory trends observed with the two techniques provide a reliable statistical basis for evaluating the flavor profiles of the samples.



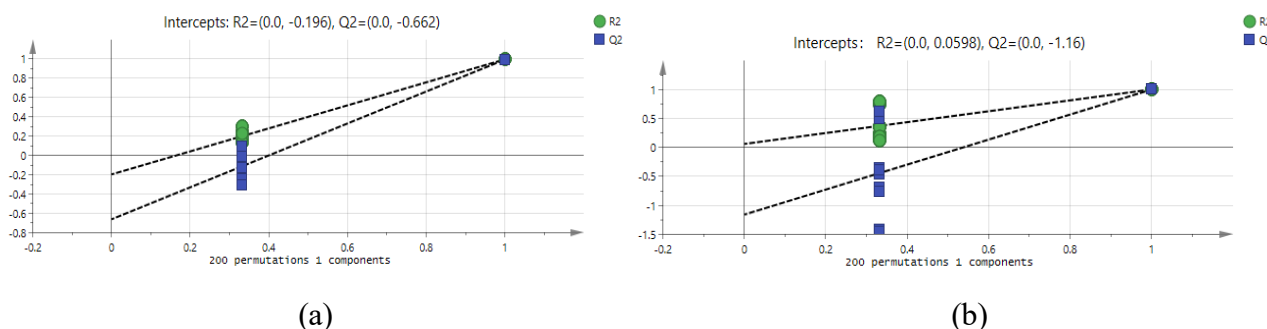
**Figure 9.** PCA Analysis of SYB and SYB Jerky (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky)

PLS-DA is a supervised analysis method that maximizes inter-group differences according to predefined classifications, offering superior separation performance compared to PCA. Using the relative contents of VOCs from the two techniques as the dependent variable and sample

classification as the independent variable, a PLS-DA model was constructed (Figure 10). The results show that for the GC-IMS model,  $R^2X = 0.851$ ,  $R^2Y = 0.999$ ,  $Q^2 = 0.999$ ; for the GC-MS model,  $R^2X = 0.901$ ,  $R^2Y = 0.999$ ,  $Q^2 = 0.998$ . All models exhibited  $R^2$  and  $Q^2$  values close to 1, indicating excellent goodness of fit and predictive capability. Validation through 200 permutation tests (Figure 11) confirmed that the intercepts of  $R^2$  and  $Q^2$  for both models were close to 0, with negative  $Q^2$  intercepts, demonstrating that the models are stable, reliable, and free from overfitting. Thus, they can effectively distinguish the flavor characteristics between raw materials and finished products.

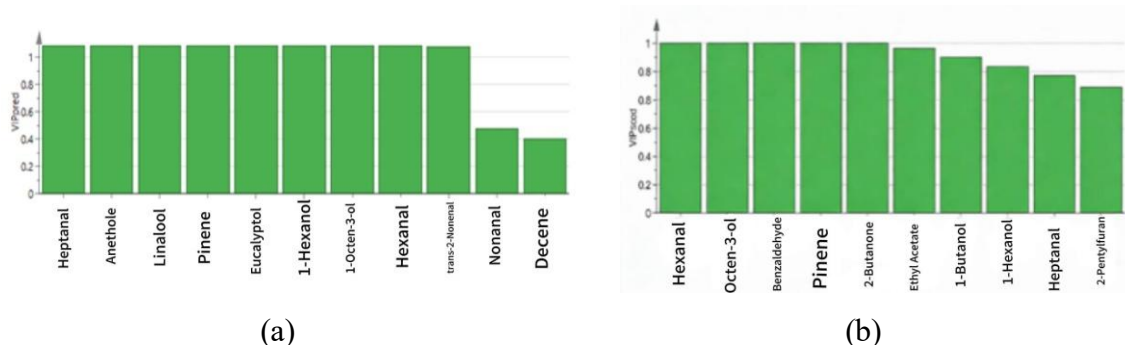


**Figure 10.** PLS-DA analysis of SYB and SYB Jerky (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky)



**Figure 11.** Permutation test for VOCs of SYB and SYB Jerky (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky)

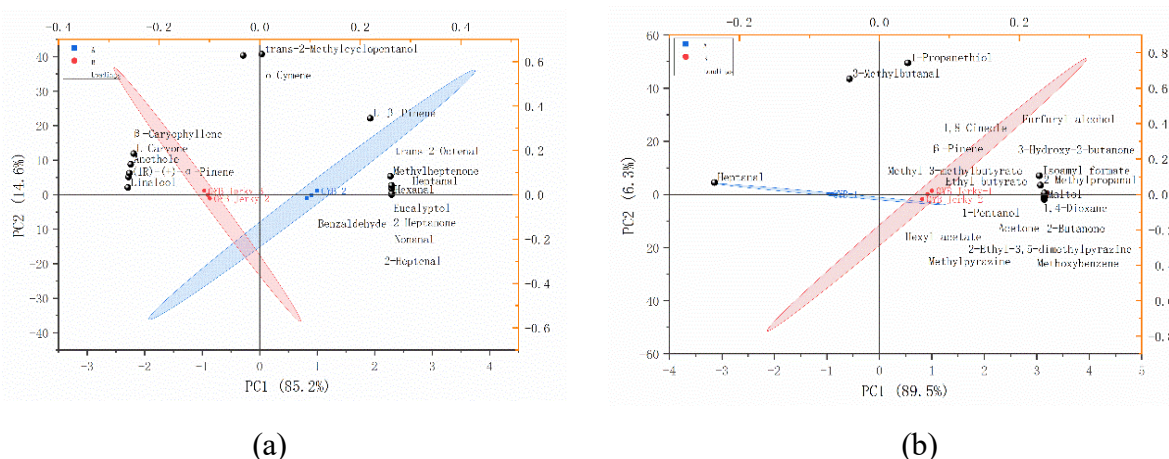
Based on the PLS-DA model, variable importance in projection (VIP) values were calculated, and significantly different compounds were screened using the criteria  $VIP > 1$  and  $P < 0.05$  (Figure 12). GC MS identified 11 differential compounds with  $VIP > 1$ , which, in descending order of VIP values, are heptanal, anethole, linalool, octanal, eucalyptol, 1 hexanol, 1 octen 3 ol, hexanal, trans 2 nonenal, nonanal, and pinene. The VIP values of the first nine differential compounds are close to 1, while the last two are also greater than 0.3, indicating that all contribute significantly to flavor discrimination between the two sample groups and serve as key indicators of flavor differences. GC IMS identified 10 differential compounds with  $VIP > 1$ , which, in descending order of VIP values, are hexanal, 1 octen 3 ol, benzaldehyde, furfural, 2 butanone, ethyl acetate, 1 propanol, 1 hexanol, heptanal, and 2 pentylfuran. The VIP values of all these differential compounds are significantly greater than 1, demonstrating extremely high contributions to the flavor discrimination between SYB and SYB Jerky.



**Figure 12.** SYB and SYB Jerky VIP Plot (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky)

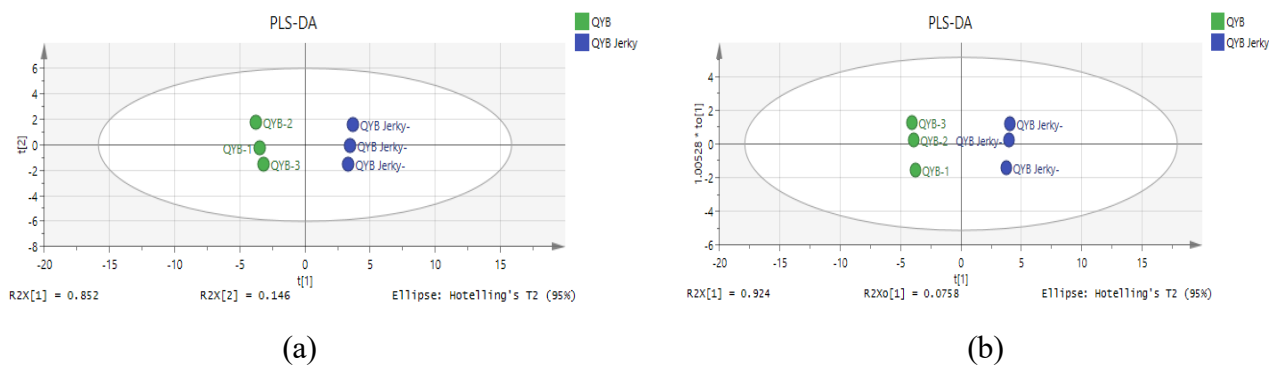
### 3.4.2. Screening of Volatile Compounds in Qinghai Yak Meat and Dried Qinghai Yak Meat

Principal component analysis (PCA) was performed on the data from the two detection techniques, and the results are shown in Figure 13. The cumulative contribution rate of the first two principal components in the GC-IMS model reached 95.8%, while that in the HS-SPME-GC-MS model reached 99.8%. QYB and QYB Jerky were completely separated in both models, indicating significant differences in their volatile components. The PCA model can effectively distinguish the flavor attributes of the raw materials and the finished product.

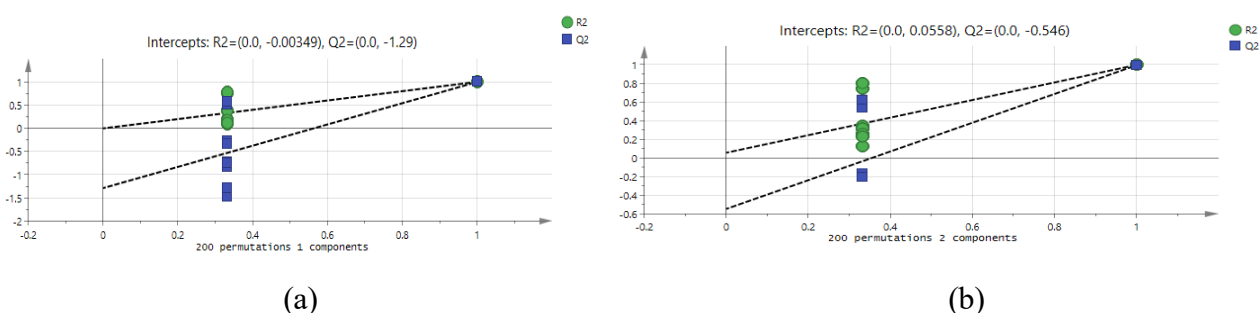


**Figure 13.** PCA analysis of QYB and QYB Jerky (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

A PLS-DA discriminant model was constructed (Figure 14). The results showed: in the GC-MS model, the cumulative explained variance of the two principal components  $R^2X = 0.998$ , the cumulative group explanation rate  $R^2Y = 0.999$ , and the cross-validated predictive power  $Q^2 = 0.997$ ; in the GC-IMS model, the cumulative explained variance of the two principal components  $R^2X = 1.000$ , the cumulative group explanation rate  $R^2Y = 0.999$ , and the cross-validated predictive power  $Q^2 = 0.998$ . Both models demonstrated excellent fit and predictive ability without overfitting. The results of 200 permutation tests (Figure 15) indicated that neither model exhibited overfitting, both were stable and reliable, and could effectively differentiate the flavor differences between raw materials and finished dried meat of Qinghai yak. The 200 permutation test results (Figure 15) show that both models are free from overfitting, stable, and reliable, and can be used to discriminate the flavor differences between raw and finished products of Qinghai yak meat.

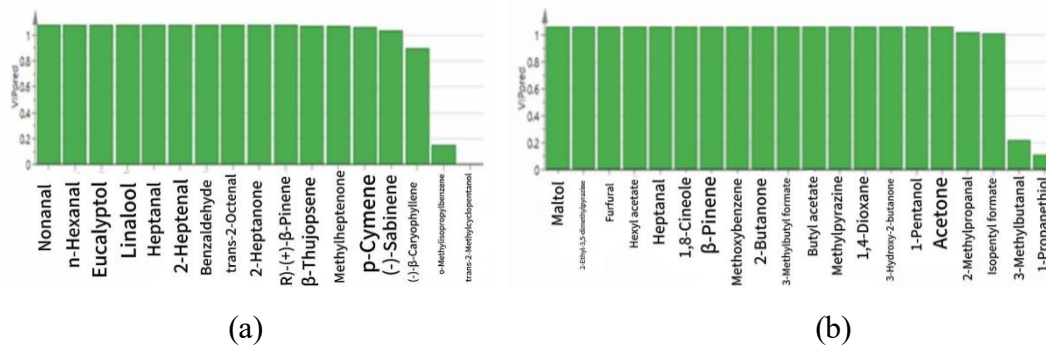


**Figure 14.** PLS-DA analysis of QYB and QYB Jerky (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)



**Figure 15.** Permutation test for VOCs of QYB and QYB Jerky (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

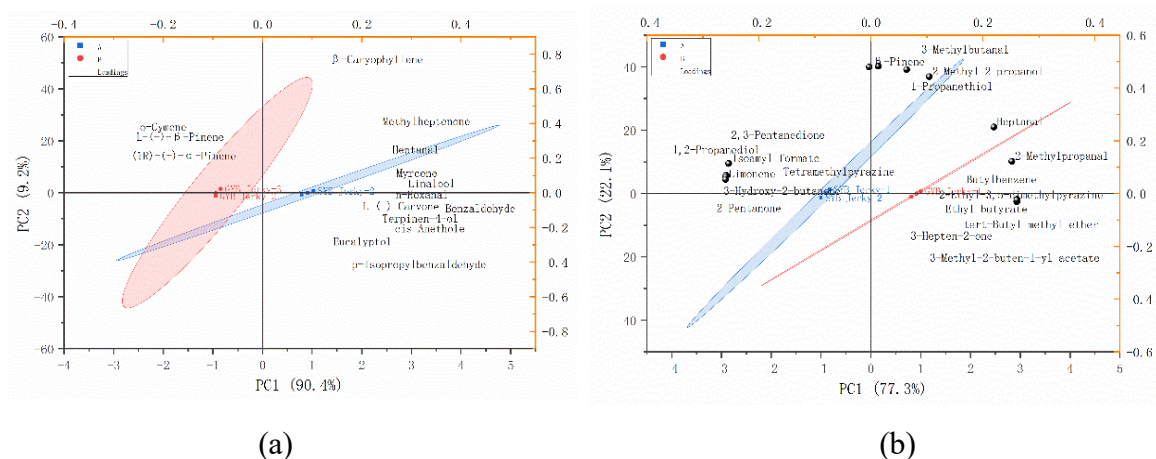
The volatile compounds detected by GC-MS are ranked in descending order of VIP value: nonanal, hexanal, eucalyptol, linalool, heptanal, 2-heptenal, 2-heptanone, (1R)-(+)- $\beta$ -caryophyllene, 6-methyl-5-hepten-2-one, anethole, L-carvone, L- $\beta$ -pinene, 2-methyl-5-(1-methylethyl)phenol, and trans-2-methylcyclopentanone. The differential compounds screened by GC-IMS primarily include maltol, 2-ethyl-3,5-dimethylpyrazine, furaneol, hexyl acetate, among others, with VIP values of all compound categories showing significant differences. Among these, aldehydes such as hexanal, nonanal, and heptanal mainly originate from lipid oxidation. Terpenes like linalool, eucalyptol, and  $\beta$ -caryophyllene are mostly natural aromatic substances inherent to Qinghai yak meat raw materials. In contrast, compounds such as maltol and pyrazines are closely associated with the Maillard reaction during processing. These compounds collectively constitute the core markers distinguishing the flavor differences between QYB raw materials and QYB jerky products.



**Figure 16.** QYB and QYB Jerky VIP Plot (QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

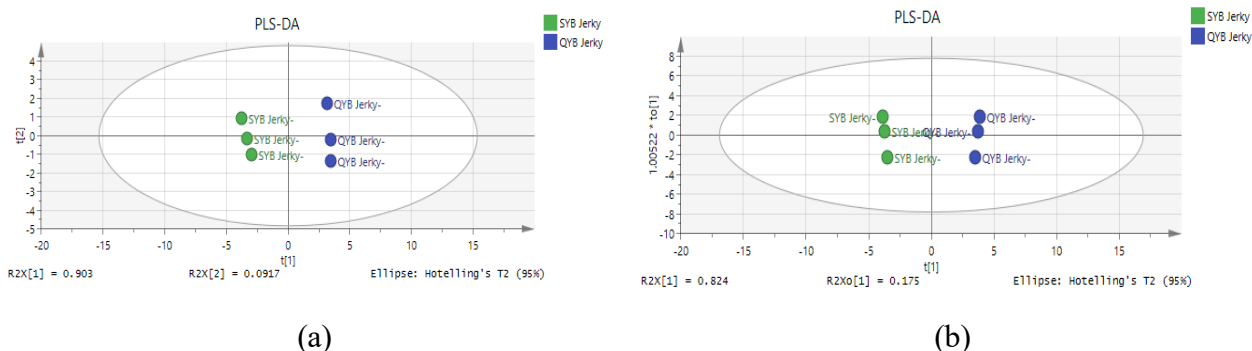
### 3.4.3. Screening of volatile compound differences between Shandong yellow beef jerky and Qinghai yak beef jerky

The data from the two detection techniques were analyzed using PCA dimensionality reduction, and the results are shown in Figure 17. The cumulative contribution rate of the first two principal components in the GC-IMS model reached 100%, while that in the HS-SPME-GC-MS model was 99.6%. The two types of dried meat samples were completely separated in the models, indicating significant differences in the composition and content of volatile compounds between them. The PCA model can effectively distinguish the flavor attributes of dried beef from different varieties.

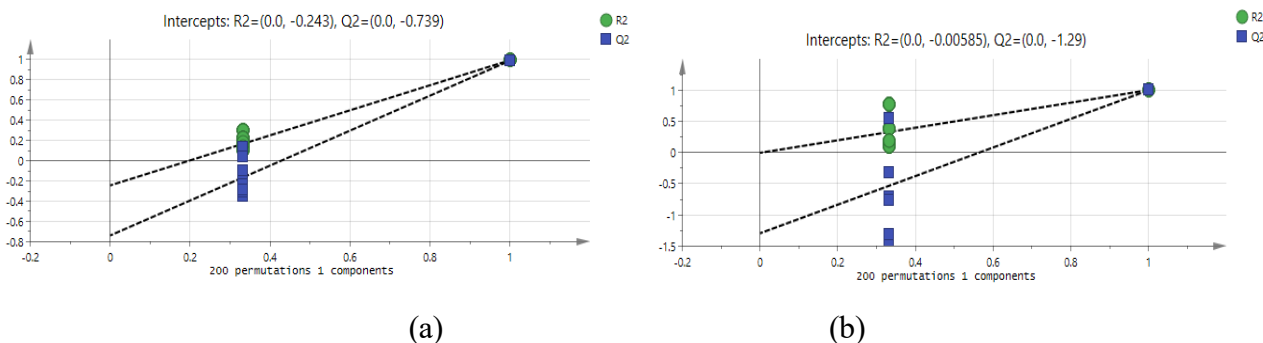


**Figure 17.** PCA analysis of SYB Jerky and QYB Jerky (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

Construct a PLS-DA discriminant model (Figure 18). The results show that for the GC-MS (MS) model,  $R^2X = 0.903$ ,  $R^2Y = 0.999$ ,  $Q^2 = 0.998$ ; for the GC-IMS (IMS) model,  $R^2X = 0.824$ ,  $R^2Y = 0.999$ ,  $Q^2 = 0.997$ . All models have  $R^2$  and  $Q^2$  values close to 1, indicating excellent fit and predictive capability. After 200 permutation tests (Figure 19), the intercepts of  $R^2$  and  $Q^2$  for both models are close to 0, with the  $Q^2$  intercept being negative, confirming that the models are stable, reliable, and free from overfitting. The models effectively distinguish the flavor characteristics of Shandong yellow beef jerky and Qinghai yak beef jerky.



**Figure 18.** PLS-DA analysis of SYB Jerky and QYB Jerky (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

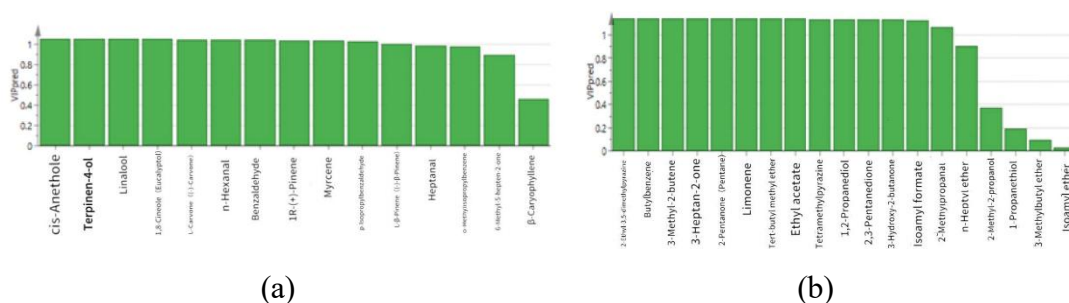


**Figure 19.** Permutation test for VOCs of SYB Jerky (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

Based on the PLS-DA model, the Variable Importance in Projection (VIP) was calculated (as shown in Figure 20).

GC-MS data: The volatile compounds involved are ranked in descending order of VIP values as follows: *cis*-anethole, terpinen-4-ol, linalool, eucalyptol, L-carvone, *n*-hexanal, benzaldehyde, (1*R*)-(+)-myrcene, *p*-isopropylbenzaldehyde, L-beta-pinene, heptanal, *o*-methylisopropylbenzene, methylheptenone, beta-caryophyllene. Among these, aldehydes such as *n*-hexanal and heptanal mainly originate from lipid oxidation, while terpenes like linalool, eucalyptol, and beta-caryophyllene are mostly natural aroma compounds inherent to raw Qinghai yak meat. Compounds such as *cis*-anethole, benzaldehyde, and L-carvone are closely associated with the processing techniques and flavor characteristics of Shandong yellow beef jerky.

GC-IMS data: The selected differential compounds are ranked in descending order of VIP values as follows: 2-ethyl-3,5-dimethylpyrazine, butylbenzene, 3-methyl-2-buten-3-hepten-2-one, 2-pentanone, limonene, tert-butyl methyl ether, ethyl butyrate, tetramethylpyrazine, 1,2-propanediol, 2,3-pentanedione, 3-hydroxy-2-butanone, isoamyl formate, 2-methylpropanal, heptanal, 2-methyl-2-propanol, 1-propanethiol, beta-pinene, 3-methylbutanal. Among these compounds, pyrazines, ketones, and others are largely associated with the Maillard reaction, while thiols, esters, and similar compounds are closely linked to flavor formation during processing.



**Figure 20.** SYB Jerky and QYB Jerky VIP Plot (SYB Jerky: Shandong Yellow Beef Jerky QYB Jerky: Qinghai Yak Beef Jerky)

### 3.5. Identification of Key Aroma Components

Based on the flavor analysis results of four samples—Shandong yellow cattle beef, Qinghai yak beef, Shandong yellow cattle beef jerky, and Qinghai yak beef jerky—the relative odor activity value (ROAV) was used to analyze and evaluate the volatile flavor components of each sample. When the ROAV of a volatile flavor compound is  $\geq 1$ , it is considered a key flavor-contributing compound for the corresponding sample; within a certain range, the higher the ROAV, the greater the compound's contribution to the overall flavor of the sample.

Aldehydes are one of the important sources of the characteristic aroma of beef. These compounds generally have low odor thresholds and contribute significantly to the flavor of meat products. [16] According to Table 3, combined with the relative odor activity value and thresholds, aldehydes are identified as the core flavor-contributing category for both types of raw fresh meat, each with clear benchmark flavor compounds.

In fresh Shandong yellow cattle beef, trans-2-nonenal has an ROAV of 100.00, making it the core benchmark compound. Its aldehyde aroma is concentrated and highly recognizable, supplemented by key flavor compounds with ROAV  $\geq 1$ , such as octanal, 1-octen-3-ol, and hexanal, collectively forming a base flavor dominated by fatty and meaty notes.

In fresh Qinghai yak beef, nonanal has an ROAV of 100.00, ranking first in the raw meat samples and serving as the core benchmark compound. Octanal, hexanal, and heptanal are the main supplementary compounds, providing a pure aldehyde base. P-cymene acts as a flavor modifier, adding a faint herbal aroma.

Terpenes are the core flavor-contributing category for both types of finished beef jerky. Aldehydes, due to volatilization and degradation during processing, are significantly reduced in proportion, with only a small amount remaining to serve as the flavor base.

In Shandong yellow cattle beef jerky, linalool has an ROAV of 26.43, ranking first in the jerky group and serving as the core flavor-contributing compound. Pinene acts as a flavor modifier. The combination of terpenes and residual aldehydes after processing forms a complex flavor profile, making it the most flavor-rich group among the four samples.

In Qinghai yak beef jerky, linalool has an ROAV of 19.81, serving as the core of the jerky group. D-limonene and eucalyptol are key supplementary compounds with ROAV  $\geq 1$ , imparting citrus and cool herbal aromas, respectively. Anethole and pinene act as flavor modifiers. The ROAVs of other flavor compounds are the lowest among the four samples, with fewer types of flavor-contributing substances, resulting in a relatively simple overall flavor composition.

**Table 3.** ROAV of Volatile Flavor Compounds in Four Samples

Category Key Flavor Compounds	Category Key Flavor Compounds	CAS Number	Odor threshold (mg/kg)	ROAV			
				SYB	SYB Jerky	QYB	QYB Jerky
Aldehydes	Hexanal	66-25-1	0.005	9.72	0.51	21.1	21.72
	Heptanal	111-71-7	0.0028	2.3	0.2	10.59	0.13
	Octanal	124-13-0	0.0007	17.38	-	44.26	-
	Nonanal	124-19-6	0.0011	11.66	100	100	100
	trans-2-Octenal	2548-87-0	0.003	-	-	2.74	-
	trans-2-Decenal	3913-81-3	0.0003	-	-	4.92	-
	trans-2-Nonenal	18829-56-6	0.00019	100	-	-	-
Alcohols	Linalool	78-70-6	0.037	-	26.43	4.95	19.81
	1-Hexanol	111-27-3	0.0056	1.3	-	-	-
	1-Octen-3-ol	3391-86-4	0.001	30.28	-	-	-
	Eucalyptol	470-82-6	0.023	-	-	-	2.12
Terpenes	Pinene	7785-70-8	0.041	-	0.2	-	0.72
	D-Limonene	5989-27-5	0.034	-	-	-	9.82
	p-Cymene	535-77-3	0.00586	-	-	0.91	-
Ethers	Anethole	104-46-1	0.05	-	-	-	0.23

Note: The "-" in the table indicates that volatile substances were not detected.

Based on the relative content of volatile flavor compounds, ROAV, and aroma attribute classification, the aroma characteristics of the four samples are analyzed as follows:

SYB is characterized by an aldehyde-dominated aroma profile. Key contributors include trans-2-nonenal, nonanal, and octanal, which impart rich fatty and nutty notes. These are complemented by the mushroom-hay scent of 1-octen-3-ol and the subtle fruity aroma of hexanal, resulting in an overall impression of "prominent aldehydic notes, mellow fatty aroma, and a pure, highly recognizable flavor base."

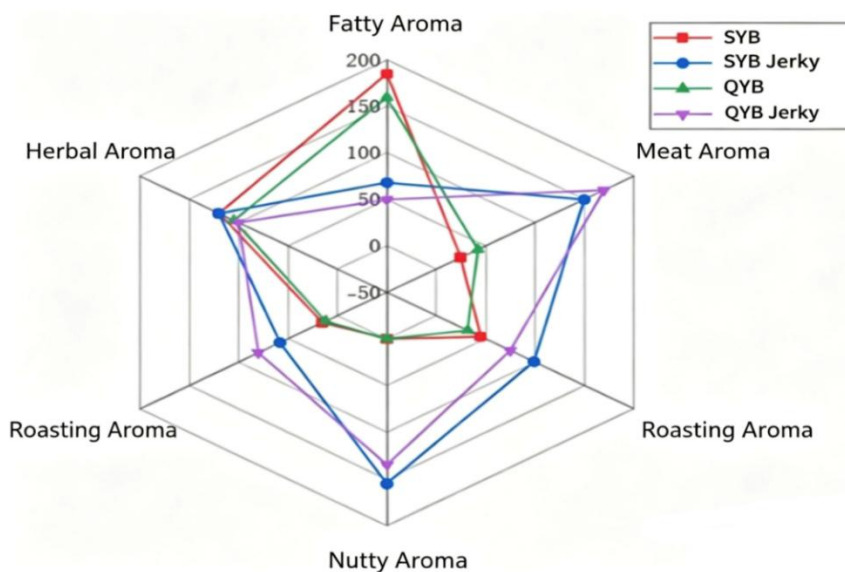
SYB Jerky exhibits a terpene-roasting composite profile. Terpenes (linalool, pinene) contribute fresh herbal-floral notes, while Maillard reaction products (such as tetramethylpyrazine) provide a rich roasted aroma. The fatty base from nonanal is retained, and esters (e.g., 3-methyl-2-buten-1-ol acetate) add fruity layers, creating a complex and multi-dimensional flavor.

QYB also features an aldehyde-dominated aroma profile. Heptanal, nonanal, and octanal contribute a pure fatty and waxy aroma, accompanied by the piney-cool herbal notes of terpenes (such as 1- $\beta$ -pinene and 1,8-cineole) and the flavor base of 1-propanethiol. The overall impression is "pure aldehydic notes, elegant terpene aroma, combined with herbal nuances, refreshing and non-greasy."

QYB Jerky centers on a terpene-dominated aroma. Among the 12 terpenes, linalool and D-limonene contribute intense herbal-citrus fruity notes, enhanced by the roasted-nutty aromas of Maillard reaction products (such as 2-ethyl-1,3,5-trimethylpyrazine and furfural) and fruity accents from esters (e.g., ethyl acetate), while retaining the fatty base of nonanal.

In this study, key terpene aroma compounds in the jerky samples, such as linalool, eucalyptol, and D-limonene, are primarily derived from the compound spices added during marination [17]. These compounds impart fresh herbal and citrus fruity notes, enhance the richness of meat aroma through flavor synergy, and mitigate the decline in meat aroma and accumulation of burnt flavors during processing and storage [18]. Key flavor compounds identified in this study, such as nonanal, hexanal, and 1-octen-3-ol, serve as characteristic markers of lipid oxidation in beef jerky [19]. Moderate oxidation enriches flavor layers, while excessive oxidation can lead to rancid off-notes. Terpene components from spices, together with these oxidation markers, help ensure the flavor stability of beef jerky. This finding confirms that terpene components from spices are key factors regulating the flavor characteristics and stability of beef jerky, and also explains why both types of jerky exhibit terpenes as core flavor compounds.

Drawing on the common attribute lexicon in meat flavor research, this study identified six aroma attributes to describe beef and beef jerky: fatty, herbal, nutty, roasted, fruity, and meaty. By calculating the cumulative contribution of ROAV values for key aroma compounds, radar maps illustrating the aroma characteristics of different samples were generated (see Figure 21). The results indicate that among the aroma attributes of the four samples, the response value for the fatty dimension in raw meat samples (SYB, QYB) is significantly higher than the other five aroma dimensions. In contrast, the herbal dimension exhibits the highest response value in jerky samples (SYB Jerky, QYB Jerky). In terms of meaty and nutty dimensions, the Shandong Yellow Cattle group (SYB, SYB Jerky) shows significantly higher values than the Qinghai Yak group (QYB, QYB Jerky). However, the SYB sample demonstrates higher intensity in fatty, meaty, and nutty notes. SYB Jerky is characterized by more prominent roasted and nutty attributes. QYB is distinguished by its unique herbal aroma and mild fatty notes, while QYB Jerky exhibits relatively stronger herbal and fruity dimensions, resulting in an overall fresher and more refreshing flavor profile.



**Figure 21.** Radar Chart of Aroma Characteristics of Four Samples (SYB: Shandong Yellow Beef SYB Jerky: Shandong Yellow Beef Jerky QYB: Qinghai Yak Beef QYB Jerky: Qinghai Yak Beef Jerky)

## 4. CONCLUSION

This study systematically characterized the flavor profiles of Shandong yellow beef (SYB), Qinghai yak beef (QYB), and their corresponding cured and dried jerky using HS-SPME-GC-MS and GC-IMS combined with PCA, PLS-DA multivariate statistical analysis, and ROAV calculation. The results showed that a total of eight major classes of volatile flavor compounds were detected in the four samples by GC-MS. Aldehydes, terpenes, and Maillard reaction products were identified as the core flavor-contributing substances. In the fresh meat stage, both SYB and QYB exhibited aldehydes as the core flavor components, while the proportion of aldehydes significantly decreased in the processed jerky, with terpenes becoming the dominant flavor compounds. Additionally, roasted aroma characteristic Maillard reaction products, such as pyrazines, were newly formed. Multivariate statistical analysis confirmed that PCA could effectively differentiate the flavor characteristics of different raw materials and their corresponding products. The PLS-DA model demonstrated goodness-of-fit ( $R^2Y$ ) and predictive ability ( $Q^2$ ) both close to 1, validating the significant flavor differences among the samples. Based on ROAV, nonanal, linalool, and 2-ethyl-1,3,5-trimethylpyrazine were identified as key odor markers. Both SYB and QYB raw materials exhibited an aldehyde-dominated aroma, while SYB Jerky displayed a terpene-roasted compound aroma, and QYB Jerky showed a terpene-dominated refreshing aroma. This study clarified the evolution of volatile flavor compounds during the processing of the two types of beef and the characteristic differences in the final products, providing data support for the targeted regulation of jerky flavor, quality evaluation, and rapid screening of raw material quality before processing. Notably, changes in the content of key flavor compounds, such as nonanal and octanal, can sensitively reflect quality fluctuations during raw material processing, offering a new perspective for rapid quality screening of raw materials before jerky production [20].

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compounds of Shandong Yellow beef and Qinghai yak beef, whether it was optimizing the sample curing and drying processes, finalizing the GC-MS and GC-IMS detection protocols, or subsequent data analysis and interpretation of the flavor compounds, whenever I encountered experimental bottlenecks or difficulties in writing, Professor He was always able to pinpoint the issues and clarify my thinking with precision. His rigorous academic attitude and solid expertise have been a tremendous source of learning for me. I also extend my thanks to my laboratory colleagues for their enthusiastic assistance in experimental operations and data processing. Furthermore, I am grateful to all the teachers and peers who have instructed and supported me throughout my academic journey. Finally, I would like to offer my heartfelt thanks to everyone who has provided me with help and encouragement. I also extend my appreciation to myself for the perseverance and dedication maintained along this research path.

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