



Quality enhancement in refrigerated red drum (*Sciaenops ocellatus*) fillets using chitosan coatings containing natural preservatives

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ABSTRACT

The present work was undertaken to examine the influence of grape seed extract (GS) and tea polyphenols (TP), as natural preservatives, combined with chitosan (Ch), on the quality of red drum (*Sciaenops ocellatus*) fillets during refrigerated storage. Two different treatments (Ch + GS and Ch + TP) and a control were prepared. The samples were stored (4 ± 1 °C) for 20 days and the sampling was done at 0, 4, 8, 12, 16, 20 days. Microbiological, physicochemical and sensory attributes were periodically assessed. The results indicated that the two pretreatments could more effectively maintain quality and could extend the shelf life by 6–8 days compared with the control group during refrigerated storage.

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1. Introduction

Red drum (*Sciaenops ocellatus*) is one of the most important marine-cultured fish species in China as it is fast-growing, strong in resisting virus and germs, easy to manage and adaptable to large-scale sea-resistant net casing aquaculture (Li, 2008). Red drum is an appreciated fish, not only for its culinary taste, but also for its health properties. Nevertheless, raw fish are highly perishable products and deteriorate due to protein degradation, lipid oxidation or decomposition caused by microbial or endogenous enzymes, resulting in a short shelf-life (Maqsood & Benjakul, 2010).

The use of edible coating could have a beneficial effect on the preservation of seafood products, since they function as a barrier against moisture and oxygen penetration (Pereira de Abreu, Losada, Maroto, & Cruz, 2010; Pereira de Abreu, Maroto, Villalba Rodríguez, & Cruz, 2012; Rodríguez-Turiénzo et al., 2011). Chitosan, a cationic polysaccharide mainly made from crustacean shells, is a well-known film-forming biopolymer with strong antimicrobial and antifungal activities (Aider, 2010; Duan, Cherian, & Zhao, 2010), which has been widely applied to the preservation of seafood products (Duan et al., 2010; Fan et al., 2009; Ojagh, Rezaei, Razavi, & Hosseini, 2010).

Natural preservatives, such as grape seed extract or tea polyphenols, have been broadly used in the food industry (Li, Hu, et al., 2012; Li, Li, et al., 2012; Moradi et al., 2012; Yi, Zhu, Fu, & Li, 2010). The preservative effect of grape seed extract and tea polyphenols is mainly due to their ability to scavenge free radicals (Fan, Chi, & Zhang, 2008; Mielnik, Olsen, Vogt, Adeline, & Skrede, 2006). In addition, grape seed extract and tea polyphenols also possess antimicrobial properties (Adams, Sivarooban, Hettiarachchy, & Johnson, 2005; Yi et al., 2010).

Development of natural preservatives and edible coatings with high antioxidant and antibacterial activities that prolong the shelf-life of food are valuable (Siripatrawan & Harte, 2010; Siripatrawan & Noipha, 2012). The objective of the present study was to evaluate the effects of grape seed extract and tea polyphenols combined with chitosan coating on the shelf-life of refrigerated red drum fillets by using microbiological, physicochemical and sensory assessment.

2. Materials and methods

2.1. Fish sample preparation

Fresh whole red drums (*Sciaenops ocellatus*) with an average weight of 570 ± 10 g were purchased from Dalian Aquatic Market during January and February 2012 (Dalian, Liaoning province, China). They were transferred to Food Processing Laboratory of Dalian Nationalities University within 0.5 h and kept alive before

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being processed. The fish were killed by slurry ice. After that they were decapitated and filleted by hand. Two fillets were obtained from each fish and kept at 0 °C until use.

2.2. Natural antioxidants and chemicals

Tea polyphenols were purchased from Zhejiang University Tea Scientific Co., Ltd (purity $\geq 98\%$, Hangzhou, Zhejiang province, China). Grape seed extracts were obtained from Shanxi Ruikang Biotechnology Company. (purity $\geq 95\%$, Xian, Shanxi province, China). The two extracts were packed in polyethylene bags and stored at 4 ± 1 °C until use. Chitosan, with a relative molecular weight (Mw) of 1.6×10^5 and 85% degree of deacetylation, was purchased from Ningbo Haixin Co., Ltd. (Ningbo, Zhejiang province, China).

2.3. Preparation of coating solutions and treatment of fish samples

Chitosan solution was prepared with 1.5% (w/v) chitosan in 1% (v/v) acetic acid. To obtain complete dispersion of chitosan, the solution was stirred at room temperature for 2 h. The solution in beakers was placed on a hotplate/magnetic stirrer, and glycerol was added to chitosan with an addition of 25% glycerol (w/w chitosan) as a plasticiser and stirred for 10 min. The resultant chitosan coating solution was filtered through a Whatman No. 3 filter paper to remove any undissolved particles. Fillet samples (6×13 cm) were randomly assigned into three groups including the control (uncoated) group and two groups treated with the following coating solutions: the fish fillets were given dip treatment in 0.2% (w/v) grape seed extracts solution (4 °C) or in 0.2% (w/v) tea polyphenols (4 °C) for 30 min and then drained well. After that, they were individually coated by immersing in chitosan solution for 10 min, and then fish fillets were removed and permitted to drain for 0.5 h at 4 °C on a sterile metal net, in order to form the edible coatings. For each group, approximately 30 red drum fillets were used. After that, they were packed in air-proof polyethylene pouches and stored at 4 ± 1 °C for subsequent quality assessment. Microbiological, physicochemical and sensory analyses were performed at 4-day intervals to measure the quality of fish.

2.4. Proximate composition analyses

A proximate composition analysis was performed on 5 fish on day 0 of storage. Proximate analyses (moisture content, total crude protein, ash content and lipid content) of the fish were based on procedures set by the AOAC (1997).

2.5. Microbiological analyses

Fish samples were taken aseptically in a vertical laminar-flow cabinet and 10 g were transferred to a stomacher bag; 90 ml of 0.1% peptone water with salt (NaCl, 0.85%, w/v) were added and the mixture was homogenised for 60 s with a stomacher. From this dilution, other decimal dilutions were obtained and 1 ml of three dilutions was transferred in triplicate to petri dishes containing 15 ml commercial plate count agar (PCA, Base Bio-Tech, Hangzhou, China). Total viable counts (TVC) were determined by counting the number of colony-forming units after incubation at 25 °C for 48 h.

2.6. Chemical analyses

2.6.1. Determination of pH

The pH values were measured to analyse the hygienic standard of fishery products according to the GB/T of the Chinese standard (GB/T 5009.45-2003). Minced fish sample (10.0 g) of red drum was mixed with 90 ml of distilled water and the mixture was

filtered. After 30 min, the pH values of the filtrate were measured using a digital 320 pH meter (Mettler Toledo, Zurich, Switzerland).

2.6.2. Total volatile basic nitrogen (TVB-N)

The total volatile basic nitrogen (TVB-N) values were evaluated by the FOSS method (2002). TVB-N values were measured with a Kjeltac 2300 (FOSS, Hiller, Denmark). TVB-N values were expressed in mg nitrogen kg^{-1} (mg N kg^{-1}) sample.

2.6.3. K-value

ATP and its breakdown products were analysed according to the modified method of Özogul, Taylor, Quantick, and Özogul (2000). Five grams of minced fish meat without skin were extracted with 25 ml of 0.6 M perchloric acid using an Ultra-Turrax (T25 basic; IKA-Werke, Staufen, Germany) for 1 min in an ice bath. The extraction mixture was centrifuged at 3000g for 10 min and then the supernatant was quickly neutralised to pH 6.5–6.8 with 1 M KOH using a digital 320 pH meter (Mettler Toledo, Zurich, Switzerland). The neutralised supernatant was allowed to stand for 30 min in an ice bath to precipitate most of the potassium perchlorate, which was then removed by centrifuging at 3000g for 10 min. The supernatant solution was made up to 50 ml and then stored at -80 °C until HPLC analysis. The identification of nucleotides, nucleosides, and bases was made by comparing their retention times with those of commercially obtained standards and by adding or spiking of standards. The K value was calculated as the percent amounts of HxR and Hx to the sum of ATP and degradation products as follows:

$$K\text{-value}(\%) = (\text{HxR} + \text{Hx}) / (\text{ATP} + \text{ADP} + \text{AMP} + \text{IMP} + \text{HxR} + \text{Hx}) \times 100.$$

2.6.4. Thiobarbituric acid reactive substances (TBARS)

The thiobarbituric acid value was determined colorimetrically by the method of Porkony and Dieffenbacher, as described by Kirk and Sawyer (1991). A portion (200 mg) of sample was weighed into a 25-ml volumetric flask. An aliquot (1 ml) of 1-butanol was added to dissolve the sample. The mixture was made to volume with 1-butanol and mixed. A portion (5.0 ml) of the mixture was pipetted into a dry stoppered test tube and 5 ml of TBA reagent (prepared by dissolving 200 mg of 2-TBA in 100 ml 1-butanol, filtered, stored at 4 °C for not more than 7 days) were added. The test tubes were stoppered, vortexed and placed in a water bath at 95 °C for 120 min, then cooled. Absorbance (A_s) was measured at 530 nm against water blank. A reagent blank was run and absorbance (A_b) recorded. TBA value (mg of malonaldehyde equivalents/kg of tissue) was obtained by the formula:

$$\text{TBA} = \frac{50 * (A_s - A_b)}{200}$$

2.7. Hardness and sensory evaluation

Texture profile analyses (TPA) were carried out according to Sigurgisladottir et al. (1999). The TA.XT texture analyser (Stable Micro Systems Ltd., Godalming, UK) was used. A flat-ended cylinder that simulated the human finger was used. Constant penetration depth of 2.5 mm was applied on fillets of about 15-mm thickness after testing penetrations in the range of 2–5 mm. This penetration depth was chosen as the maximum distance which could be applied without influencing the muscle structure by damaging it and leaving a mark on the fillet. Three sampling points were selected on each fillet [dorsal, tail (8 mm from the edge of tail) and between dorsal and tail]. Double compression was applied to construct the texture profile analyses (TPA) parameters. The flat-ended

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