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Effect of chitosan-based edible coating on preservation of white shrimp during partially frozen storage

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ABSTRACT

Chitosan and chitooligosaccharides are preservatives with proven antibacterial activity, while glutathione has antioxidant activity. This study investigated the effects of chitosan coating combined with chitooligosaccharides and glutathione (0.8% glutathione + 1% chitooligosaccharides + 1% chitosan) on preservation of white shrimp (*Penaeus vannamei*) during partially frozen storage. Chitosan-based coating treatments effectively inhibited bacterial growth, reduced total volatile basic nitrogen and malondialdehyde, and basically maintained the sensory properties of white shrimp (*P. vannamei*) during partially frozen storage. Therefore, chitosan-based edible coating combined with chitooligosaccharides and glutathione could be a promising antimicrobial and oxidant method to prevent metamorphism of white shrimp with extended shelf life.

1. Introduction

Shrimps are an important seafood with considerable nutritional and economic value in many countries worldwide [1]. Due to their high water content and nutritional components; shrimps are highly perishable; and they start deteriorating immediately after death. In view of their highly perishable nature; the fresh shrimps have to be processed to extend their shelf life for off-season use. Among the various methods employed for preservation; partially frozen is one of the common methods used for shrimps.

Chitosan, a polymer composed primarily of β-(1 → 4)-2-amino-2-deoxy-D-glucose (D-glucosamine) monomers, is derived by deacetylation of naturally occurring biopolymer chitin [2]. It is nontoxic and biodegradable and possesses a broad spectrum of antimicrobial activities against both gram-positive and gram-negative bacteria [1]. The inhibition effect on *Listeria monocytogenes* growth in meat, poultry, and seafood products by chitosan-based coatings has been explored [3–6]. Chitooligosaccharides, oligomers primary of β-1,4-linked 2-amino-2-deoxy-D-glucopyranose (GlcN), have many special biological, chemical, and physical properties, such as antifungal, antibacterial, and antitumor activities, which are different from the ordinary chitosan [7]. Glutathione has antioxidant activity and is known for its ability to inhibit the activity of PPO and browning [8]. Thus, chitosan-based coating combined with chitooligosaccharides and glutathione could have antibacterial and antioxidant activities and have the ability to inhibit the activity of PPO and browning.

Therefore, the aim of this study is to investigate the inhibitory effects of chitosan-based coating combined with chitooligosaccharides and glutathione on preservation of white shrimp (*Penaeus vannamei*) during partially frozen storage.

2. Methods and materials

2.1. Materials

Live white shrimps, which were from the same sources, and had similar properties, were purchased from a local agricultural market, Xinpu, China. Chitosan, as initial material from shrimp shells, was obtained from Nantong Biochemical Co. (Nantong, Jiangsu, China). The M_W and the degree of deacetylation were 41×10^4 Da and 93.5%, respectively. Glutathione were purchased from Fuchen Chemical reagents Co. (Tianjin, China). All other chemicals were of reagent grade.

2.2. Preparation of chitooligosaccharides

The chitooligosaccharides were prepared according to the methods described by Wu with a slight modification [9]. The raw chitosan was dissolved in 1% (v/v) aqueous acetic acid to a concentration of 1% (w/w) and the pH was adjusted to 5.5 using 1 M NaOH. A 40 mg mass of α-amylase was added into a reactor containing 500 mL of chitosan solution, maintained in a thermostatic water bath at 55 °C for 4 h, and then heated to 95 °C for 15 min to terminate the reaction. The hydrolysates were neutralized with

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1 M NaOH, filtered, concentrated to ~16% (w/v), precipitated with 5 volumes of ethanol, and dried at 60 °C for 3 h to yield a white powder.

2.3. Dipping and partly frozen storage conditions of shrimps

Dipping solutions of glutathione, chitooligosaccharides, and chitosan were: (1) control (deionized water) and (2) 0.8% glutathione + 1% chitooligosaccharides + 1% chitosan. The shrimps were shocked with crushed ice first and dipped into the two dipping solutions fully immersed for 5 min. Residual solutions on the shrimps were dripped off for 1 min, and the shrimps were kept at 5 °C until the excess of water was drained. Then these shrimps were placed on plastic-coated wire racks inside plastic containers at −3 °C for 30 days.

2.4. Microbiological analyses

For determining total viable count (TVC), minced white shrimp samples were homogenized with sterile phosphate-buffered saline solution to prepare sample suspensions. Pour-plate method using plate count agar was used to determine the total plate counts in samples. The inoculated agar plates were incubated at 30 °C for 48 h before counting the colonies [10].

2.5. pH measurement

The pH value was measured as described by Duan, Cherian, and Zhao with a digital pH meter (Model number PHS-3C Digital Precision PH Meter, Leici, Shanghai, China) [11].

2.6. Total volatile basic nitrogen (TVB-N)

For determination of TVB-V, partially frozen samples were thawed overnight in a refrigerator and then homogenized using a blender. A 10 g sample was washed into the distillation flask and 1 mg magnesium oxide (Merck, Darmstadt, Germany) was added with a drop or two of silicone antifoam solution (Fluka, Steinheim, Germany). Samples were boiled and distilled into 10 mL of 0.1 N HCl (Merck) solutions in a 500 mL conical flask with added tashiro-indicator (Riedel-de Haen, Seelze, Germany). After distillation, the contents of conical flask were titrated with 0.1 N NaOH (Merck) [12].

2.7. Sensory evaluation

Sensory evaluation of shrimp samples was performed by a group of six trained people on each sampling day. Panelists were asked to evaluate appearance, odor, texture, flavor, and overall acceptability of shrimp samples. A rating was assigned separately for each parameter on a 1-to-9 descriptive hedonic scale, with 9 as the highest-quality sample. A sample scored below 5 for a sensory attribute was considered unacceptable [1].

2.8. Statistical analysis

All experiments were replicated 3 times, with triplicate samples prepared for each experimental trial. All data are presented as mean ± S.D. *t*-Test was used to compare the means of two groups of the rat study. Statistical significance at the 95% and 99% probability levels were set at $P < 0.05$ and $P < 0.01$, respectively. Microsoft Excel (Microsoft Corporation, USA) was used for statistical analysis.

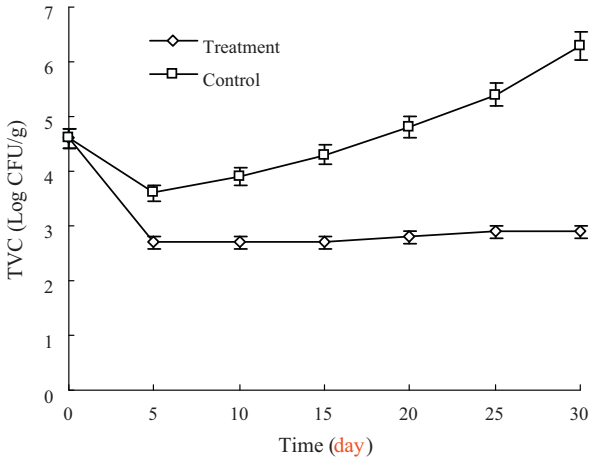

Fig. 1. Effect of chitosan-based coatings on the total viable count (TVC) of white shrimp (*Penaeus vannamei*) during partially frozen storage. Bars represent the standard deviation ($n = 3$).

3. Results and discussion

3.1. Effect of chitosan coating combined with chitooligosaccharides and glutathione on TVC during partially frozen storage

Fig. 1 illustrates the antibacterial effect of chitosan coating combined with chitooligosaccharides and glutathione and cryogenic freezing against bacteria in shrimp during storage. The initial TVC in shrimp was approximately 4.6 ± 0.21 log CFU/g. Both the treatment and control group significantly ($P < 0.05$) reduced TVC by 1.9 ± 0.08 and 1.0 ± 0.04 log CFU/g in 5 days of storage, respectively. This was expected as antibacterial effect of the chitosan coating combined with chitooligosaccharides and glutathione and ice crystals for the treatment group and antibacterial effect of ice crystals for the control group. However, TVC for the control group significantly ($P < 0.05$) increased up to 6.3 ± 0.29 CFU/g after 30 days of storage, while TVC for the treatment group remained unchanged.

3.2. Effect of chitosan coating combined with chitooligosaccharides and glutathione on pH during partially frozen storage

Though the treatment group had a higher initial pH value than the control group, the control group showed higher pH values than treatment group after 10 days of the storage (Fig. 2). For the control group, pH values significantly ($P < 0.01$) increased during storage (Fig. 2). In the case of the treatment group, pH values increased slightly. It was reported that the pH increased with time and there was a relationship between pH and acceptability for shrimps [13]. It was also reported that shrimp pH of 7.7 or less indicated prime quality, 7.70–7.95 showed poor but acceptable quality and 7.95 or greater represented unacceptable quality [14]. The increased pH could be related to the formation of volatile amines from microbial activity and enzymatic ammonia production during partially frozen storage of shrimp.

3.3. Effect of chitosan coating combined with chitooligosaccharides and glutathione on TVB-N during partially frozen storage

TVB-N is one of important indicators of chemical spoilage. The increase in TVB-N was related to the activity of spoilage bacteria and endogenous enzymes [15]. The changes in TVB-N values as affected by chitosan coating combined with chitooligosaccharides