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ORIGINAL ARTICLE

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The effect of different pretreatments on the quality of ready-to-eat jellyfish *Rhopilema esculentum* Kishinouye products

Xiuping Dong[1,2] · Xinru Fan[1,2] · Yang Wang[1,2] · Libo Qi[1,2] · Shuang Liang[1,2] · Lei Qin[1,2] · Chenxu Yu[2,3] · Beiwei Zhu[1,2]

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Abstract

The aim of this study was to characterize the effects of different pretreatments on ready-to-eat jellyfish products. Salted jellyfish umbrella edges were used as raw materials. After desalting, samples were blanched under different temperatures for different periods of time. Moisture contents and color characteristics of the pretreated samples were determined, and texture characterized. Masson staining was used to image microstructures, and sensory evaluation was performed. Microbial growth at 37 °C was monitored for samples which underwent the different treatments. Moisture content and whiteness decreased with extended heating, and the higher the temperature, the faster these changes occurred. The moisture content of samples with no heat treatment was 97.89 ± 0.23%, which was nearly 3% higher than that of samples which had undergone heating. The 70 °C heat treatment had a greater effect on the whiteness of jellyfish. Samples exposed to the 60 °C treatment for 1 min had the highest sensory scores, and in conjunction with other results, this temperature was chosen to be the optimal heat treatment. The combination of heating, acid soaking, ultraviolet sterilization and potassium sorbate treatment was shown to prolong the shelf life of jellyfish products effectively.

Keywords Heating · Sensory score · Hurdle technology · Shelf life

Introduction

The jellyfish *Rhopilema esculentum* Kishinouye is a traditional seafood in a number of Asian countries, including China and Japan. It has been reported that in China alone 205,453 t of jellyfish was captured from the wild, and 79,848 t of cultivated jellyfish produced in 2016 (Fishery Bureau of the Ministry of Agriculture of the People's Republic of China 2017 2017). Liaoning Province cultivates the largest amount of jellyfish in China, and is a major trading market for jellyfish products. Jellyfish produced in Liaoning Province has a brittle texture and relatively high content of collagen, hence it is favored by consumers. Jellyfish has a high commercial value, but due to its high moisture content, it is prone to microbial growth, hence the preservation of jellyfish has always been a challenge.

Ready-to-eat jellyfish products are emerging as consumer favorites. European law defines ready-to-eat products as food intended by the producer or the manufacturer for direct human consumption without the need for cooking or other processing to eliminate or reduce to an acceptable level contaminating microorganisms of concern (Atanassova

Chemical compounds studied in this article: acetic acid [PubChem compound identifier (CID) 176], potassium sorbate (PubChem CID 23676745), acid fuchsin (PubChem CID 5464362), Ponceau S (Pubchem CID 6508672), orange G (PubChem CID 9566064), brilliant green (PubChem CID 12449), ethyl alcohol (PubChem CID 702), xylene (PubChem CID 7809).

✉ Beiwei Zhu
zhubeiwei@163.com

Xiuping Dong
dxiuping@163.com

[1] School of Food Science and Technology, Dalian Polytechnic University, Qinggongyuan 1, Ganjingzi District, Dalian 116034, China

[2] National Engineering Research Center of Seafood, Qinggongyuan 1, Ganjingzi District 116034, China

[3] Department of Agricultural and Biosystems Engineering, Iowa State University, Ames, IA 50011, USA

et al. 2014). The growth of microorganisms can affect the acceptance and safety of ready-to-eat products, and lead to the formation of undesirable odors and changes in both quality and structure of food (Costa et al. 2014). Therefore, the number of microorganisms in ready-to-eat jellyfish is a critical control factor, and is of great concern in the quality control of jellyfish products.

Heating is a common method in the processing of aquatic products. Appropriate thermal treatment improves the quality, safety and acceptability of products (Khan et al. 2014a, b), and can produce a better texture by altering the microstructure of the products (Øiseth et al. 2013). Proper thermal treatment also imparts a better color (Schubring 2006), favorable sensory characteristics and a special flavor (Chiou et al. 2004) to the final products. In order to ensure the safety of the products, heating can also be combined with other techniques, such as high pressure processing, organic acid soaking, vacuum packaging, cold storage, or sterilization via irradiation (Moure et al. 2006; Khan et al. 2014b) to inhibit microbial growth.

Conventional preservation technologies such as thermal processing ensure the safety and shelf life of fresh products, but can result in the loss of physicochemical and nutritional qualities. High temperature sterilization inevitably causes losses of valuable nutrients due to heat-induced degradation. In addition, jellyfish are susceptible to heat-induced structural degradation, and they tend to lose their original shape under high temperature sterilization. Therefore, high temperature sterilization is not an optimal method for jellyfish processing. Heat treatment at a relatively low temperature combined with other processing techniques [e.g., acidic soaking, ultraviolet (UV) treatment etc.] may be a good alternative for jellyfish sterilization.

Hurdle technology is used in food manufacturing and preservation throughout the world, and is an important process in the modern food industry. Hurdle technology is based on the regulation of food processing by various effective factors, which interact or show a synergetic effect on the control of growth of contaminating organisms, and are used to improve food quality and safety (Leistner 2000). Synergy of multiple hurdle factors in food processing has been shown by Thippeswamy et al. (2011). Hurdle technology has been widely used in meat processing, such as the production of chicken breast fillets (Rodríguez-Calleja et al. 2012), pork sausages (Thomas et al. 2008), buffalo products (Malik and Sharma 2014), and beef (Tango et al. 2014). It has also been used for aquatic products such as trout *Oncorhynchus mykiss* (Křížek et al. 2012), and for fruit (Abadias et al. 2011; Gómez et al. 2011) and vegetables such as sliced carrots (Koide et al. 2011) and peppers (Horvitz and Cantalejo 2013).

In this study, salted jellyfish from Yingkou, Liaoning Province was investigated as a raw material. The aim of our study was to investigate the effects of different heat treatments on the sensory qualities, structural characteristics, color, and moisture content of desalted jellyfish. Heating, acetic acid and/or potassium sorbate soaking, and UV irradiation were analyzed as hurdle factors to investigate the control of microbial growth during storage. The ultimate objective was to provide a theoretical basis for optimizing the processing and storage of ready-to-eat jellyfish products.

Materials and methods

Materials and sample preparation

Salted jellyfish umbrellas were purchased from Rongfa (Yingkou Liaoning; 1.15 ± 0.1 kg each, diameter 0.51 ± 0.03 m). The umbrellas were divided into different parts according to Iwatani et al. (2010), and the edges were cut into 7.0-cm × 2.0-cm × 1.0-cm pieces as samples for subsequent analysis. The following groups of samples were prepared:

1. Desalted samples, which served as the control.
2. Desalted samples that were then heated in a water bath for 0, 0.5, 1, 3, 5, 10, and 30 min, respectively at 50, 60, and 70 °C, which are temperatures commonly used in jellyfish processing.
3. After heating, samples were soaked in 2 mL/L acetic acid (Zhengqiang, Shanghai) solution for 5 min.
4. After heating and acetic acid soaking, samples were subjected to UV sterilization for 1 h.
5. After heating and acetic acid soaking, samples were subjected to UV sterilization for 1 h in potassium sorbate (Wang Long, Ningbo, Zhejiang) at a concentration of 1.0 g/kg.

All the samples were packaged under atmospheric pressure. All five groups of samples were kept in an incubator at 37 °C for 10 days, and evaluated for microbial growth on days 0, 3, 7, 10.

Sensory evaluation

Fourteen panelists evaluated the sensory attributes of the samples, according to the criteria listed in Table 1, with color, hardness, toughness, flavor, and juiciness as evaluation indexes. The grades of samples are shown in Table 1. The qualities of the samples were evaluated according to three categories (poor 1–4, medium 5–7, and good 8–10), and scores for each sample recorded. The overall sensory score (OSS) was based on the overall evaluation for each sample by each panelist.

Table 1 Criteria used by the sensory panel for jellyfish evaluation

Index	Score		
	10–8	7–5	4–1
Color	Light white, natural gloss	Color turns dark, reduced transparency	Color turns yellow with no gloss
Hardness	Good hardness	Hardness decreased	Tissue turns soft, becomes stretchy
Toughness	Hard to tear	Toughness decreased	Little toughness, easy to tear
Flavor	Natural seafood flavor, no unusual smell	Deteriorated flavor, still acceptable	Flavor unacceptable, a strong ammonia smell
Juiciness	Well hydrated, juicy	With water loss, still acceptable	Significant water loss, dry mouth feel, fibrous

Texture analysis

Heated samples were cooled to room temperature, then cut into pieces of 5.0 cm × 0.70 cm × 0.50 cm. A TA.XT Plus texture analyzer (Stable Micro Systems, Surrey, UK) fitted with a P5 probe was used for the textural analysis. The texture of the jellyfish was characterized by instrumental texture profile analysis for hardness, springiness, chewiness, cohesiveness and resilience. Each sample underwent two cycles of compression analysis with a strain level of 75% (Zhu et al. 2011).

Color measurement

Heated samples were cooled to room temperature, the red coats of the samples removed and the outer umbrella color determined. A white board was used as the background for color determination, and the test mode was set as small-hole reflection (Chiou et al. 2004). An Ultra Scan PRO color photometer (Hunter Lab, USA) was used. The whiteness (W) score is defined as:

$$W = 100 - \sqrt{100 - L^2 + a^2 + b^2}$$

Where L is the lightness (0–100%), a is the color ranging from green (−) to red (+), and b is the color ranging from blue (−) to yellow (+). The whiteness score provides a good measure of the relatively small difference between white and slightly yellowish (Skipnes et al. 2011), and has been used as a parameter in determining surimi quality (Benjakul et al. 2004).

Moisture content determination

Moisture content of each sample was determined by its weight loss when dried at 105 °C until a constant weight was reached (Chiou et al. 2004).

Histological study

Samples were cut into 0.50-cm × 0.50-cm × 0.50-cm cubes, embedded in tissue freezing medium (optimal cutting temperature compound) at − 30 °C. These blocks were sliced into sections of 10-μm thickness using a low-temperature slicing machine (CM1950, Leica, Germany). The sections were then mounted on a glass slide and stained with Masson staining. Light micrographs were taken with an Olympus BX51 optical light microscope (BX51, Olympus, Japan) (Xin et al. 2003).

Microbiological characterization

Aseptically packed samples (10 g) were comminuted and homogenized with 90 mL (0.85% w/v) NaCl solution and incubated at 30 °C for 72 h for the determination of total aerobic count on Plate Count Agar through serial dilutions. Colonies are expressed as log colony-forming units (CFU) per gram (Miguéis et al. 2015).

Statistical analysis

Results are presented as mean values ± SD. Differences were tested for significance by ANOVA to determine any significant effects at $p < 0.05$. Bivariate correlations were tested using SPSS (version 8.0) adapted to a personal computer (Khan et al. 2014b). A principal component analysis (PCA) was performed to study the variations in the multivariate data sets of the textural characterization of the jellyfish samples. Seven variables for 19 jellyfish samples were used for PCA analysis. PCA was done using Unscrambler software version 9.7 (CAMO, Trondheim, Norway) (Liu et al. 2015).

Results

Sensory evaluation

The final results of the sensory evaluation of samples under different heating conditions are shown in Table 2. The most suitable temperature treatment led to an improvement of sample color and texture, and samples that underwent heating at 60 °C for 1 min exhibited the highest OSS

Table 2 Sensory scores of jellyfish samples that underwent different heat treatments

Treatments	Color	Hardness	Toughness	Juiciness	Flavor	OSS
0[a]	6.79 ± 1.01 A,b,c,2,3	6.50 ± 0.85 A,b,c,2	6.64 ± 1.39 A,b,c,d,3,4	7.64 ± 1.00 A,b,3	6.21 ± 1.19 A,a,1	7.00 ± 0.96 A,c,4
50[b]-*t*1[c]	6.71 ± 0.83 A	6.21 ± 0.97 A	6.71 ± 1.44 A	7.00 ± 0.88 A	6.50 ± 0.94 A	7.29 ± 0.99 A
50-*t*2	7.29 ± 0.73 A	6.00 ± 1.11 A	7.29 ± 1.20 A	7.00 ± 0.78 A	6.07 ± 1.14 A	6.93 ± 0.92 A
50-*t*3	6.86 ± 1.23 A	6.00 ± 1.04 A	6.79 ± 1.12 A	7.14 ± 1.10 A	6.00 ± 1.96 A	6.93 ± 0.73 A
50-*t*4	6.43 ± 1.02 A	6.21 ± 0.80 A	6.64 ± 1.60 A	7.07 ± 0.73 A	6.21 ± 1.48 A	7.00 ± 1.03 A
50-*t*5	6.57 ± 1.22 A	6.14 ± 0.86 A	6.79 ± 2.00 A	6.93 ± 1.33 A	6.43 ± 0.76 A	7.00 ± 0.78 A
50-*t*6	6.36 ± 1.55 A	6.00 ± 1.04 A	7.21 ± 1.37 A	6.93 ± 1.44 A	6.14 ± 1.66 A	7.07 ± 0.92 A
60-*t*1	7.14 ± 0.86 c,d	6.93 ± 0.73 c,d	7.21 ± 1.25 d	7.07 ± 1.14 b	6.00 ± 1.1 a	7.43 ± 0.76 c,d
60-*t*2	7.86 ± 0.77 d	7.57 ± 0.85 d	6.93 ± 1.54 c,d	6.93 ± 0.83 b	6.36 ± 0.84 a	7.79 ± 0.70 d
60-*t*3	6.64 ± 1.28 b,c	7.00 ± 0.78 c,d	7.00 ± 0.96 c,d	7.00 ± 0.96 b	6.21 ± 0.89 a	7.29 ± 0.91 c,d
60-*t*4	6.27 ± 1.03 b	6.93 ± 0.96 c,d	6.13 ± 1.19 a,b,c	6.87 ± 0.99 b	6.13 ± 1.06 a	6.93 ± 0.88 c
60-*t*5	4.00 ± 0.78 a	5.86 ± 0.95 b	5.71 ± 1.38 a,b	5.64 ± 1.55 a	6.21 ± 1.19 a	5.79 ± 0.97 b
60-*t*6	3.50 ± 1.61 a	5.14 ± 1.29 a	5.29 ± 1.33 a	5.21 ± 1.12 a	5.93 ± 0.83 a	4.86 ± 0.86 a
70-*t*1	7.07 ± 1.21 3	6.14 ± 0.77 2	7.43 ± 1.34 4	7.00 ± 1.52 3	6.21 ± 1.19 1	6.93 ± 1.21 4
70-*t*2	6.64 ± 1.28 2,3	5.86 ± 0.77 2	6.21 ± 0.89 3	6.00 ± 1.04 2	6.14 ± 1.35 1	5.93 ± 1.14 3
70-*t*3	5.79 ± 1.31 2	4.50 ± 1.02 1	5.64 ± 1.65 2,3	5.79 ± 1.19 2	6.07 ± 1.07 1	5.86 ± 1.17 3
70-*t*4	4.36 ± 1.55 1	4.14 ± 0.95 1	5.14 ± 1.29 1,2	5.71 ± 1.44 2	5.71 ± 1.20 1	4.93 ± 1.00 2
70-*t*5	3.54 ± 1.56 1	3.92 ± 0.86 1	4.69 ± 1.37 1,2	4.46 ± 1.39 1	5.46 ± 1.61 1	4.23 ± 0.73 1,2
70-*t*6	3.79 ± 1.37 1	3.86 ± 0.77 1	4.14 ± 1.29 1	4.36 ± 1.55 1	5.00 ± 0.96 1	4.00 ± 0.78 2

OSS Overall sensory score

Capital letters indicate significant differences for each parameter in samples heated at 50 °C for different periods ($p < 0.05$). *Lowercase letters* indicate significant differences for each parameter in samples heated at 60 °C for different periods ($p < 0.05$). *Different numbers* indicate significant differences for each parameter in samples heated at 70 °C for different periods ($p < 0.05$)

[a]Unheated control jellyfish strip

[b]Temperatures (50, 60, 70 °C)

[c]Heating time (*t*1 0.5 min, *t*2 1 min, *t*3 3 min, *t*4 5 min, *t*5 10 min, *t*6 30 min)

(7.79 ± 0.70). However, the OSS decreased significantly when the heating time was longer than 10 min. The lowest OSS was observed in samples that underwent 70 °C heating for 30 min, suggesting excessive heating has an adverse effect on the sensory qualities of jellyfish, and should be avoided in jellyfish processing. Samples with the highest OSS also had higher scores for hardness and color (Table 2).

The correlation analysis between sensory attributes is shown in Table 3. Each index correlates well with the samples' OSS ($p < 0.01$); the textural indices (hardness and toughness) correlated the best with OSS, followed by color and juiciness scores. Flavor was the least correlated factor with the other attributes, including OSS. Since the flavor of jellyfish is mostly determined by its chemical composition, not by its physical structure, this observation seems is reasonable (Kumazawa et al. 2013; Park et al. 2014). The samples differed mainly in textural quality, color and juiciness, while flavor contributed the least to the OSS of jellyfish. Color, toughness and juiciness scores correlated well with each other, and all of them are related to moisture content (Chiou et al. 2004).

Table 3 Correlations between color, hardness, toughness, juiciness, flavor and OSS of samples that underwent different heat treatments

	Color	Hardness	Toughness	Juiciness	Flavor	OSS
Color		0.688**	0.833**	0.792**	0.466*	0.740**
Hardness			0.658**	0.712**	0.663**	0.893**
Toughness				0.730**	0.497*	0.762**
Juiciness					0.457*	0.740**
Flavor						0.681**
OSS						

* $p < 0.05$, ** $p < 0.01$ (two-tailed)

Principal component analysis

The matrix of normalized mean scores for the attributes of texture (hardness, springiness, cohesiveness and resilience) across different temperatures and times were analyzed by PCA (Fig. 1). The first two principal components explained more than 87% of the total variance within

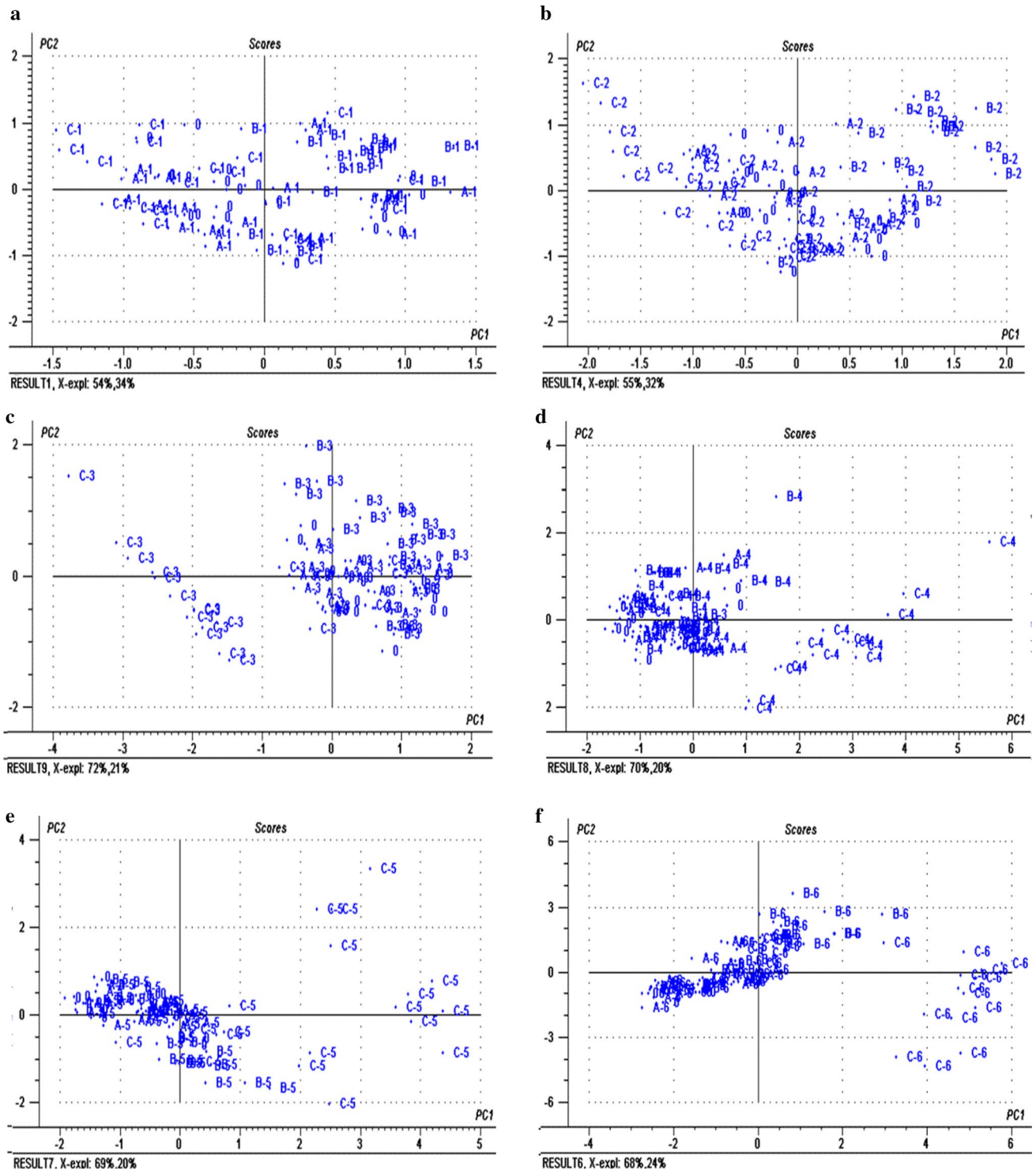

Fig. 1 Principal component analysis of instrumental texture measurements combined for different samples heated for 0.5 min (**a**), 1 min (**b**), 3 min (**c**), 5 min (**d**), 10 min (**e**), 30 min (**f**). *0* Unheated, *A* 50 °C, *B* 60 °C, *C* 70 °C

the data set of all the samples under heating, and were used to characterize the samples.

For the PC1 (54–72%) vs. PC2 (34–20%) plots, at the shortest heating time (i.e., 30 s), regardless of temperature, most data points form one loosely overlapping cluster. As the heating time increased, the group heated at the highest temperature started to emerge as a distinguishable group, and this trend became very clear when the period of heating exceeded 3 min, suggesting that more intensive heating at higher temperature caused more significant difference in samples and enabled clearer differentiation between them. However, it remained difficult to differentiate groups A and B (i.e., heating at 50 and 60 °C, respectively) based on their textural attributes, even when heated for over 30 min.

There are reasons to believe that after heating, the microstructure of jellyfish protein changes, resulting in dehydration of the sample (Zhu et al. 2011). The fiber becomes denser, which may lead to increased hardness of the sample (Zhu et al. 2011). It is believed that heating at 70 °C caused more rapid denaturation of collagen proteins, which led to more dramatic changes in the jellyfish samples compared to samples heated at lower temperatures. It appears that a threshold temperature may exist between 60 °C and 70 °C, where the heat-induced changes in the jellyfish become more significant and time-sensitive.

Instrumental color analysis

The effect of different heat treatments on the whiteness of the outer umbrella of jellyfish is shown in Fig. 2. The whiteness score tended to decrease with an extended heating time. For samples that underwent the 50 °C treatment, the differences from unheated samples were significant ($p < 0.05$) only after 30 min heating. However, as the temperature increased, significant differences were observed much earlier [1 min for 60 °C (B-2), and 0.5 min for 70 °C (C-1)]. It appeared that the higher the temperature, the faster the color change. Overall, the jellyfish outer umbrella tended to lose its whiteness with extended heating, and gradually turned yellow. When the color of the outer umbrella of jellyfish changed, it became less favorable according to the sensory panel (Table 2). The correlation analysis showed that the changes in whiteness were positively correlated to the color score in the sensory evaluation ($p < 0.05$). In general, the lighter the color, the more favorable samples were to consumers (i.e., in this case, the sensory panel).

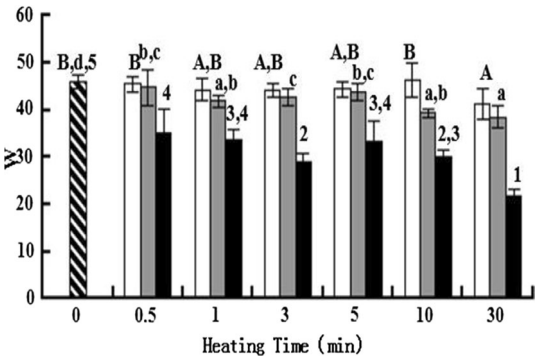

Fig. 2 Change in whiteness scores (*W*) of jellyfish outer umbrellas with heating [unheated (*hatched bar*), 50 °C (*white bar*), 60 °C (*gray bar*) and 70 °C (*black bar*)]. *Different capital letters*, *lowercase letters* and *numbers* indicate significant differences between samples exposed to 50 °C, 60 °C and 70 °C, respectively ($p < 0.05$). Five replicates were performed, and the results are presented as mean value ± SE

Jellyfish have a particular chemical composition, comprising mainly water and collagen. The change of opacity of jellyfish during cooking is mainly caused by protein aggregation. The optical path length of the flesh is reduced due to this increase in opacity, and as a result, the light that enters the surface is less selectively absorbed (Larsen et al. 2011). In addition, the water holding capacity of protein decreases after heating, and the loss of water leads to the darkening of the jellyfish (Skipnes et al. 2011). The correlation analysis results suggest that the moisture content is positively correlated to jellyfish whiteness ($p < 0.01$).

Moisture content

The effect of different heat treatments on the moisture content of jellyfish samples is shown in Fig. 3. The change of moisture content may lead to changes in the textural quality, color and sensory quality (Skipnes et al. 2011). Moisture contents in samples gradually decreased with extended heating time. At 50 °C, little change in moisture content was recorded. In comparison with unheated samples, 50 °C heating for 30 min caused the moisture content to change by 1.77%, and the same level of change was reached at 60 °C for 5 min. The higher the temperature, the earlier protein denaturation occurred (Chiou et al. 2004). Thermal

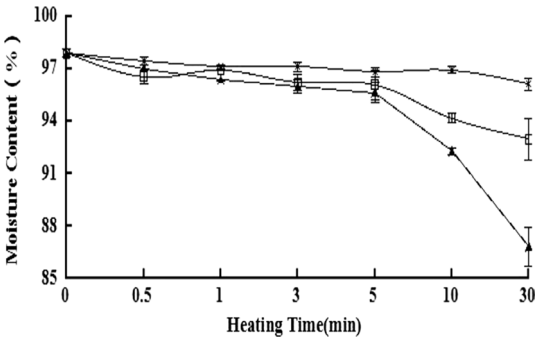

Fig. 3 Moisture content of jellyfish heated to 50 °C (*asterisk*), 60 °C (*open square*), 70 °C (*filled triangle*). Data are expressed as means of two replicates ± SD

denaturation of protein causes jellyfish to gradually lose their water holding capacity. The higher the temperature, the faster water is lost. The correlation analysis showed that juiciness is strongly correlated with the moisture content ($p < 0.01$).

Histological analysis

Light micrographs of jellyfish that underwent different temperature treatments are shown in Fig. 4. Myofibrils and collagen fibers are stained red and green, respectively. As shown in Fig. 4a, in unheated jellyfish samples, collagen fibers comprise the primary structure, as evidenced by the strong green color; a small amount of red is shown, indicating the secondary role of myofibrils in the jellyfish structure.

Heating, however, has a significant effect on the jellyfish microstructures. As shown in Fig. 4a, in unheated raw jellyfish, collagen fibers are connected in a randomly dispersed manner; Heating at 50 °C did not cause a significant color change; however, collagen fibers appeared to gradually shrink and form bundles with extended heating (Fig. 4b, c, d).

Previous reports showed that collagen in duck meat denatures at 60–65 °C (Khan et al. 2014b). As collagen denatures, the fiber structure may degrade, and allow smaller dye molecules to penetrate the interior; also the denaturation may expose the hydrophobic groups that are hidden inside protein molecules, and render staining with xylidine Ponceau to become more effective. A color change from green (intact collagen fibers) to red (myofibrils and denatured collagen proteins) is expected upon collagen denaturation.

As shown in Fig. 4e, f, g, h, when heated at 60 °C for 5 min, a color change from green to red becomes conspicuous, and the degradation and rupture of the collagen fibers also become visible with the formation of small holes (Fig. 4f), which grow in size with extended heating (Fig. 4f, g, h).

When the temperature was increased to 70 °C, a dramatic color change from green to red was observed at 0.5 min, as shown in Fig. 4i, j, k, l. This suggests that the denaturation of collagen in jellyfish is extremely temperature sensitive; at 70 °C, the denaturation is rapid, and the changes in the collagen structure occur almost immediately.

Thermal denaturation may also contribute to changes in texture and color (Kimura et al. 1983; Porturas et al. 1993; Skipnes et al. 2011), through heat-induced protein denaturation and degradation. The heating process disrupts hydrogen bonds, which in turn leads to the disintegration of the three strands of collagen. The higher the temperature, the greater the damage to the structure of collagen. In the early stage of low temperature heating, hydrogen bonds within collagen molecules are disrupted, and peptide chains contract, which

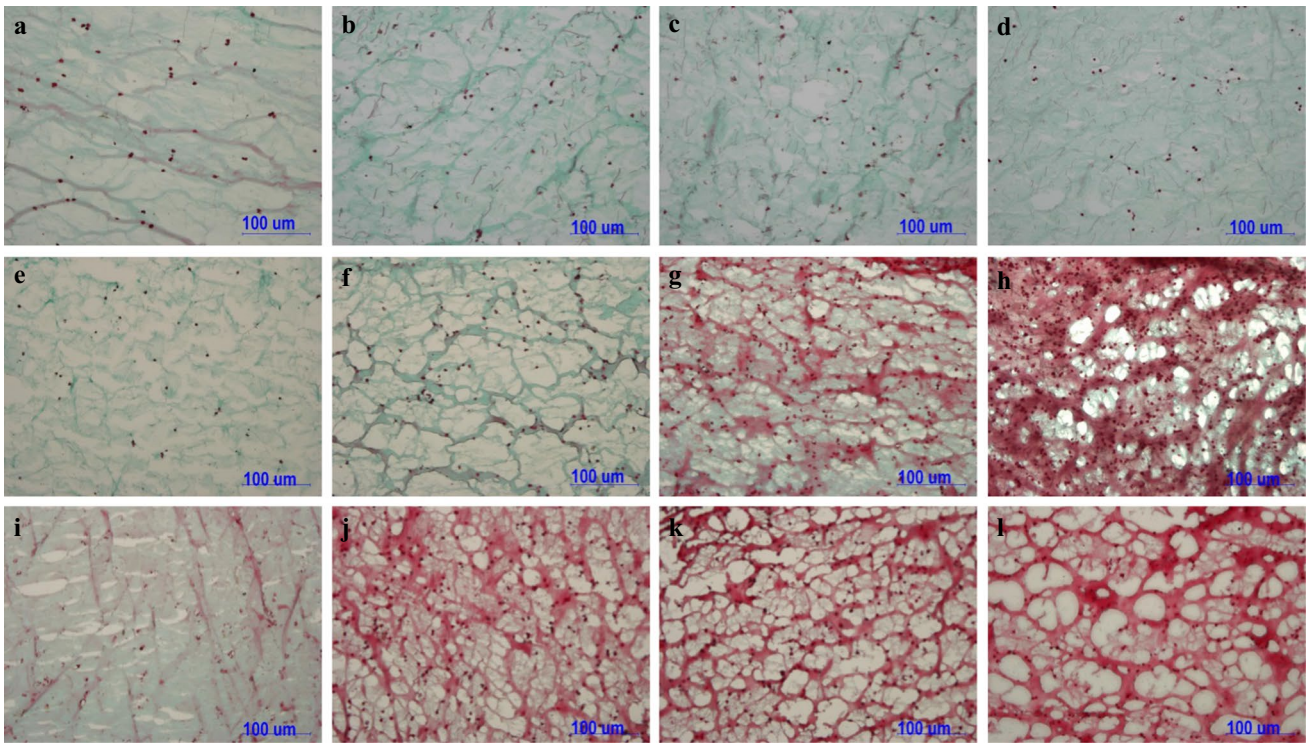

Fig. 4a–l Microstructural changes of jellyfish samples. **a** Unheated; **b–d** 50 °C heating for 1, 5, 10 min, respectively; **e–h** 60 °C heating for 1, 3, 5, 30, respectively; **i–l** 70 °C heating for 0.5, 3, 5, 10 min, respectively

causes the hardness of jellyfish to increase. When heating is prolonged, intramolecular hydrogen bonds are disrupted, resulting in a further decrease in hardness. At high temperatures, the hydrogen bonds between collagen molecules and within collagen molecules are disrupted, which destroys the stability of the collagen structure, hence, the hardness of the jellyfish is rapidly reduced.

Effect of different hurdle factors on microbial growth during storage

Samples which underwent the different treatments were stored at 37 °C for up to 10 days. The total number of microorganisms was determined through plate counts on days 0, 3, 7, and 10, respectively. Currently, there is no established legal standard for microbiological indicators in ready-to-eat jellyfish products. Chinese legislation allows manufacturers to establish their own control limits according to their specific production processes. Most local enterprises in Liaoning Province set a limit for the total bacterial counts of ready-to-eat jellyfish products at 5000 CFU/g [3.7 lg (CFU)/g]. Hence, for samples with significant microbial growth, our storage experiments were set to stop once the bacterial counts reached this limit. For samples in which microbial growth was under control, the storage experiments were carried out until day 10. During the experiments, especially in groups 1 and 2, with an increase of the microbial population, the textural characteristics of instant jellyfish changed, accompanied by the emergence of a peculiar smell. Similar results have been reported in the processing of ready-to-eat sea cucumber and freshly cut salads (Hou et al. 2014; Tsironi et al. 2017).

Figure 5 shows the initial microbial counts for the different groups in the different treatments. Heating alone only decreased the initial microbial count slightly. Figure 6 shows the time course of microbial growth during storage for the various sample groups. Samples in groups 1 and 2 reached

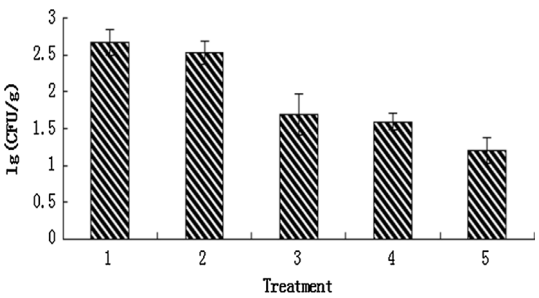

Fig. 5 Influence of different treatments on initial number of jellyfish samples. Control (*1*), heating (*2*), heating + acetic acid (*3*), heating + acetic acid + ultraviolet (UV) sterilization (*4*), heating + acetic acid + UV sterilization + potassium sorbate (*5*). Data are shown as means of three replicates ± SD. *CFU* Colony-forming unit

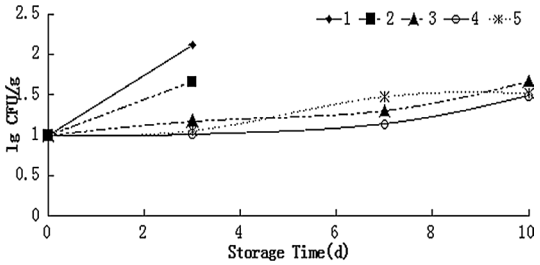

Fig. 6 Influence of different treatments on total number of bacteria during storage at 37 °C. Control (*filled diamond*), heating (*filled square*), heating + acetic acid (*filled triangle*), heating + acetic acid + UV sterilization (*open circle*), heating + acetic acid + UV sterilization + potassium sorbate (*asterisk*). Data are expressed as means of three replicates ± SD. *d* Days

the microbial growth limit (i.e., 5000 CFU/g) on day 3, while the microbial growth in other groups was significantly inhibited. Since the common factor in these groups was acid soaking, it appears that this is a main contributing factor to bacterial growth inhibition. Most organic acids, as small molecules showing some degree of hydrophobicity, can diffuse through cell membranes. Acid molecules undergo dissociation as soon as they enter the cellular cytoplasm, slowing down growth kinetics (Cássio et al. 1987). Acetic acid may also inhibit microbial growth by lowering the cytoplasmic pH to an inhibitory level (Stratford et al. 2009). Most microorganisms are susceptible to organic acids, and such inhibitory effects increase while the pH decreases (Conner et al. 1990). For instance, acetic acid was reported to lower bacterial growth rate and prolong the period before bacteria started to grow (León et al. 2012).

Microbial growth in samples of group 5 was effectively inhibited even on day 10, indicating the synergistic effect of all the factors further increased the effectiveness of the preservation treatment. In addition, the combination of acid and potassium sorbate treatments can ensure that the textural quality of the jellyfish is maintained during storage (Pérez-Díaz et al. 2008).

Discussion

In this study, the impacts of different heating conditions on the sensory, textural, color, and moisture content of jellyfish were evaluated. Histological study was used to determine the mechanism of these changes. Hurdle technology was used in the products' preservation, and its effectiveness was demonstrated. It was shown that proper heat treatment can lead to better sensory and textural qualities of jellyfish, while extended heating at above 60 °C may result in more water loss, deterioration of whiteness and diminished acceptability of the products to consumers. The adverse effect of high

temperature were correlated to the fast denaturation and degradation of collagen structures, which in turn affected the hardness and toughness of the jellyfish. All attribute indexes in the sensory evaluation were well correlated, and hardness (0.893, $p < 0.01$) and toughness (0.762, $p < 0.01$) contribute greatly to the overall sensory quality; this was evidenced by their strong correlation to the OSS, followed by color (0.740, $p < 0.01$) and juiciness (0.740, $p < 0.01$). Samples that underwent 60 °C heating for 1 min ranked the highest in terms of the sensory evaluation. The sensory evaluation results also correlated well with the instrumental analyses, i.e., texture and color scores. Harder and whiter samples consistently received more favorable sensory evaluation scores. The moisture content of samples correlated positively with the color, texture and juiciness of the products. Acetic acid (2 ml/L) treatment was the most effective in inhibiting bacterial growth, thus could be a crucial method for ready-to-eat jellyfish product processing.

In conclusion, the combination of heat, acetic acid, UV sterilization and potassium sorbate is highly effective in prolonging the shelf life of ready-to-eat jellyfish *Rhopilema esculentum* Kishinouye products.

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