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Research Article

Effects of sturgeon oil and reheating methods on the qualities of sturgeon surimi gels

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Abstract: The addition of oil played a significant role in the gelling properties and flavor qualities of the ready-to-eat surimi gels. In order to investigate the effects of sturgeon oil and reheating methods on the qualities of sturgeon surimi gels, the prepared sturgeon surimi gels containing sturgeon oil (with the direct addition named SO and with the Pickering emulsion named PPE) were reheated by boiling, steaming, microwave, and frying, respectively. The gel properties, non-volatile and volatile flavor substances and digestibility were determined. Boiling and steaming made gels higher brightness value (L^*) (82.07–84.16), with lower cooking loss (7.11%–8.83%), relatively intact protein network structures and strong protein digestibility. The greatest lipid oxidation of the sturgeon surimi gels was induced by frying, followed by microwave. Furthermore, the gels reheated by microwave showed the highest total flavor-presenting nucleotides, while the steamed ones had the highest total percentage of fresh and sweet amino acids. The addition of sturgeon oil (direct addition and Pickering emulsion) decreased the textural properties of the reheated gels, but significantly improved the L^* and protein digestibility of boiled and steamed gels ($P < 0.05$). The gastric digestibility and gastrointestinal digestibility of the SO and PPE groups were 68.60%–76.33% and 80.69%–85.05%. Moreover, the addition of sturgeon oil also changed the ratio of non-volatile and volatile flavor compounds. Compared with the direct addition of sturgeon oil, the preparative Pickering emulsion was more conducive to the retention of better textural properties as well as the more flavor-presenting nucleotides and volatile flavor substances. The above results enrich the theory of quality difference of sturgeon surimi products caused by different reheating methods, and provide reference for the development of high-quality products.

Keywords: surimi gel; reheating; lipid oxidation; flavor substances; protein digestibility

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1 Introduction

In recent years, surimi products have been well received by consumers due to their unique texture, rich nutrition, delicious taste and convenient consumption[1]. Rinsing, as a key step in the preparation of surimi, washed away water-soluble proteins, pigments and bad odors from the fish to improve the quality and preservation of surimi products. However, the removal of a large amount of fish fat in the process of rinsing would lead to roughness in the texture of surimi products, reduction in the flavor and nutritional values[2–3], which was often improved by adding exogenous fats and oils. At present, fats and oils with direct addition and in the form of emulsions were the two common methods added to surimi[4].

China was the world's largest sturgeon (Acipenseridae) aquaculture country, whose production amounts accounted for about 86% of the global[5]. Perennial large sturgeon was mainly used to obtain caviar. Simultaneously, the by-products containing fish meat, fish head, fins, and swim bladder were not fully utilized, which caused great waste of resources and environmental pollution[6]. Sturgeon meat, as a by-product of sturgeon caviar processing, is commonly processed or cooked by the method of frying or boiling in China[7]. Sturgeon meat was rich in nutrients and had no thorns between the muscles, which made it a good raw material for preparing surimi[5]. Steak, surimi, and sausage are the common products made of sturgeon meat[8]. Furthermore, the rest of the by-products, such as fish head, were rich in oil. Therefore, in the present study, surimi gels were prepared from sturgeon meat, and sturgeon oil (extracted from fish head) was added to improve the flavor and nutritional value.

Surimi products on the market were mainly in the form of frozen pre-cooked products, which were usually reheated before consumption by consumers[9]. Different reheating methods may result in different responses in terms of product appearance, texture and flavor due to differences in heat transfer media[10]. During reheating, oils and fats will not only change the physic-chemical properties such as color and viscosity, but also produce a series of volatile substances through oxidative decomposition[11–12]. In addition, lipid oxidation will induce protein oxidation, leading to protein degradation and reduced digestibility[13]. Regarding the effect of oil on the quality of surimi gels, many researches mainly focused on the ready-to-eat surimi gels after two-stage heating. However, there were few reports on the changes in the quality of surimi gels after reheating and the quality differences caused by different reheating methods. The application of hot processing methods,

including steaming, boiling, roasting, and microwaving, has been observed in the food processing of eel, European sea bass and red mullet. These methods have been demonstrated to exert a considerable influence on the flavor and consumer preferences associated with these foods[8]. In China, the primary methods of processing sturgeon meat products are frying and steaming[7].

Therefore, in this study, sturgeon oil extracted from sturgeon head was added to the surimi by different methods (direct addition and Pickering emulsion), and reheating treatments (boiling, steaming, microwave, and frying) were carried out. The effects of oil addition and reheating methods on the gel characteristics, non-volatile, and volatile flavor substances and protein digestibility of sturgeon surimi gels were studied to provide theoretical guides to the development of high-quality surimi products.

2 Materials and methods

2.1 Materials and reagents

Frozen sturgeon meat and head were provided by Quzhou Sturgeon Aquatic Food Technology Development Co., Ltd. (Quzhou, China) and stored at −20 °C. Pea isolate protein was purchased from Jinkangtai Biotechnology Co., Ltd. (Shaanxi, China).

2.2 Preparation of the sturgeon oil

The sturgeon head was completely thawed at 4 °C and added with distilled water (1:1, *m*/*m*). The neutral protease (50 U/mg) and papain (≥ 6 000 U/mg) (1.5% by weight of fish head) were added after a preheating water bath (50 °C, 30 min), in which the ratio of the two enzymes was 1:2 (*m*/*m*) and hydrolyzed for 4 h with stirring. After that, the reaction was deactivated at 95 °C for 10 min. The centrifugation was carried out at 10 000 × *g* for 15 min and the upper layer was removed to obtain the crude sturgeon oil. The extraction rate was 85.26% (*m*/*m*). The crude fish oil was then refined. 1) Degumming: add 0.5% (*m*/*m*) of oil weight of phosphoric acid (85%, *V*/*V*) to the crude fish oil, heat at 60 °C for 40 min with stirring, cool and centrifuge at 6 000 r/min for 8 min to obtain the top layer of degummed oil. 2) Deacidification: add 7% (*m*/*m*) of the oil weight of NaOH (1 mol/L) to the degummed oil, stir the reaction at 60 °C for 30 min, cool and centrifuge at 6 000 r/min for 8 min to obtain the top layer of deacidified oil. 3) Decolourisation: 5% (*m*/*m*) oil weight of activated white clay was added to the decolourised oil, stirred continuously at 60 °C for 30 min, cooled, and centrifuged at 6 000 r/min for 8 min to obtain the top layer of decolourised oil. 4) Deodorisation: the decolourised oil was placed in a round-bottomed flask and deodorised in a water bath at 70 °C for 20 min under vacuum using a rotary evaporator, and then the refined fish oil was obtained. The purification rate was estimated to be about 70% (*m*/*m*).

2.3 Preparation of Pickering emulsions

The method of Cao et al.[14] was referred to and slightly modified. Pea isolate protein (5 g) was dispersed in 100 mL of distilled water and placed in a refrigerator at 4 °C for 12 h. The concentration of NaCl in the protein suspension was adjusted to 300 mmol/L. The solutions were heated (95 °C, 30 min) to obtain protein nano-dispersions which then immediately cooled to room temperature in ice water. The pH values of the dispersions were adjusted to 7.0, and homogenized (11 000 r/min, 3 min) and mixed with sturgeon oil (1:2, *m*/*m*) to produce Pickering emulsion.

2.4 Preparation of surimi gels

The white sturgeon meat was cut into small pieces and washed twice with 5 times the volume of distilled water and NaCl solution (2.5 g/L)[15], which was centrifuged at 10 000 × *g* for 20 min to remove excess water. Then 200 g of meat were chopped for 2 min then 3% (*m*/*m*) NaCl was added, and the moisture content was adjusted to 80% (*m*/*m*). Subsequently, sturgeon oil (8%, *m*/*m*) or Pickering emulsion (12%, *m*/*m*) was added, and the surimi pastes were chopped for another 5 min. The obtained substance was filled into a plastic sausage jacket (diameter 3.8 cm) and then processed by two-stage heating (40 °C-60 min, 90 °C-30 min). The fish intestines were immediately cooled in ice water and stored in a refrigerator at 4 °C overnight. Finally, they were cut into cylinders (20 mm high) and placed in self-sealing bags and frozen at −18 °C for 5 d. The samples were divided into three groups: the control group (without sturgeon oil or emulsion), the SO group (with sturgeon oil added directly) and the PPE group (with Pickering emulsion).

2.5 Reheating treatment

The surimi gels were completely thawed in a refrigerator at 4 °C, then reheated by boiling (4 min), steaming (4 min), microwave (500 W, 2.5 min) and frying (about 180 °C, 3 min). All of the samples reached a final center temperature of 85–90 °C.

2.6 Measurement of cooking loss

The mass of the surimi gels before and after reheating was recorded and denoted as m_1 (g) and m_2 (g), respectively[16]. The cooking loss was calculated by the equation (1):

$$\text{Cooking loss}\ (\%) = \frac{m_1 - m_2}{m_1} \times 100 \quad (1)$$

2.7 Color measurement

The surimi gel was cut into 2.5 cm high cylinders and white calibrated before the color was measured using a portable colorimeter. The brightness (L^*), redness (a^*), and yellowness values (b^*) of different samples were obtained[17]. Six replicates were set for each group.

2.8 Measurement of textural properties

The textural properties were determined by the method of Xiong et al.[18]. A texture analyzer (TA.XT Plus, Stable Micro Systems, UK) equipped with a 5 kg load cell was used. The probe type used was a cylindrical P/50 probe. TPA was tested under the following conditions: the trigger force was 5.0 g, each sample was set two compression cycles, the compression level was 40%, and the cycle interval was 5 s.

2.9 Microstructure observation

Microstructural properties were observed using confocal laser scanning microscopy[19] (Leica TCS SP5, Leica Ltd., England). Sections (1 mm thick) were taken from the center of the samples and stained with Nile red ethanol solution (1 mg/mL) protected from light for 10 min, followed by solid green solution (1 mg/mL) for 30 s, and the excess dye was subsequently washed away with distilled water. The excitation wavelengths of Nile red and solid green were 528 and 633 nm, respectively.

2.10 Determination of lipid oxidation

The maximum absorption of malondialdehyde (MDA) and thiobarbituric acid (TBA)-colored matter was observed at a

wavelength of 532 nm. The absorbance value of the colored matter is employed as a measure of the degree of oxidation of the oil. The lipid oxidation was measured according to the method of Song et al.[12] with slight modifications. The wavelength was set at 532 nm. Surimi (5 g) was mixed with 50 mL of 7.5 g/100 mL trichloroacetic acid (TCA) solution, followed by incubation at 50 °C for 30 min with shaking. Next, 5 mL of filtrate was blended with 5 mL of TBA (0.02 mol/L) solution, followed by incubating at 90 °C for 30 min. The absorbance value at 532 nm was measured on the spectrophotometer. MDA content in muscle samples was calculated according to the standard curve. Thiobarbituric reactive substances (TBARS) value was expressed as the mass of MDA equivalents per kilogram of gel (mg/kg).

2.11 Determination of flavor-presenting nucleotide content

Five grams of chopped gel sample was added with 20 mL of pre-cooled perchloric acid (1.20 mol/L) and homogenized for 1 min. The supernatant was collected after a centrifugation at 4 °C and 10 000 r/min for 15 min. The above steps were repeated for three times and the supernatants were combined together. The pH of the supernatant was adjusted to 5.8 with KOH (1.00 mol/L), and the nucleotides were determined by high performance liquid chromatography (Agilent 1260, Agilent Technology Co., Ltd., USA) after filtration through a filter membrane (0.22 μm).

2.12 Determination of free amino acids

The method reported by Zhang et al.[20] was used for the determination of free amino acids. Briefly, 5 g chopped gel samples were weighed and placed in a 50 mL centrifuge tube, 25 mL sulfosalicylic acid (0.14 mol/L) was added, homogenized for 1 min, centrifuged at 4 °C and 10 000 r/min for 15 min, and the supernatant was collected. The supernatant was centrifuged again for 15 min (4 °C, 10 000 × *g*) and the filter membrane (0.22 μm) was filtered and the free amino acids were determined by an automatic amino acid analyzer (S-433D, Sykam, Eresing, Germany).

2.13 Determination of volatile flavor substances

The method reported by Gao et al.[21] was referred to and slightly modified, and the differences in volatile compounds among samples were determined and analyzed by headspace-solid phase microextraction (HS-SPME) coupled with gas chromatography-mass spectrometry (GC-MS). The chopped 2 g sample and 500 μL of 2,4,6-trimethylpyridine (100 mg/L) were added to a 20 mL headspace injection vial. The sample was heated and equilibrated in a solid-phase microextraction device (TQ8040, Shimadzu Enterprise Management Co., Ltd., China) at 60 °C for 15 min, extracted at 50 °C for 30 min and resolved at 250 °C for 4 min with a DVB/PDMS-65 μm extraction head. ADB-WAX capillary column (30 m × 0.25 mm, 0.25 μm) was used and held at 40 °C for 3 min. The temperature was increased to 240 °C at a rate of 9 °C/min for 6 min, and the flow rate of the carrier gas (He) was 1.0 mL/min without splitting. MS conditions: electron ionization source; electron energy of 70 eV; ion source temperature of 230 °C; mass scan range of 30–500 *m*/*z*.

2.14 Determination of protein digestibility

According to the method of Wen et al.[22], 4 g sample was dissolved in 16 mL PBS (10 mmol/L, pH 7.0) and homogenized (10 000 r/min, 30 s) twice under ice bath conditions and the pH of the mixture was adjusted to 2.0 ± 0.1 with HCl (1 mol/L). Then, 4 mL pepsin premix (0.032 g/mL) was added, and the reaction was carried out in a shaker at 37 °C with uniform shaking for 2 h. The pH of the mixture was adjusted to 7.5 ± 0.1 with NaOH (1 mol/L). 4 mL of the mixture was taken out to be the product of the pepsin hydrolysis. The remaining mixture was added with 4 mL of trypsin premix (0.024 g/mL) and placed in a shaker at 37 °C with uniform shaking for 2 h. The enzymatic reaction was terminated at 95 °C for 10 min. 4 mL of the mixture was taken as the two-step hydrolysis product. All the hydrolyzed products were added to 12 mL of anhydrous ethanol respectively, and left to stand in the refrigerator at 4 °C overnight, then centrifuged (10 000 × *g*, 15 min). The protein content (m_3/g) of the pre-digestion and the protein content (m_4/g) of the digested precipitates were determined using the biuret method with three parallels in each group. The protein digestibility was calculated by the following formula:

$$\text{Protein digestibility}\ (\%) = \frac{m_3 - m_4}{m_3} \times 100 \quad (2)$$

2.15 Data analysis

All experiments were replicated at least for three times. The results were showed as mean ± standard deviation. The data were performed by PASW Statistics 18 (SPSS Inc., USA) for one-way analysis of variance. A significance level of $P < 0.05$ was applied to evaluated the differences between treatments using Duncan's multiple comparison test. The plots were generated utilizing the Origin 2024 software (Origin Lab Co., USA).

3 Results and discussion

3.1 Cooking loss

As illustrated in Figure 1, the cooking loss of the boiled and steamed samples ranged from 7.11% to 8.83%. The highest cooking loss was observed in the boiled SO group, which was significantly higher than those in the control and PPE groups ($P < 0.05$). However, no significant difference was observed between the control and PPE groups ($P > 0.05$). The cooking loss of the microwave and fried samples ranged from 12.70% to 17.42% and 25.75% to 29.06%, respectively, with the SO group significantly higher than the control and PPE groups ($P < 0.05$). The addition of sturgeon oil and different addition methods had no significant effect on the cooking loss of surimi gel after steaming ($P > 0.05$). Boiling and steaming caused less cooking loss to the surimi gel, while frying caused the highest cooking loss, followed by microwave.

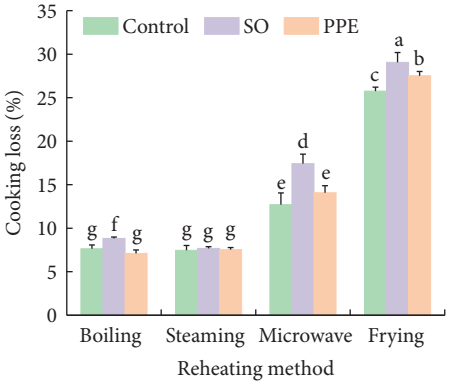

Figure 1 Effects of sturgeon oil addition and reheating methods on the cooking loss of sturgeon surimi gels. Different lowercase letters represent significant differences ($P < 0.05$).

Heating usually resulted in the loss of juices from the surimi gel, which contributed to a reduction in its lightness. The gel samples remained in contact with water throughout the boiling process and were always surrounded by water vapor during steaming. Consequently, even if the juices lost, the samples were still able to absorb some of the water from the environment, thereby reducing cooking loss. The samples were not exposed to water vapor during the process of microwave or frying. The higher temperature resulted in a more pronounced contraction of the protein network, the loss of water due to the decrease in the water holding capacity of the sample, leading to increased cooking loss. Furthermore, the loss of water during frying was accompanied by penetration of the less dense vegetable oil into the sample, which also resulted in higher cooking loss in fried samples.

3.2 Color

The changes in L^*, a^*, and b^* of gel samples from different treatment groups were shown in Table 1. Different ways of fish oil addition and reheating resulted in different chromaticity values of surimi gels. Boiling and steaming conferred higher L^* to that of other samples. Frying increased the a^* and b^* of the samples, with the lowest L^* (59.41–59.77) and the highest b^* (33.94–34.39). In the boiling and steaming groups, the L^* in both the SO and PPE groups was significantly higher than that in the control group ($P < 0.05$), and the b^* in the PPE group was significantly decreased ($P < 0.05$). In the microwave group, the L^* and a^* of the samples in the SO and PPE groups were significantly higher than those in the control group ($P < 0.05$), with the b^* no significant change ($P > 0.05$). Furthermore, there was no clear difference ($P > 0.05$) in the color among the three groups after frying.

Table 1 Effect of sturgeon oil addition and reheating methods on the color of sturgeon mince gels.

Reheating method	Group	L^*	a^*	b^*
Boiling	Control	82.15 ± 0.29^{b}	3.62 ± 0.23^{bc}	9.28 ± 0.25^{c}
	SO	84.16 ± 0.84^{a}	4.05 ± 0.28^{b}	8.60 ± 0.20^{cd}
	PPE	83.82 ± 0.84^{a}	4.00 ± 0.07^{b}	8.05 ± 0.34^{d}
Steaming	Control	82.07 ± 0.39^{b}	3.54 ± 0.05^{bc}	9.50 ± 0.14^{c}
	SO	83.49 ± 0.30^{a}	3.69 ± 0.12^{bc}	8.55 ± 0.16^{cd}
	PPE	83.42 ± 0.11^{a}	3.65 ± 0.12^{bc}	8.19 ± 0.26^{d}
Microwave	Control	75.88 ± 0.17^{d}	3.40 ± 0.10^{d}	11.16 ± 0.98^{b}
	SO	76.88 ± 0.15^{c}	3.63 ± 0.21^{bc}	10.89 ± 0.81^{b}
	PPE	77.45 ± 0.57^{c}	3.69 ± 0.11^{bc}	10.86 ± 0.76^{b}
Frying	Control	59.55 ± 0.78^{e}	11.78 ± 0.32^{a}	33.94 ± 0.56^{a}
	SO	59.41 ± 1.00^{e}	11.74 ± 0.55^{a}	34.09 ± 0.72^{a}
	PPE	59.77 ± 0.32^{e}	11.64 ± 0.63^{a}	34.39 ± 0.72^{a}

Note: In the same column, different lowercase letters represent significant differences ($P < 0.05$).

The color of a food was closely related to its composition. On the one hand, the addition of oil enhanced the light scattering effect on the gel surface of surimi and contributed to the brightness of the gel surface[23]. On the other hand, the higher the moisture content in surimi gels, the higher the reflectance of their surfaces, resulting in higher L^*[10]. As a result, the gels of the groups with fish oil (SO and PPE groups) showed higher L^* and an overall bright white color after boiling and steaming compared to those without fish oil. During frying, the surface of the gels formed a golden-yellow shell due to the occurrence of the Maillard reaction, which led to a sharp increase in the a^* and b^* ($P < 0.05$).

3.3 Textural properties

As shown in Table 2, different ways of fish oil addition and reheating affected the texture of surimi gels. In the boiling and steaming group, the values of all texture indexes of the samples in the SO group were significantly lower than those of the control samples ($P < 0.05$), and the cohesion and springiness in the PPE group were not significantly different from those of the control group ($P > 0.05$). In the microwave and frying groups, the hardness in the SO group was lower, while the elasticity and cohesion were higher comparable to those of the control. The PPE group showed a higher degree of similarity in textural indexes with the control, and only cohesion and gumminess decreased after frying. Among the four reheating methods, the hardness, gumminess and chewiness of the samples in the boiling and steaming groups were lower and the elasticity was higher, while the hardness in the microwave group was the highest, with (4.69 ± 0.05) kg of the control group, (4.55 ± 0.06) kg of the SO group and (4.70 ± 0.02) kg of the PPE group, respectively.

Different ways of adding oil and different ways of reheating produce samples with very different textures, facing different audiences and consumer markets. The direct addition of lipids might hinder the cross-linking between matrix proteins and weakened the textural properties of gels[2–3]. As a result, all samples in the SO group had the lowest hardness, but their softer texture may be more acceptable to the elderly and children. The four reheating methods resulted in different effects on the textural properties of the gel samples due to differences in temperature, heat transfer rate, heat transfer medium and maturation mechanism[10]. In this experiment, the hardness of SO group was negatively correlated with the cooking loss under different reheating methods, which demonstrated that there was a close relationship between the hardness and the water content. During the boiling and steaming process, the food was always in contact with water, and had a

Table 2 Effect of sturgeon oil addition and reheating methods on gel texture of sturgeon surimi gels.

Reheating method	Group	Hardness (kg)	Cohesion	Resilience	Gumminess (kg)	Chewiness (kg)
Boiling	Control	4.22 ± 0.04^{e}	69.48 ± 0.47^{a}	90.91 ±0.77^{a}	2.93 ± 0.01^{c}	2.67 ± 0.03^{bcd}
	SO	3.79 ± 0.06^{h}	67.56 ± 0.41^{cd}	89.42 ± 0.98^{bc}	2.56 ± 0.04^{e}	2.29 ± 0.05^{f}
	PPE	4.09 ± 0.04^{g}	69.12 ± 0.67^{ab}	89.99 ± 1.01^{ab}	2.83 ± 0.05^{d}	2.54 ± 0.04^{e}
Steaming	Control	4.23 ± 0.04^{e}	69.02 ± 0.87^{ab}	90.57 ± 1.04^{ab}	2.92 ± 0.04^{c}	2.64 ± 0.04^{bcd}
	SO	3.83 ± 0.08^{h}	67.60 ± 0.39^{cd}	88.54 ± 0.77^{c}	2.59 ± 0.06^{e}	2.29 ± 0.07^{f}
	PPE	4.13 ± 0.03^{f}	69.40 ± 0.39^{ab}	90.68 ± 1.29^{ab}	2.87 ± 0.03^{cd}	2.60 ± 0.03^{de}
Microwave	Control	4.69 ± 0.05^{a}	67.20 ± 0.58^{d}	85.15 ± 0.43^{e}	3.15 ± 0.00^{a}	2.68 ± 0.01^{abc}
	SO	4.55 ± 0.06^{b}	66.75 ± 0.47^{d}	86.03 ± 0.68^{de}	3.02 ± 0.03^{b}	2.60 ± 0.04^{de}
	PPE	4.70 ± 0.02^{a}	67.72 ± 0.80^{cd}	86.05 ± 0.57^{de}	3.19 ± 0.03^{a}	2.74 ± 0.04^{a}
Frying	Control	4.53 ± 0.03^{c}	69.28 ± 0.70^{ab}	86.44 ± 0.50^{de}	3.14 ± 0.05^{a}	2.71 ± 0.03^{ab}
	SO	4.40 ± 0.03^{d}	68.34 ± 1.09^{bc}	86.84 ± 0.41^{d}	3.01 ± 0.07^{b}	2.61 ± 0.05^{cde}
	PPE	4.50 ± 0.05^{c}	67.70 ± 0.56^{cd}	86.83 ± 0.7^{d}	3.04 ± 0.05^{b}	2.64 ± 0.06^{bcd}

Note: In the same column, different lowercase letters represent significant differences ($P < 0.05$).

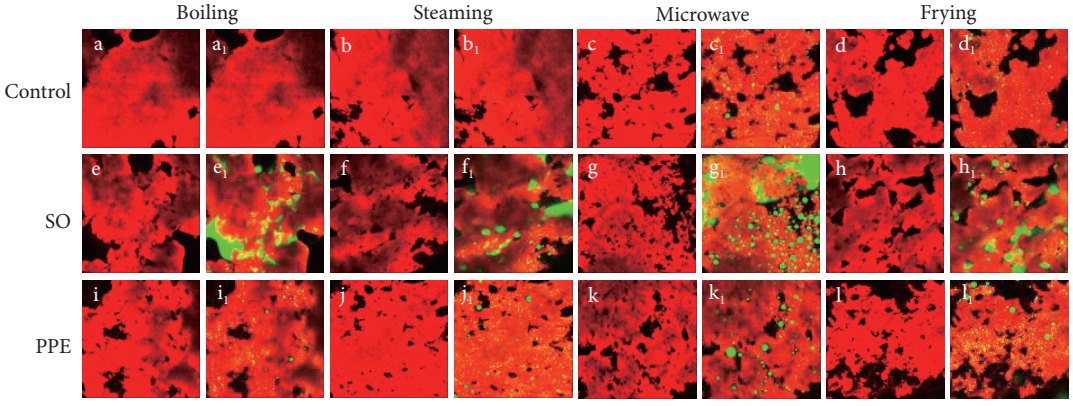

Figure 2 Effects of sturgeon oil addition and reheating methods on the microstructure of sturgeon surimi gels. (a–l) Protein network structure. (a_1–l_1) Gel complex structure. The protein network and oil droplets were indicated in red and green color, respectively.

higher moisture content and therefore a softer texture. It also tends to be the consumer market of the elderly or children. Microwave and frying exposed the samples to intense heat, leading to faster and greater water loss and shrinkage of the internal structure, resulting in a dryer and harder texture with less elasticity. This may be more acceptable to younger consumers or those who prefer hard food.

3.4 Microstructure observation

As shown in Figure 2, the control group exhibited the most complete protein network structure in the boiling and steaming groups. The SO group showed a substantial area of unevenly distributed oil phase, whereas the PPE group samples displayed smaller sized oil droplets distributed within the gel matrix. Additionally, the network structure of the samples was also more complete than that of the SO group. The protein networks of the microwave and frying samples exhibited increased numbers of pores. In contrast, the distribution of oil droplets in the PPE group remained relatively uniform and stable. These findings indicated that boiling and steaming maintained the integrity of the internal structure of the gel to a greater extent. However, the addition of fish oil resulted in a deterioration of the gel network structure. Conversely, the addition of fish oil in the form of Pickering emulsion reduced the degradation of the protein network structure and enhanced the stability of the oil droplets.

The direct addition of elevated levels of fats and oils to a protein mixture may result in incomplete emulsification by the matrix protein, leading to the retention of these substances in a free state. This was associated with an increased susceptibility to migration and aggregation when the samples were subjected to heating[24]. It can be surmised that the oils in the SO group may be in a free state or in the form of large particles, forming irregular and continuous oil-phase regions. This not only caused damage to the protein network structure, but also provided additional channels for the loss of water and non-aqueous substances to a certain extent, which in turn led to an increased cooking loss[25]. In contrast, the oils and fats using Pickering emulsions as the carriers resulted in a reduction in oil droplet sizer and the formation of uniformly and stably embedded oil droplets within the gel matrix[17]. The results of the present experiment demonstrated that fats and oils added in the form of emulsion could still maintain high stability in the gel matrix after reheating treatments. Furthermore, the network structure of the PPE group was more complete than that of the SO group, although it was still not as well-preserved as that of the control group. In the four reheating methods, boiling and steaming were more soothing, and the network structure could remain relatively intact because the samples were cooked and their internal microstructures have been formed. Additionally, Chen et al.[26] observed that boiling and steaming exerted minimal influence on the microstructure of tilapia meat. The application of microwave and frying techniques resulted in the samples being subjected to intense heat and high loss of water, which led to the shrinkage and dry cracking of the protein network, and the appearance of a greater number of densely distributed pores. This was a significant factor contributing to the observed increase in hardness (Table 2). In the control samples after microwave and frying, the more pronounced oil droplets were also observed, which might be attributed to the migration and aggregation of the oil present in the sturgeon surimi.

3.5 Lipid oxidation analysis

As shown in Figure 3, the process of reheating resulted in a range of degrees of lipid oxidation in all samples. The addition of fish oil resulted in a notable elevation ($P < 0.05$) in the TBARS value, while no discernible discrepancy ($P > 0.05$) was observed between the SO and PPE groups. The highest degree of lipid oxidation was observed in the samples subjected to frying, followed by microwave. Similar results were also observed by Luo et al.[27], who reported that in the case of frying treatment, more unsaturated fatty acids were adsorbed by the gel samples during frying, and the unsaturated fatty acids were prone to oxidation, leading to a higher fat oxidation degree in frying samples versus the other samples.

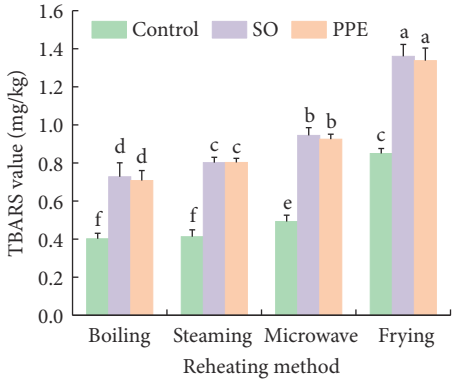

Figure 3 Effects of sturgeon oil addition and reheating methods on lipid oxidation of sturgeon surimi gels. Different lowercase letters represent significant differences ($P < 0.05$).

MDA is one of the end products of the oxidation reaction that occurs in unsaturated fatty acids. The degree of lipid oxidation was reflected in the increase of TBARS value[28], which served as a measure of MDA concentration. The TBARS value in the SO and PPE groups were significantly higher ($P < 0.05$) than those in the control group, indicating that these oils had undergone extensive oxidation and produced a considerable quantity of MDA. The degree of lipid oxidation in the microwave group was significantly higher than that of the cooking and steaming groups ($P < 0.05$), which may be attributed to the dielectric heating effect of microwaves. The penetration performance of microwave is strong, the heating and heat transfer efficiency is high, and microwave energy will promote the generation of single-linear oxygen[29]. The reaction rate of single-linear oxygen was faster than that of normal oxygen when the lipid oxidation triggered, resulting in a higher degree of lipid oxidation. During the frying process, the lipids were oxidized by the exogenous lipids absorbed by them (frying oil), resulting in the highest degree of lipid oxidation in the samples.

3.6 Analysis of flavored nucleotide content

Table 3 revealed the presence of three distinct types of nucleotides in all samples: inosinic acid (IMP), adenosinic acid (AMP), and guanosinic acid (GMP), with GMP exhibiting the highest concentration. In comparison to the control group, a decline in the concentration of each nucleotide type was observed in SO and PPE groups. Furthermore, the total amount of nucleotides in the PPE group was significantly higher than that of the SO group regardless of the reheating method ($P < 0.05$). The contents of the three nucleotides in the boiling, steaming and microwave groups remained consistent, with GMP being the most abundant, followed by AMP and IMP. The frying treatment exhibited a different pattern, with GMP being the most abundant, followed by IMP and AMP. Overall, the total amount of nucleotides in the microwave and steaming was higher, with the total amount of nucleotides in the microwave-treated samples of the control group reaching as high as (130.69 ± 0.05) mg/100 g. This was higher than the total amount of nucleotides in the SO and PPE group.

Flavor-presenting nucleotides are one of the important nonvolatile flavor substances in surimi gels. AMP can impart a sweet taste to foods, GMP imparts a fresh taste to foods, and IMP enhances food flavor through interactions with other nucleotides and amino acids. The present study indicated that GMP was the main contributor to the fresh flavor of sturgeon surimi gel. Given that nucleotides were soluble in hot water, it was speculated that a considerable quantity of nucleotides might be lost with water during boiling and microwave, resulting in a lower total amount of nucleotides[30]. In contrast, the steaming and microwave samples exhibited the higher retention of nucleotides. Zhu et al.[31] similarly found that steaming resulted in the retention of a greater quantity of flavor-presenting substances in mussels, which attributed to the high heat transfer efficiency and low water solubility loss in comparison to boiling and frying. Taste activity value (TAV) is defined as the ratio between the concentration of a flavor presenting substance in a sample and its threshold. When TAV is greater than 1, it means that the flavor presenting substance contributes significantly to the sample taste. As shown in Table 3, the IMP and GMP thresholds were established at 25 and 12.5 mg/100 g, respectively. Although the GMP content in the samples of the SO and PPE groups exhibited a notable decline ($P < 0.05$), TAV was still greater than 1, indicating that it continued to exert a predominant influence on the taste of surimi gel. Both IMP and AMP have a TAV of less than 1, yet they can function synergistically with amino acids and also contribute to the flavour of surimi gels.

3.7 Free amino acids content analysis

Figure 4A illustrates the impact of sturgeon oil supplementation and reheating methods on the amino acid profile of sturgeon surimi gel. The color intensity scale, ranging from blue to red, indicates an increase in surimi gel quantity during the reheating process. As shown in Figure 4B, the total free amino acid content subjected to different reheating treatments revealed that frying resulted in the highest concentration, followed by microwave, steaming, and boiling. The total free amino acid content of the steaming, microwave, and frying samples ranged from 700 to 1 000 mg/100 g, with the control after frying exhibiting the highest total free amino acid content of (970.58 ± 6.83) mg/100 g. Among the four reheating treatments, the total free amino acid content was higher than that of the control in the SO group and the PPE group only after boiling, with the values of (558.33 ± 7.92) and (573.28 ± 7.33) mg/100 g, respectively. Figure 4C illustrated the total percentage of two fresh amino acids (aspartic acid and glutamic acid) and four sweet amino acids (threonine, serine, glycine, and alanine) in the gels. The steamed samples had the

Table 3 Effects of sturgeon oil addition and reheating methods on flavor-presenting nucleotides of sturgeon surimi gels (mg/100 g).

Reheating method	Group	IMP	AMP	GMP	Total content
Boiling	Control	19.56 ± 0.03^{f}	20.36 ± 0.00^{f}	74.94 ± 0.04^{e}	114.86 ± 0.06^{e}
	SO	18.91 ± 0.03^{i}	20.18 ± 0.01^{h}	65.61 ± 0.23^{i}	104.71 ± 0.26^{j}
	PPE	19.14 ± 0.04^{g}	20.15 ± 0.00^{i}	67.65 ± 0.06^{h}	106.93 ± 0.05^{i}
Steaming	Control	20.15 ± 0.23^{d}	20.65 ± 0.03^{c}	80.48 ± 0.10^{b}	121.29 ± 0.10^{b}
	SO	19.12 ± 0.05^{gh}	20.10 ± 0.01^{j}	69.23 ± 0.03^{g}	108.44 ± 0.06^{h}
	PPE	19.67 ± 0.05^{hi}	20.23 ± 0.00^{g}	68.63 ± 0.02^{gh}	108.52 ± 0.03^{g}
Microwave	Control	20.62 ± 0.02^{c}	21.43 ± 0.02^{a}	88.63 ± 0.04^{a}	130.69 ± 0.05^{a}
	SO	19.66 ± 0.01^{f}	20.84 ± 0.00^{b}	76.67 ± 0.02^{d}	117.16 ± 0.01^{d}
	PPE	19.80 ± 0.02^{e}	20.82 ± 0.01^{b}	77.62 ± 0.03^{c}	118.24 ± 0.03^{c}
Frying	Control	21.05 ± 0.02^{b}	20.83 ± 0.02^{b}	72.86 ± 0.04^{f}	114.74 ± 0.03^{e}
	SO	21.36 ± 0.02^{a}	20.50 ± 0.00^{e}	66.13 ± 0.04^{i}	107.99 ± 0.05^{h}
	PPE	20.72 ± 0.01^{c}	20.53 ± 0.01^{d}	68.44 ± 0.03^{gh}	109.70 ± 0.05^{f}

Note: In the same column, different lowercase letters represent significant differences ($P < 0.05$).

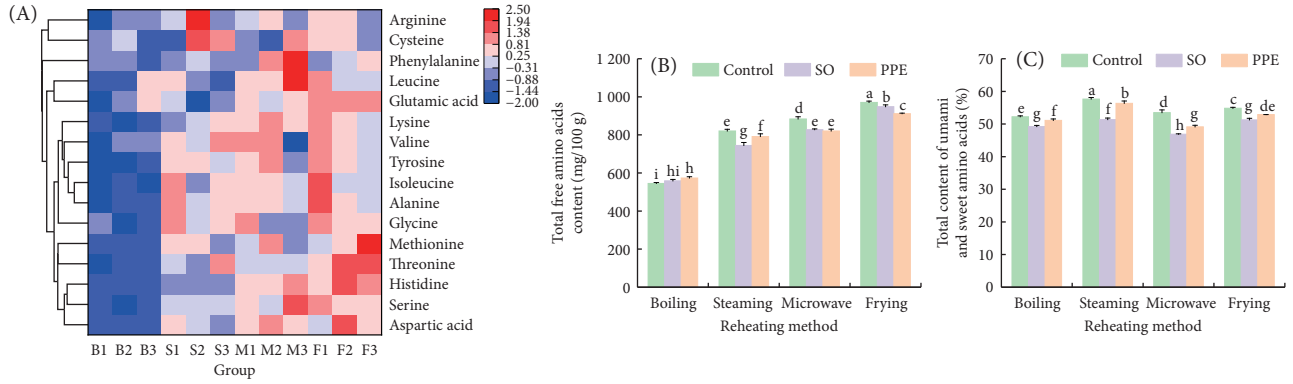

Figure 4 Effects of sturgeon oil addition and reheating methods on free amino acid content of sturgeon surimi gels. (A) Heat map of free amino acids; B: boiling; S: steam; M: microwave; F: frying; 1: control group; 2: SO group; 3: PPE group. (B) Total free amino acids content. (C) Total content of umami and sweet amino acids. Different lowercase letters represent significant differences ($P < 0.05$).

highest total percentage of six amino acids, with values of (57.67 ± 0.45)%, (51.34 ± 0.50)%, and (56.34 ± 0.70)%, respectively. Conversely, the lowest total percentage was (46.86 ± 0.12)% in the SO group after microwave.

The alteration of free amino acids in meat products is linked to the degradation of proteins, the Maillard reaction, loss of juices and their own breakdown[32]. It can be hypothesized that the samples were always in contact with water during the cooking process, which may have resulted in a significant loss of amino acids. Microwave and frying may have resulted in a greater degree of protein degradation as evidenced by the elevated amino acid content, which might be attributed to their higher thermal effect. Furthermore, frying might resulte in an elevated amino acid content in the fish[33]. In addition, the oxidation of pig backfat could promote protein degradation and elevate the concentration of free amino acids[34]. It was speculated that the higher amino acid content in the fried samples in this experiment may also be associated with oxidative protein degradation. The lower amino acid content observed in the SO and PPE groups may be attributed to two factors. Firstly, the higher cooking loss resulted in a significant loss of amino acids in the water. Secondly, amino acids might be related to the oxidative degradation of sturgeon[30]. It was postulated that amino acids may react with carbonyl compounds generated from the oxidation of sturgeon oil in a carbonyl-ammonia reaction, thereby generating more complex and varied flavor substances and conferring a superior odor profile. These findings indicated that the addition of sturgeon oil and different reheating methods altered the content and ratio of amino acids in the surimi gels, imparting distinctive flavor profiles. It might be posited that the steamed samples would be more favorably received by consumers, given that they contained the highest percentage of fresh and sweet amino acids.

3.8 Volatile flavor substances analysis

The heat map (Figure 5A) cluster analysis was capable of discerning the impact of sturgeon oil supplementation on volatile flavor variations in surimi gel samples subjected to disparate reheating techniques. The primary volatile flavor substances identified in the samples were alcohols, aldehydes, hydrocarbons, esters, ketones, acids, and others, which collectively contributed to the distinctive odor of the surimi products. The addition of sturgeon oil significantly increased the content of volatile flavors in the samples, particularly aldehydes and hydrocarbons. The steaming group demonstrated a high content of volatile flavor substances, with 4.84, 5.65, and 6.72 mg/kg of the control, PPE, and SO groups, respectively. In contrast, the frying group had a relatively high percentage of aldehydes, with 32.34%, 37.84%, and 30.99%, respectively.

Sturgeon oil was rich in unsaturated fatty acids, which were susceptible to oxidative decomposition when heated to produce a series of volatile flavor substance[35]. In addition, the incorporation of sturgeon oil may facilitate the activity of lipoxygenase within the surimi gel, thereby accelerating the oxidative decomposition of unsaturated fatty acids to generate a series of fatty acid hydroperoxides. These fatty acid hydroperoxides would undergo further cleavage to form a number of small molecule compounds, which were conducive to the formation of a richer, more volatile flavor substance[12]. The inherent network structure of surimi gel can be exploited as an effective carrier of flavor compounds, thereby stabilizing the flavor substances introduced by sturgeon oil.

Alcohols are important flavor substances in surimi gels. The alcohols detected in high levels in gel samples were *N*-pentanol, *N*-hexanol, *cis*-4-heptenol, 1-octen-3-ol, dodecanol, 1-penten-3-ol, octanol, 1-tridecanol and (*E*)-2-octen-1-ol. Among them, 1-octen-3-ol had a mushroom and earthy odor, which was mainly formed by the degradation of linoleic acid hydroperoxides, and was one of the main fishy components in freshwater fish[36]. The content of 1-octen-3-ol in the samples from the addition groups (SO and PPE) was found to be significantly higher than that in the control, with values ranging from 181 to 299 μg/kg. The primary source of hydrocarbons was the homolytic cleavage of fatty acid alkane oxidation radicals, which had a high threshold and might not contribute directly to the flavor but the overall flavor of the fish[10]. The increased hydrocarbons amounts in the oil-added group mainly included styrene, limonene, isobutylene, stilbene, basking soda, and 2,6,10-trimethyltridecane. Aldehydes were mainly derived from the oxidation and degradation of fatty acids, which contributed significantly to the flavor of surimi gels due to their low threshold. The main aldehydes detected in the samples were octanal, hexanal, nonanal, (*E*)-2-octenal and (*E*,*Z*)-2,6-nonadienal. Following treatment with a microwave or frying, the samples from the oil-added group exhibited a markedly elevated aldehyde content, which might be attributed to a heightened degree of lipid oxidation. Ketones were produced primarily by the degradation of amino acids or oxidation of unsaturated fatty acids and possessed a fruity and aromatic flavor[37]. The ketones detected in the samples were mainly 2,5-octanedione, 2-methyl-3-ketone, 3-hydroxy-2-butanone, and 7-octyl-2-oxoheptanone, with reheated accounting for a relatively low percentage of the samples. Furans were typically generated through the Maillard reaction and imparted a savory, caramelized quality to the flavor profile[9]. Higher concentrations of

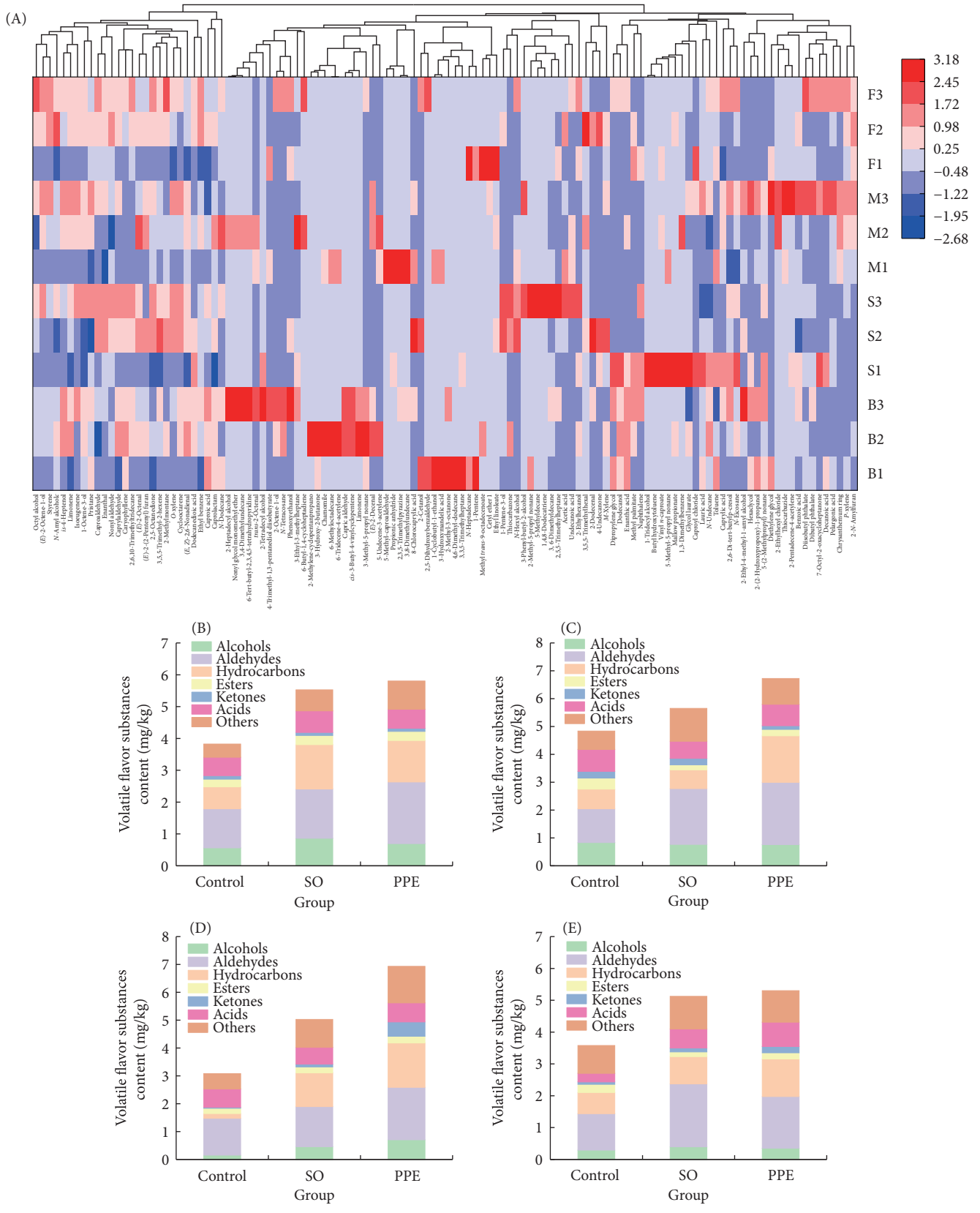

Figure 5 Effects of sturgeon oil addition and reheating methods on the volatile flavor substance content of surimi gels. (A) Thermogram. B: boiling; S: steaming; M: microwave; F: frying; 1: control group; 2: SO group; 3: PPE group. (B–E) Total volatile flavor substances, samples reheated by (B) boiling, (C) steaming, (D) microwave, and (E) frying, respectively.

2-pentylfuran were also detected in microwave and frying samples, suggesting that microwave and frying conferred a more distinctive flavor to the surimi gels. The reason for the higher total content of volatile flavors in the PPE group compared to that in the SO group could be that the Pickering emulsion was evenly distributed in the gel matrix. This might have facilitated the activation of flavor-related enzymes within the gels, including lipoxygenase, peroxidase, lyase, and others[12]. It can therefore be concluded that the addition of sturgeon oil in the form of Pickering emulsion was a more effective method of enhancing the flavor of surimi gels after reheating than the direct addition of sturgeon oil. A sensory evaluation of surimi gels may be conducted in the future to gain a more comprehensive understanding of how alterations in these flavor substances influence the overall taste and aroma of the product.

3.9 Protein digestibility analysis

As illustrated in Figure 6, the gastric digestibility of proteins in the SO and PPE groups after boiling and steaming ranged from 68.60% to 76.33%. The gastrointestinal digestibility of these samples ranged from 80.69% to 85.05%, representing significantly higher rates than those observed in the control group ($P < 0.05$). Furthermore, no statistically significant difference was observed between the SO and PPE groups ($P > 0.05$). There was no significant difference ($P > 0.05$) between the digestibility of the samples of after microwave, and frying decreased the digestibility of the SO and PPE groups. Among the four reheating methods, the digestibility of the boiled and steamed samples was relatively higher, while that of the fried samples was the lowest.

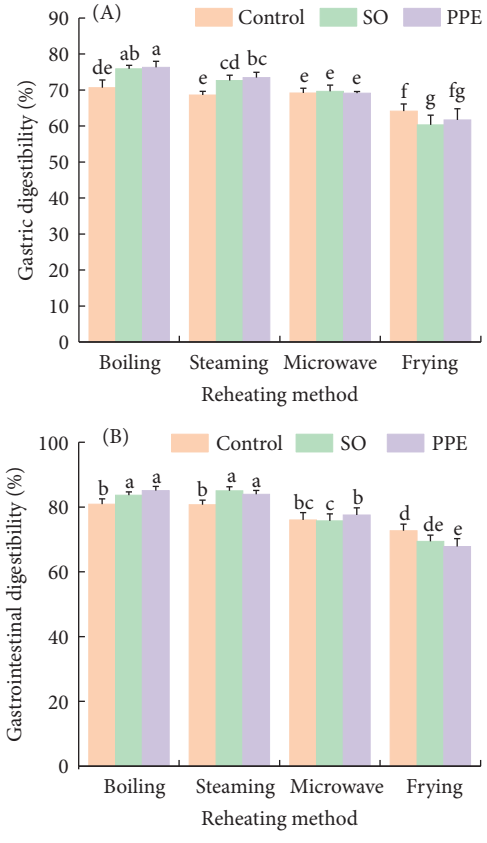

Figure 6 Effect of sturgeon oil addition and reheating methods on *in vitro* simulated digestibility of surimi gels. (A) Gastric digestibility; (B) gastrointestinal digestibility. Different lowercase letters represent significant differences ($P < 0.05$).

The higher the digestibility of a protein, the greater the proportion absorbed and utilized by the body. As the digestibility of surimi gel increases, it was more readily degraded into small peptides, thereby enhancing its nutritional value[38]. The oxidation products of lipids have been demonstrated to reduce protein digestibility by inducing oxidation and aggregation of proteins, which in turn led to a reduction in the accessibility of hydrolysis sites for digestive enzymes[39]. The addition of oil may promote the lipid oxidation of surimi gels, and to some extent may reduce the digestibility of proteins. The findings of this study demonstrated that the addition of sturgeon oil resulted in an increase in lipid oxidation across all samples. However, it did not lead to a reduction in protein digestibility in all samples. Both the boiled and steamed groups showed higher digestibility, which may be related to the lower hardness and gel strength of the samples in both groups[40]. Microwave and frying may have resulted in a reduction in protein digestibility, due to the generation of elevated levels of lipid and protein oxidation, increased hardness, and a more pronounced Maillard reaction[41]. It can be concluded that the essential amino acid composition of surimi products remains unaltered regardless of the reheating method employed[38]. However, the selection of boiling and steaming may prove a more efficacious means of utilizing and absorbing the nutrients inherent to surimi products.

4 Conclusion

The addition of sturgeon oil (direct addition and Pickering emulsion) resulted in an enhancement of the brightness and protein digestibility of the samples following reheating (boiling, steaming, microwave, frying), as well as the development of more complex volatile flavor profiles. Compared to the way of direct addition of fish oil, the way of adding Pickering emulsion proved to be a more effective method for maintaining the textural integrity of the samples after reheating, and the production of higher levels of flavor-presenting nucleotides and volatile flavor compounds. Among the four reheating methods, steaming was the most effective one, as evidenced by the samples' bright white in color, relatively intact network structure, low cooking loss and lipid oxidation, high total percentage of fresh and sweet amino acids, richness in volatile flavor compounds, and strong protein digestibility. In conclusion, the market demand for high-quality surimi products could be met by incorporating sturgeon oil (direct addition and Pickering emulsion). Furthermore, a variety of reheating methods (boiling, steaming, microwave, and frying) could be employed to create new surimi products, thereby catering to different consumer groups.

Author contributions

Yuanxiu Liu: Conceptualization, formal analysis, methodology, and writing-original draft. **Xiaomo Guo:** Methodology, investigation, and data curation. **Tong Shi:** Methodology, software, and writing-review & editing. **Zhiyu Xiong:** Validation and supervision. **Li Yuan:** Visualization and resources. **Ruichang Gao:** Funding acquisition, project administration, conceptualization, writing-review & editing, and supervision.

Conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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