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N,O-Carboxymethyl Chitosan: An Innovation in New Natural Preservative from Shrimp Shell Waste with a Nutritional Value and Health Orientation

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ABSTRACT

This research has been done to modify chitosan into its derivatives, i.e. N,O-Carboxymethyl Chitosan. The characterization of chitosan and N,O-Carboxymethyl Chitosan, which includes analysis using FTIR, SEM, and XRD, showed that the a natural preservative N,O-Carboxymethyl Chitosan had formed. Our data indicated that addition of N,O-Carboxymethyl Chitosan to samples of chicken meat could be regarded as a solution to increase fiber contents, resilience of food storage, and stability of nutrients (lowering levels of dry substances), lower ash contents, increase protein contents, keep fat contents, as well as increase levels of Nitrogen-Free Extract. Therefore, we conclude that the N,O-Carboxymethyl Chitosan can be used as a preservative which also orients towards nutritional values and health.

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Key Words: N,O-Carboxymethyl Chitosan, Preservatives, Chicken Meat, Food, Shrimp Shell

Introduction

Food is the most essential basic need for humans to sustain life and living. Food which functions as a source of nutrients such as carbohydrates, fats, proteins, vitamins and minerals serves as the main base for people to achieve health and well-being throughout the life cycle, starting from fetuses, infants, toddlers, children, adolescents, adults and the elderly need food in accordance with

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the nutritional requirements to survive, grow and develop as well as achieve working achievement [1] [2]. Proteins are one of the nutrients necessary for humans to grow, develop, and stay healthy. The cheapest sources of animal proteins are eggs and chicken meat [3].

Several important things to worry in relation to products of animal origin are the presence of contamination or microbial contamination, residues of veterinary medicines such as biological products, pharmaceuticals, premix, and chemicals as well as the use of certain preservatives that harm consumers [4]. The number of cases of food poisoning that currently occurs in the society indicates a mistake in the processing and the preservation of food consumed. The fundamental issues in food processing performed by the society is mainly due to a culture of food processing that is less oriented toward nutritional values. In addition limited knowledge and the economic pressure at the same time also making the problems on food fulfillment and food processing neglected. One way to overcome the problems of food processing is to develop compounds that can serve as a preservative and orient toward nutritional values and health.

One of substances that is known to be a food preservative is chitosan. Indonesia is a country rich in natural resources. The Indonesian fisheries sector is a sector with a good prospect. It can be seen from the increase in the export value of fishery products of the nation. As reported by the Ministry of Maritime Affairs and Fisheries, the export value of Indonesia fisheries based on the total commodities from January to November 2013 reached U.S. $ 3.77 billion or increased approximately up to 7.0% from U.S. $ 3.53 billion in 2012. During that period, shrimps became the main commodity of the Indonesian fisheries export with a value of U.S. $ 1.280 million. Shrimp export increased by 25.46% from the previous year with the largest contribution value from frozen shrimps by U.S. $ 1.121 million. Along with the increased shrimp production, the waste produced from the shrimp processing has also increased. The amount of waste generated, if not processed immediately, will cause environmental pollution. So far, the processing of shrimp waste is used only as a substance to make kerupuk (shrimp crackers), terasi (a condiment made from fermented shrimp paste), and supplements for animal feed. In fact, shrimp shell waste is very potential due to its chitin content that reaches approximately 99.1% [5]. Thus, the use of shrimp waste by processing it into chitin and its derivatives can be one alternative to solve environmental problem caused by shrimp industry.

Chitin, after further processing, will result in chitosan that can be used as a product preservative and stabilizer. Chitosan can be used as a preservative both for food and drinks due to its nature of inhibiting the growth of adverse microorganisms while at the same time coating the preserved product so that results in minimal interaction between the product and its environment [6].
In addition to the potential as a preservative for food or drink, chitosan has been widely used as drug coating intended to optimize the absorption of the drug in the target cell. These properties of chitosan such as biodegradable, biocompatible, non-immunogenic, non-carcinogenic make chitosan suitable for use in food technology. However, the use of chitosan has a drawback, i.e. it can be dissolved only in an acidic solution (pH < 6) and cannot be dissolved in a physiological environment (pH = 7.4) [7]. To overcome it, this study modified the chitosan into its derivatives, i.e. N,O-Carboxymethyl Chitosan. N,O-Carboxymethyl Chitosan is a derivative of chitosan that is hydrophilic in nature, making it dissolved in either an acid, neutral, or alkaline condition.

To determine the effectiveness of N,O-Carboxymethyl Chitosan as a preservative which also orients toward nutritional values and health, this study performed a variety of tests to examine the application of N,O-Carboxymethyl Chitosan in the attempt to improve fiber contents and maintain the quality of chicken meat. Meanwhile, the characterization of chitosan and N,O-Carboxymethyl Chitosan was performed using the spectroscopy method which included FTIR to analyze the existing functional groups, XRD to examine or confirm whether the N,O-Carboxymethyl Chitosan had been formed, and SEM to investigate the topography surface. This study is expected to generate a safer preservative that can be easily used by the general community.

Materials and Methods

Materials

The materials that this research used consisted of shrimp shells (powder), NaOH, NaOCl, HCl, acetone, mono chloro acetic acid, isopropyl alcohol, glacial acetic acid, methanol, ethanol, and distilled water. The Equipment employed in this study consisted of glass tools commonly used in organic chemistry labs and Ostwald viscometer. The spectroscopic identification of compounds was performed using Scanning Electron Microscope (SEM) with FEI Brand, Type: Inspect-S50, Fourier Transform Infrared (FTIR) with Perkin Elmer Brand, and X-Ray Diffraction (XRD) with Philip Brand, Type: E’xpert Pro.

Methods

Chitin Manufacturing

Deproteinization

This process was carried out at a temperature of 60-70°C using 1M NaOH solution with the comparison between shrimp shells (powder) and NaOH = 1:10 (g powder / mL NaOH) while stirring for 60 minutes. Then the mixture was filtered to separate and take the sediment.
Washing and Drying
Sediment washing was done using distilled water up to the neutral pH. Then, it was filtered and dried in order to take the sediment.

Demineralization
Removal of minerals was done at a temperature of 25-30°C using 1M HCl solution with a comparison between the sample and the HCl solution = 1:10 (g powder / mL HCl) while stirring for 120 minutes. Then the mixture was filtered to separate and take the sediment.

Decolorization
Sediment resulted from demineralization was extracted using acetone and bleached with 0.315% NaOCl (w/v) for 5 min at room temperature. The solid and solvent comparison equals to 1:10 (w/v).

Washing and Drying
Sediment washing was done using distilled water up to the neutral pH. Then, it was filtered and dried. The result of this drying process is called chitin.

Chitin Deacetylation into Chitosan
Chitin generated in the previous process were included in NaOH solution with a concentration of 50% at a temperature of 90-100°C and stirred at a constant speed for 60 minutes. This stage generated slurry that had to be filtered first. After filtration, the obtained sediment was washed using distilled water and then added with dilute HCl solution to make the pH neutral then it was dried. The result of this drying process was called chitosan.

Synthesis of N,O-Carboxymethyl Chitosan
A total of 5 grams of chitosan was suspended in 50 mL of isopropyl alcohol and the resulting slurry was stirred in a 200 mL flask at room temperature. 13 mL of 10N NaOH solution, divided into five equal portions, was then added to the slurry which had been stirred for 25 minutes. This alkaline slurry was then stirred again for 30 minutes. Then, as many as 30 grams of mono chloro acetic acid were added, to the five equal portions, with a 1-minute addition interval. The reaction mixture was then heated up to a temperature of 60°C and stirred at this temperature for 3 hours. The reaction mixture was filtered and the product in the form of solids (N,O-Carboxymethyl Chitosan) was washed with methanol. N,O-Carboxymethyl Chitosan was then dried in an oven at a temperature of 60°C [8].

Characterization of Chitosan and N,O-Carboxymethyl Chitosan
To examine or confirm whether the N,O-Carboxymethyl Chitosan had been formed, the characterization of the chitosan and N,O-Carboxymethyl Chitosan which included the analysis of
Examining the Variation of N,O-Carboxymethyl Chitosan Concentration in Comparison with Fiber Contents and Quality of Chicken Meat

In this test, there were two types of treatments. In the first treatment which served as a control, no N,O-Carboxymethyl Chitosan was added in the chicken meat. While in the second treatment, the chicken meat was added with N,O-Carboxymethyl Chitosan with the following concentration: 100 ppm, 250 ppm, 500 ppm, and 1000 ppm. Each treatment was then examined to determine the fiber content and quality of the chicken meat. To determine the quality of the chicken meat, this research performed the examination of dry matter contents, protein contents, fat contents, ash contents, and Nitrogen-Free Extract contents.

Examining the Variation of N,O-Carboxymethyl Chitosan Concentration in Comparison with the Shelf-Life of Chicken Meat

During this part of our research the conditions for the control and tested samples were same as described in the previous section. Each treatment was examined to determine the shelf-life for 7 days by looking at the organoleptic properties of the chicken meat sample. The organoleptic properties examined included odor, texture, and color.

Results and Discussion

Synthesis of N,O-Carboxymethyl Chitosan

Synthesis of N,O-Carboxymethyl Chitosan in the research was performed by reacting chitosan with mono chloro acetic acid in an alkaline condition [9]. In general, the reaction which occurred in the synthesis of N,O-Carboxymethyl Chitosan from chitosan is given as follows.

![Figure 1. Synthesis of N,O-Carboxymethyl Chitosan from Chitosan](image)

Based on the above synthesis reaction of N,O-Carboxymethyl Chitosan from chitosan, it can be seen that substitution by the carboxymethyl group on the hydroxyl group (-OH) and the amino group (-NH$_2$) occurs. To examine whether N,O-Carboxymethyl Chitosan, which is the target
product, has been formed, the solubility can be examined first. Based on the existing literature [8] [10], N,O-Carboxymethyl Chitosan is dissolved in distilled water. Therefore, this study performed the solubility test to the synthesized product by taking a few samples of the product resulted from synthesis and adding distilled water to the product. Based on this test of solubility, it can be seen that the synthesized product is dissolved in distilled water, consistent with the solubility test results according to the literature. Thus in accordance with the results of this solubility test, it can be assumed that the product resulted from the synthesis is N,O-Carboxymethyl Chitosan or it can be said that N,O-Carboxymethyl Chitosan has been formed. N,O-Carboxymethyl Chitosan solution in distilled water indicates that the carboxymethyl group contained in chitosan improves the hydrofility of chitosan. To ensure that N,O-Carboxymethyl Chitosan which is the target product has really been formed, a number testing and characterization are necessary.

![Figure 2](image_url)  
**Figure 2.** (a) Chitosan; (b) N,O-Carboxymethyl Chitosan

**Characterization of Chitosan and N,O-Carboxymethyl Chitosan**

**FT-IR Analysis**

The infra-red absorption spectra of chitosan and N,O-Carboxymethyl Chitosan is shown in Figures 3 and 4. The peaks that appeared in the infra-red spectrum of chitosan and N,O-Carboxymethyl Chitosan are identical. In the infra-red spectrum of chitosan, the peak observed at a wave number 3437.64 cm\(^{-1}\) is the stretching vibration of -NH\(_2\) and -OH groups. The peak at wave number 1643.97 cm\(^{-1}\) is the bending vibration of -NH\(_2\) group.

In the infra-red spectrum of N,O-Carboxymethyl Chitosan, carboxymethylation is indicated with the presence of peaks at wave numbers 1625.79 cm\(^{-1}\) and 1399.87 cm\(^{-1}\) [11]. This wave number 1625.79 cm\(^{-1}\) itself is actually a characteristic peak of N,O-Carboxymethyl Chitosan that indicates the presence of carboxylic acid salt (the stretching vibration of -COO\(^{-}\) antisym) [8]. And to
ensure that N,O-Carboxymethyl Chitosan has actually been formed, it can be done by examining from the presence of peaks at wave numbers 1257.52 cm\(^{-1}\) and 1083.62 cm\(^{-1}\) which are the C-O-C stretching vibration. In addition, the peak seen at a wavelength of 3432.06 cm\(^{-1}\) which continues widening and has increasingly small transmittance intensity also indicates that carboxylation has been formed in both groups, i.e the amino group and the primary hydroxyl group found in the structure of chitosan [11].

![Figure 3. The infra-red absorption spectra of Chitosan](image)

![Figure 4. The infra-red absorption spectra of N,O-Carboxymethyl Chitosan](image)
XRD Analysis

In this study, XRD analysis was conducted to test or confirm whether N,O-Carboxymethyl Chitosan had been formed. XRD analysis was conducted using a Cu Kα tube targets with an angle length of 5 to 80° (2θ).

Figure 5. The results of the XRD diffractogram for Chitosan obtained using Cu Kα tube targets with an angle length of 5 to 80° (2θ)

Figure 6. The results of the XRD diffractogram for N,O-Carboxymethyl Chitosan obtained using Cu Kα tube targets with an angle length of 5 to 80° (2θ)
Based on the obtained results of XRD diffractogram, it can be seen that all the sharp peaks that exist on chitosan appear at different angles when compared to the sharp peaks found in N,O-Carboxymethyl Chitosan. It is also using this same XRD diffractogram that chitosan can be recognized, i.e. with the emergence of sharp characteristic peaks at an angle of 20°. While N,O-Carboxymethyl Chitosan can be recognized by the appearance of characteristic peaks that are less sharp than the characteristic peaks of chitosan appearing at an angle of about 33° [11].

The results of XRD diffractogram obtained in this study both for chitosan and N,O-Carboxymethyl Chitosan show the existence of sharp characteristic peaks at certain angles as has been mentioned in the literature. In this study, the characteristic peaks of chitosan appeared at an angle of 19.81729°. While the characteristic peaks of N,O-Carboxymethyl Chitosan appeared at an angle of 32.12226°. In addition, the diffractogram of N,O-Carboxymethyl Chitosan also suggests that the characteristic peaks that appeared are indeed less sharp than the characteristic peaks of chitosan. This finding is also supported or can be seen from the obtained peak height value. The peak height value of chitosan’s characteristic peaks appeared at an angle of 19.81729° was 136.83. While the peak height of N,O-Carboxymethyl Chitosan’s characteristic peaks appearing at an angle of 32.12226° was 14.08. Based on the XRD diffractogram data available, it can be said that N,O-Carboxymethyl Chitosan has been formed or has been successfully synthesized.

**SEM Analysis**

In this study, SEM analysis was conducted to determine the surface characteristics of both chitosan and N,O-Carboxymethyl Chitosan. Based on this SEM analysis, it is revealed that chitosan has a rather flat and not porous surface. While the N,O-Carboxymethyl Chitosan has a characteristic of an uneven surface and the surface displays the existence of a new formation that attaches to the surface of chitosan. SEM analysis results for chitosan and N,O-Carboxymethyl Chitosan are shown in Figures 7 and 8.
**Figure 7.** SEM images of Chitosan with a magnification of (a) 1000 times, (b) 2000 times, and (c) 3000 times

**Figure 8.** SEM images of N,O-Carboxymethyl Chitosan with a magnification of (a) 1000 times, (b) 1600 times, (c) 2000 times, and (d) 5000 times
The Results for the Examination of the Variation of N,O-Carboxymethyl Chitosan Concentration in Comparison with Fiber Contents and Quality of Chicken Meat

By adding N,O-Carboxymethyl Chitosan at several concentrations, it can be seen that the fiber contained in the sample of chicken meat has increased significantly. Animal food products such as meat, fish, milk, eggs, and dairy products contain a very measly amount of fiber making consumption of such food necessary to be balanced with the consumption of food sources that contain fiber [12]. Therefore, addition of N,O-Carboxymethyl Chitosan can be assumed to constitute a solution to increase the fiber content in a certain diet. A certain type of food can be considered good if the fiber content of this food continues to increase. The benefits from chitosan and its derivatives including N,O-Carboxymethyl Chitosan as a dietary fiber are to increase fecal mass, reduce the glycemic response of the food, and lower the cholesterol level [13]. In which the main role of dietary fibers is to bind water, cellulose, and pectin. Fibers help speed up the remnants of food through the digestive tract to be secreted out. Without the help of fibers, faeces with a low water content will stay longer in the intestinal tract and has difficulty passing through the intestine to be secreted out because the peristaltic movements of the colon become slower.

Table 1. The results for the analysis of the examination of dry matter contents, protein contents, fat contents, ash contents, and Nitrogen-Free Extract contents in chicken meat added with N,O-Carboxymethyl Chitosan at various concentrations

<table>
<thead>
<tr>
<th>N,O-Carboxymethyl Chitosan Concentration Added to Chicken Meat</th>
<th>ANALYSIS RESULT (%)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Dry Matter</td>
</tr>
<tr>
<td>0 ppm</td>
<td>26.5329</td>
</tr>
<tr>
<td>100 ppm</td>
<td>24.1804</td>
</tr>
<tr>
<td>250 ppm</td>
<td>24.7459</td>
</tr>
<tr>
<td>500 ppm</td>
<td>24.7235</td>
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<tr>
<td>1000 ppm</td>
<td>24.021</td>
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</tbody>
</table>

The quality of chicken meat can be determined by performing several types of tests such as tests of dry matter contents, protein contents, fat contents, ash contents, and Nitrogen-Free Extract contents. The determination of this dry matter content refers to a way to estimate the resilience of food storage and stability of existing nutrients. The content of dry matters that must exist in order to result in the resilience of food storage and stability of existing nutrients is maximally 86% [14]. With the various concentrations of N,O-Carboxymethyl Chitosan added, it can be seen in Table 1
that the dry matter content contained in the samples of chicken meat tends to decline as the concentration of N,O-Carboxymethyl Chitosan increases. It indicates that the higher the concentration of N,O-Carboxymethyl Chitosan added, the better the resilience of food storage and stability of nutrients.

![Figure 9. Chicken meat added with N,O-Carboxymethyl Chitosan in different concentrations; (a) control (0 ppm), (b) 100 ppm, (c) 250 ppm, (d) 500 ppm, (e) 1000 ppm](image)

The ash content of certain food indicates the mineral content of the food. Ash itself is a non-volatile organic substances, resulted from the combustion or oxidation process. Measurement of the ash content found in chicken meat was done to see the overall mineral content instead of certain minerals and thus the types of minerals contained in the chicken meat are unknown. If the ash content of certain food is more than 3%, it suggests that the mineral content is relatively high in amounts. However, it is possible that the meat contains certain minerals harmful to the body. It is therefore safe to eat food with an ash content below 3% [15]. With addition of different concentrations of N,O-Carboxymethyl Chitosan, it can be seen in Table 1 that the ash content that the chicken meat samples contain tends to decline with the increasing concentration of N,O-Carboxymethyl Chitosan. With the decreasing ash content of chicken meat after the addition of N,O-Carboxymethyl Chitosan, it can be said that the chicken meat is becoming increasingly safe for consumption. In addition, the decrease in the ash content occurs presumably because of the nature of N,O-Carboxymethyl Chitosan which has the ability to pull heavy metal ions.

Chicken meat belongs to food that contains proteins. Proteins are one of the essential nutrients for the body, which are responsible for cell growth and replacement of damaged cells as
well as serve as a fuel for human body. Therefore, protein deficiency can cause interference in humans. Based on the examination results, it is known that the level of proteins found in chicken meat increases with increasing addition of N,O-Carboxymethyl Chitosan concentration. The increase in the protein content of chicken meat found after the concentration of N,O-Carboxymethyl Chitosan added to it is increased occurs presumably because of the Nitrogen element in the amina group of the N,O-Carboxymethyl Chitosan that is included in the calculation of the total N content, used to determine the protein content of the product. In addition, increased levels of protein in chicken meat caused by N,O-carboxymethyl chitosan has properties that can interact or bind to proteins in foodstuff and N,O-carboxymethyl chitosan has N group capable of forming amino compounds which are components in the formation of proteins.

Fats are found in nearly all food in different levels of content. The addition of fats in food processing aims to increase energy, improve the texture and taste of the food. Based on the analysis of the fat content of chicken meat added with N,O-Carboxymethyl Chitosan at various concentrations, it can be seen that fat contents do not significantly change after a higher concentration of N,O-Carboxymethyl Chitosan is added. The function and role of fats in the diet are as a potential source of energy, sources of essential fatty acids, a flavor enhancer, and a carrier of fat-soluble vitamins as well as to improve feed efficiency. During storage, fats encounter several processes, such as the process of rancidity caused by the activity of enzymes (lipolytic) and oxidation [16].

Meanwhile, analysis of Nitrogen-Free Extract contents suggests that the greater the concentration of N,O-Carboxymethyl Chitosan added, the higher the Nitrogen-Free Extract content. The Nitrogen-Free Extract content itself is soluble carbohydrates that include monosaccharides, disaccharides, and polysaccharides that are easily dissolved in acid and alkaline solutions and have a high level of digestibility [17]. The higher the level of the Nitrogen-Free Extract contained in certaine food, the better the quality of the food.

Based on the findings of this research, it can be seen that the addition of N,O-Carboxymethyl Chitosan serves a function as a natural preservative that can perform coating. This coating prevents the dietary contents to come out. In addition, the benefits derived from the use of chitosan and its derivatives including N,O-Carboxymethyl Chitosan as a preservative with a nutritional value and health orientation are that N,O-Carboxymethyl Chitosan can act as an antibacterial agent, replacement of blood vessels, anti-tumor, or a coagulant of leukemia cells [13].
**Table 2.** Observation results for chicken meat added with N,O-Carboxymethyl Chitosan at various concentrations

<table>
<thead>
<tr>
<th>N,O-Carboxymethyl Chitosan Concentration</th>
<th>Day</th>
<th>Odor</th>
<th>Texture</th>
<th>Color</th>
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<td>0 ppm</td>
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<td>2</td>
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Description:

**Odor:** ++++ (the chicken meat still smells fresh); +++ (the chicken meat has already smelled a bit unfresh); ++ (the chicken meat has already smelled bad); + (the chicken meat has already smelled bad)

**Texture:** ++++ (the texture is still good and springy); +++ (the texture is still good and springy); ++ (the texture begins to be mushy and fungi start to grow on it); + (the texture is mushy and fungi grow on it)

**Color:** ++++ (the color is still fresh); +++ (the color is still fresh); ++ (it is brownish in color); + (the color changes into brown)
The Results for the Examination of the Variation of N,O-Carboxymethyl Chitosan Concentration in Comparison with the Shelf Life of Chicken Meat

Based on the examination result for the variation of N,O-Carboxymethyl Chitosan concentrations over the shelf life of chicken meat, it is revealed that the greater the concentration of N,O-Carboxymethyl Chitosan added to the chicken meat, the longer the shelf life of the chicken meat.

Chitosan and its derivatives including N,O-Carboxymethyl Chitosan are highly potential for use as an antimicrobial material because they contain the lysosim enzyme and the aminopolysacharida group that can inhibit microbial growth and the inhibition efficiency of N,O-Carboxymethyl Chitosan against bacteria depends on the dissolution concentration of N,O-Carboxymethyl Chitosan. The ability to suppress bacterial growth is resulted from N,O-Carboxymethyl Chitosan which has a positive polikation just like chitosan which can inhibit the growth of bacteria and mold. The antibacterial property of N,O-Carboxymethyl Chitosan is greater than that of chitosan. It is due to the value of the degree of substitution of the carboxymethyl group in N,O-Carboxymethyl Chitosan that is higher compared to chitosan which contains no carboxymethyl groups, and this is it which lead to increased antibacterial activities [18].

One mechanism that may occur in food preservation is that the molecules of N,O-Carboxymethyl Chitosan have the ability to interact with compounds on the surface of bacterial cells which are then absorbed forming a kind of layer which inhibits the transport channels of the cells making them lack substances necessary to grow and lead to the death of the cells. In addition to meeting the microbiological standards, chitosan and its derivatives are also chemically not harmful since in this process, N,O-Carboxymethyl Chitosan simply needs distilled water to make it dissolved and form a homogeneous N,O-Carboxymethyl Chitosan solution that is relatively safer. Damaged food can be identified in several ways, First, using the organoleptic test, i.e. by looking for signs of damage such as changes in texture or the springy condition, viscosity, color, odor, slime formation, and others. Based on those tests of damaged food, the test which is considered fairly simple to be applied in areas armed with simple equipment facility is: the microbiological test, by counting the number of microbes [19].

The rot or damage of meat is characterized by the formation of malodorous compounds such as ammonia, H₂S, indole, and amen, which are resulted from protein breakdown by microorganisms. Damaged meat shows organoleptic changes, i.e smell, color, the springy condition, appearance, and taste. Changes in off-odor of the meat usually occur if the total bacteria on the surface of the meat reach 107.0 to 7.5 koloni/cm², followed by the formation of slime on the surface of the meat if the number of bacteria reaches 107.5 to 8.0 colony/cm².
The longer shelf life of chicken meat along with the increasing concentration of N,O-Carboxymethyl Chitosan added is consistent with the results for the dry matter content determination test. Where this test shows that dry matters that the meat contains tend to decrease as an increasing concentration of N,O-Carboxymethyl Chitosan is added. The results for the determination test of the dry matter content is in fact supported and evidenced by this shelf life test that had been done. Finally, it can be concluded that the resilience of food storage (shelf life) and the stability of the nutrients are becoming increasingly better with the addition of N,O-Carboxymethyl Chitosan in higher concentrations.

Conclusions

Synthesis of N,O-Carboxymethyl Chitosan to be used as a preservative can be done by reacting chitosan from shrimp shell waste with mono chloro acetic acid in alkaline conditions. Based on the results of the test, which included examination of fiber contents, dry matter contents, ash contents, protein contents, fat contents, as well as Nitrogen-Free Extract contents over chicken meat, it can be said that N,O-Carboxymethyl Chitosan can be used as a preservative with a nutritional value and health orientation. Where the addition of N,O-Carboxymethyl Chitosan to samples of chicken meat can increase the fiber content, the resilience of food storage, and the stability of the nutrients (lower the dry matter content), lower the ash content, increase the protein content, maintain the fat content, as well as increase the Nitrogen-Free Extract content. Further research on the use of N,O-Carboxymethyl Chitosan as a preservative for food is required, given that this study is a new innovation in the development of a safe preservative that is also oriented towards the nutritional value and health. In addition, further research is also necessary regarding the examination of N,O-Carboxymethyl Chitosan as a safe preservative that is also oriented towards the nutritional value and health such as the microbiological test, the LC$_{50}$ test, and the LD$_{50}$ test.

References


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