# Comparative analysis of characteristic volatile compounds in Chinese traditional smoked chicken (specialty poultry products) from different regions by headspace–gas chromatography−ion mobility spectrometry

Wensheng Yao,[*,†] Yingxuan Cai,[*,†] Dengyong Liu,[*,‡,1,2] Zhinan Zhao,[*,‡] Zhenghao Zhang,[†] Shuangyu Ma,[*,‡] Mingcheng Zhang,[*,‡] and Hao Zhang[§]

*National & Local Joint Engineering Research Centre of Storage, Processing and Safety Control Technology for Fresh Agricultural and Aquatic Products, Bohai University, Jinzhou 121013, China; †College of Chemistry and Chemical Engineering, Bohai University, Jinzhou 121013, China; ‡College of Food Science and Technology, Bohai University, Jinzhou 121013, China; and §Shandong Hanon Scientific Instrument Co., Ltd., Jinan, Shandong 250101, China

**ABSTRACT** This article presents investigation of the flavor profile on 5 different regional Chinese smoked chicken samples using gas chromatography−ion mobility spectrometry analysis methods. Five batches of samples were obtained over the course of 6 mo. A total of 34 flavor substances were identified in the 5 smoked chicken samples, including 10 aldehydes, 7 alcohols, 4 ketones, 2 hydrocarbons, 3 heterocyclic compounds, 4 esters, 2 ethers, and 2 phenolic compounds. The whole spectral fingerprint visually displayed flavor differences and relations in 5 types of smoked chicken with local characteristics. Moreover, the orthogonal projections to latent structures discriminant analysis model revealed that these samples could be separately classified into 5 groups. Multivariate statistical analysis showed that 20 chemicals with higher Variable Importance for the Projection values were the key contributors to the differences of flavor in these 5 kinds of smoked chicken. N-nonanal, heptanal, n-nonanal, heptanal, furfurol, and hexanal were the main common flavor compounds in the 5 types of Chinese smoked chicken, whereas linalool, alpha-terpineol, 1,8-cineole, and anethole were the main characteristic flavor compounds of Goubangzi chicken (No. 1); gamma-butyrolactone, 2-acetylfuran, 2-methoxyphenol, 2-acetylpyrrole, and limonene were determined as the key flavor compounds of Liaocheng chicken (No. 2); the concentration of octanal and n-nonanal was higher in Tangqiao chicken (No. 3); butyl acetate was the key contributor to the flavor compounds of Jinshan chicken (No. 4). 2-Heptanone and 2-pentylfuran had a high correlation with Zhuozishan chicken (No. 5). The different raw materials and ingredients used, especially the method of preparation and cultural differences, in different regions of the country in China were the main reasons leading to the similarities and differences of volatile compounds in the 5 kinds of Chinese traditional smoked chicken.

**Key words:** HS–GC–IMS, smoked chicken, poultry product, volatile compound

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## INTRODUCTION

Smoked meat products refer to meat products that are processed from the livestock and poultry meat, mainly through the smoking process. In ancient times, smoking was one of the oldest methods used to preserve meat (Tongo et al., 2017). In modern times, smoking is also no longer the main storage method (Damaziak et al., 2016). The purpose of modern smoke processing has gradually turned to adding a unique fragrance to food. As harmful pollutants of smoking, the healthy smoked materials are encouraged to decrease the concentration of polycyclic aromatic hydrocarbons (Rozentāle et al., 2015). The smoking process not only gives the products a good texture but also gives them an attractive color and a unique flavor. The smoky flavor is mainly produced by heating and decomposing the smoked material



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[1]Present address: College of Food Science and Technology, Bohai University, No. 19, Keji Rd., New Songshan District, Jinzhou 121013, Liaoning province, China.
[2]Corresponding author: jz_dyliu@126.com

to produce volatile flavor substances (Jerković et al., 2007). Therefore, the research studies related to flavor compounds of smoked meat attracted the attention of people.

Smoked chicken is one of the typical representatives of traditional Chinese chicken meat products. In the process of more than 5,000 yr, the Chinese nation has fostered a diverse kind of smoked chicken owing to the vast territory (Song and Cho, 2017), especially widely distributed in the north and southeast coastal areas of China. There are significant regional flavor differences of these products in the different regions in China based on the different geographical environments and eating habits. However, there are still some similarities among them on account of the same Chinese nation they belong to.

Flavor is an important indicator for evaluating the quality of smoked meat products (Radovcic et al., 2016). Researchers have paid more attention to exploring different analytical methods to predict the quality of food by testing the flavor compound in food products. One of the main standard techniques for analyzing the flavor compound is gas chromatography (**GC**)–mass spectrometry (MS) (Creaser et al., 2004; Cumeras et al., 2015; Sun et al., 2020; Wang et al., 2020), and it has been remaining as the dominant technique for measurement of flavor compounds (Chen et al., 2006; Mayr et al., 2015; Hopfer et al., 2016; Qi et al., 2018; Wang et al., 2019).

In recent years, there has been a rapid rise in the application of headspace–gas chromatography−ion mobility spectrometry (**HS–GC–IMS**) as an inexpensive and powerful analytical technique for detection of flavor compounds at ambient pressures and temperatures (Cumeras et al., 2015). Ion mobility spectrometry is an analytical technique used to separate and identify ionized molecules at ambient pressures and temperatures based on their mobilities under an electric field. The technique was initially used for military and security purposes, such as detecting drugs and explosives (Creaser et al., 2004). Headspace–gas chromatography−ion mobility spectrometry, due to need of no complex pretreatment and its high sensitivity, has been widely used in the detection of flavor fingerprints of food (Hernández-Mesa et al., 2017). Egg products' freshness was also identified through HS–GC–IMS (Cavanna et al., 2019). Volatile biogenic amines in muscle food products were monitored by HS–GC–IMS (Karpas et al., 2002; Liu et al., 2017). Although some research studies have been carried out on determination of flavor compounds in smoked meat products, however, few studies have examined the differences of smoked chicken from different regions, especially for famous Chinese brands.

Considering the aforementioned reason, this article selected 5 types of smoked chicken from Liaoning province, Shandong province, Zhejiang province, Chifeng city, and Zhuozi county in Inner Mongolia. The five famous brands come from 5 regions located in the north of China and southeast coastal areas, which enjoy great popularity across China and abroad owing to their extraordinary good taste. The primary aim of this article is to obtain the whole fingerprint spectrum of flavor differences and relations in the 5 types of smoked chicken. Comparative analysis of smoked chicken in different regions in China can fully understand the main characteristics of smoked chicken and the quality differences of smoked chicken in different regions, which could provide reference for consumers to purchase products. In addition, the study was particularly focused on principal component analysis (**PCA**), partial least squares discriminant analysis (**PLS-DA**), and orthogonal partial least squares discriminant analysis (**OPLS-DA**) technique; a procedure was suggested to further evaluate the flavor of smoked chicken and provide theoretical basis for improving the flavor and quality of smoked chicken.

## MATERIALS AND METHODS

### Experimental Samples

As previously mentioned, 5 kinds of smoked chicken with local characteristics were selected from 5 regions. Goubangzi chicken (No. 1), Liaocheng chicken (No. 2), Jinshan chicken (No. 3), Zhuozishan chicken (No. 4), and Tangqiao chicken (No. 5) were collected on the manufacturing date. They were vacuum packed and immediately delivered to the laboratory via low-temperature cold chain transports. All samples were transferred to a refrigerator where it was stored at 4°C or less and tested within 2 d. Five batches of samples were obtained over the course of 6 mo. One of each type was collected in each batch. Five samples of each type of chicken were used. Four parallel tests were performed on each sample.

For each brand of smoked chicken, the chicken thighs were selected for testing as samples. The chicken thigh skin was carefully peeled away, and the meat and the skin were separately cut into small pieces (length = 2.0 mm, width = 2.0 mm, thickness = 2.0 mm approximately). The thighs and skin were mixed together in the proportion of 4:1. A 2.0-g sample (1.6 g of chicken and 0.4 g of chicken skin) was directly transferred into a 20-mL headspace vial that was subsequently incubated at 65°C for 20 min. Then, the samples were directly injected for analysis. For the evaluation of method reproducibility, the samples were prepared and injected in triplicate.

### Reagents

All the reagents used in this study were of analytical grade. Analytical standards of nonanal, octanal, caproaldehyde, heptaldehyde, 1-octen-3-ol, 2-pentylfuran, and (+)-limonene were purchased from Shanghai Aladdin Bio-Chem Technology Co. Ldt, Shanghai, China. 2-Butanone, 2-pentanone, 2-hexanone, 2-heptanone, 2-octanone, and 2-nonone were purchased from Sinopharm Chemical Reagent Co., Ltd., Beijing, China.

### Instrumentation and Data Treatment

Analyses of the smoked chicken samples were performed using an IMS commercial instrument (FlavourSpec) from Gesellschaft für Analytische Sensorsysteme mbH (Dortmund, Germany), equipped with a low-radiation tritium $^{3}H$ source of 5.68 keV. The chromatographic separation was performed on a FS-SE-54-CB-1 (5% diphenyl, 95% dimethylpolysiloxane) capillary column (15 m × 0.53 mm, 1-μm film thickness) kept at 60°C. Two grams of the sample was incubated at 65°C for 20 min. Then, 500 μL of headspace was automatically injected by means of a heated syringe (85°C) into the heated injector (85°C). Nitrogen gas was used as a carrier gas with the flow ramp starting at 2 mL/min for 2 min, then increased to 15 mL/min in 8 min, increased to 100 mL/min in 10 min, and finally increased to 150 mL/min in 10 min. The total GC runtime was 30 min. Moreover, nitrogen gas was also as the drift gas at a rate of 150 mL/min.

Gas chromatography–ion mobility spectrometry data were obtained in positive mode using LAV software, and unknown chemical compounds of the samples were identified by using GC–IMS library search software supplied by Gesellschaft für analytische Sensorsysteme mbH. Some flavor compounds were further confirmed by using standard chemicals. Moreover, unsupervised PCA, supervised PLS-DA, and OPLS-DA were performed using SIMCA-P software version 14.1 (Umetrics, Umeå, Sweden; (https://umetrics.com/products/simca)).

## RESULTS AND DISCUSSION

### Optimization of HS–GC–IMS Parameters

Some experimental variables of the GC–IMS system were initially optimized in accordance with the aim of this work. When the temperature rises, the volatile organic compounds with high boiling points usually are easily released. The incubation temperature of 65°C is mainly based on the ideal serving temperature of smoked chicken (65°C). To examine the influence of sample incubation time, the groups of incubation time were selected: 10 min, 20 min, and 30 min. The results showed that no matter the sampling amount (2 g, 3 g, or 5 g), the signal intensities of volatile compounds in 20-min incubation samples increased comparing with those in 10-min samples, but did not change much between 20 min of hatching and 30 min of hatching. Therefore, an optimum value of 20 min was chosen (Supplementary Figure 1). Then, under the 20-min optimum value, the effect of the amount of the sample was studied: 2 g, 3 g, and 5 g for optimization of conditions (Supplementary Figure 2). An amount of 2 g was selected as optimum because no significant differences were obtained when a higher amount, i.e., 3 g, was used, and the content of volatile compounds decreased with the 5 g sampling amount. After condition optimization, the incubation time was chosen to be 20 min, and the sample volume was 2 g.

### Headspace–Gas Chromatography–Ion Mobility Spectrometry Topographic Plots of Five Kinds of Chinese Traditional Smoked Chicken

The HS–GC–IMS method was applied to obtain global IMS information from the samples of the smoked chicken, with the aim of helping to identify the differences of flavor substances in different chickens and with the aim of helping to identify the flavor substances and change rules in the process. A GC–IMS analysis resulted in a topographic plot, as shown in Figure 1A. The vertical coordinate represents the retention time of gas chromatography, and the horizontal coordinate represents the ion migration time. The background of the whole figure is blue, and the red vertical line at abscissa 1.0 is the reactant ion peak (**RIP**, after normalization). Each point on both sides of the RIP peak represents a flavor substance. The color represents the concentration of the substance; white means lower concentration, red means higher concentration, and the darker the color, the higher the concentration. The three-dimensional spectra of volatile organic compounds in chicken samples of five different 5 smoked chickens are also shown in Figure 1B. The differences between the volatile organic compounds in different types of smoked chicken samples can be obtained clearly. It is because of the differences of dietary culture in different nationalities and regions and because the formula and technology of smoked chicken are different, which results in certain differences in flavor compounds of smoked chicken.

### Qualitative Analysis of HS–GC–IMS

Qualitative analysis results of GC–IMS with the selected markers are presented in Figure 2, and their corresponding GC–IMS global area set integration parameters and qualitative analysis results can be seen in Table 1. The markers in Figure 2 were labeled with the corresponding numbers, which are in accordance with volatile compounds in Table 1. And the corresponding dimers formed in the IMS drift tube are represented by the symbol *. There are a large number of published studies that described the principles, instrumentation, and applications of IMS (Creaser et al., 2004; Lanucara et al., 2014; Cumeras et al., 2015). The molecules of the samples that are ionized in the ionization region depend on using a β-source from a small foil of radioactive nickel-63. Nitrogen and oxygen may be ionized depending on the ionization technique and result in the formation of a series of reactions of positive/negative ions of $N_2^+$ and $O^{2-}$. The reactant ions $[(H_2O)\ n\ (H_3O)^+]$ can be generated, while the ions of $N_2^+$ and $O^{2-}$ react with water vapor molecules, and the corresponding peak in IMS chromatograms is called the RIP. The analyte molecule M would be ionized into positive $M^+$ and/or negative $M^-$. Furthermore, a series of different reactions take place between $M^+$ (and/or $M^-$) and the reactant ions, if the gas-phase proton affinity of the sample molecule is larger than that of water (691 kJ $mol^{-1}$); moreover, further hydration reaction

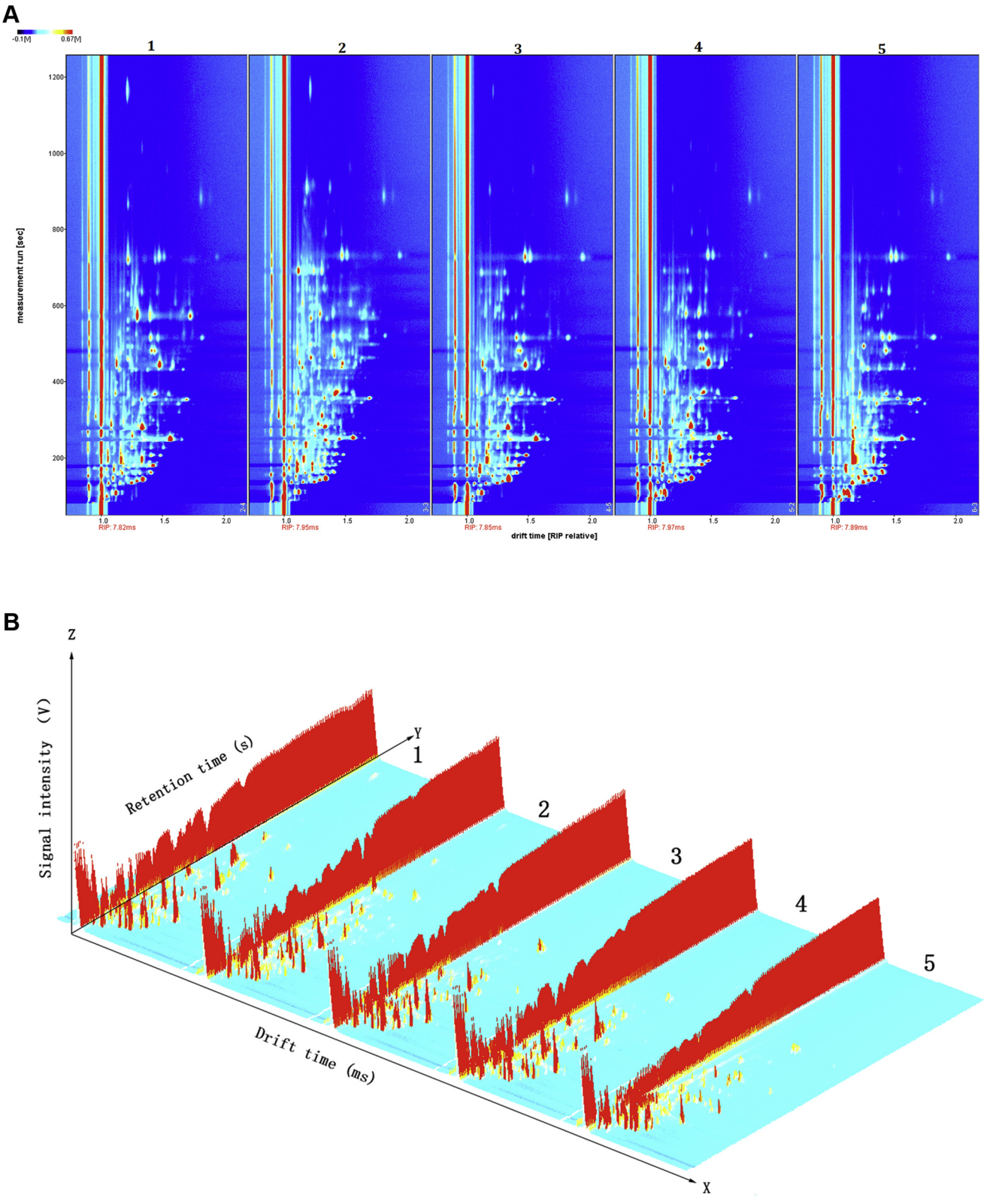


**Figure 1.** Volatile compounds in 5 kinds of Chinese traditional smoked chicken. 1: Goubangzi chicken; 2: Liaocheng chicken; 3: Jinshan chicken; 4: Zhuozishan chicken; 5: Tangqiao chicken. Abbreviation: RIP, reactant ion peak. (A) Topographic plot of GC–IMS spectra; (B) 3D-topographic in different samples.

will lead to the formation of $MH^+$, $(M\text{-}H)^-$, $MO^{2-}$, proton-bound dimers $M_2H^+$, or even trimmers $M_3H^+$ at high concentrations of the analyte. Therefore, the remarkable proton monomer and the proton-bound dimer of these flavor compounds could be seen in Table 1.

A total of 34 flavor substances were detected from the 5 smoked chicken samples, including 10 aldehydes, 7 alcohols, 4 ketones, 2 hydrocarbons, 3 heterocyclic compounds, 4 esters, 2 ethers, and 2 phenolic compounds. In terms of volatile compound development, some major common

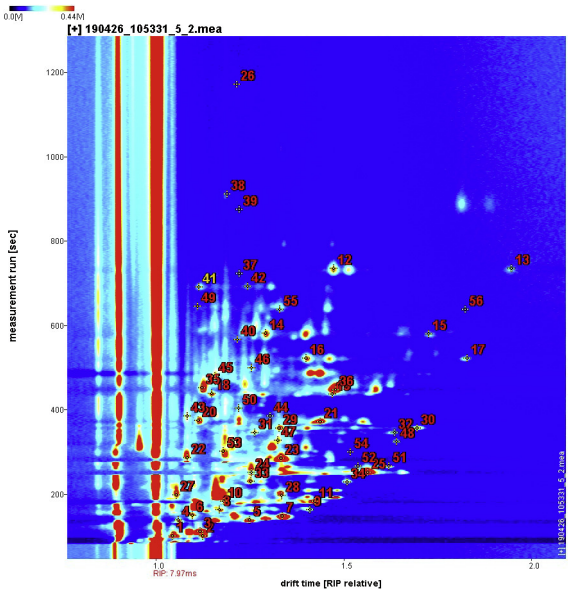


**Figure 2.** Topographic plots of GC–IMS spectra with the selected markers obtained for Tangqiao chicken. Abbreviations: GC–IMS, gas chromatography−ion mobility spectrometry; RIP, reactant ion peak.

reactions may be considered: lipid degradation (oxidative reactions), Maillard reactions, Strecker degradation, and thiamine degradation. These factors are interrelated and interconnected. Usually, alcohols, ketones, and aldehydes are considered minor contributors owing to their high odor threshold values, while furans, pyrroles, and sulfur compounds were key aroma contributors (Flores, 2018).

The analytical device proved attractive owing to its lower detection limits, extraordinary sensitivity, and ease of use. These advantages have dramatically improved the application ranges of the IMS instrument: medical diagnostics (Kyle et al., 2016), environmental analysis (Márquez-Sillero et al., 2011), and so on.

## *The Whole Spectrum Analysis by HS–GC–IMS*

Smoked chickens are widely manufactured in China; however, in terms of manufacturing process, many major different factors may be considered owing to the different national and regional differences in eating culture. The differences in raw material, ingredients, additives, and the manufacture process caused the differences in flavor, especially the differences in smoking materials (Morey et al., 2012; Huang et al., 2019). Therefore, a variety of products with different tastes and flavors are manufactured, such as Tangqiao chicken (No. 5) with sweet taste in the south and Gubangzi chicken (No. 1) and Liaocheng chicken (No. 2) with salty taste in the north. They are all famous for its unique color, aroma, taste, and shape. Their consumer groups are relatively stable and have a broad development prospect.

A global overview of the spots identified in the 5 traditional smoked chicken samples is reported in Figure 3. The volatile compounds in 5 types of Chinese traditional chicken were significantly different. Each kind of smoked chicken contained its own characteristic flavor substance; the quality of these characteristic flavor compounds was much higher than that of others. In smoking, the burning materials play the most vital role. The five types of Chinese chicken used the following ingredients respectively: sugar and fruit tree wood shavings (No. 1), fruit tree wood shavings (No. 2), brown sugar and cypress wood shavings (No. 3), sugar and cypress shavings (No. 4), and tea leaves, brown sugar, and rice (No. 5). A lot of smoky substances in the products appeared using these materials. In the area labeled with a red rectangle, there were some specific flavor substances in the smoked chicken of No. 1 sample, including anethole, α-terpineol, 1,8-cineole, and linalool. These substances were all derived from the spice. Sugar besides wood was used in local areas in the north of China, and Goubangzi chicken (No. 1) was the most typical product. It is a famous traditional chicken in Liaoning province, produced in Beizhen city, Liaoning province, which was founded in 1889. Its essence lies in the use of traditional white sugar as smoking materials, which gives the smoked chicken golden color and the characteristic of aroma. The green rectangle highlighted the most specific flavors in Liaocheng chicken (No. 2), including α-pinene, limonene, γ-butyrolactone, 2-methoxyphenol, 4-methylguaiacol, and 2-acetylpyrrole. α-Pinene and limonene were usually derived from plant-based flavors; it was also important to remark that in many cases, 2-methoxyphenol and 4-methylguaiacol were considered contributors that give the meat a nice smoky flavor. Liaocheng chicken (No. 2) is produced in Liaocheng, Shandong province. It has a history of nearly 200 yr. In Liaocheng products processing, fruit tree wood were commonly used for smoking.

As shown in the purple and yellow rectangle (Figure 3), No. 3 and No. 4 smoked chicken also contained a small amount of specific flavor substances, such as butyl acetate in No. 4; the substance 116# and 86# in No. 3 may be very important contributors to No. 3 smoked chicken, although they had not been identified in this work. Neimeng Jinshan smoked chicken (No. 3) is produced in Jinshan town, Chifeng city, and the Inner Mongolia Autonomous Region. It is a century-old brand. It has been famous for its legends and has been well received by consumers. In the smoking process of Jinshan chicken, first smoked with cypress for a few minutes, then change to brown sugar for smoking.

Zhuozishan chicken (No. 4) is a traditional and famous cuisine in Zhuozi county, Inner Mongolia, which uses a mixture of sugar and cypress shavings as smoking materials. The material in the blue box was the specific flavor substance in No. 5 smoked chicken, including 2-pentylfuran. Tangqiao smoked chicken (No. 5) is produced in Tangqiao town, Zhejiang province. Brown sugar, tea leaves, and rice gave off caramel fragrance and adhere to the surface of cured chicken.

**Table 1.** Gas chromatography−ion mobility spectrometry global area set integration parameters obtained from the Chinese traditional smoked chicken.

| Count | Compound | CAS# | Formula | MW | RI | Retention time (s) | Drift time (ms) | Comment | Identification approach |
|---|---|---|---|---|---|---|---|---|---|
| 1 | Ethanol | C64175 | $C_2H_6O$ | 46.1 | 474.9 | 101.04 | 1.0461 | Monomer | RI, DT, STD |
| 1* | Ethanol | C64175 | $C_2H_6O$ | 46.1 | 472.6 | 100.454 | 1.1257 | Dimer | RI, DT |
| 2 | Acetone | C67641 | $C_3H_6O$ | 58.1 | 509.8 | 110.704 | 1.12 | | RI, DT |
| 3 | Ethyl methyl ketone | C78933 | $C_4H_8O$ | 72.1 | 596.0 | 138.673 | 1.0607 | Monomer | RI, DT |
| 3* | Ethyl methyl ketone | C78933 | $C_4H_8O$ | 72.1 | 597.2 | 139.112 | 1.2511 | Dimer | RI, DT |
| 4 | Ethyl acetate | C141786 | $C_4H_8O_2$ | 88.1 | 624.0 | 149.215 | 1.0981 | Monomer | RI, DT |
| 4* | Ethyl acetate | C141786 | $C_4H_8O_2$ | 88.1 | 620.3 | 147.751 | 1.3385 | Dimer | RI, DT |
| 5 | 3-Methylbutanal | C590863 | $C_5H_{10}O$ | 86.1 | 659.9 | 163.892 | 1.1705 | Monomer | RI, DT |
| 5* | 3-Methylbutanal | C590863 | $C_5H_{10}O$ | 86.1 | 658.9 | 163.459 | 1.4112 | Dimer | RI, DT |
| 6 | Pentanal | C110623 | $C_5H_{10}O$ | 86.1 | 699.4 | 182.752 | 1.1819 | Monomer | RI, DT |
| 6* | Pentanal | C110623 | $C_5H_{10}O$ | 86.1 | 700.1 | 183.185 | 1.4254 | Dimer | RI, DT |
| 7 | n-Nonanal | C124196 | $C_9H_{18}O$ | 142.2 | 1105.2 | 735.041 | 1.4743 | Monomer | RI, DT STD |
| 7* | n-Nonanal | C124196 | $C_9H_{18}O$ | 142.2 | 1104.9 | 734.462 | 1.9479 | Dimer | RI, DT |
| 8 | 1,8-Cineole | C470826 | $C_{10}H_{18}O$ | 154.3 | 1035.9 | 581.368 | 1.2929 | Monomer | RI, DT |
| 8* | 1,8-Cineole | C470826 | $C_{10}H_{18}O$ | 154.3 | 1035.1 | 579.729 | 1.7269 | Dimer | RI, DT |
| 9 | Octanal | C124130 | $C_8H_{16}O$ | 128.2 | 1005.4 | 522.038 | 1.4014 | Monomer | RI, DT, STD |
| 9* | Octanal | C124130 | $C_8H_{16}O$ | 128.2 | 1005.2 | 521.71 | 1.8311 | Dimer | RI, DT |
| 10 | Benzaldehyde | C100527 | $C_7H_6O$ | 106.1 | 956.4 | 438.439 | 1.1495 | Monomer | RI, DT |
| 10* | Benzaldehyde | C100527 | $C_7H_6O$ | 106.1 | 955.8 | 437.555 | 1.472 | Dimer | RI, DT |
| 11 | 2-Acetylfuran | C1192627 | $C_6H_6O_2$ | 110.1 | 911.7 | 373.692 | 1.1147 | Monomer | RI, DT |
| 11* | 2-Acetylfuran | C1192627 | $C_6H_6O_2$ | 110.1 | 910.4 | 372.0 | 1.4391 | Dimer | RI, DT |
| 12 | Furfurol | C98011 | $C_5H_4O_2$ | 96.1 | 834.7 | 286.263 | 1.085 | Monomer | RI, DT |
| 12* | Furfurol | C98011 | $C_5H_4O_2$ | 96.1 | 833.3 | 284.962 | 1.3353 | Dimer | RI, DT, |
| 13 | Hexanal | C66251 | $C_6H_{12}O$ | 100.2 | 796.5 | 251.217 | 1.2551 | Monomer | RI, DT, STD |
| 13* | Hexanal | C66251 | $C_6H_{12}O$ | 100.2 | 797.2 | 251.828 | 1.5649 | Dimer | RI, DT, STD |
| 14 | Anethole | C104461 | $C_{10}H_{12}O$ | 148.2 | 1279.8 | 1171.513 | 1.2163 | | RI, DT |
| 15 | Acetoin | C513860 | $C_4H_8O_2$ | 88.1 | 723.3 | 197.604 | 1.0561 | Monomer | RI, DT |
| 15* | Acetoin | C513860 | $C_4H_8O_2$ | 88.1 | 721.9 | 196.677 | 1.3366 | Dimer | RI, DT |
| 16 | Heptanal | C111717 | $C_7H_{14}O$ | 114.2 | 898.2 | 356.115 | 1.329 | Monomer | RI, DT, STD |
| 16* | Heptanal | C111717 | $C_7H_{14}O$ | 114.2 | 898.5 | 356.496 | 1.6976 | Dimer | RI, DT |
| 17 | 2-Heptanone | C110430 | $C_7H_{14}O$ | 114.2 | 890.0 | 345.88 | 1.2653 | Monomer | RI, DT |
| 17* | 2-Heptanone | C110430 | $C_7H_{14}O$ | 114.2 | 889.3 | 345.083 | 1.6375 | Dimer | RI, DT |
| 18 | 1-Pentanol | C71410 | $C_5H_{12}O$ | 88.1 | 770.1 | 230.269 | 1.2539 | Monomer | RI, DT |
| 18* | 1-Pentanol | C71410 | $C_5H_{12}O$ | 88.1 | 768.1 | 228.753 | 1.5114 | Dimer | RI, DT |
| 19 | 5-Methylfurfural | C620020 | $C_6H_6O_2$ | 110.1 | 964.6 | 451.397 | 1.125 | Monomer | RI, DT |
| 19* | 5-Methylfurfural | C620020 | $C_6H_6O_2$ | 110.1 | 962.4 | 447.882 | 1.48 | Dimer | RI, DT |
| 20 | Linalool | C78706 | $C_{10}H_{18}O$ | 154.3 | 1099.1 | 723.104 | 1.2229 | | RI, DT |
| 21 | 4-Methylguaiacol | C93516 | $C_8H_{10}O_2$ | 138.2 | 1185.7 | 911.203 | 1.1903 | | RI, DT |
| 22 | Alpha-Terpineol | C98555 | $C_{10}H_{18}O$ | 154.3 | 1170.4 | 874.835 | 1.223 | | RI, DT |
| 23 | Limonene | C138863 | $C_{10}H_{16}$ | 136.2 | 1028.7 | 566.769 | 1.2179 | | RI, DT, STD |
| 24 | 2-Methoxyphenol | C90051 | $C_7H_8O_2$ | 124.1 | 1084.9 | 691.129 | 1.1162 | Monomer | RI, DT |
| 24* | 2-Methoxyphenol | C90051 | $C_7H_8O_2$ | 124.1 | 1085.3 | 691.989 | 1.2456 | Dimer | RI, DT |
| 25 | Gamma-butyrolactone | C96480 | $C_4H_6O_2$ | 86.1 | 919.9 | 384.855 | 1.0845 | Monomer | RI, DT |
| 25* | Gamma-butyrolactone | C96480 | $C_4H_6O_2$ | 86.1 | 920.1 | 385.184 | 1.3056 | Dimer | RI, DT |
| 26 | 1-Octen-3-ol | C3391864 | $C_8H_{16}O$ | 128.2 | 981.6 | 479.817 | 1.1581 | | RI, DT, STD |
| 27 | 2-Pentylfuran | C3777693 | $C_9H_{14}O$ | 138.2 | 992.6 | 499.011 | 1.2557 | | RI, DT, STD |
| 28 | n-Hexanol | C111273 | $C_6H_{14}O$ | 102.2 | 873.5 | 326.894 | 1.3255 | Monomer | RI, DT |
| 28* | n-Hexanol | C111273 | $C_6H_{14}O$ | 102.2 | 871.3 | 324.449 | 1.642 | Dimer | RI, DT |
| 29 | 2-Acetylpyrrole | C1072839 | $C_6H_7NO$ | 109.1 | 1065.7 | 645.849 | 1.1122 | | RI, DT |
| 30 | Alpha-pinene | C80568 | $C_{10}H_{16}$ | 136.2 | 933.3 | 403.717 | 1.2215 | | RI, DT |
| 31 | Butyl acetate | C123864 | $C_6H_{12}O_2$ | 116.2 | 813.3 | 266.037 | 1.6224 | | RI, DT |
| 32 | Ethyl 2-hydroxypropanoate | C97643 | $C_5H_{10}O_3$ | 118.1 | 814.5 | 267.149 | 1.539 | | RI, DT |
| 33 | 2-Hexen-1-ol | C2305217 | $C_6H_{12}O$ | 100.2 | 850.5 | 302.2 | 1.1811 | Monomer | RI, DT |
| 33* | 2-Hexen-1-ol | C2305217 | $C_6H_{12}O$ | 100.2 | 848.3 | 299.947 | 1.5195 | Dimer | RI, DT |
| 34 | (E)-2-octenal | C2548870 | $C_8H_{14}O$ | 126.2 | 1062.3 | 638.145 | 1.3312 | Monomer | RI, DT |
| 34* | (E)-2-octenal | C2548870 | $C_8H_{14}O$ | 126.2 | 1061.9 | 637.167 | 1.8245 | Dimer | RI, DT |

Abbreviations: RI, retention indices calculated in the experiment; DT, confirmed by a drift time (ms); STD, further confirmed by analytical standard chemicals; MW, relative molecular weight.

*Dimer of the flavor compounds.

## *Differences of Flavor Compounds Based on Multivariate Data Analysis*

With the aim of further investigating the flavor differences and relations in the 5 types of smoked chicken, PCA, PLS-DA, and OPLS-DA techniques were applied to the data. Principal component analysis is an unsupervised technique for deriving a set of low-dimensional features from a largest set of variables. It is popularly used in cluster analysis and to visualize higher dimensional data, while still preserving as much variance as possible (Aleixandre-Tudo et al., 2015). Partial least squares is a latent variable regression method based on covariance between the predictors and the response. The OPLS algorithm was introduced by Trygg and Wold (2002) to model separately the variations of the predictors correlated and orthogonal to the response.

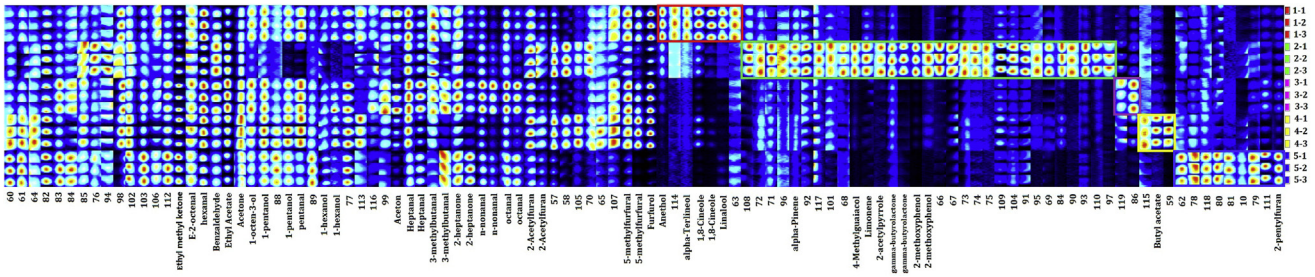


**Figure 3.** Global overview of the spots identified in Chinese traditional smoked chicken. A zone of each topographic plot, which contains most of the important data, is labeled with a colorful rectangle, respectively. 1. Goubangzi chicken (the red rectangle); 2: Liaocheng chicken (the green rectangle); 3: Jinshan chicken (the purple rectangle); 4: Zhuozishan chicken (the yellow rectangle); 5: Tangqiao chicken (the blue rectangle).

Data were divided in the training set (80% of samples) and validation set (20% of samples). First, PCA was applied over the training set to visualize any possible grouping of samples. A distinction between No. 2 and No. 5 was observed (Figure 4A); No. 1, No. 3, and No. 4 also belonged to one cluster, respectively (Figure 4a); however, to a certain extent, all categories were dispersed. Figure 4B/b showed that the 5 kinds of smoked chicken can be clearly distinguished based on the PLS-DA model compared with the results from the PCA. However, compared with both results obtained with PCA and PLS-DA, OPLS-DA exhibited the best separation effect of 5 kinds of smoked chicken (Figure 4C/c). The model resulted in cross-validated predictive ability Q2 (Y) = 94.5%, total explained variance R2 (X) = 89.0%, and R2 (Y) = 95.2%. No. 2 smoked chicken samples were mostly located toward the positive side of PC2, and No. 5 samples were located toward the negative side of PC1. Moreover, a differentiation in No. 1, No. 3, and No. 4 samples was observed in Figure 4C.

Each type of smoked chicken samples obviously had an aggregate trend, whereas 5different types of smoked chicken samples were well separated from each other (Figure 4C/c). Figure 5 was a Variable Importance for the Projection (**VIP**) diagram of the OPLS-DA model. Twenty chemical volatile substances with the VIP value of the OPLS-DA model higher than 1.0 were identified, as shown in Figure 5. They could be considered important in the given model, which was in combination with the results of Figure 2. N-nonanal, heptanal, n-nonanal, heptanal, furfurol, and hexanal were the main flavor compounds for Chinese smoked chicken in common. The contents of 4 compounds including linalool, alpha-terpineol, 1,8-cineole, and anethole was the highest in No. 1 smoked chicken. Among these marker compounds, gamma-butyrolactone, 2-acetylfuran, 2-methoxyphenol, 2-acetylpyrrole, and limonene were the abundant compounds in No. 2 smoked chicken. The content of octanal and n-nonanal was higher in No. 3 smoked chicken. Butyl acetate was the key contributor to the flavor compounds of No. 4 smoked chicken. 2-Heptanone and 2-pentylfuran had a high correlation with No. 5 smoked chicken. Multivariate data analysis revealed that the 5 types of regional smoked chicken can be clearly distinguished based on the OPLS-DA model.

## CONCLUSION

In this article, the potential of GC–IMS to examine the flavor differences and relations in the 5 types of smoked chicken has been demonstrated. The GC–IMS technique made it possible to visually display the results by the whole spectral fingerprint in a color contour image. A total of 106 volatile compounds were detected including the dimers of some chemicals. Among them, 34 flavor substances were identified, mainly including aldehydes, alcohols, ketones, hydrocarbons, heterocyclic compounds, esters, ethers, and phenolic compounds. Twenty chemicals with higher VIP values were the key contributors to the differences of flavor in these 5 types of smoked chicken. N-nonanal, heptanal, n-nonanal, heptanal, furfurol, and hexanal were the main common flavor compounds in the 5 types of Chinese smoked chicken. Besides, each smoked chicken had its own characteristic flavor substances. The flavor compounds of Chinese regional smoked chicken were distinctive from one another, which mainly result from different raw materials, ingredients, preparation methods, and cultural differences in different regions of the country in China. In other respects, they were also connected with each other to some extent owing to being Chinese national cuisine from the same country. To the best of our knowledge, this is the first time that GC–IMS has been used to evaluate the characteristic flavor compounds in Chinese regional smoked chicken. The results showed that GC–IMS was a reliable and relative low-cost method. Gas chromatography–ion mobility spectrometry technique is therefore a very useful tool to ensure the application in food laboratories. This study provided scientific data and laid the foundations to study the formation pathways of smoked chicken flavor substances and their interaction mechanisms. The analysis of the flavor profile was also helpful for product identification and new product development.

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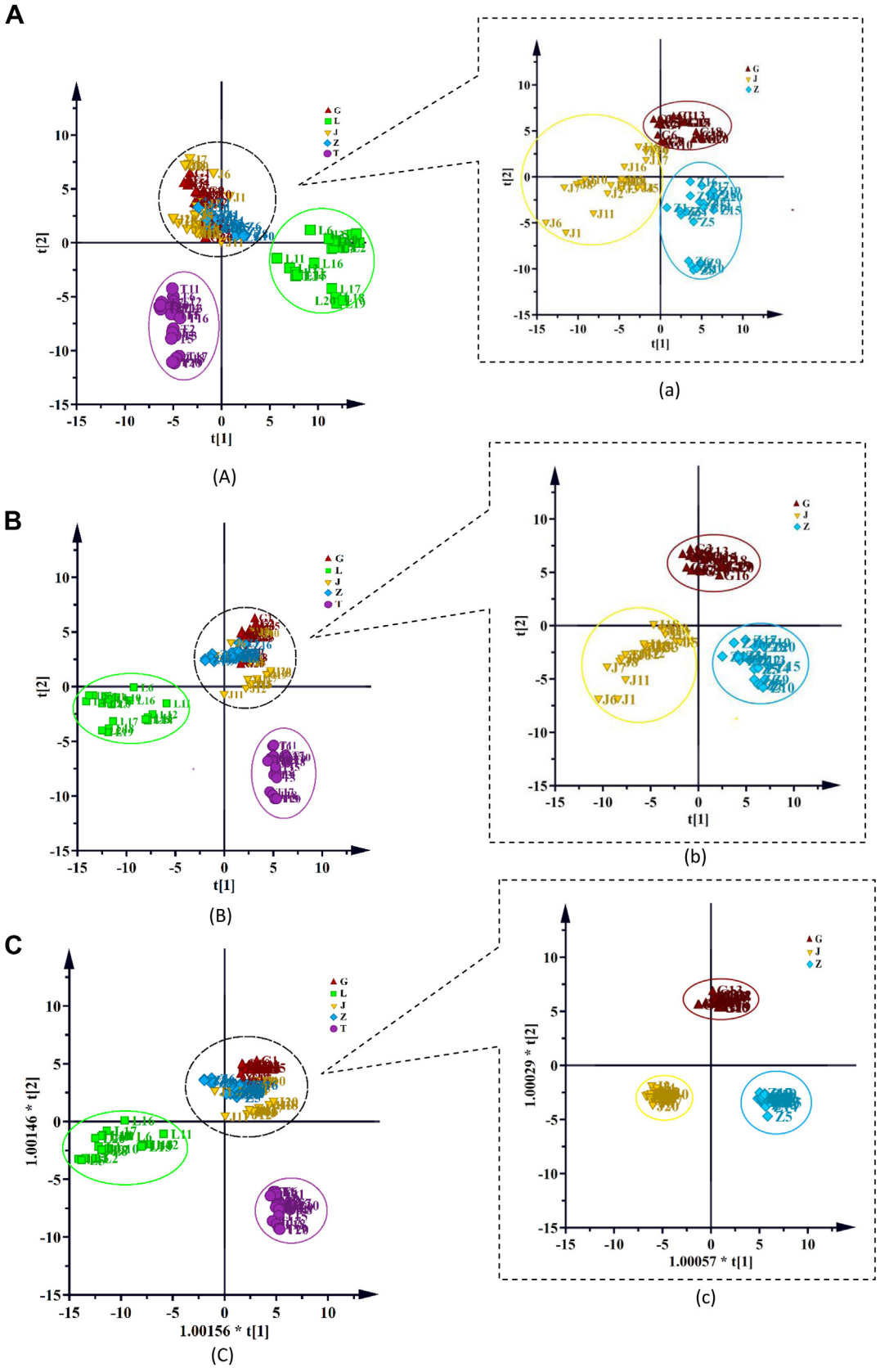


**Figure 4.** (A, B, C) PCA, PLS-DA, and OPLS-DA plot for the 5 types of smoked chicken. (a, b, c) PCA, PLS-DA, and OPLS-DA plot for 3 types chicken. 1: Goubangzi chicken (▲G); 2: Liaocheng chicken (■L); 3: Jinshan chicken (▼J); 4: Zhuozishan chicken (◆Z); 5: Tangqiao chicken (●T). Abbreviations: OPLS-DA, orthogonal partial least squares discrimination analysis; PCA, principal component analysis; PLS-DA, partial least squares discrimination analysis.

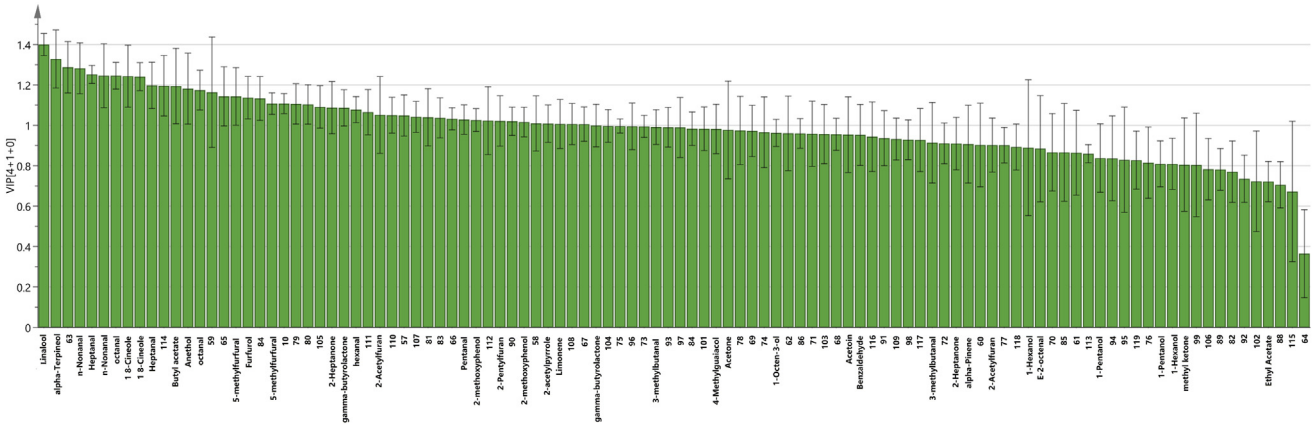


**Figure 5.** Variable Importance for the Projection (VIP) diagram of the OPLS-DA model. Abbreviation: OPLS-DA, orthogonal partial least squares discriminant analysis.

Liaoning Province (LNSPXKBD2020204, LNSPXKBD2020306).

STATEMENT: Our manuscript does not have any research which involves experimenting on live animals.

## DISCLOSURES

The authors declare that they have no known competing financial interests.

## SUPPLEMENTARY DATA

Supplementary data associated with this article can be found in the online version at https://doi.org/10.1016/j.psj.2020.09.011.

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