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# Effect of sodium alginate-based edible coating containing different anti-oxidants on quality and shelf life of refrigerated bream (*Megalobrama amblycephala*)

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ABSTRACT

The effect of alginate-based edible coating containing Vitamin C (Vc) and tea polyphenols (TP) for shelf-life extension of bream (*Megalobrama amblycephala*) was evaluated over a 21-day storage at refrigerated temperature ($4 \pm 1$ °C). Bream were left untreated (CK), or were treated with alginate–calcium coating (T1), alginate–calcium coating incorporating 5% Vc (T2), or alginate–calcium coating incorporating 0.3% TP (T3). The fish samples were analyzed periodically for water loss, microbiological (total viable count), chemical (pH, total volatile basic nitrogen (TVB-N), thiobarbituric acid (TBA), *K*-value) and sensory characteristics. The results indicated that coating treatments retarded the decay of fish compared to uncoated bream. T2 more efficiently inhibited the growth of total viable counts than did T1 or T3 ($p < 0.05$). Coating treatments predominantly reduced chemical spoilage, reflected in TVB-N, pH, and TBA, retarded water loss ($p < 0.05$) and increased the overall sensory quality of fish compared to uncoated bream.



## 1. Introduction

Bream (*Megalobrama amblycephala*) is native to China, distributed originally to affiliated lakes of middle and lower reaches of the Yangtze River in China. In 1964 it was domesticated successfully, and gradually extended due to aquaculture. Now it has become one of the main farmed freshwater species in China. Endowed with excellent biological characteristics for rearing (fast growth rate, easy cultivation and high feed efficiency ratio), its farming in China is currently increasing. Statistical data show that more than 1,000,000 tons of bream were caught in China in 2009. Bream has high nutritional values, so it has been shown to be a convenient specie for commercial production, aiding in the diversification of freshwater aquaculture. It also has become a popular edible fish in China now. However, bream is usually more perishable than most other foodstuffs. Each year about 50,000 tons of bream are wasted. After it is harvested, its storage period is limited. Though low temperature can delay the rate of fish deterioration and also extend the shelf life of fish to some extent, the quality of fish muscle will still deteriorate during cold storage. Microbial activity and enzymes contained in fish tissues also degrade the muscle protein resulting in the quality loss of bream.

Deterioration of fish muscle mostly occurs in the fat-containing portions. The proportion of unsaturated fatty acids in bream fat is approximately 70%. These fatty acids are affected by the environmental oxygen that oxidizes and spoils the fish meat (Kilincceker, Dogan, & Kucukoner, 2009). So taking some measures to delay the decline of fish quality and extend the preservation life of fish through inhibiting, or retarding the growth of microorganisms and reducing the rate of lipid oxidation is necessary. Coating the foods with edible materials has been researched as an effective method to improve the food quality (Matuska, Lenart, & Lazarides, 2006).

Hydrophilic edible films are good barrier for oxygen and carbon dioxide and possess suitable mechanical properties at low relative humidity. Many studies have shown that edible coatings made of protein, polysaccharide, and oil-containing materials help to prolong the shelf life and preserve the quality of fish (Artharn, Prodpran, & Benjakul, 2009; Fan, Sun & Chen, 2009; Jeon, Kamil, & Shahidi, 2002; Sathivel, 2005; Stuchell & Krochta, 1995). But there is little literature about bream preservation so far.

Alginate is a salt of alginic acid, a polymer of D-mannuronic acid and L-guluronic acid, and is isolated from brown algae (Lu, Liu, & Ye, 2009). Alginate has unique colloidal properties and can form strong gels or insoluble polymers through cross-linking with $Ca^{2+}$ by post-treatment of $CaCl_2$ solution. Such biopolymer-based films can keep good quality and prolong shelf life of foods by increasing water barrier, preventing microbe contamination, maintaining the flavor,

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reducing the degree of shrinkage distortion and retarding fat oxidation. Alginate is a GRAS substance (FDA). Coating fish, shrimp, scallop and pork with sodium alginate showed that it can prolong their shelf life, reducing thawing loss, cooking loss, weight loss and maintaining the functional properties during frozen storage (Wanstedt, Seideman, & Donnelly, 1981; Wang, Liu, & Teng, 1994; Yu, Li, & Xu, 2008; Zeng & Xu, 1997). Moreover, the coatings may serve as carriers for antimicrobial compounds and antioxidant in order to maintain high concentrations of preservatives on the surface of foods. A few antimicrobial agents and antioxidant have been incorporated into edible coatings to suppress quality changes during storage (Chidanandaiah, Keshri & Sanyal, 2009; Fan, Chi & Zhang, 2008; Haque, Shon, & Williams, 2009; Kang, Jo, & Kwon, 2007). Furthermore , to meet consumers demands for safe foods, numerous studies are currently focused on using natural ingredients instead of synthetic preservatives (Gennadios, Milford, & Hanna, 1997). Vitamin C (Vc) and Tea polyphenols (TP) are both well-known natural anti-oxidants. They play an important role in fat oxidation and enzyme inhibition and demonstrate potential for their use as the preservatives and the anti-oxidants in food industry especially in the field of the preservation of manufactured meat. Vc can scavenge singlet oxygen and reduce oxygen- and carbon-centered radicals, resulting in dehydro-ascorbic acid formation (Gregory, 1996). Vc can also chelated metal ions. TP is a kind of Polyhydroxy organic matter extracted from tea leaves; it is a good hydrogen donor, can remove the original free radicals, resulting in the formation of relative stable free radical intermediate substances.

This research was to identify whether the addition of anti-oxidants into alginate–calcium coating solutions can keep good quality and extend the shelf life of bream through suitable coating formulations.

## 2. Materials and methods

### 2.1. Materials

Breams, (*Megalobrama ambycephala*; weight and length: $381.34 \pm 35.30$ g and $29.19 \pm 1.78$ cm, respectively) were purchased from a local market (Huilongguan market) in Beijing, China and transported to the laboratory alive, then immediately stunned, scaled, gutted and washed in water.

Food-grade sodium alginate (Ri-xing Seaweed Industrial Co., Ltd., Qingdao City, China) was used for the coating formulations. Calcium chloride (Yuanli Chemical Co., Ltd., Tianjin City, China) was used to induce the cross-linking reaction. Vc (Biodee Biotechnology co., Ltd., Beijing, China) and TP (Keyi Chemical Co., Ltd., Zhengzhou City, China) were added as anti-oxidants.

### 2.2. Preparation of the coating-forming solutions and treatments

Bream were divided into four coating formulations to which the following treatments were randomly assigned.

CK: control, untreated.
T1:1.5% sodium alginate/10% glycerin
T2: 1.5% sodium alginate/10% glycerin/5% Vc
T3: 1.5% sodium alginate/10% glycerin/0.3% TP

Sodium alginate solution was prepared by mixing 30 g of alginate with 1000 ml of distilled water and stirred at a controlled temperature of 80 °C until the mixture became clear. 500 ml solution containing nothing or 100 g Vc or 6 g tea polyphenols and 200 ml glycerin were mixed with the prepared sodium alginate solution and stirred thoroughly. Then the well-mixed solution was made up to 2000 ml with distilled water. Two percent (w/v) calcium chloride was also prepared. All solutions were cooled to room temperature prior to surface application onto fishes. Bream were immersed in the solutions for 1 min, air-dried for 1 min and then immersed in two percent (w/v) calcium chloride to gel for 1 min. They were then packed in polyethylene bags, tied off, and stored at $4 \pm 1$ °C for 21 days. Fish samples were taken randomly at intervals for analyzing.

### 2.3. Microbiological analysis

Total viable counts (TVC) were determined in plate count agar by the pour-plate method (AOAC, 2002). 25 g of fish portion was aseptically weighed and homogenized with 225 ml of sterile 0.1% peptone water for 1 min using a stomacher (FM200,fluko co., Ltd., Shanghai, China) at a speed of 6000 rpm. The homogenized samples were serially diluted (1:10) in sterile 0.1% peptone water. Samples (1 ml) of serial dilutions were plated onto plate count agar and then incubated at 35–37 °C for 48 h. Microbiological data were transformed into logarithms of the number of colony-forming units (CFU/g). All counts were performed in duplicate.

### 2.4. Water loss analysis

The water loss was estimated as described by Lu et al. (2009).The percentage weight loss relative to the initial weight was calculated by weighing the samples every 2 days in triplicate.

### 2.5. Chemical analysis

Proximate composition was determined by AOAC (2002) method.

#### 2.5.1. Determination of pH

10 g sample of fish flesh was dispersed in 100 ml of distilled water and stirred for 30 min, and then the mixture was filtered. pH value of filtrate was measured using a digital pH meter (Mettler toledo FE20/EL20, Shanghai, China)

#### 2.5.2. Determination of total volatile basic nitrogen (TVB-N)

The micro-titration method was employed to analyze TVB-N. 10 g sample of fish flesh was dispersed in 100 ml of distilled water and stirred for 30 min, and then the mixture was filtered. After the addition of 5 ml MgO (10 g/l) to 5 ml filtrate, the mixture was to distill through Kjeldahl Apparatus (KDY-9820, Beijing, China). The distillate was absorbed by 20 ml aqueous solution of boric acid (2%) containing a mixed indicator produced from dissolution of 0.1 g of methyl red and 0.1 g of methylene blue to 100 ml of ethanol. Afterward, the boric acid solution was titrated with a 0.01 mol/l hydrochloric acid (HCl) solution. TVB-N value was determined according to the consumption of hydrochloric acid.

#### 2.5.3. Determination of thiobarbituric acid (TBA) value

Thiobarbituric acid reactive substances (TBARS) were determined as described by Buege and Aust (1978) with some modifications. Fish flesh (5 g) was dispersed in 20 ml of thiobarbituric acid solution (0.375% thiobarbituric acid, 15% trichloroacetic acid and 0.25 mol/l HCl). The mixture was heated in boiling water for 10 min, cooled with water and centrifuged at 3600 g for 20 min at room temperature. The absorbance of the supernatant was measured at 532 nm (UV-2600, Shanghai, China). The standard curve was prepared using malondialdehyde (MDA) and TBARS were expressed as mg MDA/kg sample.

#### 2.5.4. Determination of K value

*K* value (as a percentage of the ratio between Ino + Hx to the total ATP and its degradation products) was determined as