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Effect of radio frequency combined with nisin on the physicochemical properties and volatile compounds of stir-fried sliced pork with *Agaricus bisporus*

Mengqi Huang,†[a] Guoqiang Zhang,†[b] Wuyue Li[c] and Jicheng Xu *[a]

This study aimed to comprehensively compare the effects of radio frequency combined with nisin (RF-nisin) and conventional high-pressure steam (HPS) sterilization on multiple aspects of stir-fried sliced pork with *Agaricus bisporus* (*A. bisporus*), a popular Chinese dish. The microbial survival, flavor, and physicochemical properties were systematically investigated. The flavor-related volatile compounds were analyzed by gas chromatography-ion mobility spectrometry (GC-IMS), the microstructure was observed *via* scanning electron microscope, and the temperature distribution during treatment was monitored using an infrared thermal imager. The results indicated that RF-nisin pasteurization outperformed HPS in terms of flavor retention, physicochemical properties, and taste parameters. Specifically, after 10 minutes treatment, the relative content of heptaldehyde decreased by 21.3% and 31.3% for RF-nisin and HPS respectively, while 5 minutes RF-nisin pasteurization had minimal impact on aldehydes. Both treatments damaged the cell wall and tissue structure of the samples, but RF-nisin treatment showed a more uniform temperature distribution throughout the sample, eliminating the local overheating "corner effect". This research demonstrates that 10 minutes RF-nisin treatment is a mild and effective pasteurization method, which has the potential to significantly enhance the quality of stir-fried sliced pork with *A. bisporus*, providing a new approach for the preservation and quality improvement of traditional Chinese cuisine.

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Sustainability spotlight

Every year, it is estimated that one-third of the total food output, which is equivalent to 1.3 billion tons of food, rots in the trash cans of consumers and retailers, or deteriorates due to improper transportation and harvest. Sustainable consumption and production aim at "reducing consumption, increasing consumption and improving quality", that is, while improving the quality of life, it can increase the net welfare income of economic activities by reducing resource consumption and environmental pollution throughout the life cycle. This work acts on foods with short shelf life through high technology, reducing quality damage and promoting efficient use of resources and energy. This process needs the participation of many parties, including enterprises, consumers, researchers and development cooperation institutions.

1 Introduction

Stir-fried sliced pork with *Agaricus bisporus* is a traditional Chinese dish, which is popular because of its fragrant and delicious taste. Stir-fried sliced pork with *A. bisporus* is rich in nutritional value (rich in protein, vitamins, minerals, *etc.*), and has the effects of regulating the spleen and stomach, promoting muscle growth, maintaining heart health and enhancing immunity.[1] At present, there are two main types of fried meat with mushrooms on the market.[2] A canned form, after HPS treatment, has a long shelf life but has many negative effects, such as: reducing nutritional value and sensory quality; the other is vacuum packaging, cold chain transportation and storage, which have potential safety hazards and waste. Therefore, a mild pasteurization technology is needed to prolong the shelf life on the premise of preserving the flavor and nutrition of food.

Radio frequency (RF) is a novel heat treatment and non-heat treatment technology, which has electromagnetic waves of 10–300 MHz. Compared with microwave food processing, RF heating has better uniformity, and corners are not easy to dry or even burn.[3,4] In addition, RF has strong penetration, and can pasteurize eggs or packaged products with shells to extend the shelf life of food.[5] In the study, it was investigated that although

[a]*College of Biological and Food Engineering, Anhui Polytechnic University, 241000, Wuhu, Anhui, China. E-mail: xujicheng@ahpu.edu.cn; Fax: +86 13855303015; Tel: +86 13855303015*
[b]*Wuhu Green Food Industry Research Institute, 241000, Wuhu, Anhui, China*
[c]*College of Life Sciences, Shanxi University, 237016, Taiyuan, Shanxi, China*
† Co-first authors.

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RF can effectively kill bacteria, it can not inactivate high-temperature-resistant spores, resulting in food spoilage.[6]

In order to ensure the safety of dishes, we consider adding an efficient antibacterial agent-nisin when RF processing food. At present, nisin has been widely used in dairy products, meat products, beverages and other food fields to extend the shelf life of food and ensure food safety. Among many antimicrobial peptides, nisin has strong inhibitory and bactericidal effects on *Salmonella* and *Listeria*, and nisin can destroy the cell membrane of bacteria, leading to the outflow of small molecules, resulting in the lysis of cell spores.[7] Compared with other commonly used antibacterial agents, nisin has unique advantages, which not only can stably exert antibacterial effect in high temperature environment, but also has high safety, can be decomposed by protease in human body, does not remain in human body like some chemically synthesized antibacterial agents, and has no potential harm to human health, which makes it popular in the field of food preservation.[8] Theoretically, it is feasible to extend the shelf life of stir-fried sliced pork with *A. bisporus* by combining nisin and RF with high temperature resistance.

RF is a mild pasteurization technology, nisin can kill heat-resistant spores, and the combination of them can theoretically be used to extend the shelf life of stir-fried sliced pork with *A. bisporus*. Therefore, the main purpose of this study is to develop a sterilization method for processing stir-fried sliced pork with *A. bisporus* by RF-nisin and evaluate the effect of RF-nisin on the quality and volatile flavor changes of stir-fried sliced pork with *A. bisporus* during shelf life.

2 Materials and methods

2.1. Materials

Fresh lean pork, *A. bisporus* and green peppers are all purchased from the local central vegetable market of Jinghu New Town, Jiujiang District, Wuhu City. Soybean oil, salt, soy sauce and other condiments are all purchased from the local Runsheng Supermarket in Jiujiang District, Wuhu City.

2.2. Preparation of stir-fried sliced pork with *A. bisporus* dishes

Clean the chilled pork and *A. bisporus*, cut them into 5 mm slices, and cut the green pepper into 2 cm dices. Boil the water in the pot and add *A. bisporus* for 2 min, then take it out for later use. Then stir-fry the pork with soybean oil for 6 min in a pan, and stir-fry it with *A. bisporus* and green pepper for 3 min. Each piece of stir-fried sliced pork with *A. bisporus* consists of 60 g of *A. bisporus*, 20 g of green pepper and 20 g of pork slices. After the stir-fried sliced pork with *A. bisporus* are cooled to room temperature, 0.25 g kg^{-1} nisin dispersion is added, evenly mixed, vacuum packed (100 g per bag), and then the subsequent sterilization treatment is carried out.

2.3. Stir-fried sliced pork with *A. bisporus* sterilization treatment

The packaged samples of stir-fried sliced pork with *A. bisporus* were put into a radio frequency instrument, and the distance between the plates of the radio frequency instrument was set to 12.5 cm, and the prepared samples were sterilized for 5 min, 10 min and 15 min respectively, and were named RFN 5, RFN 10 and RFN 15. Non-sterilized samples and samples treated by high pressure steam sterilization (HPS, 121 °C, 30 min) were used as controls.

2.4. Determination of sterilization temperature uniformity

An infrared thermal imaging camera is used to measure the surface temperature of the sample. Samples were taken out at 5 min, 10 min and 15 min after RF-nisin sterilization, and the surface temperature was measured immediately. The measured sample is located at 50 cm from the lens of the imaging camera. The whole measurement process is completed in 3 s.

2.5. Determination of total bactericidal colonies

Weigh 10 g samples of 5 g *A. bisporus* and 5 g meat slices, and dilute them with normal saline at 1 : 10. According to the national standard (GB 4789.2-2022), the total number of microorganisms was slightly modified. 25 g samples were put into 225 mL physiological saline, homogenized in a homogenizer for 75 s, and diluted ten times continuously at this ratio. 1 mL samples of each sample diluent were evenly coated on a plate counting agar in triplicate, and finally put upside down in a constant temperature incubator at 37 °C for 48 h, and the total number of selected colonies was 30–300. Expressed as logarithm of colony forming unit (log CFU g^{-1}) g^{-1}.

2.6. Sensory evaluation of sterilized stir-fried sliced pork with *A. bisporus* slices

Sensory evaluation was conducted on a 9-point scale, and the higher the score, the more the sensory evaluator liked the sample (5 points was the lowest acceptable limit).[9] Appearance, smell, taste, texture and overall acceptability were analyzed by a sensory evaluation team composed of 20 laboratory teachers and graduate students. Reheat stir-fried sliced pork with *A. bisporus* in 60 °C water bath for 5 min before sensory evaluation. Before each sensory evaluation, all sensory evaluators are required to rinse their mouths with pure water for 30 s and thoroughly clean their mouths to avoid mutual influence between the two samples. The sensory evaluation standard of stir-fried sliced pork with *A. bisporus* is shown in the Table 1.

2.7. Determination of color difference of stir-fried sliced pork with *A. bisporus* slices after sterilization

After correcting the black-and-white plate with a color difference meter, select a smooth surface and aim the standard light source vertically at the tested sample. The results were expressed by L^* (brightness/darkness), a^* (redness/greenness) and b^* (yellowness/blueness), and each sample was repeated three times.[10] In addition, the calculation formula for total chromatic aberration ΔE is as follows:

$$\Delta E = \sqrt{\left(L^* - L_0^*\right)^2 + \left(a^* - a_0^*\right)^2 + \left(b^* - b_0^*\right)^2} \quad (1)$$

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Table 1 Sensory evaluation standard of stir-fried sliced pork with *A. bisporus*

Evaluation projects	Evaluation criterion	Score
Appearance	Pork, mushrooms and green peppers are bright in color	7–9
	Pork, mushrooms and green peppers are slightly darker in color	4–6
	Colors of pork, mushrooms and green peppers	1–3
Smell	It has a pleasant aroma that should be present in a mushroom stir-fry	7–9
	Moderate aroma, no odor	4–6
	Unscented, odorless	1–3
Taste	It has the proper flavor of mushroom stir-fry with a long-lasting aftertaste in the mouth	7–9
	Medium flavor, medium aftertaste in the mouth	4–6
	Poor taste and aftertaste in the mouth	1–3
Texture	Good chewiness, firm meat with no loose ends, excellent side dish texture	7–9
	Moderately chewy, slightly loose meat, slightly soft texture of side dishes	4–6
	Poor chewiness, loose and soft meat, side dish texture	1–3

2.8. Determination of texture of sterilized stir-fried sliced pork with *A. bisporus* slices

Texture profile analysis (Tane Plus, Baoshan Industrial Development Co., Ltd, Shanghai, China) was adopted by texture analyzer. *A. bisporus*: the pre-test speed was 5.0 mm s^{-1}, the test speed was 1.0 mm s^{-1}, the return speed was 5.0 mm s^{-1}, the strain compression ratio is 50%, the secondary compression interval was 5.0 s, and the trigger force was 0.05 N. Sliced pork: the speed and trigger force before, during and after the test were set to 3.00, 1.00, 5.00 mm s^{-1} and 0.05 N respectively. The firmness of sample was the maximum force obtained from the force *vs.* time graphs (Newtons). Each sample was compressed twice.[11]

2.9. Determination of total soluble solids in stir-fried sliced pork with *A. bisporus* after sterilization

Before measurement, the digital Abbe refractometer is calibrated according to the current room temperature, and then 2–3 drops of grinding liquid made of 3 g samples are added to the sample cell to read the indicator.[12]

2.10. Determination of protein content in sterilized stir-fried sliced pork with *A. bisporus*

The protein content of stir-fried sliced pork with *A. bisporus* was determined by *Coomassie* brilliant blue method. Take 400 μL sample soup, add 1600 μL distilled water and 10 mL *Coomassie* brilliant blue solution, mix well, and let stand at room temperature for 2 min. Subsequently, the absorbance was measured at the wavelength of 595 nm, and the protein content was calculated according to the standard curve of bovine serum albumin.[13] For quantification of protein, bovine albumin serum was taken as a standard (0.1 mg mL^{-1}).

2.11. Determination of microstructure of sterilized stir-fried sliced pork with *A. bisporus*

The sample was sliced and placed in a vacuum freeze-drying oven for 48 h. The samples cut into 5 × 5 mm are evenly spread on the scanning electron microscope sample table with carbon conductive adhesive, and after checking that there is no flying powder, they are sprayed with gold. Finally, the microstructure of *A. bisporus* was found under field emission scanning electron microscope. Observed at an accelerating voltage of 3 kV, a working distance of 10 mm and a magnification of 500–800 times.[14]

Table 2 Analysis conditions of GC-IMS systems

FlavorSpec®	
Analysis time	15 min
Column type	MXT-5 (0.53 mm)
Chromatographic column temperature	70 °C
Carrier gas flow	10 mL min^{-1}
Drift gas flow	75 mL min^{-1}
Carrier gas/drift gas	N_2
IMS temperature	45 °C
Syringe temperature	85 °C
Automatic headspace sampling unit	
Sample volume	500 μL
Incubation time	20 min
Culture temperature	70 °C

2.12. Determination of volatile flavor of stir-fried sliced pork with *A. bisporus* after sterilization

Add 0.5 g *A. bisporus*, 0.5 g sliced meat and 1 g soup to the empty bottle in turn. Non-shunt mode was adopted in the experiment, and the injection needle was purged with high-purity nitrogen for 15 minutes. Detailed parameters are shown in the Table 2.[15,16]

2.13. Data processing and analysis

In the experiment, each item of data was conducted in biological triplicates. The experimental data were processed by Excel 2021 software (Microsoft, Washington, America), and these results were expressed as the average plus or minus standard error of three data. SPSS software (California, America) was used for significant difference analyses and correlation analyses, with $P < 0.05$ indicating significant difference. Origin 2021 software (Northampton, America) was used for image processing of the data.

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3 Results and discussion

3.1. Effect of RF-nisin on the surface temperature of stir-fried sliced pork with *A. bisporus*

The temperature uniformity during the processing of stir-fried sliced pork with *A. bisporus* is shown in Fig. 1. When the radio frequency processing time (RT) was from 5 min to 15 min, the temperature of the sample increased with the increase of heating time, and the surface average center temperature of the mushroom fried meat quickly rised to 74.3 °C, and then kept stable at 73.0 °C. From the temperature distribution inside the material, the temperature gradually decreases from the center to the edge. In addition, RF treatment reduces the corner effect during heating, and the heating is more uniform.[17] Secondly, it can be observed that the effect of RF treatment on high-temperature resistant vacuum retort pouch heating was poor, but the part containing materials was selectively heated. The reason may be that the water content in stir-fried sliced pork with *A. bisporus* was higher than that in vacuum retort pouch, and it had a higher dielectric loss factor, so RF would generate more heat for food materials, which can prevent food packaging and materials from being heated together, causing harmful substances to volatilize and posing a threat to food safety.[18]

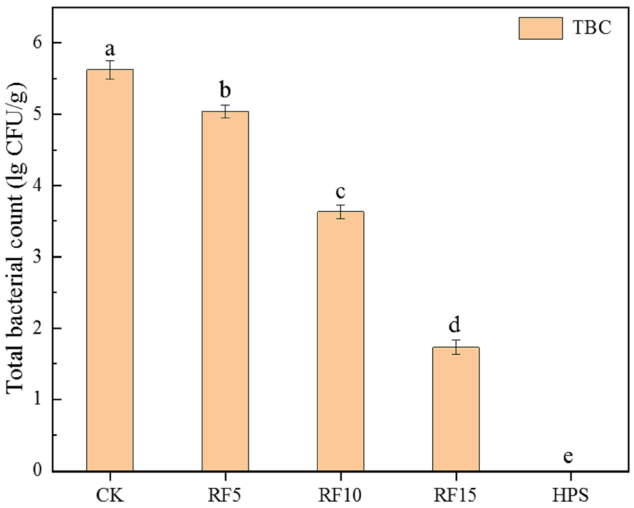

Fig. 2 Impact of various methods on bacterial growth in sliced pork stir-fried with mushrooms. Note: Vertical bars indicate standard error (±SE). Different letters indicate significant differences according to Duncan's test ($p < 0.05$). CK represents without any treatment, RF 5, RF 10, and RF 15 represent that the RF treatment time in the RF-nisin treatment is 5, 10 and 15 min, respectively; HPS represents high pressure steam sterilization 30 min. The same below.

3.2. Effect of RF-nisin on the total colonies of stir-fried sliced pork with *A. bisporus*

The sterilization effect of *A. bisporus* fried meat was improved by adjusting RF treatment time. Fig. 2 shows the effect of RF-nisin treatment on the total number of colonies. In all treatment methods, HPS had the best bactericidal effect, and no bacteria were detected. In the RF-nisin treatment, the antibacterial effect was the worst when RF treatment was carried out for 5 min, and the total number of colonies decreased by 0.58 lg CFU g^{-1}. The longer the RF working time, the better the effect of killing bacteria would be. When the RF treatment lasts for 10 min, it met the requirements of Chinese national standard GB 29921-2021 for the total number of colonies. This shows that the effect of RF pasteurization is related to RF processing time. Xu *et al.*,[19] reported similar results. By prolonging the RF treatment time, the sterilization effect of stir-fried sliced pork with *A. bisporus* was improved.

3.3. Effect of RF-nisin on sensory evaluation of stir-fried sliced pork with *A. bisporus*

Sensory evaluation is one of the important means to evaluate consumer acceptance.[20] The sensory evaluation (a) and general evaluation (b) of stir-fried sliced pork with *A. bisporus* after RF-nisin and HPS treatment are shown in the Fig. 3. After different sterilization treatments, the smell, texture, appearance, taste and acceptability of stir-fried sliced pork with *A. bisporus* were reduced to varying degrees. RF treatment for 5 min had little effect on the texture but the texture change was the most obvious after HPS treatment. For consumers, stir-fried sliced pork with *A. bisporus* dishes should have basic palatability, compact texture and rich taste, so as to stimulate consumers' taste buds and appetite.[21] In this study, with the increase of RF-nisin treatment time, the total score of sensory evaluation was lower than that of the CK group, which was 36 points, 31.7 points and 28.7 points respectively,

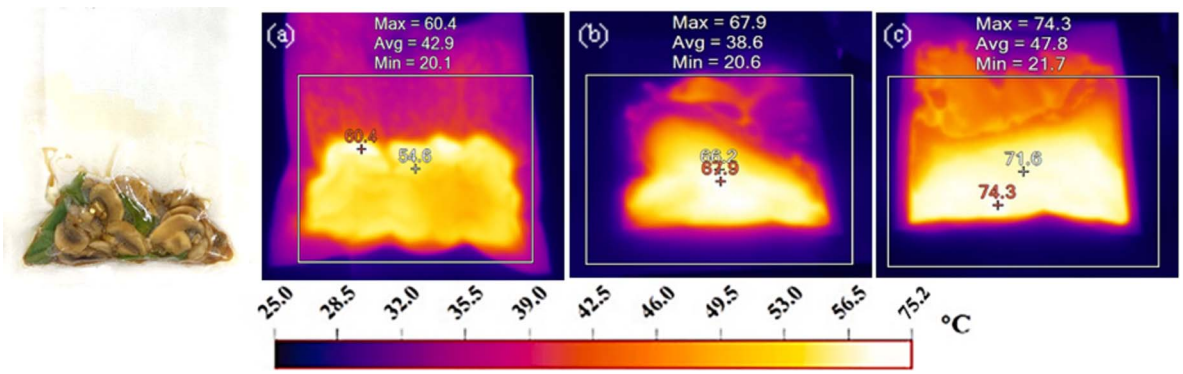

Fig. 1 Changes of surface temperature distribution of stir-fried sliced pork with *A. bisporus* with heating time of RF combined with nisin treatments. Note: (a–c) Represent RF-nisin treated stir-fried sliced pork with mushrooms (RFN 5, RFN 10, and RF 15 respectively).

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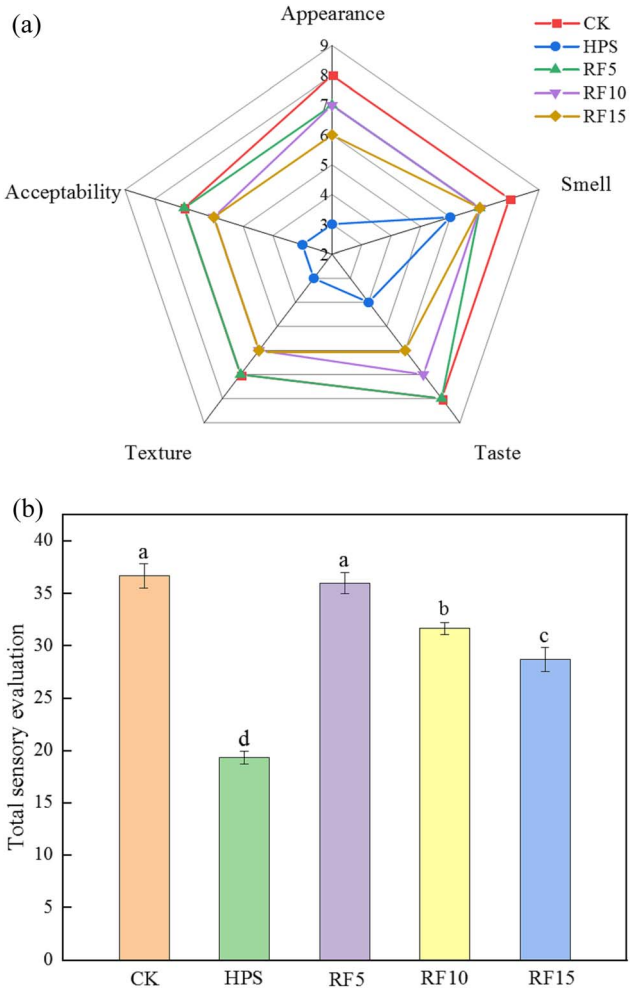

Fig. 3 Impact of various methods on sensory quality (a) and total sensory evaluation (b) of stir-fried sliced pork with mushrooms.

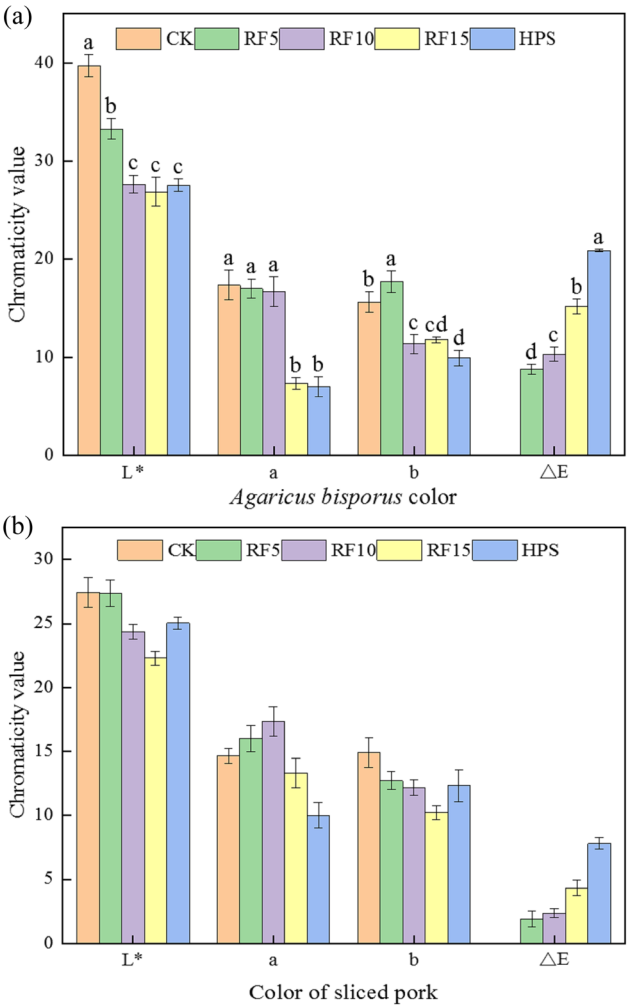

Fig. 4 Impact of various methods on color difference of stir-fried sliced pork with *A. bisporus*. (a) *A. bisporus*, (b) pork slices.

while that of the HPS group was only 19.3 points, indicating that HPS treatment affected the sensory quality of products.

3.4. Effect of RF-nisin on the color difference and texture of stir-fried sliced pork with *A. bisporus*

In order to study the effect of RF-nisin on the color of stir-fried sliced pork with *A. bisporus*, the color of *A. bisporus* and lean meat slices were determined respectively, as shown in Table 3 and Fig. 4. The results showed that the L^*, a^*, b^* and ΔE of RF treatment changed significantly, indicating that RF-nisin treatment had an effect on the color of stir-fried sliced pork with *A. bisporus*. The values of L^*, a^* and b^* in *A. bisporus* samples decreased significantly with the extension of RF-nisin treatment time, which may be due to the decrease of enzyme activity and browning of *A. bisporus* after being stewed and then treated by RF.[22] During the processing of meat slices, due to frying and RF heat treatment, the values of L^*, a^* and b^* all decreased significantly with the extension of RF working time. In other studies, it has also been found that similar heat treatment leads to this phenomenon in goose meat,[23] which may be due to the denaturation of myoglobin and

Table 3 Impact of various methods on color difference of stir-fried sliced pork with *A. bisporus*

Different samples	*A. bisporus* color				Color of sliced pork			
	L^*	a^*	b^*	ΔE	L^*	a^*	b^*	ΔE
CK	39.7 ± 1.11^{a}	17.33 ± 1.53^{a}	15.6 ± 1.05^{b}	—	27.43 ± 1.17^{a}	14.67 ± 0.57^{bc}	14.93 ± 1.15^{a}	—
RF 5	33.27 ± 1.07^{b}	17 ± 1^{a}	17.73 ± 1.10^{a}	8.77 ± 0.49^{d}	27.37 ± 1.01^{a}	17.33 ± 1.15^{a}	12.73 ± 0.72^{b}	1.9 ± 0.62^{c}
RF 10	27.63 ± 0.92^{c}	16.67 ± 1.53^{a}	11.77 ± 0.31^{c}	10.3 ± 0.7^{c}	25.03 ± 0.47^{b}	16 ± 1^{ab}	12.33 ± 1.26^{b}	2.33 ± 0.35^{c}
RF 15	27.53 ± 0.64^{c}	7.33 ± 0.58^{b}	11.33 ± 1.00^{cd}	15.2 ± 0.75^{b}	24.37 ± 0.55^{b}	13.33 ± 1.15^{c}	12.17 ± 0.60^{b}	4.3 ± 0.61^{b}
HPS	26.87 ± 1.46^{c}	7 ± 1^{b}	9.9 ± 0.8^{d}	20.87 ± 0.12^{a}	22.3 ± 0.56^{c}	10 ± 1^{d}	10.23 ± 0.55^{c}	7.8 ± 0.46^{a}

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Table 4 Impact of various methods on hardness, TSS content and protein content of stir-fried sliced pork with *A. bisporus*

Different samples	Hardness of *Agaricus bisporus* (N)	Hardness of sliced meat (N)	Total soluble solids (%)	Protein content (mg mL^{-1})
CK	31.61 ± 1.32^{a}	53.09 ± 1.12^{a}	13.13 ± 0.80^{a}	11.65 ± 1.14^{a}
RF 5	26.55 ± 0.91^{b}	45.66 ± 0.58^{b}	12.30 ± 0.95^{ab}	11.62 ± 0.63^{a}
RF 10	23.53 ± 0.97^{c}	30.88 ± 0.66^{c}	11.80 ± 0.92^{ab}	10.79 ± 1.46^{ab}
RF 15	23.06 ± 0.99^{cd}	27.22 ± 0.36^{d}	11.20 ± 0.70^{b}	9.87 ± 0.56^{ab}
HPS	21.46 ± 0.93^{d}	26.26 ± 0.34^{d}	10.73 ± 0.55^{b}	9.41 ± 1.08^{b}

other proteins, and the light reflection caused by denatured opalescence scattering increases and the brightness decreases, resulting in the decrease of redness. With the increase of RT, ΔE is greater than 1.5, indicating that there is a significant difference between the RF treated samples and the CK group.[24]

Texture is the most direct way for consumers to evaluate the quality of fresh-cut fruits and vegetables.[25] As shown in Table 4, it was not difficult to find that the texture of the sample decreased significantly with the increase of RT, and RF treatment for 5 min had little effect on the texture. Among them, the effect of RF on the texture of pork slices was higher than that of *A. bisporus*, which may be because the cell wall structure of lean meat loses its support after heating. Secondly, from the two treatment methods of RF and HPS, the effect of RF treatment for 15 min on the texture of pork slices was lower than that of HPS, which showed that HPS treatment was not conducive to the maintenance of the three-dimensional network structure and moisture in protein, resulting in the loss of chewing performance of pork.[26] The above results showed that with the extension of radio frequency treatment time, the color and texture of samples change, which is mainly due to the changes of enzyme activity, protein denaturation and tissue structure destruction caused by heating.

3.5. Effect of RF-nisin on total soluble solids of stir-fried sliced pork with *A. bisporus*

In order to judge the nutritional value of stir-fried sliced pork with *A. bisporus*, the advantages of the quality of stir-fried sliced pork with *A. bisporus* after different treatments were measured by the content of soluble solids, an important nutrient in food.[27] However, long-term high temperature treatment would cause the decomposition of soluble solids, which would lead to the decrease of their edibility. As can be shown from Table 3, with the increase of RT, the content of soluble solids gradually decreased, and when RT reached 15 min, the total soluble solids (TSS) content was 11.2%. Obviously, HPS treatment has the greatest influence on soluble solids. According to the research results of Cai *et al.*,[28] the change of TSS was related to water loss, which is consistent with the findings of this study.

3.6. Effect of RF-nisin on protein content in stir-fried sliced pork with *A. bisporus*

Pork protein in stir-fried sliced pork with *A. bisporus* played an important role in human health, which can maintain muscle growth, supplement essential amino acids and control hunger. The abundant protein in pork was deeply loved by people.[29] Table 4 shows the effects of different treatment methods on the content

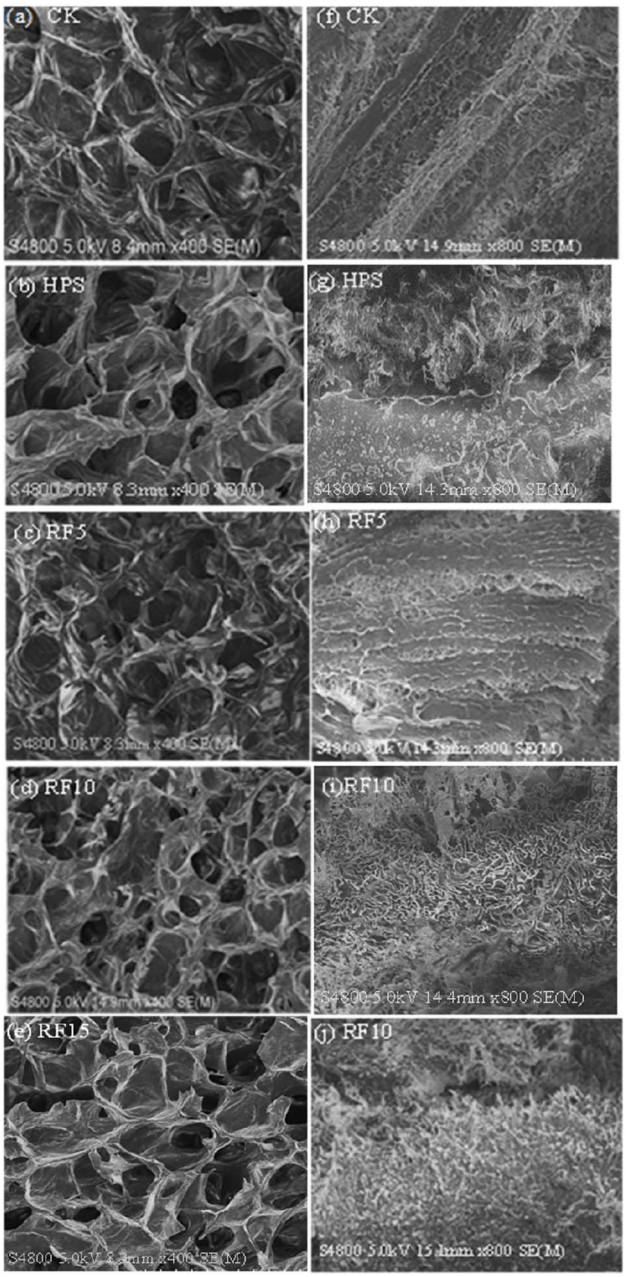

Fig. 5 Impact of various methods on microstructure of stir-fried sliced pork with mushrooms (*A. bisporus* 400 times; pork slices 800 times). Note: (a–j) represent untreated samples, samples sterilized by high-pressure steam and samples treated by RF-Nisin for 5 minutes, 10 minutes and 15 minutes respectively.

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of protein in stir-fried sliced pork with *A. bisporus*. Compared with the CK group, the protein content in pork slices can be retained to the maximum extent when RF treatment last for 5 min and 15 min, which were 11.62 mg mL^{-1} and 10.79 mg mL^{-1} respectively. When RT was 15 min or HPS treatment, the content of protein in pork decreased significantly, which may be due to the denaturation and hydrolysis of protein in pork caused by high temperature, thus leading to the loss of protein in pork.[30] Therefore, HPS treatment of stir-fried sliced pork with *A. bisporus* not only affected its texture, taste and flavor, but also reduced its nutritional value.

3.7. Effect of RF-nisin on microstructure of stir-fried sliced pork with *A. bisporus*

In order to judge the surface microstructure changes of stir-fried sliced pork with *A. bisporus* under different treatment methods, *A. bisporus* and pork were observed under different times of SEM (*A. bisporus* 400 times, pork slices 800 times). According to the Fig. 5, it was found that the surface structure of *A. bisporus* and pork changed obviously with the increase of treatment time under the same conditions of RF-nisin treatment. In Fig. 5(a–e), it can be found that the cell wall structure of *A. bisporus* has changed from original structural integrity to

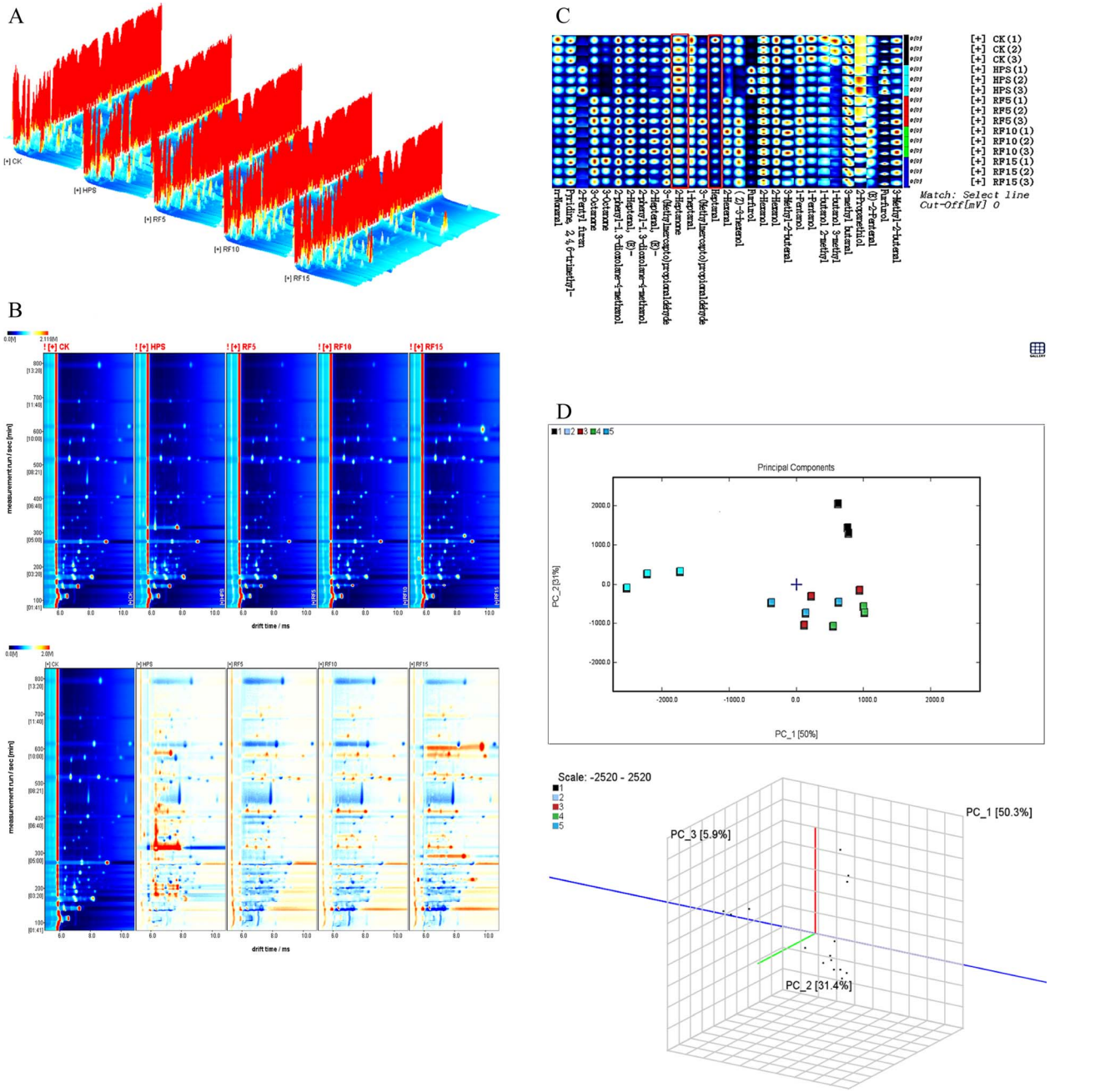

Fig. 6 (A) 3D Figure (B) topographic map (C) finger-print map (D) PCA diagram of VOCs in stir-fried sliced pork with mushrooms by different processing methods.

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deformation and rupture, and cavitation appears; Fig. 5(f–j) shows that the muscle fiber of pork has changed from compact and orderly, with bright bands and dark bands independent obviously, to loose structure, increased gap, disappearance of bright and dark bands, and even rupture,[31] which also explained why meat products after long-term heating treatment by HPS lose toughness. Therefore, with the increase of RF-nisin treatment time, the structure of stir-fried sliced pork with *A. bisporus* will change, and when RF-nisin treatment was 5 min, the structure had little effect. After RF-nisin treatment for 15 min, the structure was observed to be destroyed. Among them, HPS was the most seriously damaged, *A. bisporus* showed large-scale cavitation and cell wall deformation and distortion, and pork muscle fibers were broken and loose and swollen.

3.8. Effect of RF-nisin on volatile flavor of stir-fried sliced pork with *A. bisporus*

In order to study the effects of RF-nisin and HPS treatments on stir-fried sliced pork with *A. bisporus*, the changes of volatile organic compounds (VOCs) components under different treatments were studied by GC-IMS. The Fig. 6A shows the peak intensity of VOCs in different treatment methods. For each point, its position in the 3D image was determined by drift time, retention time and peak intensity, which reflected the characteristics of detected VOCs. It can be clearly seen from the Fig. 6B that the stir-fried sliced pork with *A. bisporus* can release more VOCs after HPS treatment, which was similar to the research results of Li *et al.*[32] High temperature treatment would lead to further lipid oxidation of pork and produce more VOCs. The distribution of VOCs in the stir-fried sliced pork with *A. bisporus* treated by RF-nisin is similar to that of CK, but the peak intensity was different, that was, the concentration of VOCs was different. In order to further evaluate the differences in VOCs in the stir-fried sliced pork with *A. bisporus* treated by RF-nisin and HPS, the ion migration time and retention time of gas chromatography can be mapped to the horizontal and vertical coordinates of the chart by using the difference comparison model for further data analysis and comparison. As shown in the Fig. 6B, the topographic map of the CK group was taken as the reference group, and the CK group was subtracted by RF-nisin and HPS treatment, respectively. If the VOCs decreased, the background after subtraction was blue, while the red color represents that the concentration of the substance was higher than that of the CK group.[33] The results showed that there were more red and blue in HPS group within the retention time range of 300–350 s, which

Table 5 Peak height of twenty-nine typical volatile compounds identified from stir-fried sliced pork with *A. bisporus* with different treatments[a]

Count	Compound	Peak height				
		CK	RF 5	RF 10	RF 15	HPS
1	*n*-Nonanal	0.59 ± 0.06^{a}	0.37 ± 0.08^{bc}	0.30 ± 0.04^{c}	0.44 ± 0.11^{b}	0.42 ± 0.06^{bc}
2	2,4,6-Trimethyl-pyridine	0.74 ± 0.01^{a}	0.71 ± 0.11^{a}	0.75 ± 0.07^{a}	0.75 ± 0.06^{a}	0.57 ± 0.05^{b}
3	2-Pentyl furan	0.16 ± 0.01^{b}	0.23 ± 0.02^{b}	0.20 ± 0.01^{b}	0.20 ± 0.03^{b}	0.69 ± 0.14^{a}
4	3-Octanone-M	0.78 ± 0.05^{b}	1.07 ± 0.04^{a}	0.98 ± 0.05^{a}	1.05 ± 0.10^{a}	0.39 ± 0.06^{c}
5	3-Octanone-D	0.24 ± 0.05^{bc}	0.58 ± 0.13^{a}	0.45 ± 0.08^{ab}	0.58 ± 0.17^{a}	0.06 ± 0.01^{c}
6	2-Phenyl-1.3-dioxolane-4-methanol	1.56 ± 0.01^{a}	1.52 ± 0.03^{ab}	1.52 ± 0.02^{ab}	1.47 ± 0.10^{ab}	1.42 ± 0.03^{b}
7	2-Heptenal, (*E*)-M	1.09 ± 0.04^{b}	1.17 ± 0.04^{ab}	1.28 ± 0.01^{a}	1.04 ± 0.14^{b}	0.61 ± 0.13^{c}
8	2-Phenyl-1.3-dioxolane-4-methanol	1.32 ± 0.08^{a}	1.25 ± 0.26^{a}	1.17 ± 0.12^{a}	1.29 ± 0.11^{a}	1.05 ± 0.11^{a}
9	2-Heptenal, (*E*)-D	0.57 ± 0.04^{b}	0.66 ± 0.08^{b}	0.87 ± 0.05^{a}	0.51 ± 0.17^{b}	0.22 ± 0.06^{c}
10	3-(Methylmercapto)propionaldehyde-M	1.18 ± 0.23^{bc}	1.31 ± 0.11^{ab}	1.44 ± 0.02^{a}	1.32 ± 0.01^{ab}	1.01 ± 0.10^{c}
11	2-Heptanone	0.67 ± 0.03^{ab}	0.49 ± 0.09^{c}	0.58 ± 0.05^{bc}	0.56 ± 0.02^{c}	0.73 ± 0.03^{a}
12	1-Heptanal	1.01 ± 0.04^{a}	0.82 ± 0.10^{b}	0.80 ± 0.07^{b}	0.82 ± 0.03^{b}	0.85 ± 0.06^{b}
13	3-(Methylmercapto)propionaldehyde-D	0.45 ± 0.20^{bc}	0.57 ± 0.17^{ab}	0.75 ± 0.05^{a}	0.57 ± 0.02^{bc}	0.3 ± 0.07^{c}
14	Heptanal	0.49 ± 0.07^{a}	0.25 ± 0.09^{b}	0.23 ± 0.05^{b}	0.24 ± 0.03^{b}	0.33 ± 0.07^{b}
15	2-Hexenal	0.51 ± 0.01^{b}	0.57 ± 0.07^{b}	0.70 ± 0.05^{a}	0.45 ± 0.10^{b}	0.21 ± 0.03^{c}
16	(*Z*)-3-Hexenol	0.50 ± 0.03^{ab}	0.47 ± 0.07^{b}	0.56 ± 0.04^{a}	0.49 ± 0.05^{ab}	0.11 ± 0.01^{c}
17	Furfurol	0.39 ± 0.05^{c}	0.73 ± 0.16^{b}	0.76 ± 0.05^{b}	0.89 ± 0.07^{b}	3.04 ± 0.15^{a}
18	2-Hexanol-M	1.29 ± 0.02^{a}	1.28 ± 0.01^{a}	1.30 ± 0.02^{a}	1.28 ± 0.03^{a}	1.16 ± 0.04^{b}
19	2-Hexanol-D	2.69 ± 0.03^{a}	2.32 ± 0.15^{b}	2.37 ± 0.07^{b}	2.26 ± 0.12^{b}	2.23 ± 0.11^{b}
20	3-Methyl-2-butenal-D	0.69 ± 0.01^{b}	0.69 ± 0.16^{b}	0.94 ± 0.12^{b}	0.67 ± 0.05^{b}	0.27 ± 0.07^{c}
21	1-Pentanol-M	1.26 ± 0.01^{a}	1.11 ± 0.79^{bc}	1.12 ± 0.04^{b}	1.09 ± 0.04^{bc}	1.0 ± 0.06^{c}
22	1-Pentanol-D	0.45 ± 0.01^{a}	0.28 ± 0.06^{b}	0.27 ± 0.04^{b}	0.25 ± 0.03^{b}	0.27 ± 0.03^{b}
23	1-Butanol 2-methyl	0.44 ± 0.05^{a}	0.35 ± 0.06^{b}	0.33 ± 0.03^{b}	0.30 ± 0.01^{b}	0.34 ± 0.06^{b}
24	1-Butanol 3-methyl	0.45 ± 0.03^{a}	0.17 ± 0.03^{a}	0.19 ± 0.01^{a}	0.19 ± 0.01^{a}	0.16 ± 0.01^{b}
25	3-Methyl butanal	1.44 ± 0.01^{a}	1.43 ± 0.01^{b}	1.45 ± 0.02^{b}	1.44 ± 0.02^{b}	1.34 ± 0.07^{b}
26	2-Propanethio	0.23 ± 0.01^{b}	0.45 ± 0.12^{a}	0.53 ± 0.01^{a}	0.51 ± 0.04^{a}	0.47 ± 0.09^{a}
27	(*E*)-2-Pentenal	0.52 ± 0.04^{bc}	0.61 ± 0.58^{ab}	0.72 ± 0.20^{a}	0.50 ± 0.13^{bc}	0.45 ± 0.04^{c}
28	Furfurol	1.07 ± 0.51^{c}	1.36 ± 0.08^{b}	1.40 ± 0.02^{b}	1.45 ± 0.03^{ab}	1.50 ± 0.39^{a}
29	3-Methyl-2-butenal-M	1.17 ± 0.01^{ab}	1.21 ± 0.08^{ab}	1.34 ± 0.04^{a}	1.14 ± 0.13^{b}	0.73 ± 0.11^{c}

[a] The repeated compounds in the table represent dimers of their monomers. Each value is expressed as the mean ± standard deviation. Means with different lowercase letters within a row indicate significant differences ($p < 0.05$).

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indicated that after HPS treatment, although the concentration of some VOCs increased, the signals of some sensitive compounds also decreased, while RF-nisin treatment did not significantly change the VOCs in the stir-fried sliced pork with *A. bisporus*.

In order to deeply analyze the differences between RF-nisin and HPS processing, all the analyzed signals were generated by GC-IMS plug-in Gallery Plot (Fig. 6C), with each row representing a processing mode, each column representing a VOCs, dots representing the substance concentration, and red indicating a higher concentration.[34] In this experiment, 34 kinds of VOCs were identified by GC-IMS technology, among which 11 kinds of VOCs were not matched with specific substances due to the limitation of the database, and the rest compounds could be divided into 12 aldehydes, 7 alcohols, 2 ketones, 1 furan and 1 aromatic organic compound according to their categories. Secondly, some high concentrations of VOCs can form a variety of signals, which were dimers or trimers. In this study, seven dimers were identified. 2-Heptanone was the product of linoleic acid oxidation. The contents of 2-heptanone and furfural in the samples treated by HPS are higher than those treated by blank and RF-nisin, while the contents of *trans*-2-pentenal glycerol acetal, 3-methyl-2-crotonaldehyde, (*E*)-2-heptene aldehyde, hexenal and 3-octanone were reduced. Both RF-nisin and HPS reduce heptanal. During processing, lipids (mainly unsaturated fatty acids in surface free fats) were oxidized to form volatile carbonyl substances, such as aldehydes and ketones. Therefore, the volatile aldehydes and ketones produced in the samples can be used as indicators of the degree of oil oxidation. The relative content of VOCs can be calculated according to the peak height derived by the system, as shown in the Table 5. Compared with the CK group, HPS, RF 5, RF 10 and RF 15 reduced the content of heptaldehyde by 21.3%, 28.7%, 31.3% and 29.3% respectively, and RF-nisin reduced the content of heptaldehyde more obviously when treated with RF for 10 min. The peak height of 3-octaneone-M in CK group is 0.78 ± 0.05^{b}, and that in RF 10 group is 0.98 ± 0.05^{a}. With the increase of concentration, it has mushroom aroma, which will make the mushroom flavor of fried *A. bisporus* slices more intense.

PCA was an unsupervised pattern recognition method, which could explain the differences between and within experimental samples.[35] Generally speaking, if the contribution rate of PC1 and PC2 exceeds 80%, there was a significant difference. The Fig. 6D shows the PCA results of untreated, RF-nisin and HPS treatments on the VOCs of stir-fried sliced pork with *A. bisporus*, and the cumulative contribution rates of PC1 and PC2, which accounted for 50% and 31% respectively, which indicated that different treatments of RF-nisin and HPS have significant differences on VOCs.

4 Conclusion

Both RF-nisin and HPS could affect the quality, microstructure and VOCs of stir-fried sliced pork with *A. bisporus*. Among them, RF-nisin treatment was to change the treatment time under the premise of a certain RH. The results showed that when it was treated for 10 min, it could meet the sterilization requirements, and had little effect on the quality, microstructure and volatile flavor of stir-fried sliced pork with *A. bisporus*. In addition, RF-nisin treatment for 10 min can maintain soluble solids and protein content. This study would provide a new idea for prolonging the shelf life of dishes.

Data availability

Data supporting the findings of this study are available upon request from the corresponding author.

Author contributions

Mengqi Huang: methodology, investigation, writing – original draft. Guoqiang Zhang: conceptualization. Wuyue Li: supervision. Jicheng Xu: conceptualization, supervision, writing – review and editing, all authors have read and approved the content of the manuscript.

Conflicts of interest

The authors declare no competing interests.

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