

# The changes of astaxanthin content and chemical characteristics of tiger prawn (*Penaeus monodon*) due to processing: boiling, smoking and frying

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**Abstract.** Food processing using high temperatures can cause changes in pigment color and chemical characteristics in food stuffs, including prawn. The aim of this research was to evaluate the changes in pigment and chemical characteristics of tiger prawn caused by boiling, smoking and frying. Ten kg of tiger prawn was boiled, smoked and fried at the temperature of  $\pm 100\text{ }^{\circ}\text{C}$  for  $\pm 10$  min. The results showed that boiling, smoking and frying gave a significant effect ( $P < 0.05$ ) on the astaxanthin pigment, pH, moisture, protein, salt content, Aw and color. The content of astaxanthin pigments in fresh prawn, boiled prawn, smoked prawn and fried prawn was:  $132.79 \pm 1.5\text{ }\mu\text{g}\cdot\text{g}^{-1}$ ;  $82.89 \pm 0.92\text{ }\mu\text{g}\cdot\text{g}^{-1}$ ;  $78.28 \pm 0.1\text{ }\mu\text{g}\cdot\text{g}^{-1}$  and  $91.35 \pm 2.59\text{ }\mu\text{g}\cdot\text{g}^{-1}$ , respectively. The value of  $^{\circ}\text{Hue}$  on fresh prawn, boiled prawn, smoked prawn and fried prawn was:  $87.85^{\circ}$ ;  $52.5^{\circ}$ ;  $55.94^{\circ}$  and  $53.98^{\circ}$ . The tiger prawn processed by the smoking method has preferable by panelist rather than processed by boiling and frying.

## 1. Introduction

Tiger prawn is a popular seafood in many countries and it becomes a favorite seafood choice by consumers because of its specific taste and aroma. Although it has a tangy smell in its raw condition, after processing, distinct flavor and specific delicious taste will arise. Based on data from the Ministry of Marine Affairs and Fisheries in 2013, Indonesia exported shrimp to various destination countries reaching a total volume of 162,000 tons. Production of shrimp aquaculture during the last five years (2010–2014) has a tendency to continue rising. The average increase of shrimp production during the last five years is 13.83 % per year. In 2012 shrimp production of 415,703 tons was reached while in 2013 amounted to 645,955 tons [1].

Food processing by heating which is also known as the cooking process is the process of heating food at a temperature of  $100\text{ }^{\circ}\text{C}$  or more. The main purpose is to get a better taste, better aroma and softer texture; moreover, it also kills microbes and deactivates all enzymes. Cooking can be done by boiling, smoking and frying. The use of heat in the cooking process is very influential on the nutritional value of the foods including boiling, smoking and frying. The goals of the salt-boiled process for shrimp are to reduce a load of microorganisms to an acceptable level and to improve the flavor of shrimp. A suitable boiling process is important to get a suitable condition to minimize loss of muscle protein and maintain the red color. Smoking by using liquid smoke can produce smoked fish that is higher in quality when compared with the traditional smoking process. Smoking by using liquid smoke may affect the color and sensory quality of smoked fish products [2]. Frying is a fast and simple culinary technique employed worldwide that consists of cooking food above the boiling point



of water, by means of partial (shallow-frying) or total (deep-frying) immersion in liquid edible oils or fats at frying temperature. Although fried foods are highly appreciated for their sensory characteristics, they must also satisfy health-related consumer concerns.

High-temperature processing can also affect the change of pigment and chemical characteristics in foodstuffs, including prawn. Prawn color turns red because it contains astaxanthin, a pigment that gives the characteristic of orange to red. Changes in pigment can also indicate that there has been a change in chemical characteristics of prawn. The purpose of this research was to evaluate the effect of processing treatments on the changes of astaxanthin content and chemical characteristics in tiger prawn.

## 2. Materials and Methods

### 2.1. Materials

The material used in this study was  $\pm 10$  kg of tiger prawn with a size of  $\pm 20$  cm in length and an average weight of  $\pm 150$  g. Cooking oil, the food grade liquid smoke made of coconut shell, boiling water and salt as a flavor enhancer.

### 2.2. Methods

This research was conducted experimentally using completely randomized design with duplication. After washed thoroughly, the prawns were dipped into 5 % liquid smoke and 5 % of brine solution. Then they were processed by boiling (at  $\pm 100$  °C), smoking (at  $\pm 100$  °C) and frying (at  $\pm 100$  °C) for  $\pm 10$  min [3].

### 2.3. Measurement of astaxanthin content

Astaxanthin content was measured following Becerra et al. [4], a total of 10 g samples was extracted with 40 mL 0.05 % BHT solution (in acetone) with a digital homogenizer for 2 min. The samples were cooled during the homogenization process to prevent the sample from heating up. After extraction, the sample was centrifuged at 4,000 rpm, temperature of 40 °C for 5 min. To separate the water-soluble compound, the acetone extract from the sample was transferred to a 250 mL separating funnel with 40 mL n-hexane. Next, 100 mL of aquadest containing 0.5 % (w/v) of sodium chloride was added to the mixture. After continuous shaking, phase separation was achieved and then the top layer was separated and transferred into 50 mL measuring flask. The absorbance spectrum of a water-soluble compound was measured at a wavelength of 472 nm using a spectrophotometer. The astaxanthin stock solution ( $0.2 \text{ mg} \cdot \text{mL}^{-1}$ ) of 0.5; 1.0; 1.5; 2.0 and 3.0 mL were respectively diluted with 10 mL n-hexane. Then the absorbance spectrum of each solution was recorded with a wavelength of 472 nm. The standard curve was obtained by plotting the concentration on absorbance.

### 2.4. Measurement of pH

The sample was ground and weighed as much as 1 g in a cup glass. Then, 10 mL of aquadest was added and stirred. Subsequently, pH level of the sample in the container was measured by using a calibrated pH meter with a buffer solution of pH 4 and pH 7. The pH value was obtained based on the reading at pH meter until the digital number indicated the constant number [5].

### 2.5. Measurement of moisture content

The determination of moisture content was based on the different weight of samples before and after drying. The method used was a drying technique for approximately 24 h at 105 °C until the sample reached a stable weight. Percentage of moisture contents (wet weight) was calculated by using the following formula [5]:

$$\text{Moisture Content (\%)} = \frac{B-C}{B-A} \times 100 \%$$

**Information:**

A: empty vial weight (g)

B: cup weight + initial sample (g)

C: cup weight + dry sample (g)

**2.6. Measurement of protein content**

Determination of protein content was done based on AOAC [5]. Basically, it could be divided into three stages namely the process of destruction, distillation and titration. In the process of destruction the sample was heated with concentrated  $\text{H}_2\text{SO}_4$  thus it broke down into their respective elements. The destruction process was completed when the solution became clear. The distillation stage resulted in the broken down of ammonium sulfate into ammonia by adding alkalis NaOH and heated. The formed ammonia which was accommodated in concentrated  $\text{H}_3\text{BO}_3$  added with BCG and methyl red indicator. The amount of  $\text{H}_3\text{BO}_3$  reacted with ammonia could be known by its reaction with 0.02 M HCl. The end of the titration was marked by the change of solution color from green to pink. Blank treatment was performed to determine the nitrogen from the reagents being used.

**2.7. Measurement of  $A_w$** 

The instrument was calibrated by inserting the  $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$  liquid and closed for 3 min to reach the reading scale of 0.9. Then, samples were inserted into  $A_w$  meter for 3 min and  $A_w$  scale was recorded. If the temperature scale reached above 20 °C, the scale reading was added as much as the excess temperature multiplied by the correction factor of 0.002 °C, as well as the temperature below 20 °C [6].

**3. Results and Discussion****3.1. Astaxanthin content**

**Table 1.** Astaxanthin content tiger prawn before and after the heating process.

Treatment	Astaxanthin ( $\mu\text{g} \cdot \text{g}^{-1}$ )
Fresh	$132.79 \pm 1.5^a$
Boiled	$82.89 \pm 0.92^b$
Smoked	$78.28 \pm 0.1^c$
Fried	$91.35 \pm 2.59^d$

Note: Value from average of duplicate  $\pm$  standard deviation.

\*Value with different lower case notation shows a significant difference ( $p < 0.05$ ).

Table 1 shows that the content of astaxanthin in tiger prawn decreased after heat processing. The heating processes led to the reduction of astaxanthin levels as the proteins protecting astaxanthin had been denatured by heat, resulting in the decomposition of astaxanthin. In boiling treatment, this reduction could be related to denaturation and partial solubilization of the carotene protein complex. In this regard, a significant decrease in proteins content (myofibrillar, sarcoplasmic and stroma) in boiled shrimp, suggesting that part of these proteins could have been dissolved in the salt solution [7]. The reduction of astaxanthin due to heat is also reported in drying treatment. Astaxanthin content in cooked shrimp decreased by 75 % after four days of direct sun drying, reaching the concentration of  $25.0 \pm 2.7 \mu\text{g} \cdot \text{g}^{-1}$  [4]. The authors noted that the decline in astaxanthin content in shrimp during sun drying mainly affects the color of the product.

### 3.2. Moisture content

**Table 2.** Moisture content, protein content and water activity of tiger prawn before and after the heating process.

Treatment	Moisture content (%)	Protein content (%)	A <sub>w</sub>
Fresh	88.72 ± 0.54 <sup>a</sup>	27.6 ± 1.5 <sup>a</sup>	0.93 ± 0.0007 <sup>a</sup>
Boiled	72.7 ± 0.32 <sup>b</sup>	14.17 ± 1.4 <sup>b</sup>	0.83 ± 0.0007 <sup>b</sup>
Smoked	59.84 ± 0.59 <sup>c</sup>	22.57 ± 0.23 <sup>c</sup>	0.72 ± 0.0014 <sup>c</sup>
Fried	57.6 ± 1.17 <sup>d</sup>	14.3 ± 0.68 <sup>d</sup>	0.62 ± 0.002 <sup>d</sup>

Note: Value from average of duplicate ± standard deviation.

\*Value with different lower case notation shows a significant difference ( $p < 0.05$ ).

The lowest moisture content was found in fried prawn at  $57.6 \pm 1.17$  % with a reduction of 35.076 %, followed by the smoked prawns with a reduction of 32.55 %, and boiled prawn with a reduction of 18.05 %. The reduction in water content as a consequence of processing related to the temperature being used, the higher the temperature the higher the water content decreases. It happened because of the process of steaming and boiling used temperature of 90–100 °C, while the frying process used a temperature of 200–205 °C. As a comparison study by previous researchers [8], showed that the largest reduction in water content occurred in soybean cake (20.13 %) followed by chicken (16.74 %), mackerel (16.05 %) and the smallest was in tofu (7.66 %).

Moisture content is related to the presence of microorganisms that grow in the body of prawn since water is a medium that supports the growth of microorganisms. The lower the water content, the lower microorganisms decompose. Water is the largest content in fish [9]. Water is a means of microorganisms to thrive. The moisture content reduction in fish can extend the shelf life of the products. Salting process is capable of losing water in fish because salt has a higher concentration than water, so the water will come out of the tissue or called plasmolysis. The decrease of the moisture content was the most prominent change in fish fillet after drying. Protein, fat and ash content increased significantly in the processed sample [10].

### 3.3. Protein content

Protein content in prawn decreased after treated with boiling, smoking or frying process. Protein content in both boiled and fried prawn had almost the same value of  $14.17 \pm 1.4$  % and  $14.3 \pm 0.68$  %, respectively, with a decrease of 48.19 %. The lowest reduction in protein content occurred in smoked prawn (18.22 %) with a protein content of  $22.57 \pm 0.23$  %. The reduction of protein content was directly proportional to the used processing temperature, meanwhile, in the process of frying and boiling the used temperature was more than 100 °C hence the protein was damaged by heat. Other researchers reported that the reduction of protein content in boiled bloated fish was 3.12 % whereas fried bloated fish was 5.24 % [8]. During cooking, heat induces protein denaturation and aggregation, causing actin, myosin and collagen shrinkage, which leads to water expulsion from muscle cells and the consequent water loss. During boiling, protein content also decreased. Water and protein losses are called cooking loss [7].

Processing with high temperatures can lead to proteins damage because the protein bonds have undergone changes and breaks. It also deals with the solubility of proteins, water-soluble proteins and faster salts lost due to heat. Protein solubility decreased with increasing temperature, it indicated changes in protein solubility [3]. Denaturation of proteins related to protein solubility, as a result of the heat, the secondary or tertiary structures of the protein was damaged, hence it transformed into a primary structure. Salt-soluble and water-soluble proteins were more easily lost in the primary structure rather than those in secondary or tertiary structures. Boiling, smoking and frying maintained and concentrated the crude protein content. After storage, there was a reduction in crude protein content of the fried fish rather [11].

### 3.4. Water activity

$A_w$  of fresh prawn was 0.93. The lowest  $A_w$  occurred in fried prawn with a value of 0.62. While the  $A_w$  of smoked and boiled prawns were 0.72 and 0.83, respectively. Processing with high temperatures could decrease the value of  $A_w$ . It was in line with the decrease in water content. The relationship of water content with water activity ( $A_w$ ) was indicated by the tendency that the higher the water content the higher the  $A_w$  value would be and vice versa [12].

**Table 3.** The value of pH and salt content of tiger prawn before and after the heating process.

Treatment	pH	Salt content (%)
Fresh	$7.6 \pm 0.14^a$	$0.78 \pm 0.04^a$
Boiled	$7.55 \pm 0.07^b$	$1.4 \pm 0.16^b$
Smoked	$6.45 \pm 0.07^c$	$4.2 \pm 0.12^c$
Fried	$7.3 \pm 0.14^d$	$3.4 \pm 0.07^d$

Note: Value from average of duplicate  $\pm$  standard deviation.

\*Value with different lower case notation shows a significant difference ( $p < 0.05$ ).

### 3.5. pH value

The pH value of all samples decreased from the initial pH of fresh prawn with a value of 7.6. The value of pH on boiled prawn and fried prawn were 7.55 and 7.3, respectively. The lowest pH value occurred in smoked prawns with a value of 6.45. The pH value in smoked prawn was acidic, whereas the pH value of other treatments indicated that the pH was neutral. The low pH of the smoked prawns occurred as the effect of the addition of liquid smoke prior to processing. This acidic pH trend was caused by the absorption of the liquid organic liquids into the sample. The longer the immersion of liquid smoke, the more organic acids such as acetic acid, formic acid would enter into the fish meat during smoking process and resulted in a reduction of pH value. The smoke attached to the fish meat caused the fish meat to be more acidic therefore the pH value tends to be lower [13].

### 3.6. Salt content

Samples were dipped into 5 % of brine before heat processing. Salt content in all samples decreased significantly after processing. The decrease in salt content was caused by salt dissolution into processing media such as oil and water. The highest salt content was found in smoked prawns with a value of 3.4 %, while the lowest was found in boiled prawn with a value of 1.4 %. The salt content of smoked catfish ranges from 2.62 to 5.83 % [3]. In this study, cooking loss observed was 32 %, which is higher than losses reported by previous researchers [7] for boiled shrimp in a salt solution (4 %, w/v) for 7 min. The greater cooking loss we found can be attributed to the longer boiling time (15 min vs. 7 min). On the other hand, the NaCl content in shrimp showed a significant increase after boiling in brine.

### 3.7. Colour analysis

Processing affected the color change in prawn. The value of  $^{\circ}\text{HUE}$  of boiled prawn reduced 35.35  $^{\circ}\text{HUE}$  into the yellow-red color category, the smoked prawn reduced 31.91  $^{\circ}\text{HUE}$  and fried prawn reduced 33.87  $^{\circ}\text{HUE}$  into the red color category (table 4). Changes in astaxanthin are marked in red color on prawn. Colour changes from reddish pink to orange in prawn are formed by the release of astaxanthin from protein-bound due to proteolysis [14].

**Table 4.** Colour analysis of tiger prawn before and after the heating process.

Treatment	Colour			°HUE
	L ( <i>Lightness</i> )	+a* ( <i>Redness</i> )	+b* ( <i>Yellowness</i> )	
Fresh	35.64 ± 0.7 <sup>a</sup>	0.2 ± 0.01 <sup>a</sup>	5.32 ± 0.66 <sup>a</sup>	87.85 <sup>d</sup>
Boiled	46.57 ± 0.49 <sup>c</sup>	14.77 ± 0.68 <sup>c</sup>	19.23 ± 0.71 <sup>c</sup>	52.5 <sup>b</sup>
Smoked	48.24 ± 0.22 <sup>c</sup>	15.63 ± 0.44 <sup>c</sup>	23.12 ± 0.08 <sup>c</sup>	55.94 <sup>a</sup>
Fried	41.32 ± 0.33 <sup>b</sup>	12.25 ± 0.45 <sup>a</sup>	16.85 ± 1.75 <sup>b</sup>	53.98 <sup>f</sup>

Note: Value from average of duplicate ± standard deviation.

\*Value with different lower case notation shows a significant difference ( $p < 0.05$ ).

### 3.8. Sensory value of fresh prawn

The results of the sensory value of fresh prawn show that appearance, odor, and texture covered average values of  $8.73 \pm 0.45$ ;  $8 \pm 0.51$  and  $8.8 \pm 0.41$ , respectively. Fresh prawn had a specific prawn's texture of elastic, compact and solid. Based on these specifications, the prawns used in this research were still in the fresh category. The sensory of fresh prawn fulfill requirements of sensory of fresh fish based on Indonesian National Standard for fresh fish which minimal sensory value was seven.

### 3.9. The prawn preferences test

**Table 5.** Preferences test of tiger prawn after heat processing treatment.

Treatment	Hedonic parameter			
	Appearance	Flavour	Odor	Texture
Boiled prawn	6.5 ± 0.51 <sup>a</sup>	6.6 ± 0.56 <sup>a</sup>	7.2 ± 0.48 <sup>a</sup>	6.23 ± 0.5 <sup>a</sup>
Smoked prawn	8.57 ± 0.5 <sup>c</sup>	8.1 ± 0.31 <sup>b</sup>	8 ± 0.49 <sup>b</sup>	8.57 ± 0.5 <sup>c</sup>
Fried prawn	7.57 ± 0.5 <sup>b</sup>	8.1 ± 0.42 <sup>b</sup>	7.3 ± 0.48 <sup>a</sup>	7.87 ± 0.57 <sup>b</sup>

Note: Value from an average of 10 panelists ± standard deviation.

\*Value with different lowercase notation shows a significant difference ( $p < 0.05$ ).

**3.9.1. The appearance.** The panelists gave the highest score for the appearance of the smoked prawn. The results of the appearance were as follows: surface quality prawn was bright, shiny, invisible dirt in the form of dried blood and the specific color of prawn. Chemical reactions among substances contained in the smoke could occur in smoked fishery products. The chemical reaction between phenol and oxygen could change the color of the smoke product to be a brownish yellow. Phenol, the major carbonyl component in smoke reacting with proteins and fats in fish, has an important role in affecting the appearance of smoked fish [9].

**3.9.2. Flavors.** The panelist's preference score in the flavor of both smoked and fried prawn was the highest with a similar average value of 8.1. The taste specifications on fried prawn were savory on the contrary the smoked prawn had a distinctive taste of smoke products. In addition, boiled prawn had a slightly bland taste when compared with the other two processing treatments. The acceptance of panelists on taste parameters was based on not only the maturity level but also taste of "umami" in the processed prawn. Processing techniques with heating were able to produce products that have extraordinary taste compared with other techniques [15].

**3.9.3. Odor.** The highest panelists preference score of the odor was in the smoked prawn based on the specification that they had the distinctive aroma of smoke. It was the effect of the increase of phenol levels due to heating. The increase of phenol level occurred because of temperature and heating time. The higher the temperature and curing time resulted to the higher the phenol production [9]. In fact, the aroma of boiled prawn and fried prawn was still included in a favorite category with a value of 7.2



$\pm 0.48$  and  $7.3 \pm 0.48$ . The aroma of a food that undergone high-temperature processing was caused by the volatile compounds. The aroma produced in food after processing affected by the compound in food and the way of cooking [15].

**3.9.4. Texture.** The highest score for texture preference was in the smoked prawns with a value of  $8.57 \pm 0.5$ . The texture of smoked prawn was solid, compact and not fragile. The addition of liquid smoke might affect the texture of prawn result from the reaction of liquid smoke components such as phenol. The main carbonyl component in the smoke played an important role. The smell, taste and texture of smoked fish were the results of the reaction of carbonyl groups in the smoke reacting with proteins and fats in fish. Smoke plays an important role in the formation of color, texture and taste [9].

#### 4. Conclusion

The different prawn processing by boiling, smoking or frying affected pigment concentration changes and chemical characteristics in tiger prawn. Further research on the influence of various types of oil in frying, various types of liquid smoke and different boiling temperature on the changes in pigment concentration and chemical characteristics of prawn needs to be conducted in the future.

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