

EFFECT OF CHITOSAN COATINGS ON PRESERVATION OF RED SNAPPER (*Lutjanus argentimaculatus* Forsskal, 1775) DURING LOW TEMPERATURE STORAGE

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Abstrak

Pengaruh pelapisan tipis tiga konsentrasi chitosan yang berbeda (0, 1 dan 2% b/v) terhadap kualitas mikrobiologi (angka lempeng total (ALT)), kualitas kimia (pH dan *total volatile basic-nitrogen* (TVB-N)), dan karakteristik sensorik ikan kakap merah (*Lutjanus argentimaculatus* Forsskal, 1775) selama penyimpanan dingin (4-7°C) dievaluasi secara berkala. Kualitas mikrobiologis ikan kakap merah yang dilapisi dengan kitosan lebih rendah dibandingkan kontrol, sedangkan pH dan TVB-N lebih rendah saat mendapatkan perlakuan pelapisan tipis kitosan. Skor keseluruhan evaluasi sensorik ikan dengan lapis tipis kitosan konsentrasi 1% (b/v) lebih tinggi jika dibandingkan perlakuan yang lain. Studi ini menunjukkan bahwa lapisan tipis kitosan sebelum penyimpanan dan distribusi ikan kakap merah dapat memperpanjang umur simpan ikan segar meskipun kualitas mikrobiologis ikan tergantung pada kualitas awal produk sebelum perlakuan pelapisan tipis kitosan.

Kata kunci: Kitosan, Pelapisan Tipis, Penanganan Ikan, Umur simpan

Abstract

The effect of three different concentration of chitosan (0, 1 and 2 % w/v) on microbiological (aerobic plate count), chemicals (pH and total volatile basic nitrogen (TVB-N)), and sensory properties of red snapper (*Lutjanus argentimaculatus* Forsskal, 1775) during cold storage (4-7°C) was evaluated periodically. The microbiological quality of red snapper coated with chitosan was lower compared to the control, whereas pH and TVB-N were lower when treated with chitosan coating. The overall score of sensory evaluation of chitosan with concentration of 1% (w/v) was higher compared to remaining treatments. This study indicated that coating of chitosan prior to storage and distribution of red snapper prolong the shelf life of the fish but the microbial quality during storage is depend on initial quality of the fish.

Keywords: Chitosan, Coating, Fish handling, Shelf life

INTRODUCTION

Red snapper (*Lutjanus argentimaculatus* Forsskal, 1775) is one of the economically important fish species in Indonesia. The quality of fish during storage remains important issues in tropical countries such as Indonesia due to the higher rate of spoilage in the room temperature. Chilling is an effective and efficient method to reduce the rate of deterioration as well as to prolong shelf life of fish [1]. Various methods have been reported which consist of thermal and non-thermal

processing. Thermal processing such as pasteurization [2] and canning is commonly applied. However, the eating quality of the products is change due to denaturation of fish protein. Non-thermal processing such as hydrostatic pressure, irradiation, ozonization, and modified atmosphere packaging was developed to overcome the problem. However, the high capital for equipment is needed. Therefore, the simple and cheap method to extend shelf life of fresh fish is needed.

Chitosan is a natural polymer (poly (1→4)-2-amino-2-deoxy-D-glucopyranosyl) obtained from deacetylation of chitin is mainly produced from crustacea shells [3]. Chitosan exhibit antimicrobial activity against wide range of microbes, such as bacteria, yeast and fungi [4]. Due to its non-toxic characteristic, it can be applied as food additive to extend the shelf life of foods. Chitosan can be applied to prolong the shelf life of pacific oysters (*Crassostrea gigas*) from 8-9 days to 14-15 days during storage under 5°C [5]. Application of chitosan with antioxidant to preserve grass carp during cold storage and found out that chitosan with the addition of acetic acid and tea polyphenol was effective to keep the sensory quality of fish during storage [1]. All of those study used high quality of fish prior to application of chitosan. However, fish is commonly purchased from the markets with various qualities due to transportation and distribution.

During transportation and distribution, fish is treated with the addition of ice, therefore the temperature is not constant. The main objective of this study was to evaluate the microbial, chemical, and sensory quality of red snapper during chilling storage and to confirm the potential of chitosan coating in fresh fish shelf-life.

MATERIAL AND METHOD

Materials

Chitosan with degree of deacetylation of 78% was purchased from Bogor Agriculture University. Glacial acetic acid (Sigma Aldrich, USA) for made dilution of chitosan was p.a. Fish used was purchased from Pabean Market, Surabaya and taken to the laboratory with insulator box with the addition ice (4-7°C).

Methods

1) Preparation of Chitosan Solution

The chitosan solution was prepared with method described by Fan and co-worker [6]. Briefly, for preparing 1% w/w chitosan

solution, 10 g chitosan were mixed with 900 ml of distilled water and stirred for 10 min prior to addition of 10 ml of glacial acetic acid. The solution was stirred for 2 h and added with distilled water up to 1 L. For preparing 2% w/w chitosan solution, 20 g chitosan were used with the same method described previously.

2) Preparation of Red Snapper Coating

Fresh red snapper (*L. argentimaculatus*) with average weight of 200 g were purchased from wet market of Surabaya. After being gutted and washed, fish samples were dipped in 1% and 2% chitosan solution and in 1% glacial acetic acid as control respectively for 2 h and then drained. All samples were individually packed in polyethylene bag and stored in insulated box with the addition of ice. Fish samples were taken randomly every 3 days for microbiological, chemicals and sensory evaluation.

3) Microbiological Analysis

Total viable count (TVC) was determined in plate count agar (PCA, Oxoid) by spread plate method [7].

4) pH and Total Volatile Basic Nitrogen (TVB-N) Analysis

Determination of pH was performed by homogenizing 10 g sample in 100 ml distilled water and the mixture was filtered. The pH of filtrate was measured by employing digital pH meter (Labortechnik, Germany).

For Total Volatile Basic-Nitrogen analysis, a total of 10 grams of fish samples were randomly taken and homogenized with 90 mL perchloric acid (6% v/v). It was followed by centrifugation at 4,000×g at 4°C for 5 min and the supernatant was distilled with micro distillation unit. The distillate was collected in boric acid (3% w/v) and titrated with hydrochloric acid (0.05 N). Results have been expressed in mg of nitrogen per 100 g of sample [8].

5) Sensory Evaluation

The sensory quality of fish sample was evaluated by a six member-trained panel from the student. Panelist scored for sensory

characteristics such as flavor, color, odor, general acceptability, and texture using a nine-point hedonic scale (1, dislike-extremely to 9, like-extremely).

6) Data Analysis

This experiment was conducted with two measurements. The significant effects of chitosan coating on the quality of red snapper were measured chemically and microbiologically. The data were subjected to analysis of variance by employing excel (microsoft). The statistical significance of differences between mean values was set at $P \leq 0.05$ with Duncan test.

RESULT AND DISCUSSION

1) Microbiological Analysis

The raw material used in this study had the medium initial total viable count (TVC) (Table 1). It was 4.15 log CFU/g which indicated that the freshness of the fish was reduced. This condition was due to the marine fish was usually caught and transported to the market without proper handling. The increasing of TVC in the red snapper coated with chitosan was higher compared to control (1% acetic acid). This finding was different with study done in other study [9] which applied chitosan on *Pangasius pangasius* fillet under chilling condition. They found that the total plate count of the pangasius fillets with the addition of chitosan were not exceeded log 5 CFU/g up until 9 days for 1% chitosan and 11 days for 2 and 3% chitosan. The other study showed that application of 1% (w/v) of chitosan on smoked milk fish at room temperature retained the microbiological quality up to seven days [13]. However, based on starting point of the application of chitosan was log 4 CFU/g, both result this study and Damayanti [9] were able to maintain the microbiological quality up until three days. This study indicated that the initial microbiological quality of the fish will affect the storage of the fish.

Antimicrobial activity of chitosan affected by degree of polymeration therefore, the pH of the solution will affect the activity of chitosan. The pH of the fish of this study

(higher than 6) maybe reduce the polymeration of chitosan on the fish matrix [10]. In addition, particle size of the chitosan also affect the activity of antimicrobial of chitosan.

Several possibilities have been described as the mode of action for chitosan, from binding to bacterial DNA which leads to inhibition of mRNA, to interaction with surface molecules. The ability of chitosan to bind DNA was commonly investigated for gene delivery, but the contribution of such ability in antimicrobial activity is unclear because chitosan would not reach a target in the cytoplasm [10].

Table 1. Total viable count of red snapper coated with chitosan during chilling storage

Storage time	Log CFU/g		
	0%	1%	2%
0 day	4.15	4.15	4.15
3 day	5.66	6.17	6.54
6 day	7.95	7.95	7.95
9 day	8.95	10.03	9.78

2) pH and TVB-N

The pH value of red snapper with and without application of chitosan during storage was showed in Table 2. The pH values of all samples were relatively constant, but the decreasing level of pH was exhibited on treatment 0% chitosan. Similar result was reported by other study [6]. The initial reduce of pH value may be due to dissolution of carbon dioxide from fish to the solution, while the increasing of pH is mainly due to production of volatile basic nitrogen. High pH value may affect the amino acid charge of the fish flesh that increases the possibility of microbes to degrade the amino acid on the flesh. The degradation of the fish protein produces volatile base that improve the pH of sample during storage [10].

Table 2. pH value of red snapper with and without chitosan coating during chilling storage

Storage time	pH		
	0%	1%	2%

0 day	7.47	7.47	7.47	0 day	6.70	6.70	6.70
3 day	6.66	7.06	7.39	3 day	6.22	6.18	6.22
6 day	6.70	7.02	6.80	6 day	3.78	5.30	4.74
9 day	7.20	7.79	7.08	9 day	2.46	3.00	3.46

Total volatile basic nitrogen (TVB-N) content of fish flesh during storage with and without chitosan coating showed in Table 3. TVB-N mainly consist of ammonia and amines which widely used as an indicator of fish deterioration. The increasing of TVB-N was related to the increase of microbial activity and indigenous enzyme that hydrolyzed the protein of the fish [6]. In addition, TVBN value indicated the freshness of the sample [11]. The higher TVBN value showed the lower the quality of the fish. The threshold of acceptable level of TVB-N was 35-40 mg TVB-N/100 g of samples [12].

Table 3. Total volatile basic nitrogen of snapper with and without chitosan coating during chilling storage

Storage time	mg TVB-N/100 g sample		
	0%	1%	2%
0 day	2.68	2.68	2.68
3 day	2.68	5.36	7.16
6 day	6.68	9.36	11.16
9 day	14.36	16.12	15.16

Based on Table 3, the application of chitosan inhibited the production of TVB-N. This result was in accordance with research in other research [6]. The TVB-N content of fish with and without treatment of chitosan was under acceptable level which indicated that the spoilage inhibited with the addition of acetic acid.

3) Sensory Evaluation

The fish samples sensory qualities were evaluated based on colour, texture, odour, flavour, and general acceptability based on 9 hedonic scale (9 extremely like, 1 extremely dislike).

Table 4. Overall sensory quality of fish (coated and uncoated of chitosan) during chilling storage

Storage time	Overall acceptance score		
	0%	1%	2%

Acceptable level of sensory quality of consumable fish was 4 [6]. Therefore, the treatment of 1% and 2% of chitosan were kept the sensory quality of the red snapper up until 6 days. The spoilage process of the fish produces off odour and rejected by the consumer. The prolonged of sensory qualities of the red snapper in this study may due to the activity of chitosan, i.e. antimicrobial, antioxidant and oxygen barrier.

CONCLUSION

This study indicated that the application of chitosan coating can prolong the shelf life of red snapper until six days under chilling storage. Coating of 1% chitosan was the most effective concentration to prolong the shelf-life of red snapper. However, application of chitosan as preservative agent of fresh red snapper was less effective on low quality raw materials, it showed by the TVC, pH, and TVBN value of this study. Therefore, the application of chitosan as coating agent on different raw material quality is needed.

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