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ORIGINAL PAPER

Effects of pectin/chitosan composite and bi-layer coatings combined with *Artemisia dracunculus* essential oil on the mackerel's shelf life

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Abstract

This study investigated the effects of pectin coating incorporated with chitosan (CH) and Tarragon essential oil (*Artemisia dracunculus*) (TEO) on the microbiological, physicochemical, and sensory properties of *Scomberomorus commerson* muscle during refrigerated storage. The results showed that pectin coating incorporated with CH and TEO could develop active coatings with excellent antibacterial activity to inhibit bacterial growth (mesophilic, psychrotrophic, and lactic acid bacteria). The initial mesophilic, psychrotrophic, and lactic acid bacteria ($\log_{10}$ CFU/g) in the all samples of fillet were 3.21, 4.03 and 1.23–1.72 $\log_{10}$ CFU/g. Pectin-CH coating on mackerel samples enriched with TEO could retard physicochemical properties and preserve the fish quality during refrigerated storage. There was a statistical difference between composite and bi-layer coatings (CC and BC) incorporating TEO ($P < 0.05$). The SDS-PAGE analysis of the samples during storage exhibited higher degradation of proteins (myosin heavy chain (MHC), paramyosin, actin, troponin T, and tropomyosin bands) in the control samples than in the treated samples. According to the quality attributes, the incorporation of essential oils or other biopolymers into edible coatings could decrease the bacterial and physicochemical deterioration of seafood during chilling.

Keywords *Scomberomorus commerson* · Pectin · Chitosan · Tarragon essential oil · Coating

Introduction

As an important new technology, active packaging prolongs the fish's shelf life due to delaying the oxidative degradation reactions of lipid and the growth of bacteria compared to other food packaging methods [1]. Active packaging in the food industry was of two types: edible coatings and edible films [2] introduced by forming a thin layer of edible material [3]. Various biodegradable polymers such as carbohydrates, proteins, and lipids have been used as bioactive substances to extend food products' shelf life [1]. Among carbohydrate biopolymers, polysaccharides such as pectin and chitosan (CH) are the common compounds for forming edible/biodegradable coatings [4, 5], which are used to improve antioxidant and antibacterial activities compared to single component-based coatings. Pectin, as a natural carbohydrate polymer derived from a plant cell wall [6], is a good barrier property against oxygen and lipids and can be used to make active packaging [5, 7]. This biopolymer is a non-toxic, biocompatible, and inexpensive natural polymer [7]. Pectin alone cannot reduce the growth of microorganisms and delay lipid peroxidation in seafood products because of its non-antibacterial activity and poor moisture barrier properties [8, 9]. However, several other findings also reflect the beneficial effects of pectin in food packaging [5, 9]. Antioxidant and antibacterial properties of edible pectin coatings can increase by combining other biopolymers such as CH. Pectin and CH are suitable materials to form composite and bi-layer coatings with higher performance. The mechanism of the interaction between pectin and CH is explained by electrostatic interactions between the negatively charged pectin (COO^-) and the positively charged side-chain groups in CH (NH^{3+}) [10]. CH, a linear polymer of β-(1–4)-linked D-glucosamine and N-acetyl-D-glucosamine, is a polysaccharide of natural origin derived from the deacetylation of chitin found widely in the crustacean and fungi [11]. A number of studies confirmed the use of antioxidant and antibacterial packaging systems of CH to prevent food oxidation and bacterial contamination [12–15]. However, no studied investigated the application of CH and pectin simultaneously.

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Recently, some researchers have indicated that composite and bi-layer coatings or films made of biopolymers could improve the quality of fish products [14, 16–18]. Nowzari et al. indicated no significant difference between composite coating (CC) and bi-layer coating (BC) concerning the quality properties of fillet [14]. The effectiveness of composite and bi-layer edible coatings can be enhanced by incorporating essential oils (EOs). Biopolymers are carriers of food additives such as EOs [19]. Synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) have been frequently applied in the food industry. Consumers now pay attention to preserving seafood products with natural preservatives like EOs. Incorporating EOs with biopolymers coatings or films has beneficial compounds compared to using them alone for seafood products because it helps gradually release antimicrobial and antioxidant agents to extend the fish's shelf life [20]. The addition of plant-derived EOs/extracts can help develop packaging materials to preserve food [21–24]. EOs consist of different volatile organic compounds obtained from the plant's secondary metabolism [25]. EOs could be used as potential sources of antioxidant and antimicrobial compounds on foods and as an alternative food packaging to synthetic additives. Tarragon essential oil (*Artemisia dracunculus*) (TEO) has been found to have high antioxidant and antimicrobial activities [26]. As researchers have announced, EOs cannot be used directly because of the interaction between volatile oxidation compounds and environmental factors such as light, oxygen, and heat [27]. Some scientific literature on shelf life extension and preservation of fishery products is available on applying biopolymers in hybrid systems and EOs [27–29]. The mackerel (*Scomberomorus commerson*; Scombridae), also known as "Sheer fish" in Persian, is the most popular fish with the highest economic value. Thus, this study aimed to study the effect of pectin/CH composite and bi-layer coatings containing TEO on the mackerel's shelf life during refrigerated storage (4 ± 1 °C).

Material and methods

Sample preparation

Biopolymers solutions were prepared separately. The CH solution was prepared with 1% (w/v) CH (Sigma Chemical Co., medium molecular weight, viscosity 200–800 cP) in 1% v/v acetic acid [17]. The CH solution was stirred at 25 °C to dissolve completely. Pectin from the citrus peel (Sigma–Aldrich) was dissolved in water (2% w/v). Glycerol as a plasticizer was added with the concentration of 0.75 ml/g and stirred for 10 min [17]. TEO was purchased from the Barij Essence Pharmaceutical Company (Iran).

The samples were randomly distributed into five groups, including:

(1) Control (un-coated)
(2) CC (composite coating): the fillets were immersed for 30 s in the composite CH-pectin solution and then allowed to stand for a 2-min period followed by a second immersion in the solution for 30 s.
(3) BC (bi-layer coating): the fillets were immersed for 30 s in the CH solution and then allowed to stand for a 2-min period followed by immersion in pectin solution for 30 s.
(4) CC + TEO: composite coating containing TEO.
(5) BC + TEO: bi-layer coating containing TEO.

Then, to form the edible coating, the fillets were taken out from the solution and allowed to drain at 4 °C for 1 h. The samples were preserved in plastic bags and then stored in a refrigerator for 16 days. Physicochemical, bacteriological, SDS-PAGE, and sensory analyses were performed at 4-day intervals to assess the mackerel quality.

Bacteriological analyses

The pour plate method was used in a plate count agar (PCA) to count bacteria [total mesophilic bacteria (TMB) and psychrotrophic bacteria (PTC)] that were incubated at 30 °C for 24–48 h, and 7 °C for 7 days, respectively. Lactic acid bacteria (LAB) was isolated using the pour plate method on the MRS agar by incubating at 30 °C for 72 h.

Physicochemical analysis

The total volatile bases nitrogen (TVB-N) content was examined using the distillation method based on Goulas and Kontominas's method [30]. The pH content of the fish muscle was examined using a digital pH meter (913 pH meter, Metrohm, Herisaw, Switzerland) [31]. The thiobarbituric acid reactive substances (TBARS) value was analyzed according to Siripatrawan and Noipha's method [32]. According to Woyewoda et al. [33], the free fatty acids (FFA) value of the fish samples was examined with lipid extract.

SDS–polyacrylamide gel electrophoresis (SDS-PAGE)

An amount of 0.5 g of fillet (without fat) was transferred to an appropriately labeled microfuge tube containing Laemmli sample buffer. The samples were vortexed for a few seconds and then were incubated for 5 min at room temperature to extract and solubilize the proteins. The buffer containing the extracted proteins was pipetted into a new 1.5 mL screw-cap tube. Fish protein samples were boiled, and the actin and

myosin samples were purified. Additionally, protein standards (ladder) were boiled to denature the proteins to perform electrophoresis. The polymerization changes of the modified proteins were determined by Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE). The samples were solubilized in 5% SDS (1:9, w/v) and dissolved in sample buffer with and without b-mercaptoethanol. Then, SDS-PAGE was carried out using 4% stacking gels and 10% running gels by the Laemmli procedure (1970) [34].

Sensory evaluation

Changes in sensory attributes of fillets, including color, texture, odor, and appearance, were evaluated during the storage. In this study, a five-point hedonic scale (five points: 5 = like extremely and 1 = dislike extremely) was used for sensory evaluation. Eight semi-trained members (age between 25 and 32) were selected for the sensory analysis of the samples.

Statistical analysis

The one-way analysis of variance (ANOVA) was used to determine whether there were any statistically significant differences between the means of the treatments by SPSS 16.0 for Windows (SPSS Inc., Chicago, Illinois, USA). Duncan's multiple test was applied to compare the means values of the groups. A p value of less than 0.05 was considered significant.

Results and discussion

The changes in the bacteria counts

The effects of the coating methods on the total mesophiles counts (TMC), psychrotrophic counts (PTC), and lactic acid bacteria (LAB) counts of the mackerel fillet during storage in the refrigerator are shown in Fig. 1. The mackerel fillet's initial TMC and PTC values were 3.21 $\log_{10}$ CFU/g and 4.03 $\log_{10}$ CFU/g, respectively. The low initial number of microbial loads indicated the good quality of the mackerel fillets. The result of initial bacteria is similar to that reported by Mohan et al. [35]. There was an enhancement in both mesophiles and psychrotrophic during the storage period. The growth pattern of TMC and PTC showed an increasing behavior during refrigerated storage (Fig. 1). The TMC and PTC values of the control samples increased more quickly than those in the coated samples (CC and BC), indicating that the coated samples with the CH-pectin solution inhibited the growth of bacteria. Although pectin may reduce bacterial growth, pure CH could enhance antimicrobial properties to decrease bacterial growth when added

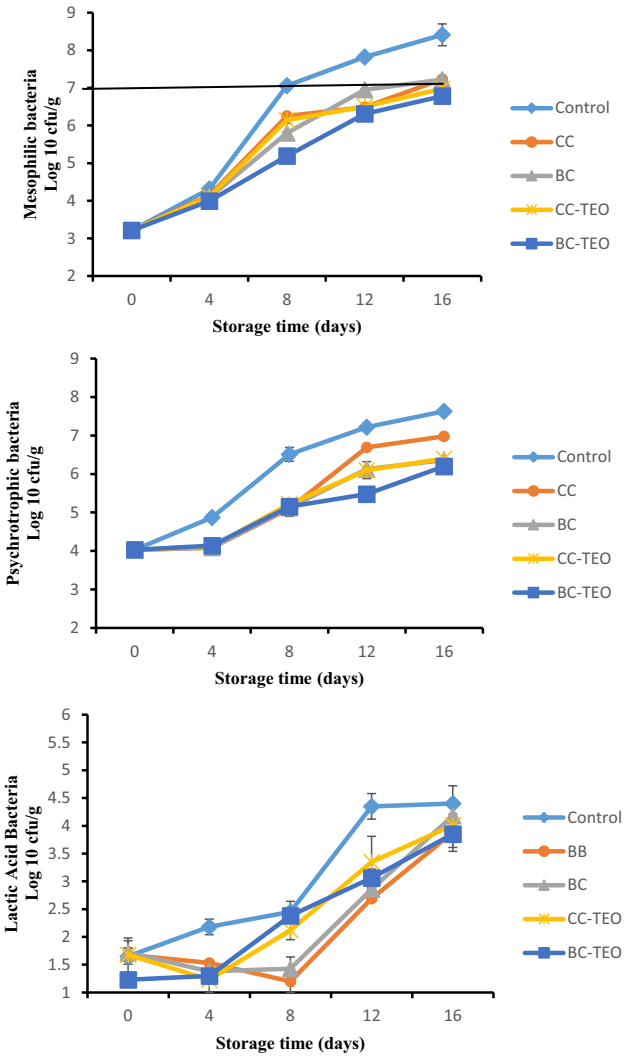

Fig. 1 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on mesophilic bacteria, psychrotrophic bacteria, and LAB counts of mackerel during storage at refrigerator

to the pectin solution. The antibacterial mechanism of CH was the interaction of positive charge on the NH_3^+ group of glucosamine monomer in CH molecules with negatively charged macromolecules on the microbial cell surface [17]. Moreover, CH acts as a barrier against oxygen transfer [36]. It was observed that the mesophilic and psychrotrophic bacterial growth rate in the CH-pectin coating solution was the same in two different coating methods (CC and BC). Similar results were reported by Nowzari et al. [14]. In line with this study, Saki et al. [16] showed that the bacteria population had no significant difference in the composite coated Belanger's croaker than in the bi-layer coated species. According to Pereda et al. [37], there was no significant difference between the antibacterial effect of blending or laminating gelatin and CH. All the treatments (BC, CC, CC-TEO, and BC-TEO) indicated a significant reduction in TMC and PTC

in mackerel compared to the uncoated samples. The lowest number of bacteria belonged to coated fillets with CH-pectin and TEO due to the synergism effect of antibacterial activity of the pectin-CH coating matrix and TEO. Therefore, incorporating TEO in the CH-pectin coating effectively delayed the growth of bacteria on mackerel fillets. TEO has strong antibacterial activity due to the presence of phenolic compounds. TEO is composed of monoterpene hydrocarbons (42.12%), oxygenated monoterpenes (5.69%), sesquiterpene hydrocarbons (4.20%), oxygenated sesquiterpenes (0.89%), aliphatic compounds (7.74%), aromatic compounds (0.24%), and other unidentified compounds (39.11%) [26]. The most abundant components of TEO are sabinene (19.19%), β-terpinene (8.94%), terpinen-4-ol (3.83%), and α-pinene (3.08%) [26]. The antimicrobial action of EOs has been attributed to: (1) the direct contact between bioactive components present in EOs and the phospholipid bilayer of the bacterial membrane that disturbed the structural integrity of the cell membrane and then influenced the cell metabolism causing cell death [38]; (2) the effects of polyphenols on hydrolytic enzymes, interactions with cell envelope transport proteins and nonspecific interactions with carbohydrates [39]; and (3) the inhibition of nucleic acid synthesis [40]. On the other hand, incorporating these bioactive compounds of TEO into a solution can improve its antibacterial activity by interacting with biopolymers and reducing the movement of antimicrobial agents into foods [27]. In agreement of with this result, Shahbazi and Khezrian and Shahbazi [41, 42] reported that incorporating EO into polymers reinforced antibacterial activity to decrease bacterial growth. According to Scudeler et al. [43], adding EOs into coating solutions can decrease seafood spoilage. Generally, the seafood's shelf life is estimated by TMB and TPC. The acceptable level for TMB of raw seafood is 7 log CFU/g [44]. From the bacteriological point of view, samples coated with BC-TEO and CC-TEO coatings were acceptable up to 16 days of storage. By day 12 of storage, TMC in the mackerel fillet became more than 7 $\log_{10}$ CFU/g for the BB and BC samples. TMB in the uncoated mackerel fillet was higher than the suggested limit (about 7 $\log_{10}$ CFU/g) in fresh fish after 8 days of storage, showing a bacterial shelf life of about 7 days for the uncoated groups.

The initial LAB count for all the groups ranged from 1.23 to 1.68 $\log_{10}$ CFU/g (Fig. 1). The LAB count was lower for the control and coated samples than for the other bacteria analyzed in the current study during the storage. The LAB count increased slowly in all the samples during the storage. Adding TEO into the CH/pectin solution did not provide a further reduction in the LAB of the treated samples, and there was no significant difference between the control and coated mackerel fillets during storage in a refrigerator. According to Burt and Kostaki et al. [38, 45], LAB has a higher resistance to EOs due its ability to face osmotic stress.

The changes of TVB-N value

TVB-N is a spoilage index to estimate raw fish's freshness [46]. TVB-N includes different compounds, including ammonia, primary, secondary and tertiary amines. Figure 2 shows changes in the TVB-N values of the control and coated samples during refrigerated storage. The initial content of TVB-N in all the samples was determined as 9.67 ± 1.26 mg/100 g in the fresh samples. The TVB-N value of the uncoated and treated groups was progressively enhanced with prolonging the storage time ($P < 0.05$), which could be attributed to the degradation of protein and non-protein nitrogenous compounds by the activity of the internal enzymes and microbial activity [47]. The TVB-N value was much lower in fillets coated with CH/pectin and TEO (CC-TEO and BC-TEO) than in those treated with CH/pectin (CC and BC). There was no difference between CC and BC, but a significant difference was observed between CC-TEO and BC-TEO ($P < 0.05$). The highest TVB-N content was shown in the uncoated sample, followed by fillet coated with CC and BC ($P > 0.05$). According to the European Commission [48], the highest recommended level of TVB-N is 35 mg/100 g. The TVB-N value of mackerel fillets treated with CC-TEO and BC-TEO remained below 29 mg/100 g at the end of storage. Khezrian and Shahbazi (2018) [42] reported that adding natural compounds to nanocomposite CH and carboxymethyl cellulose films prolonged the increase in TVB-N. The lowest TVB-N content of pectin/CH-TEO might be because of the oxygen barrier of pectin and CH and the antioxidant activities of CH and TEO, which reduced the protein decomposition caused by decreasing the growth of microorganisms (such as H2S-producing bacteria and *Pseudomonas* spp.) and the ability of bacteria for the oxidative de-amination of non-protein compounds [49]. In this study, the synergistic and additive effects of simultaneous incorporating TEO into the pectin/CH-based solution decreased the TVB-N content. Therefore, the pectin-CH edible coating added with TEO could effectively retard the spoilage of mackerel fillets.

The changes in pH value

At day 0, the initial pH was 6.37 ± 0.11, and it increased with storage time (Fig. 3). The pH value of the control and coated samples significantly increased over time ($P < 0.05$). Generally, after the fish's death, the pH value of fish fillet decreases due to the conversion of glycogen to lactic acid [50]. Various studies have reported that the pH value significantly increases by endogenous and microbial enzymes, which produce alkaline compounds such as ammonia, trimethylamine, and volatile basic compounds [51]. On the other hand, the seafood protein is rapidly decomposed under sufficient oxygen, which increases the pH value. The pH

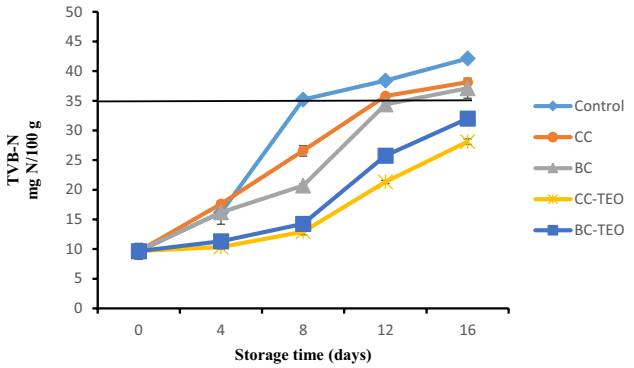

Fig. 2 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on TVBN value of mackerel during storage at refrigerator

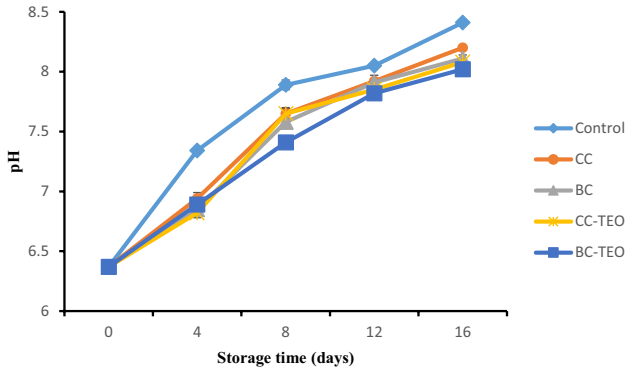

Fig. 3 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on pH value of mackerel during storage at refrigerator

content of the control, CC, BC, CC-TEO, and BC-TEO was 8.41, 8.20, 8.11, 8.08, and 8.02, respectively, on the last day of storage. The control sample indicated higher pH content compared to the other treated samples. In our study, the pH value of the treated samples did not significantly increase during the earlier storage ($P > 0.05$), which may be attributed to the limited microorganisms that can degrade proteins to produce volatile nitrogenous substances [52]. However, the pH value of the control sample rapidly increased to 8.41 on the 16th day. The lowest amounts of the pH value were enumerated in samples coated with CC-TEO, and BC-TEO ($P < 0.05$). The simultaneous incorporation of natural preservative agents (TEO) and CH into pectin-based coatings indicated the synergistic antimicrobial effects against bacterial growth and the decomposition of nitrogenous compounds. Similar results were reported by Xiong et al. [5]. Therefore, this study results confirmed that the bacteria population in the samples affected pH values, and thus, the accumulation of volatile bases by bacteria might be attributed to the increase of pH in the mackerel samples. Moreover, fresh mackerel can prolong its quality if coated with pectin/CH-TEO.

The changes in TBARS value

TBARS is used as an essential index to determine secondary compounds (Malondialdehyde (MDA)) produced from the oxidation of unsaturated fatty acids in the fillet [36], resulting in the unpleasant (rancid) taste and smell in seafood products and shortening of their shelf life.

A high level of dark muscle and mono and poly-unsaturated fatty acids of mackerel fillets led to fat oxidation. Changes in the TBARS value of the control and treated samples are presented in Fig. 4. All the samples' initial TBARS values were 0.25 ± 0.00 mg malonaldehyde equivalents/kg, which was similar to those reported by Silbande et al. and Kannaiyan et al. [53, 54]. The TBARS values of all the samples fluctuated during the storage period. The TBARS contents of all the samples continuously increased ($P < 0.05$) with prolonging the storage time. The TBARS content of control, CC, BC, CC-TEO, and BC-TEO was 2.25, 1.92, 2.02, 1.48, and 1.52 mg malonaldehyde equivalents/kg, respectively, at the end of the storage period. TBARS values were higher for the control samples than for the treated samples.

CH or pectin can significantly reduce the lipid oxidation of fillets due to oxygen barrier properties. However, pectin as biopolymers needs to improve antioxidant properties. Pure pectin coatings can combine with other biopolymers, or incorporate antioxidant agents such as EOs [5]. A significant difference in the TBARS value was observed when the CH was added to pectin treatments. It has been reported that CH has an effective antioxidant mechanism by chelating action metal ions and mixing with lipids [38] or forming a stable fluorosphere with the bind of MDA and primary amino groups of CH. This result suggested that lipid oxidation in the fillet could be decreased using EOs combined with CH due to the antioxidant properties. The antioxidant mechanism of TEO might be through the strong antioxidant activity with radical scavenging capacities with the antioxidant agent (polyphenolics compounds such as flavonoids and cinnamic acid derivatives) [55]. These results suggested that EOs supplied better conservation than pectin or CH on oxidation. The antioxidant activity of pectin or CH coating could be improved by adding TEO. Xiong et al. [5] reported that fresh pork loin coated with pectin + oregano EO had a lower TBARS value than the control sample during refrigerated storage [55]. Nisar et al. [56] showed a significant effect of the treatment in preventing lipid oxidation of bream coated with pectin stored at 4 °C for 15 days. According to Sallam and Alsaggaf et al. [57, 58], the maximum level of the TBARS value is 8 mg malonaldehyde/kg as the acceptable limit of fresh seafood

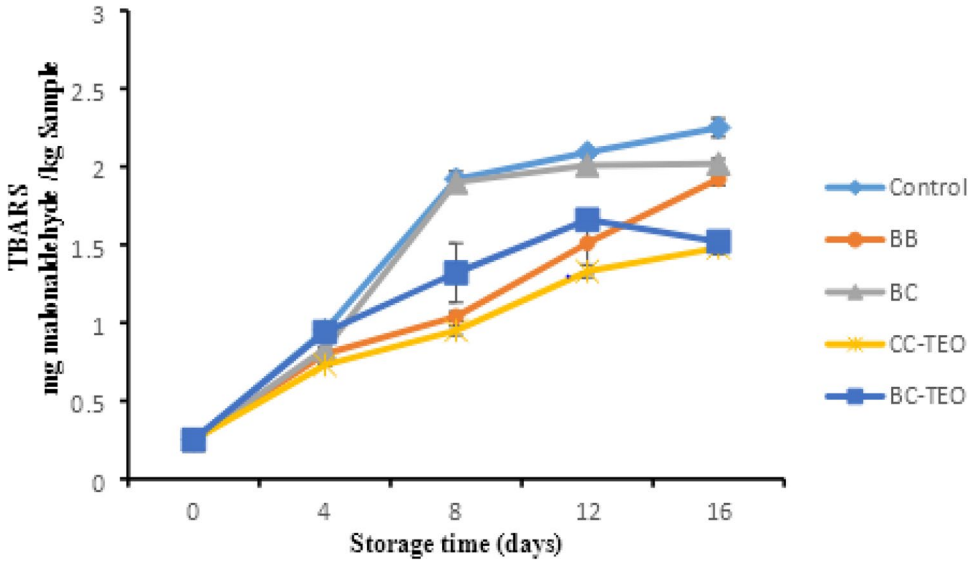

Fig. 4 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on TBARS value of mackerel during storage at refrigerator

products. Based on this study, samples coated with CC, BC, CC-TEO, and BC-TEO had lower contents than the recommended level at the end of the storage period, indicating the good quality of fillets regarding lipid oxidation during storage in ice. However, TBARS values were not in acceptable levels in the control samples, pectin treatments, and CH treatments. This result suggested that pectin incorporated with CH and EO could protect mackerel fillets from lipid oxidation.

The changes in FFA value

Changes in the FFA contents of mackerel fillets during storage are depicted in Fig. 5. The initial FFA of all the samples was $1.28 \pm 0.01\%$ oleic acid. The FFA content of control, CC, BC, CC-TEO, and BC-TEO was 25.41%, 16.52%, 15.70%, 11.96% and 11.88% of oleic acid, respectively, on the 16th day of storage. Generally, an increasing trend of FFA is shown during storage. The lowest FFA content was noticed in the treated groups compared to the uncoated groups. The reduction of the FFA value in the coated samples could be due to the powerful antioxidant and antibacterial activity of TEO, which reduced the growth and reproduction of PTC, especially *Pseudomonas* spp., and also, the inhibition of the enzymatic hydrolysis of the esterified lipids in the muscle. Significant synergistic antimicrobial and antioxidant effects were observed between TEO and CH against spoilage bacteria and lipid oxidation. The effects of plant extract on pectin coatings are in agreement with those of Xiong et al. [5], who found that the addition of oregano EO into pectin coatings reduced the FFA value in the pork loin due to improving the antioxidant properties of the coating.

SDS- polyacrylamide gel electrophoresis (SDS-PAGE) pattern

Changes in fish protein affect functional and textural characteristics. Such changes and the reduction of myofibrillar proteins during storage in the refrigerator are the evidence of proteolytic activity in the muscle [59]. Major muscle proteins are myosin and actin, contributing to most of the functional properties [35]. Thus, a reduction in myofibrillar proteins during storage is the evidence of proteolytic activity in the muscle. As shown in Fig. 6, five major protein bands, corresponding to myosin heavy chain (MHC), paramyosin, actin, troponin T, and tropomyosin bands, were observed in the mackerel muscle. The band intensity of these proteins in all the treatments decreased with increasing the storage time. After 16 days of storage, the protein band intensities of MHC, paramyosin, actin, troponin T, and tropomyosin in samples treated with CC-TEO and BC-TEO were markedly higher than those obtained for the samples CC and BC. TEO can postpone the oxidation of proteins due to the excellent antioxidant role of TEO in fish preservation.

Sensory analysis

The results of sensory attributes such as taste, color, odor, and overall acceptability evaluations of uncounted and coated mackerel fillets are shown in Fig. 7. The parameters were scored from 0 to 5, based on the observed differences in their characteristics. The sensory parameters are essentially helpful in giving a suitable description of changes in fillets during refrigerated storage. Sensory scores of > 3 of groups were observed to be acceptable for human consumption. Some spoilage signs could be

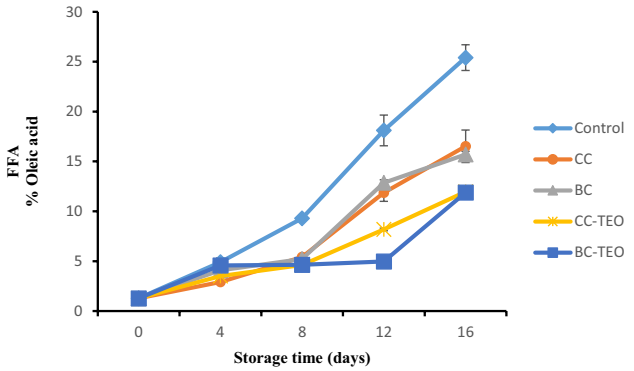

Fig. 5 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on FFA value of mackerel during storage at refrigerator

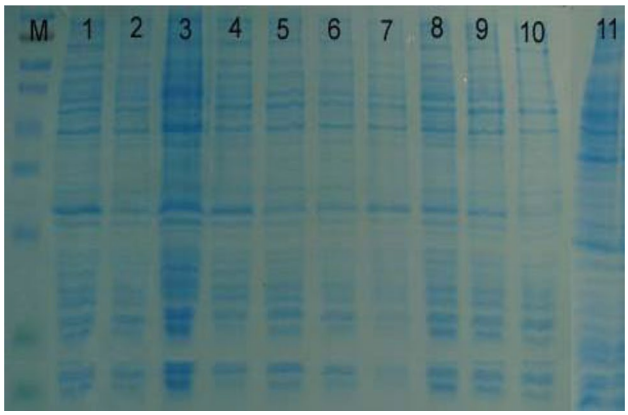

Fig. 6 Electrophoresis patterns of SDS-PAGE gels of actomyosin from mackerel fillets during refrigerated storage. *MHC* myosin heavy chain, *A* actin, *TM* tropomyosin, *MLCs* myosin light chains, *MW* molecular weight markers. 10 µg of protein (actomyosin) was loaded in each lane of the gel. M: Marker; 1: Control (Day 16); 2: Control (Day 8); 3: Control (Day 0); 4: CC (Day 8); 5: CC (Day 16); 6: BC (Day 8); 7: BC (Day 16); 8: CC-TEO (Day 8); 9: CC-TEO (Day 16); 10: BC-TEO (Day 8); 11: BC-TEO (Day 16)

noticed, such as putrid odor, no shiny color, and overall unacceptability of seafood fillet. Some researchers showed that a linear correlation between sensory parameters and storage time in chilling might be used to readily predict the remaining shelf life due to subsequent microbiological analysis. The scores given by the semi-trained panelists displayed that mackerel fillets have high quality (at 0 to 3 days of refrigerated storage). The oxidation of lipid and proteins with haemo groups (hemoglobin and myoglobin), non-enzymatic browning reactions between lipid oxidation products and the amine groups in proteins, and bacterial spoilage could cause color loss [32]. A significant difference in overall acceptability was not observed between pectin coating and the control. Incorporating CH and TEO into the pectin solution led to an increase in the panelists' scoring of flavor and smell of the treated samples compared to those of the untreated samples during the storage. Pectin/CH + TEO coating best maintained overall acceptability compared to others. Nisar et al. [56] showed that adding EOs to pectin-based coatings could extend the overall acceptability of bream fillets. Khezrian and Shahbazi and Saki et al. [16, 42] showed that the use of polysaccharide-based antibacterial coatings incorporated with the pomegranate peel extract led to a significant enhancement in the sensory attributes of meat. This sensory evaluation result is in good correlation with those of bacterial counts, TVB-N contents, and lipid oxidation. The least appropriate treatments for long-term storage were with pectin/CH + TEO coating, suggesting that pectin and CH coatings along with TEO could decrease the growth of off-odors producing bacteria.

Conclusion

It was observed that incorporating TEO as a natural antioxidant and an antibacterial agent into the pectin/CH solution effectively extended the mackerel fillet's shelf life by 3 days compared to the uncoated samples. The pectin/CH composite and bi-layer coatings combined with TEO improved various physicochemical (TBARS, FFA, pH, and TVB-N) and bacterial (TMC and PTC) parameters during the storage. The results suggested that biopolymer composite coating containing EOs is a good bioactive packaging that can be employed as a natural preservative for seafood during refrigerated storage.

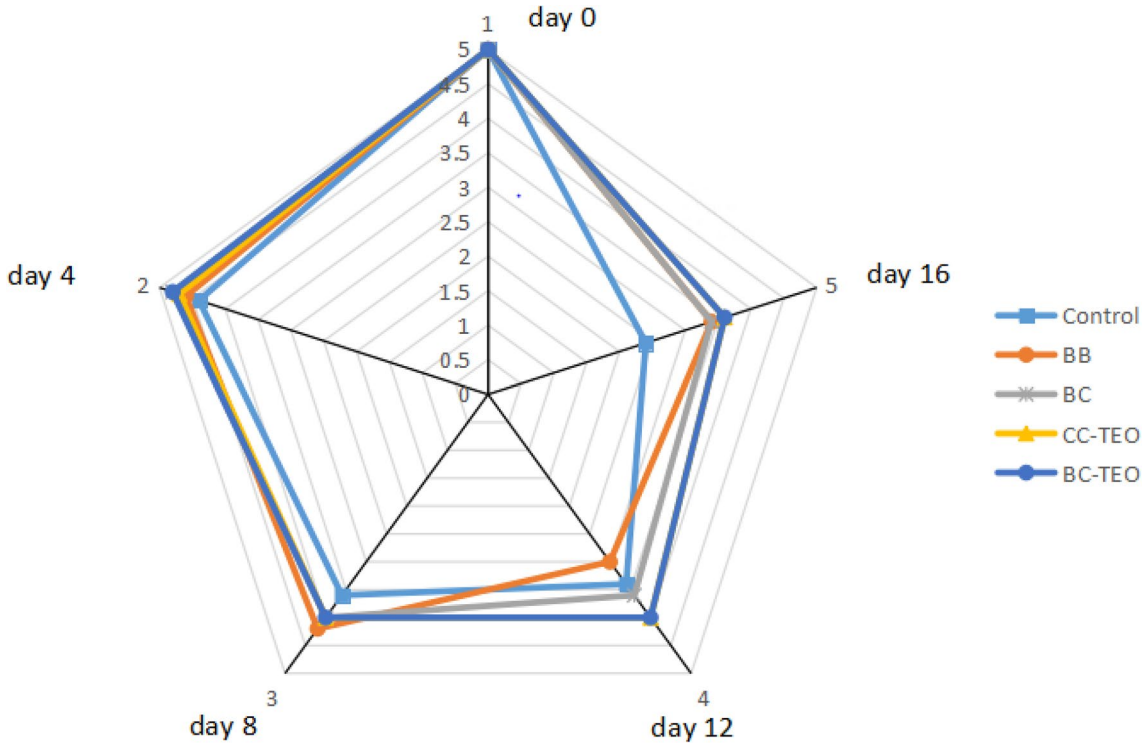

Fig. 7 Effect of pectin/chitosan composite and bi-layer coatings incorporating TEO on sensory analysis of mackerel during storage at refrigerator

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Declarations

Conflict of interest The authors declared that they have no conflicts of interest to this work.

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