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# Characterization of aroma characteristics of silver carp mince glycated with different reducing sugars

Junya Liu<sup>a,b</sup>, Min You<sup>c</sup>, Xueshen Zhu<sup>a,\*\*</sup>, Wenzheng Shi<sup>b,\*</sup>

<sup>a</sup> School of Life Science and Chemistry, Key Laboratory of Innovative Applications of Bioresources and Functional Molecules of Jiangsu Province, Jiangsu Second Normal University, Nanjing 211200, China

<sup>b</sup> College of Food Science and Technology, Shanghai Ocean University, Shanghai 201306, China

<sup>c</sup> Technology Center, China Tobacco Jiangsu Industrial Co., Ltd., Nanjing 210019, China

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#### ABSTRACT

The purpose of this study was to investigate the volatile flavor changes in silver carp mince (SCM) gel glycated with different reducing sugars (glucose, L-arabinose, and xylose) based on *E*-nose, GC-IMS, and sensory evaluation. These results showed that glycation reduced the fishy smell of SCM gel and increased the meaty, toasty, and burnt smell. A total of 10 volatile compounds were considered as characteristic flavor compounds and potential markers. Among them, the main contributors of fishy included hexanal, heptanal, n-nonanal, octanal, etc. Toasty and burnt were mainly related to the production of 3-methylbutanal and furfurol. These results heralded that glycation could be used to improve the volatile flavor of SCM. This research provided a theoretical basis and technical support for glycation in aquatic food flavor quality control, aquatic pre-made food development, and aquatic leisure food processing.

# 1. Introduction

China is a large country in freshwater fish breeding, but because of its intense fishy smell, the processing efficiency of cultured freshwater fish is generally not high at present. To solve this predicament, scientists and food manufacturers have developed various methods, such as saline soaking, spices pickling, Maillard reaction, fermentation, and enzymatic hydrolysis. However, the current methods have some problems, such as poor fishy-removing effect and narrow application range, which can no longer meet people's increasing pursuit of healthy, nutritious, and delicious fish products. Therefore, it is still necessary to develop a new flavor enhancement technology to promote the development and utilization of fish products.

Glycation is one of the protein molecular modification techniques and usually occurs in the early stages of Maillard Reaction (Pirestani, Nasirpour, Keramat, & Desobry, 2017). Glycation is the covalent crosslinking reaction between the free amino group in the side chain of food proteins and the free carbonyl group in reducing sugar through the Maillard reaction to form glycoprotein (Chen, Tume, Xu, & Zhou, 2015). Silver carp (*Hypophthalmichthys molitrix*) is low in price and large production, which is a typical freshwater fish in China. In our early study, the effects of different amounts of L-arabinose on the sensory quality and digestibility of sausages prepared with silver carp mince (SCM) were analyzed. The results showed that glycation could effectively improve the smell, taste, and texture of silver carp sausage, and enhance the overall acceptability of the product without changing the protein digestibility (Liu, Huang, Feng, Luo, & Shi, 2022). In addition, we also investigated the changes in protein structure, protein nutritional value, glycation site, and volatile compounds of silver carp mince under different heating times with and without glucose (Liu, Hu, Wei, & Shi, 2022; Liu, Shen, Xiao, Jiang, & Shi, 2022). Based on the above studies, we found that different reducing sugars (L-arabinose and glucose) may have different effects on the odor characteristics of silver carp mince, which may be related to the degree of glycation, which could be regulated by the type of reducing sugar. However, the effect of different types of reducing sugar on the flavor characteristics of silver carp mince under a single variable was not clear. The types of characteristic flavor compounds were also unclear. Therefore, it is necessary to explore the above issues clearly through some technical means, such as electronic nose (E-nose), headspace-gas chromatography-ion mobility spectrometry (HS-GC-IMS), and quantitative description analysis (QDA).

Compared with oligosaccharides or polysaccharides,

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<sup>\*</sup> Correspondence to: W. Shi, No.999, Huchenghuan Rd, Nanhui New City, Pudong New District, Shanghai, China.

<sup>\*\*</sup> Correspondence to: X. Zhu, No.6, Xinhe West Road, Shiqiu Street, Lishui District, Nanjing City, Jiangsu Province, China. *E-mail addresses:* xueshen\_zhu@163.com (X. Zhu), wzshi@shou.edu.cn (W. Shi).

monosaccharides exhibit higher activity and efficiency in the glycation process (Ai, Xiao, & Jiang, 2021). Glucose, the most widely distributed and most important monosaccharide in nature, was selected as reducing sugar. In addition, considering the health strategy of a low-sugar diet, xylose and L-arabinose (two new sucrose substitutes) were also chosen as reducing sugar. Simulating the fish sausage processing system, SCM and reducing sugar were mixed and heated. The effect of different monosaccharides on the odor characteristics of SCM gel was analyzed by an E-nose, HS-GC-IMS, and artificial sensory evaluation (QDA). Based on multivariate statistical analysis, volatile compounds with significant differences were screened out, and their relationship with the odor characteristics of SCM gel was further analyzed, which laid the foundation for further research on flavor formation mechanisms. This research will provide a theoretical basis and technical support for glycation in aquatic food flavor quality control, aquatic pre-made food development, and aquatic leisure food processing. Moreover, this research will have important economic and social benefits to promote the high-quality and high-value processing and utilization of freshwater fish.

# 2. Materials and methods

# 2.1. Chemicals

D-(+)-glucose (C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>, MW 180.16), L-(+)-arabinose (C<sub>5</sub>H<sub>10</sub>O<sub>5</sub>, MW 150.13), and D-(+)-xylose (C<sub>5</sub>H<sub>10</sub>O<sub>5</sub>, MW 150.13) (All 99% purity) were purchased from Shanghai Macklin Biochemical Co., Ltd. (Shanghai, China). O-phthalaldehyde (OPA), disodium hydrogen phosphate, sodium dihydrogen phosphate, methanol, sodium dodecyl sulfate (SDS), borax, and  $\beta$ -mercaptoethanol were of analytical reagent grade and supplied by Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China).

# 2.2. Preparation of SCM gels added with different reducing sugars

SCM was prepared according to our previous report (Liu, Huang, et al., 2022). Silver carp (10 pieces, average weight 5 kg) were purchased from a farmer's market in Shanghai, slaughtered (blow to the head), and shipped to the laboratory. The back meat is collected uniformly and minced using a meat grinder. The chemical composition of raw minced meat was determined (moisture (78.23  $\pm$  0.15) %, protein (19.76  $\pm$  0.15) %, fat (1.02  $\pm$  0.15) %, ash (1.32  $\pm$  0.15) %, pH 6.78  $\pm$  0.15, AOAC, 2000), divided into 400 g per bag, and frozen in -80 °C refrigerator until use.

Defrost the raw SCM in a 4 °C refrigerator until the center is free of lumps. SCM was weighed (200 g) and mixed with 5% (*w*/w) glucose, L-arabinose, or xylose in a homogenizer (L13-Y91S, Joyoung, Zhejiang, China). Afterward, mixed samples were separated into 50 mL centrifuge tubes with threaded caps, and heated in a thermostatic water bath (HWS-24, Huitai, China) at 90 °C for 40 min. Another 200 g of SCM (without sugar) was performed in the same procedure as the control group. The heated SCM gel was placed in iced water for 30 min prior to overnight storage at 4 °C for further analysis.

#### 2.3. Determination of degree of grafting (DG)

DG was determined by a modified OPA method, and the OPA reagent was prepared based on our previous report (Liu, Shen, et al., 2022). When tested, 0.5 g of glycated SCM was weighed and mixed with 50 mL 50 mM phosphate buffer solution (PBS, pH 7.0, containing 0.1 M NaCl), followed by a layer of gauze filtration. Then the 200  $\mu$ L sample solution was mixed with 4 mL OPA reagents in a whirlpool oscillator and heated in a water bath at 35 °C for 2 min. The absorbance at a wavelength of 340 nm was determined by a UV-1800PC spectrophotometer (Shanghai Mapada Instrument Co., Ltd., Shanghai, China). Besides, 4 mL of OPA solution and 200  $\mu$ L of deionized water were used as a blank. DG is calculated as follows:

$$DG(\%) = (A_0 - A_t / A_0 \times 100$$
(1)

where  $A_0$  and  $A_t$  are the content of free amino groups of SCM in reaction time of 0 and t min, respectively.

#### 2.4. E-nose testing

The E-nose testing was carried out according to our previous report (Liu, Huang, et al., 2022). Approximately 4 g samples homogenously mixed with 0.18 g/mL brine (1: 1, w: v) were weighed and loaded in a 10 mL head-space vial. Samples were incubated at 60 °C for 15 min and then injected into a FOX 4000 *E*-nose odor analysis system (Alpha MOS, Toulouse, France). Injection volume: 2500  $\mu$ L. Injection temperature: 40 °C. The obtained data were analyzed by principal component analysis (PCA) using the AlphaSoft v12.0 software supported by the software of the instrument.

# 2.5. HS-GC-IMS testing

VFCs were evaluated using a GC-IMS Flavor analyzer (FlavourSpec®, Dortmund, Germany) based on Li, Wang, Yang, Dong, and Lin (2020). Two grams of sample was added into a 20 mL headspace injection vial and injected into the injector (500 µL, 85 °C, splitless mode) after incubating at 60 °C for 15 min. The chromatographic column type chosen was MXT-5 (column length 15 m, inner diameter 0.53 mm, film thickness 1.0 µm) provided by Restek Corporation (Bellefonte, Pennsylvania, USA). The heating procedure was set as follows: 2 mL/min for 2 min, 10 mL/min for 8 min, and 100 mL/min for 10 min. Drift gas (nitrogen gas) flow rate: 150 mL/min. IMS ionization chamber temperature: 45 °C. The retention index (RI) of each compound was calculated by the external standard method. Volatile compounds were identified by comparing RI and drift time (Dt) with the GC  $\times$  IMS Library Search (FlavourSpec®, Dortmund, Germany). The plug-ins of Gallery Plot and Dynamic PCA of VOCal supported by HS-GC-IMS instrument were used to draw fingerprint spectra, PCA plot, and fingerprint similarity analysis (FSA) plot.

# 2.6. Sensory evaluation

Sensory evaluation was carried out using the method of quantitative descriptive analysis based on Gomez and Lorenzo (2013) with a minor modification. The sensory panel consists of eight trained professionals (3 men and 5 women), who signed the Sensory Evaluation Consent Statement in advance and were trained for at least six months with the attributes and the scale to be used. Training details according to the methodology proposed by ISO regulations (ISO 8586-2:2008). Odor intensity and four sensory attributes (fishy, meaty, toasty, and burnt) were screened and assessed on a four-point scale: 0 = none, 1 = mild, 2= moderate, and 3 = strong. Among them, odor intensity was scored on a three-point scale: 1 = low, 2 = medium, and 3 = high. The flavor model for sensory evaluation was adopted (Table S1) as reported by Liu, Liu, He, Song, and Chen (2015) and Cai et al. (2016). Before the experiment, the sample was cut into fixed-size slices and placed on a table in the sensory evaluation analysis room (25 °C, incandescent lamp). All samples were subjected to sensory evaluation under the same conditions. Ethical permission was not required for this trial.

#### 2.7. Statistical analysis

The experiment was repeated three times. Each measurement was performed at least three times during each experiment. The results were presented as the mean  $\pm$  standard deviation (SD). Significance analysis was performed by one-way analysis of variance (ANOVA) and Duncan's multiple range tests at the level of 0.05 (P < 0.05) using SPSS 21.0

software (IBM, Armonk, NY, USA). Radar map was drawn by Origin 2019b (OriginLab Corporation, MA, USA). Orthogonal partial least squares discriminant analysis (OPLS-DA) method was carried out using SIMCA 14.1 (Umetrics Corporation, Malmo, Sweden). Partial least squares regression (PLSR) was operated by XLSTAT (Annual Version 2021.2.2, Addinsoft Inc., New York, NY).

#### 3. Results and discussion

# 3.1. Change in DG

DG represents the degree of binding between protein and reducing sugar (Pirestani et al., 2017; Wang et al., 2019). The higher the DG value, the higher the degree of glycation. As shown in Fig. 1, the DG was affected by monosaccharide types. The DG value of SCM gel glycated with xylose was the highest, followed by L-arabinose, while glucose and control group N were the lowest. Glycation extent was affected by the type of reducing sugar, the ratio of reducing sugar to protein, heating temperature, heating time, pH, moisture content, and ionic strength (Oliveira, Coimbra, De Oliveira, ZuñIga, & Rojas, 2016). The reactivity of the reducing sugar used in this experiment was: L-arabinose > Dxylose > glucose. However, The DG value of sample X was greater than sample A, indicating that the optimum conditions for the formation of maximum DG of different types of sugar were different. The most suitable reaction conditions should be determined according to the actual production needs. In this study, changes in each indicator were evaluated only based on the current reaction system.

# 3.2. E-nose analysis

E-nose technology is an effective way to distinguish discrepancies between aroma profiles for different samples. Fig. 2 shows the radar map, PCA plot and discriminant factor analysis (DFA) plot of odor profiles of SCM gels glycated with different monosaccharides. According to the response values of the *E*-nose sensors in Fig. 2A, the control group had lower response values at sensors TA/2, T40/1, T40/2, P30/2, P40/2, PA/2, T70/2, P40/1, P10/2, P10/1, and T30/1. After glycation, the response values of the above sensors in all SCM samples increased to varying degrees. The sensitive substance types corresponding to E-nose sensors are shown in Table S2 (Xie et al., 2022). As seen in Table S1, it could be concluded that glycation changed the concentration of polar compounds, ketones, amines, hydrocarbons, and other polar



**Fig. 1.** Changes in DG of SCM gels glycated with different monosaccharides. SCM: silver carp mince; N: SCM without sugar; G: SCM with glucose; A: SCM with L-arabinose; X: SCM with xylose.







**Fig. 2.** Radar map (A), PCA plot (B), and DFA plot (C) of E-nose data of SCM gels glycated with different monosaccharides. SCM: silver carp mince; N: SCM without sugar; G: SCM with glucose; A: SCM with L-arabinose; X: SCM with xylose.

compounds.

To further distinguish the difference in aroma profiles, the response value signals were analyzed by PCA and DFA. As seen in Fig. 2B, the first principal component (PC1) explained 97.414% of the sample variance, the second principal component (PC2) explained only 2.059%. This result suggested that the majority of the variation captured by PC1 for all volatile compounds allowed for the distinction between the samples. PC2 remained important in distinguishing aroma profiles of different samples, although there were notably small variations (Zhou, Chong, Ding, Gu, & Liu, 2016). Discrimination index (DI) represents the degree of sample discrimination provided by the electronic nose statistics software. A positive DI value indicates that the samples are independent of each other (Wang, Zhang, Zhu, Wang, & Shi, 2018). Moreover, the larger the DI value, the better the differentiation effect, and the DI value between 80 and 100 indicates effective differentiation. In this experiment, DI was 71, indicating that the samples could be distinguished (such as A and X) but there was similarity among the samples (such as N

and G). This result might be due to the fact that xylose and L-arabinose are five-carbon sugars and have a higher glycation reaction rate than samples N and G.

DFA is designed to further analyze data based on PCA to better distinguish samples. As seen in Fig. 2C, the contribution rates of DF1 and DF2 were 84.845% and 13.019% respectively, and the cumulative contribution rate was over 85%. Therefore, DF1 and DF2 could comprehensively represent the original information of the sample. DFA

showed that the four groups of samples could be completely distinguished and there was the similarity between samples A and X, which did not conflict with PCA results.

# 3.3. HS-GC-IMS analysis

*3.3.1. Qualitative and quantitative analysis of VFCs* HS-GC-IMS is a simple, rapid, and sensitive measurement technique



Fig. 3. Two-dimensional topographic plot (A), fingerprint spectra (B), PCA plot (C), and FSA plot (D) of volatile compounds in SCM gels glycated with different monosaccharides. SCM: silver carp mince; N: SCM without sugar; G: SCM with glucose; A: SCM with L-arabinose; X: SCM with xylose.

for VFCs and has been demonstrated to isolate and identify VFCs in complex matrices, such as aldehydes, ketones, alcohols, and other volatiles (Li et al., 2020). Fig. 3 shows the two-dimensional topographic plot, fingerprint spectra, PCA plot, and FSA plot of VFCs in SCM gels glycated with different monosaccharides. The vertical red line in Fig. 3A represents the reactive ion peak (RIP) normalized to the drift time (DT). Each dot on the two-dimensional topographic plot represents a VFC. The color of the dot represents the concentration of the VFC. Blue means lower concentration, and red means higher concentration (Li et al., 2020). As seen in Fig. 3A, the color point distribution contours of all samples were similar, that is to say, and the types of VFCs of SCM gels glycated with different monosaccharides were similar.

To further compare the differences of VFCs in the samples, the fingerprint spectra was generated in Fig. 3B. Each line in the graph represents all signal peaks selected from a sample, and each column in the graph represents the signal peak of the same VFCs in different samples (Xie et al., 2022). A total of 50 signal peaks were obtained in this study. Among them, 14 VFCs could not be identified in the migration spectrum library and were replaced by numbers (1–14). Another 36 VFCs were accurately identified (Table S3), including 18 aldehydes, 7 ketones, 8 alcohols, and 3 esters. As seen in Fig. 3B, the fingerprint spectra of VFCs in the three groups of samples G, A, and X were different from that of control group N. The concentrations of VFCs (red region 1 of Fig. 3B) in N were higher than those in other samples, mainly including aldehydes, ketones, alcohols, and esters. In addition, the concentrations of VFCs (red region 2 of Fig. 3B) in X and A were similar, and they were higher than those in other samples, mainly including: 2-butanone (monomer and dimer), 3-methylbutanal (monomer and dimer), 3methyl-3-buten-1-ol, acetone, and furfurol (monomer and dimer).

Aldehydes were mainly produced by lipid oxidation and Maillardinduced amino acid degradation (Han et al., 2020; Li et al., 2021). Ketones are another significant flavor precursor in the Maillard reaction system (K. Chen et al., 2020), and are related to the degree of lipid oxidation, especially the β-oxidation of fatty acids derived from triglycerides (Yin et al., 2021). Alcohols are mainly derived from the oxidation and degradation of lipids (Yin et al., 2021). Linear alcohols were the main lipid oxidation products, while branched alcohols were the degradation products from the corresponding branched aldehydes (Yin et al., 2021). Esters are usually produced by esterification reactions between carboxylic acids and alcohols (Lorenzo & Carballo, 2015). Therefore, the decrease in VFCs in red region 1 of Fig. 3B was related to the Maillard reaction, lipid oxidation and degradation, and their interaction. In red region 2 of Fig. 3B, 3-methylbutanal (dimer and monomer) and furfurol (dimer and monomer) are two important Maillard reaction intermediates. 3-methylbutanal participated in the Maillard reaction and Strecker degradation, related to branched amino acids (valine, isoleucine, and leucine), and usually provided the pleasant odor of peach, chocolte, malt, and bitter almonds (Luo, Nasiru, Zhuang, Zhou, & Zhang, 2021). Furfurol is another typical intermediate Maillard reaction product, formed mainly from pentose at low pH values (pH < 7.0), and provides the odor of caramel and bread (Li, Belloch, & Flores, 2021; Martins, Jongen, & Boekel, 2000). The peak area of 3-methylbutanal and furfurol increased with the increase of DG, indicating that glycation promoted the Maillard reaction. The peak area of 2-butanone (dimer and monomer) and acetone, associated with lipid oxidation, increased with the increase of DG, indicating that glycation promoted lipid oxidation. Ethanol and 3-methyl-3-buten-1-ol usually do not have an impact on the sensory characteristics in the food system owing to their high odor thresholds (Wang, Zhang, Wang, Wang, & Liu, 2019).

To further explore the effect of glycation on the overall aroma profile of samples, all VFCs were classified into four categories (aldehydes, ketones, alcohols, and esters), and the peak volume of each category is shown in Table S3. The peak volume of total aldehydes and total VFCs in control group N were significantly higher than those in other samples. After glycation, sample G with low DG had lower VFCs of all classes than control group N, except total esters. Sample X with high DG had higher VFCs of all classes than sample G. These results showed that glycation enhanced the overall aroma profile of SCM gels.

## 3.3.2. PCA and DFA of VFCs

PCA and FSA were performed to further evaluate the differences in overall aroma profile between samples. As can be seen from Fig. 3C, PC1 and PC2 could explain 72% and 18% of the variation in volatile compounds in the samples, respectively. PCA showed that four samples could be distinguished and samples X and A had similar odor profiles. In the FSA plot (Fig. 3D), the closer the distance between the samples, the smaller the difference. FSA indicated that samples X and A were similar, which was similar to the results of fingerprint spectra and *E*-nose DFA.

#### 3.3.3. OPLS-DA of VFCs

OPLS-DA is a supervised and newly developed statistical method of discriminant analysis to achieve the prediction of sample classifications (Zhang, Zhang, & Xing, 2021). To find out the significantly different VFCs in samples, the OPLS-DA model was established. Fig. 4 shows the OPLS-DA score plot, permutation plot, and variable importance for the projection (VIP) plot of VFCs in SCM gels glycated with different monosaccharides. The OPLS-DA score plot (Fig. 4A) showed the differences between samples consistent with the GC-IMS PCA plot (Fig. 3E).  $R^{2}X$  (cum),  $R^{2}Y$  (cum), and  $Q^{2}$  represent the cumulative interpretation rate of the model in the X-axis and Y-axis, as well as the cumulative prediction rate of the model in multivariate statistical analysis modeling, respectively. Larger values for R<sup>2</sup>X (cum), R<sup>2</sup>Y (cum), and Q<sup>2</sup> indicate better models. In this study, the  $R^2X$  (cum),  $R^2Y$  (cum), and  $Q^2$ were 0.987, 0.971, and 0.931 respectively, and  $R^2X$ - $R^2Y < 0.3$ , indicating that the OPLS-DA model was excellent. The permutation plot (Fig. 4B) was conducted to verify the reliability of the model by R<sup>2</sup> and  $Q^2$  values. All permuted  $Q^2$  values were lower than the original ones, and the Q<sup>2</sup> regression line had a negative value of intercept, verifying this model performed pretty well and was not overfitted.

The VIP plot was applied to screen VFCs with significant differences (Fig. 4C). When the VIP value of VFC is >1 (VIP >1), the VFC is considered as a characteristic flavor compound that can be used as a potential marker. As illustrated in Fig. 4C, 10 VFCs were considered to be characteristic flavor compounds and potential markers, including hexanal (dimer and monomer), 3-methylbutanal (dimer), ethanol, 2butanone (dimer), acetone, methyl benzoate (dimer), heptanal (dimer), n-nonanal (monomer), and octanal (monomer). Among them, hexanal (4.5 ng/g), heptanal (2.8 ng/g), n-nonanal (1.1 ng/g), octanal (0.587 ng/g), and 3-methylbutanal (0.2 ng/g) usually have a greater effect on food flavor due to their low thresholds (Zhou et al., 2016). Hexanal is derived from the oxidation of n-6 fatty acids such as linoleic acid and arachidonic acid, while heptanal, n-nonanal, and octanal are the autooxidation products of oleic acid (Luo et al., 2021). 3-methylbutanal is a typical Maillard reaction intermediate. Alcohols (ethanol), ketones (2-butanone dimer and acetone), and esters (methyl benzoate dimer) usually have high odor thresholds and do not affect the sensory characteristics of products (Wang, Zhang, et al., 2019). To sum up, five volatile compounds (hexanal, heptanal, n-nonanal, octanal, and 3-methylbutanal) should be paid more attention to in the subsequent research on the molecular mechanism of glycation affecting flavor formation in SCM gels.

## 3.4. Sensory evaluation

Mean sensory scores for SCM gels glycated with different monosaccharides are shown in Fig. 5. Glycation had a significant effect on odor intensity (P < 0.05). The fishy smell of control group N was strong, but the fishy smell of the SCM after glycation weakened. Instead, meaty, toasty, and burnt became the main aroma characteristics. In all the samples, sample A had the highest score for toasty flavor, and sample X had the highest score for burnt flavor. Meaty and toasty flavor are pleasant aromas, while burnt is the opposite (Cai et al., 2016). Meat



**Fig. 4.** OPLS-DA score plot (A), permutation plot (B), and VIP plot of volatile compounds modeling in SCM gels glycated with different monosaccharides. VIP: variable importance for the projection; SCM: silver carp mince; N: SCM without sugar; G: SCM with glucose; A: SCM with L-arabinose; X: SCM with xylose.

flavor was related to the generation of thiazoles, sulfides, aldehydes, and ketones. Toasty was mainly contributed by pyrazines, pyrroles, and miazines. In addition, toasty might be changed into disagreeable burnt owing to the caramelization of sugar or the overreaction of materials. This burnt smell could be suppressed by controlling glycation extent, which is affected by the amount of reducing sugar added, heating time, heating temperature, and other factors (Liu, Huang, et al., 2022). This result that A and X had a similar and interchangeable aroma characteristic could also explain why A and X were close in the *E*-nose PCA and DFA results. These results suggested that glycation is an effective technique for enhancing the sensory quality of SCM.



Fig. 5. Radar map of sensory scores of SCM gels glycated with different monosaccharides. SCM: silver carp mince; N: SCM without sugar; G: SCM with glucose; A: SCM with L-arabinose; X: SCM with xylose.

## 3.5. PLSR of sensory evaluation data and GC-IMS data

To further analyze the relationship between sensory attributes and odor compounds, PLSR was performed between sensory evaluation data (X matrix) and GC-IMS data (Y matrix) (Yan et al., 2024; Zhang et al., 2022). The two circles in Fig. 6A (inner circle and outer circle) represent 50% and 100% of the explanatory variance, respectively. The  $R^2X$ (cum),  $R^2Y$  (cum), and  $Q^2$  in Fig. 6B represent the cumulative interpretation rate of the model in the X-axis and Y-axis, as well as the cumulative prediction rate of the model in multivariate statistical analysis modeling, respectively. Larger values for R<sup>2</sup>X (cum), R<sup>2</sup>Y (cum), and Q<sup>2</sup> indicate better models. Moreover,  $R^2X-R^2Y < 0.3$  indicates an excellent model. As seen in Fig. 6B, the R<sup>2</sup>X, R<sup>2</sup>Y, and Q<sup>2</sup> cumulated index of Comp1 were 0.908, 0.684, and 0.394, respectively. The R<sup>2</sup>X, R<sup>2</sup>Y, and Q<sup>2</sup> cumulated index of Comp2 were 0.997, 0.932, and 0.811, respectively. The difference of  $R^2X-R^2Y$  in the two components was <0.3, indicating that the PLSR model provided an excellent quality predictive model.

According to PLSR results, there were significant correlations between sensory attributes and odor compounds in SCM gels to varying degrees. Fishy was related to heptanal (7, 8), octanal (11,12), n-nonanal (17, 18), hexanal (20), 1-pentanol (4), 1-hexanol (5), 1-pentanol (21,22), and 2-heptanone (33, 34). This result was consistent with the report of Zhou et al. (2016), which reported that the smell of fishy was mainly derived from aldehydes, ketones, and alcohols. The reduction of these substances could effectively reduce the smell of fishy. In particular, four substances (heptanal, octanal, n-nonanal, and hexanal) with lower thresholds and significant differences (in OPLS-DA) may have a more important contribution to the reduction of fishy odor.

As shown in Fig. 6A, meaty was close to the roast flavor. Toasty and burnt in the same quadrant, and showed a significant positive correlation with 3-methylbutanal dimer (30) and furfurol (1). 3-methylbutanal and furfurol are two important intermediates of the Maillard reaction. These results indicated that meaty, toasty, and burnt could be transformed into each other, and their formation was closely related to the Maillard reaction. This result did not conflict with that of OPLS-DA, which shows that 3-methylbutanal was an important characteristic flavor compound (VIP > 1). In addition, this result was also consistent with our previous reports that glycation promoted the production of more aroma substances through the Maillard reaction (Liu, Shen, et al., 2022).





Fig. 6. Correlations loadings (A) and model quality (B) of PLSR. Figs. (1-36) in Fig. 6A correspond to the number of volatile compounds in Table S3.

All in all, PLSR showed that different reducing sugar types (different degrees of glycation) have different effects on the volatile flavor characteristics of SCM gels.

#### 4. Conclusion

The aroma characteristics of SCM gels could be effectively improved by glycation with glucose, L-arabinose, and xylose. Among them, xylose and L-arabinose were effective in enhancing the volatile flavor of fish. Glycation reduced the fishy smell of SCM gel and increased the meaty, toasty, and burnt smell. PCA of *E*-nose and GC-IMS data showed that the samples could be distinguished effectively. GC-IMS showed that glycation improved the overall aroma profile of SCM gel by promoting Maillard reaction and lipid oxidation. OPLS-DA showed that a total of 10 VFCs were considered as characteristic flavor compounds and potential markers. Among them, hexanal, heptanal, n-nonanal, octanal, and 3methylbutanal had more potential to be used as markers to analyze the mechanism of glycation affecting the volatile flavor of SCM due to their low thresholds. Furthermore, PLSR showed that the main contributors of fishy included hexanal, heptanal, n-nonanal, octanal, etc. Toasty and burnt were mainly related to the production of 3-methylbutanal and furfurol. In a word, this research heralded that glycation could be used as an aroma improvement technique for developing fish products.

# CRediT authorship contribution statement

Junya Liu: Writing – original draft, Visualization, Methodology, Formal analysis, Data curation, Conceptualization. Min You: Writing – review & editing, Visualization, Formal analysis. Xueshen Zhu: Writing – review & editing, Funding acquisition, Formal analysis. Wenzheng Shi: Writing – review & editing, Project administration, Formal analysis.

# Declaration of competing interest

The authors declare that no conflict of interest exist in this research article.

# Data availability

The authors do not have permission to share data.

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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.2024.101335.

# References

- Ai, M. M., Xiao, N., & Jiang, A. M. (2021). Molecular structural modification of duck egg white protein conjugates with monosaccharides for improving emulsifying capacity. *Food Hydrocolloids*, 111, Article 106271. https://doi.org/10.1016/j. foodhyd.2020.106271
- AOAC. (2000). Offical methods of analysis of AOAC International (17th ed.). Gaithersburg, MD, USA: AOAC International.
- Cai, L. Y., Li, D. M., Dong, Z. J., Cao, A. L., Lin, H., & Li, J. R. (2016). Change regularity of the characteristics of Maillard reaction products derived from xylose and Chinese shrimp waste hydrolysates. *LWT - Food Science and Technology*, 65, 908–916. https:// doi.org/10.1016/j.lwt.2015.09.007
- Chen, K. N., Yang, Q. F., Hong, H., Feng, L. G., Liu, J., & Luo, Y. K. (2020). Physicochemical and functional properties of Maillard reaction products derived from cod (Gadus morhua L.) skin collagen peptides and xylose. *Food Chemistry*, 333, Article 127489. https://doi.org/10.1016/j.foodchem.2020.127489
- Chen, X., Tume, R. K., Xu, X. L., & Zhou, G. (2015). Solubilization of myofibrillar proteins in water or low ionic strength media: Classical techniques, basic principles, and novel functionalities. *Critical Reviews in Food Science and Nutrition*, 57(15), 3260–3280. https://doi.org/10.1080/10408398.2015.1110111
- Gomez, M., & Lorenzo, J. M. (2013). Effect of fat level on physicochemical, volatile compounds and sensory characteristics of dry-ripened "chorizo" from Celta pig breed. *Meat Science*, 95(3), 658–666. https://doi.org/10.1016/j. meatsci 2013.06.005
- Han, G., Zhang, L., Li, Q. X., Wang, Y., Chen, Q., & Kong, B. H. (2020). Impacts of different altitudes and natural drying times on lipolysis, lipid oxidation and flavour profile of traditional Tibetan yak jerky. *Meat Science*, 162, Article 108030. https:// doi.org/10.1016/j.meatsci.2019.108030
- ISO (International Organization for Standardization). (2008). General guidance for the selection, training and monitoring of assessors. Part 2: Expert sensory assessors, ISO 8586–2:2008.
- Li, L., Belloch, C., & Flores, M. (2021). The Maillard reaction as source of meat flavor compounds in dry cured meat model systems under mild temperature conditions. *Molecules*, 26(1), 223. https://doi.org/10.3390/molecules26010223
- Li, W. Q., Chen, Y. P., Blank, I., Li, F. Y., Li, C. B., & Liu, Y. (2021). GC x GC-ToF-MS and GC-IMS based volatile profile characterization of the Chinese dry-cured hams from different regions. *Food Research International*, 142, Article 110222. https://doi.org/ 10.1016/j.foodres.2021.110222
- Li, X. R., Wang, K., Yang, R. W., Dong, Y. F., & Lin, S. Y. (2020). Mechanism of aroma compounds changes from sea cucumber peptide powders (SCPPs) under different

storage conditions. Food Research International, 128, Article 108757. https://doi.org/ 10.1016/j.foodres.2019.108757

- Liu, J. B., Liu, M. Y., He, C. C., Song, H. L., & Chen, F. (2015). Effect of thermal treatment on the flavor generation from Maillard reaction of xylose and chicken peptide. LWT-Food Science and Technology, 64(1), 316–325. https://doi.org/10.1016/j. lwt.2015.05.061
- Liu, J. Y., Hu, Y., Wei, H. H., & Shi, W. Z. (2022). Effect of glycation on protein structure, amino acid composition and digestibility of silver carp mince. *International Journal of Food Science & Technology*, 57(4), 2487–2497. https://doi.org/10.1111/ijfs.15617
- Liu, J. Y., Huang, S. Q., Feng, Q., Luo, Y. X., & Shi, W. Z. (2022). Sensory quality and digestibility evaluation of silver carp sausage glycated with L-arabinose. *Journal of Food Processing and Preservation*, 46(2), Article e16220. https://doi.org/10.1111/ JFPP.16220
- Liu, J. Y., Shen, S. Y., Xiao, N. Y., Jiang, Q. Q., & Shi, W. Z. (2022). Effect of glycation on physicochemical properties and volatile flavor characteristics of silver carp mince. *Food Chemistry*, 386, Article 132741. https://doi.org/10.1016/j. foodchem.2022.132741
- Lorenzo, J. M., & Carballo, J. (2015). Changes in physico-chemical properties and volatile compounds throughout the manufacturing process of dry-cured foal loin. *Meat Science*, 99, 44–51. https://doi.org/10.1016/j.meatsci.2014.08.013
- Luo, J., Nasiru, M. M., Zhuang, H., Zhou, G. H., & Zhang, J. H. (2021). Effects of partial NaCl substitution with high-temperature ripening on proteolysis and volatile compounds during process of Chinese dry-cured lamb ham. *Food Research International*, 140, Article 110001. https://doi.org/10.1016/j.foodres.2020.110001
- Martins, S. I. F. S., Jongen, W. M. F., & Boekel, M. A. J. S. V. (2000). A review of Maillard reaction in food and implications to kinetic modelling. *Trends in Food Science & Technology*, 11(9), 364–373. https://doi.org/10.1016/S0924-2244(01)00022-X
- Oliveira, F. C. D., Coimbra, J. S. D. R., De Oliveira, E. B., ZuñIga, A. D. G., & Rojas, E. E. G. (2016). Food protein-polysaccharide conjugates obtained via the Maillard reaction: A review. *Critical Reviews in Food Science and Nutrition*, 56(7), 1108–1125. https://doi.org/10.1080/10408398.2012.755669
- Pirestani, S., Nasirpour, A., Keramat, J., & Desobry, S. (2017). Preparation of chemically modified canola protein isolate with gum Arabic by means of Maillard reaction under wet-heating conditions. *Carbohydrate Polymers*, 155, 201–207. https://doi. org/10.1016/j.carbpol.2016.08.054
- Wang, C. Y., Li, J. H., Li, X., Chang, C. H., Zhang, M. Q., Gu, L. P., ... Yang, Y. J. (2019). Emulsifying properties of glycation or glycation-heat modified egg white protein. *Food Research International*, 119, 227–235. https://doi.org/10.1016/j. foodres.2019.01.047
- Wang, H. L., Zhang, J. J., Zhu, Y. Z., Wang, X. C., & Shi, W. Z. (2018). Volatile components present in different parts of grass carp. *Journal of Food Biochemistry*, 42 (6), Article e12668. https://doi.org/10.1111/jfbc.12668
- Wang, W. L., Zhang, L., Wang, Z. Q., Wang, X. C., & Liu, Y. (2019). Physicochemical and sensory variables of Maillard reaction products obtained from Takifugu obscurus muscle hydrolysates. *Food Chemistry*, 290, 40–46. https://doi.org/10.1016/j. foodchem.2019.03.065
- Xie, Q. S., Xu, B. C., Xu, Y., Yao, Z., Zhu, B. W., Li, X. F., & Sun, Y. (2022). Effects of different thermal treatment temperatures on volatile flavour compounds of waterboiled salted duck after packaging. *LWT- Food Science and Technology*, 154, Article 112625. https://doi.org/10.1016/j.lwt.2021.112625
- Yan, X. H., Tian, Y., Zhao, F., Wang, R. F., Zhou, H. J., Zhang, N. M., ... Zhang, C. H. (2024). Analysis of the key aroma components of Pu'er tea by synergistic fermentation with three beneficial microorganisms. *Food Chemistry: X, 21*, Article 101048. https://doi.org/10.1016/j.fochx.2023.101048
- Yin, X. Y., Lv, Y. C., Wen, R. X., Wang, Y., Chen, Q., & Kong, B. H. (2021). Characterization of selected Harbin red sausages on the basis of their flavour profiles using HS-SPME-GC/MS combined with electronic nose and electronic tongue. *Meat Science*, 172, Article 108345. https://doi.org/10.1016/j.meatsci.2020.108345
- Zhang, D. N., Yang, N., Fisk, I. D., Li, J. T., Liu, Y., & Wang, W. L. (2022). Impact of cooking on the sensory perception and volatile compounds of Takifugu rubripes. *Food Chemistry*, 371, Article 131165. https://doi.org/10.1016/j. foodchem.2021.131165
- Zhang, J., Zhang, W. G., & Xing, L. J. (2021). Effects of ultrasound on the taste components from aqueous extract of unsmoked bacon. *Food Chemistry*, 365, Article 130411. https://doi.org/10.1016/j.foodchem.2021.130411
- Zhou, X. X., Chong, Y. Q., Ding, Y. T., Gu, S. Q., & Liu, L. (2016). Determination of the effects of different washing processes on aroma characteristics in silver carp mince by MMSE-GC-MS, e-nose and sensory evaluation. *Food Chemistry*, 207, 205–213. https://doi.org/10.1016/j.foodchem.2016.03.026