EFFECTS OF TEMPERATURE, SALT AND ASCORBIC ACID ON THE STRUCTURE AND YELLOW DISCOLORATION OF CUTTLEFISH FILLET DURING COOL STORAGE

ẢNH HƯỞNG CỦA NHIỆT ĐỘ, MUỐI ĂN VÀ ACID ASCORBIC ĐỐI VỚI CẤU TRÚC VÀ SỰ BIẾN VÀNG CỦA FILLET MỰC NANG TRONG QUÁ TRÌNH BẢO QUẢN LẠNH

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ABSTRACT

An observation and treatment method of texture and color modification of prefreezing cuttlefish fillet are performed based on the control of storage temperature, NaCl, ascorbic acid concentration to prevent and reduce fillet's discoloration on cuttlefish fillet. In this research, the physical result effecting on cuttlefish fillet discoloration is storage temperature. The chemical aspects strongly impacting on coloration and keeping water ability are NaCl solution, antioxidant - ascorbic acid. Soaking cuttlefish fillet in 2 ÷ 4^{o}C with 3% NaCl and 0,2% ascorbic acid solution helps restrict the yellow discoloration and improve water holding capacity and the structure of cuttlefish fillet.

Keywords: *Cutllefish; yellow discoloration; ascorbic acid; NaCl; water holding capacity.*

TÓM TẮT

Ảnh hưởng của nhiệt độ bảo quản; muối ăn và acid ascorbic đến sự biến vàng và thay đổi cấu trúc của fillet mực nang trong quá trình bảo quản lạnh đã được khảo sát. Kết quả khảo sát cho thấy, nhiệt độ bảo quản có ảnh hưởng rõ rệt đến màu sắc của fillet mực nang. Nồng độ muối NaCl trong dịch ngâm, sự bổ sung chất chống oxy hóa acid ascorbic có tác động rất lớn đối với sự thay đổi màu sắc, khả năng giữ nước của fillet mực nang. Khi ngâm fillet mực nang trong dung dịch acid ascorbic 0,2% với NaCl 3% ở nhiệt độ 2 ÷ 4℃ làm tăng khả năng giữ nước, hạn chế tối đa sự biến vàng của cơ thịt mực.

Từ khóa: *mực nang; sự biến màu vàng; ascorbic acid; NaCl; khả năng giữ nước.*

1. INTRODUCTION

Whiteness is a crucial criterion for quality classifying of cuttlefish fillet in frozen cuttlefish processing [1-4]. If the color of fillet is discolored, the grade of fillet will be declined and reducing the commercial value.

After harvest and during the preservation, processing before freezing, cuttlefish fillet color is easily changed to yellow or dark brown due to the liposome system oxidation of cuttlefish's lipid [5-6]. Lipid contained in the cuttlefish will be oxidized by the effect of enzymes and oxygen. Chemical reactions of oxidized lipids with amines, amino acids, and proteins have received considerable attention

because they are associated with changes in functional properties, nutritive value, flavor, and color of food [7-9].

Frozen cuttlefish production and consumption is increasing worldwide. Although the microbiological deterioration is effectively restricted by cool storage, various chemical reactions still take place. Yellow pigment formation sometimes occurs during frozen storage of cuttlefish, accompanied by the development of rancid odours [6,10-11] reported that the yellow/brown colour correlated with lipid oxidation of cod fillet during salting. The objective of this study is to investigate the effects of preserving temperature, sodium chlorite and ascorbic acid concentration on the structure and yellow discoloration of cuttlefish fillets (*Sepia pharaonis*) [12-14].

2. MATERIALS AND METHODS

2.1 Materials

Pure acid ascorbic was from PA, China.

Cuttlefish (*Sepia pharaonis*) of the size of 10–15 cuttlefish/kg, caught by fishing junks in the areas of Kien Giang Sea, were purchased from Tac Cau Fishing Port, Kiengiang Province. The cuttlefish were preserved by ice in an insulated container and transported to the laboratory of Department of Food Technology, Can Tho University within 3 hours. Then, cuttlefish were cleaned, and stored in a fridge at 0 °C during pending.

Samples randomly taken from the prepared material were filleted after peeling the skin and washing under tap of water.

2.2 Apparatus

Samples were soaked in cool solution by water bath Lauda RC 25 CP, Germany.

The color was measured by Color Reader CR-10, Konica Minolta, Japan, and recorded by using the color system profile of L*, a* and b* then turned into the whiteness (Kinoshita et al., 2009) [15].

Water holding ability was measured by compression on filter paper method of Grau and Hamm [16].

2.3 Methods

Examining the effects of temperature

Cuttlefish fillets were soaked in water at various different temperature of 0 ÷ 2°C; 2 ÷ 4°C; 4 ÷ 6°C and at room temperature (control sample). Soaking water rate per sample was 2 per 1. The experimental parameters (color and water holding capacity) were collected for 2 hours each time.

Examining the effects of salty concentration

Cuttlefish fillets were soaked in salty solution at various different concentrations of 2%; 3%; 4% and 0% (control sample) with optimal temperature found. Soaking water rate per sample was 2 per 1. The experimental parameters (color and water holding capacity) were recorded for 2 hours each time.

Examining the effects of the combination of salt and ascorbic acid

Samples were soaked in salty-acid ascorbic solution at various different concentrations of acid ascorbic in range of 2%; 3%; 4% and 0% (control sample) with optimal temperature and salty concentration found. Soaking water rate per sample was 2 per 1. The experimental parameters (color and water holding capacity) were recorded for 2 hours each time.

Statistical analysis

All experiments were run in triplicate. Analysis of variance (ANOVA) according to LSD testing system to conclude about the average difference between experimental

roots. The colleted data were processed with statistical Statgraphic 4.0.

3 RESULTS AND DISCUSSIONS

3.1 Effect of storage temperature on the change of structure and color of cuttlefish fillet

The change of seafood quality is influenced by various factors and the most important of which is the temperature. If raw cuttlefish are not properly preserved, exposing in ambient temperature can seriously degrade their quality [17]. In terms of trade, icing or refrigerating plays an important role in reducing the deterioration caused by bacterial and enzyme in cuttlefish muscle [18]. Low storage temperature significantly reduces the chemical reactions, particularly lipid oxidation. Oxidation reactions are the main cause of rancidity and yellow pigment formation on the cuttlefish [19].

Water holding capacity (WHC) is an indirect parameter used to evaluate the structure of cuttlefish muscle. The increase of WHC shows the maintaining of structure of muscle [14]. WHC is dependent on various factors such as storage temperature, raw material, the size of cuttlefish, in which temperatures are the core factor greatly affecting the structure. Results surveyed are summarized in Figure 1 and Table 1 in different processing temperatures.

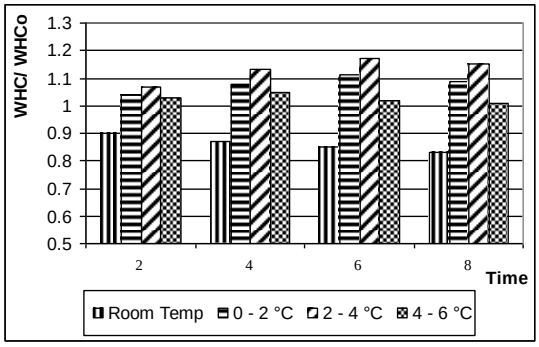

Figure 1. *The change of WHC of cuttlefish fillet by soaking time.*

Figure 1 shows that WHC of cuttlefish fillets stored at 2 ÷ 4°C temperature was the best and increased with storage time, expressed via the ascending rate of WHC/WHCo. Cuttlefish fillets stored at room temperature got worst WHC due to the dehydrate by oxygen while the chemical reactions strongly took place in the muscle, especially the lipid oxidation and hydrolysis affected the structure of muscle by formation of covalent bond with proteins presenting in cuttlefish muscle [6]. Simultaneously, the protein hydrolysis reaction was catalyzed by additional temperature leading to the change of structure characteristic on protein muscle fiber and decreasing the muscle WHC [20]. At 0 ÷ 2^{o}C preserved temperature, close to the ice-point of cuttlefish [20], little ice crystals were formed, which caused cells membrane to tear and release water, leading to the WHC lower than it was at 2 ÷ 4^{o}C. As preserving cuttlefish fillet at 4 ÷ 6^{o}C, chemical reactions still slowly took place [1] and affected the structure of cuttlefish muscle and gradually reduced the WHC as well.

Table 1 shows that there was no significant difference in the WHC of the fillet over soaked time. Meanwhile, low storage temperature was not only preventing the WHC from reducing but also improves WHC in comparison to the original samples. In range of 0 ÷ 2^{o}C, 2 ÷ 4^{o}C and 4÷ 6^{o}C, the WHC/ WHCo had a value greater than 1, proving that cuttlefish soaked in water could absorb water and stably kept water in its muscle. This could also be the cause for improvement of the bright whiteness of the raw material after processing time. Indeed, the results of fillet whiteness from Figure 2 confirmed the positive role of temperature on the cuttlefish fillet color stability.

Table 1. *Effects of preserving temperature on Water Holding Capacity of cuttlefish fillet*

Soaking temperature (ºC)	Time (hours)				Mean
	2	4	6	8	
Room temp	0,90	0,87	0,85	0,83	**0,86^{A}**
0 ÷ 2^{0}C	1,04	1,08	1,11	1,09	**1,08^{BC}**
2 ÷ 4^{0}C	1,07	1,13	1,17	1,15	**1,13^{C}**
4 ÷ 6^{0}C	1,03	1,05	1,02	1,01	**1,03^{B}**
Mean	**1,01^{a}**	**1,03^{a}**	**1,04^{a}**	**1,02^{a}**	

Different letters in the same column (capital) or the same row display the significant statistical differences at 95% reliability.

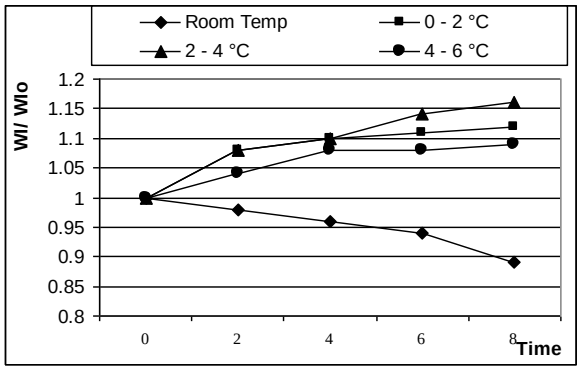

Figure 2. *The effects of whiteness of cuttlefish fillet by different temperature.*

Figure 2 shows that the fillet whiteness had the best value at 2 ÷ 4ºC and it was darker when stored at room temperature. At temperatures of 0 ÷ 2ºC, the fillet whiteness also improved but not as effectively as it did at 2 ÷ 4ºC. At room temperature, lipid oxidation reactions quickly took place and led to the formation of color compounds making fillet whiteness reduction over storage time [6]. In case of 4 ÷ 6ºC preserved temperature, fillet whiteness was also more improved than its original sample but not high. With samples preserved at 2 ÷ 4ºC, fillets' whiteness was gradually increased due to chemical reactions and bio-chemical reactions, especially lipids oxidation, were limited with poor oxygen condition in soaking water. Effects of low temperature on oxygen diminution, reducing color development under cuttlefish skin has been confirmed by [15].

3.2 Effects of salty concentrations on the change of quality of cuttlefish fillet

Appropriate low salty concentration (about 3 – 5%) [12] increased muscle WHC. Besides, salty absorption into cuttlefish muscle significantly affected muscle discoloration (Bone et al., 1981 [10]).

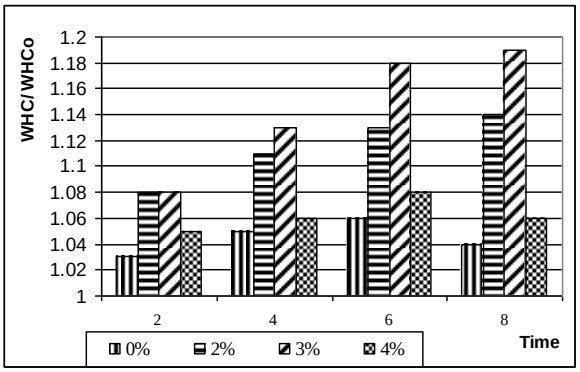

Figure 3*. Effects of saline concentrations on the water holding capacity of cuttlefish fillet*

Figure 3 shows the effects of different salty concentration on changes of fillet structure. The firmness of sample soaked in 3% salty solution had worst improvement (WHC/ WHCo increased). With samples soaked in 2% saline solution, the structure was also improved but to a lesser extent. For control samples, soaked in 0% saline solution, the water holding capacity were lowest and declining over time (WHC/WHCo decreased) due to lack of salt effects to improve the water-holding capacity. Salt also contributed significantly to cuttlefish fillet color, making the surface more light white, more transparent and reducing the fillet blurriness. The results of cuttlefish fillet discoloration are shown in Table 2.

Table 2. *Effects of saline concentrations on the change of whiteness of cuttlefish fillet*

Ascorbic acid (%)	Soaking time (hours)				Mean
	2	4	6	8	
0	1,08	1,11	1,13	1,14	**1,12^{A}**
0,1	1,15	1,18	1,19	1,22	**1,19^{A}**
0,2	1,15	1,20	1,25	1,25	**1,22^{A}**
0,3	1,10	1,14	1,15	1,16	**1,24^{A}**
Mean	**1,12^{a}**	**1,16^{ab}**	**1,18^{ab}**	**1,29^{b}**	

Different letters in the same column (capital) or the same row display the significant statistical differences at 95% reliability.

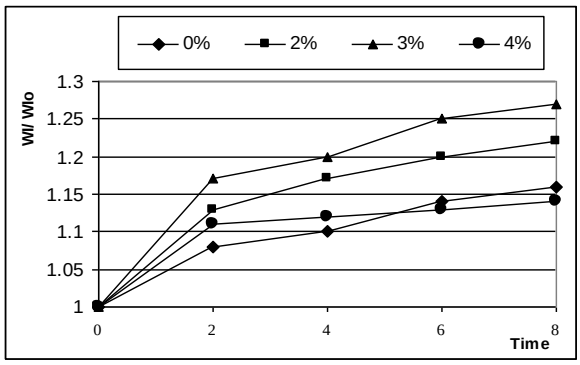

Figure 4 . *The efffects of saline concentration on the changes of cuttlefish fillet whiteness*

Figure 4 displays the effects of salty solution on the changes of cuttlefish fillet whiteness at different concentrations. It has been clearly seen that, at concentration of 3% saline, the fillet whiteness was highest. The present of salt reduced the rate of dissolved oxygen in soaked solution, inhibiting the growth of microorganisms and slowing down the lipids oxidation so cuttlefish fillet color was improved [16]. At concentrations of 2% salty solution, cuttlefish fillet whiteness was also improved but still lower than that in the case of a concentration of 3% saline. Low saline concentration, less salty absorption, could not prevent the oxidation. However, with a high salty concentration, the amount of water was significantly extracted degrades the fillet surface' whiteness. This was also the cause of meaningless difference between samples soaked 4% saline solution and the control samples.

3.3 The role of ascorbic acid combines with salt in prevention of quality degradation and discoloration on cuttlefish fillet.

Oxygen has been known as one of the core factors promoting discoloration of cuttlefish [16]. Besides controlling the use of oxygen at low temperatures combined with immersion in water (oxygen-limited environment), the use of antioxidant compounds, such as ascorbic acid, was also considered [20].

The results in Table 3 and Figure 5 show that ascorbic acid did not have effective function in improving the water holding capacity of cuttlefish fillet. It was reflected by statistical results that there was no significantly different between treatments.

Table 3. *The impacts of ascorbic acid on the water hlding capacity of cuttlefish fillets*

Ascorbic acid (%)	Soaking time (hours)				Mean
	2	4	6	8	
0	1,08	1,11	1,13	1,14	**1,12^{A}**
0,1	1,15	1,18	1,19	1,22	**1,19^{A}**
0,2	1,15	1,20	1,25	1,25	**1,22^{A}**
0,3	1,10	1,14	1,15	1,16	**1,24^{A}**
Mean	**1,12^{a}**	**1,16^{ab}**	**1,18^{ab}**	**1,29^{b}**	

Different letters in the same column (capital) or the same row display the significant statistical differences at 95% reliability.

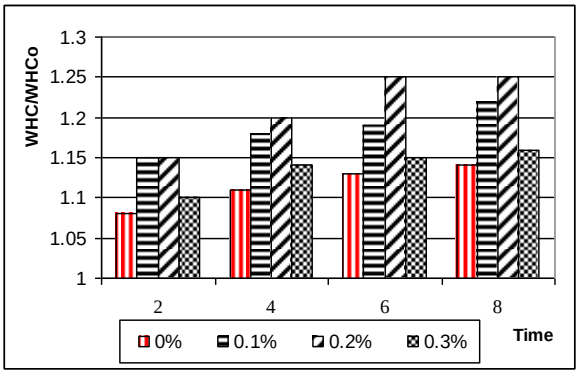

Figure 5. *The effects of ascorbic acid concentrations on the water holding capacity of cuttlefish fillets.*

Table 4 . *The effects of ascorbic acid concentration on the change of whiteness of cuttlefish fillet*

Ascorbic acic (%)	Time (hours)				Mean
	2	4	6	8	
0	1,14	1,17	1,23	1,26	**1,20^{A}**
0,1	1,19	1,22	1,32	1,33	**1,27^{BC}**
0,2	1,24	1,32	1,36	1,36	**1,32^{C}**
0,3	1,18	1,26	1,29	1,19	**1,23^{AB}**
Mean	**1,19^{a}**	**1,24^{ab}**	**1,30^{c}**	**1,29^{bc}**	

Different letters in the same column (capital) or the same row display the significant statistical differences at 95% reliability.

In terms of the impact of time, there still had an increase of WHC with the degree of change from 1.12 times to 1.29 times in comparison to original raw samples, according to the soaking time from 2 to 8 hours (Table 3) and no significant statistical difference between samples soaked in 4, 6 and 8 hours. This revealed that ascorbic acid seem had almost no effects on structure. Regarding to color change, ascorbic acid has shown the observing bleaching capability on cuttlefish fillet. This result is clearly shown in Table 4 and Figure 6.

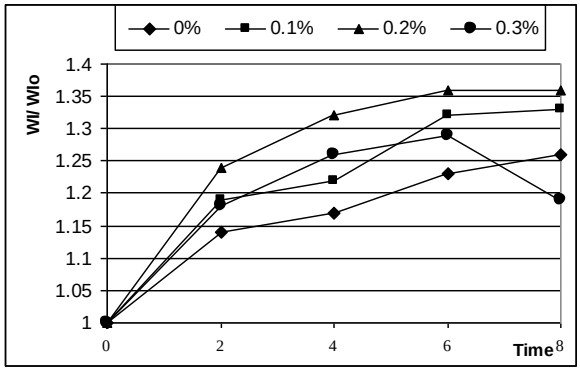

Figure 6. *The effects of ascorbic acid concentration on the change of whiteness of cuttlefish fillet*

Figure 6 shows that the whiteness of sample soaked in 0.2% ascorbic acid solution had the highest value. At concentration of 0.1% even, cuttlefish fillet whiteness was improved, significantly statistical difference compared with samples soaked in 3% NaCl solution only (the whiteness had an increase of 27 % compared to the its original value, meanwhile this increase was just 20% at samples immersed in 3% NaCl solution). Also, there was no statistical difference between samples soaking in ascorbic acid solution of 0.1% and 0.2%. However, the whiteness of samples soaked 3% ascorbic acid solution was lower and quite different compared to the samples of 0.2%, especially after 6 hours soaking. This was because of the presence of high concentration in the solution. Ascorbic acid could adhere on the fillet surface, causing the self-oxidation on the surface, making its color darker [20].

At levels of 0.2% ascorbic acid combining with the addition of 3% NaCl immersion fluid cartridge at temperatures between 2 ÷ 4^{o}C inhibited the chemical changes and the growth of microorganisms to keep freshness and quality of the original while preventing the growth of yellow spots affecting the color and product due to lipid oxidation.

4 CONCLUSION

A combination of 0.2% ascorbic acid and 3% NaCl in solution at 2 ÷ 4^{o}C well inhibited the biochemical reactions and microorganism development. Therefore, cuttlefish freshness could be preserved as its original qualities. Besides, the discoloration by lipids oxidation was prevented and the structure was significantly improved.

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