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BOVINE AND FISH GELATIN COATINGS INCORPORATING TANNINS: EFFECT ON PHYSICAL PROPERTIES AND OXIDATIVE STABILITY OF SALMON FILLETS

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A B S T R A C T

Fish gelatin provides an alternative source of gelatin for Halal and Kosher applications and is prion and zoonotic agent free. However, applications of fish gelatin have been limited due to inferior mechanical and barrier properties. The physical properties of fish gelatins can be improved by crosslinking using natural polyphenolic compounds such as tannic acid. The objectives of this study were to develop fish gelatin films incorporated with tannic acid and evaluate their antioxidant, thermal, tensile, water vapor permeability and water solubility properties. Also, the effect of tannin-incorporated gelatins on the oxidative stability of salmon fillets was examined at 4 and 10 °C. Comparative data with bovine gelatins were generated. Fish gelatin (6.75% wt/wt) films were prepared at a gelatin:tannic acid ratio (wt/wt) of 1:0.05, 1:0.10 and 1:0.15. Tensile strength of bovine gelatin control was approximately 49 MPa and that of fish gelatin was 21 MPa. Tensile strength of bovine gelatins increased with tannic acid incorporation ($P > 0.05$) and did not vary significantly for fish gelatin films. Percent elongation of films increased and elastic modulus decreased with tannic acid incorporation. Water solubility of bovine gelatin films was reduced significantly ($P < 0.05$) and there was no significant effect of tannin on the solubility of fish gelatin films. The water vapor permeability was not significantly different for both the gelatins ($P > 0.05$) and the values ranged between 1.62 and 2.01 g mm/kPa h m^2. Bovine and fish gelatin films with highest level of tannic acid showed an increase in glass transition temperature of approximately 12 and 6 °C, respectively. Films with tannic acid possessed antioxidant activity and were able to reduce oxidation (TBARS values) in gelatin coated refrigerated salmon held for 12 days.

Keywords: fish gelatin, bovine gelatin, salmon, tannin, tannic acid, antioxidant, lipid oxidation.

INTRODUCTION

Shelf life of meat and seafood can be extended if microbial growth and oxidative reactions are limited. As consumer rejection of synthetic additives is becoming more common, natural preservative systems are sought. Due to high water activity, neutral pH, presence of autolytic enzymes and relatively high concentration of free fatty acids, fish such as salmon can be spoiled easily (Duna *et al*., 2010). Storage under controlled atmosphere can be effective for fish. For instance, in muscle foods stored under modified atmosphere, the microbial counts were well within the limit for 3 weeks, but deterioration in muscle color was observed after 12 days (Antionewski *et al*., 2007). Lipid oxidation can lead to off odors due to rancid volatiles, loss of muscle color due to oxidation of oxymyoglobin and drip accumulation (liquid oozing out of stored muscle) leading to deterioration in appearance (Antionewski *et al*., 2007). Fish muscle contains polyunsaturated fatty acids (He *et al*., 1997) such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Several health benefits are associated with these fatty acids; however, these foods are easily susceptible to oxidation and strategies to control oxidation would be beneficial for extending the shelf life for these perishable foods.

Use of antioxidant systems such as plant phenolics and plant extracts on fish fillets to reduce oxidation was investigated. Direct treatment of fish fillets with antioxidants were reported in several studies. Vacuum packed sardines (rosemary extract) (Ozogul *et al*., 2010), bonito fillets (grape seed and green tea extracts)

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(Yerlikaya and Gokoglu 2009), blue sprat (tea polyphenol) (Seto *et al*., 2005) and salmon fillets (isoeugenol solution) (Tuckey *et al*., 2009) were treated with antioxidants to retard oxidation. Instant green tea powder was added to mackerel fillets (Alghazeer *et al*., 2008), and green tea extracts were added to minced mackerel muscle (He *et al*., 1997; Tang *et al*., 2001) and minced carp (Dembele *et al*., 2010) to extend fish shelf life. Solutions of chitosan with vitamin E were able to reduce oxidation in lingcod fillets (Duna *et al*., 2010).

Use of edible coatings and films incorporated with active components (such as antioxidants or antimicrobials) will result in prolonged activity of the active components on foods than direct application by dipping or spraying (Min *et al*., 2005). Edible films can act as barriers to oxygen, water and limit lipid oxidation. Muscle foods can be wrapped in a preformed gelatin film, dipped or sprayed with gelatin film forming solution. On industrial scale, dipping muscle foods in gelatin solutions seems to be more practical. Significant improvements in oxidative stability of fish fillets dipped in chitosan-megrim skin gelatin solution (Caballero *et al*., 2005) and gelatin-benzoic acid solution (Ou *et al*., 2002) have been observed. Treated plastic films are also effective. For example, low density polyethylene films with barley husk extracts were able to reduce the TBARS values of salmon fillets stored for 12 month at -20 °C (Abreu *et al*., 2010). Chitosan microcapsules with horseradish extract coated onto ethylene vinyl acetate film (Jung *et al*., 2009) resulted in an extended shelf life of Spanish mackerel stored at 5 °C for 9 days due to reduced oxidation.

Bovine and porcine gelatins are well studied for use as coatings to extend quality of meat and seafood products. Recently, fish gelatin gained importance as an alternative source to mammalian gelatin due to socio-cultural and safety/sanitary reasons (Gomes Estaca *et al*., 2009). Fish processing waste can account to about 75% of total catch and 30% of this consists of fish skin and bones with gelatin (Gomez-Guillen *et al*., 2002). However, use of fish gelatins is limited as they have inferior rheological properties due to low number of proline and hydroxyproline residues and fewer inter and intra chain crosslinks compared to mammalian gelatins (Gomez Estaca *et al*., 2009). These films are naturally hydrophilic and absorb large quantities of water, resulting in more plasticization and inferior properties, which affect the applications of these films for extending shelf life of muscle foods.

Gelatins can be crosslinked with a variety of cross-linking agents. Most of these studies are intended to reduce solubility of gelatins and enhance strength properties. Crosslinking will result in the formation of new covalent bonds between reactive groups leading to enhanced physical properties. Often, plant phenolics are a good choice as crosslinkers as they are natural compounds and also possess antioxidant activity. Tannic acid is a plant phenolic with multiple phenolic groups and can react with proteins resulting in improved film forming ability. Crosslinking bovine gelatin films with tannic acid (Zhang *et al*., 2010b) resulted in reduction of film solubility by about 80%. Equilibrium moisture uptake of crosslinked films was lower than that of the control films. However, crosslinking did not limit the water uptake of films. Other crosslinkers including enzymes like transglutaminase (Piotrowska *et al*., 2008), white grape juice and coffee (Strauss *et al*., 2004), phenolic extract from Acacia bark (Haroun and Toumy, 2010), genipin (Bigi *et al*., 2002) and caffeic acid (Zhang *et al*., 2010a) were used to crosslink food grade gelatins. These systems reduced the water solubility and altered properties of films to different extents. From the scope of the literature, use of fish gelatin coatings and films to extend shelf life of fish products has been limited. The objectives of this work is to develop gelatin coatings incorporated with tannins and investigate the effect on fish gelatin properties and suitability of these as antioxidative coatings on salmon fillets.

MATERIALS AND METHODS

Film preparation with tannic acid: Commercial cold water fish gelatin (Norland Fish Products, Cranbury, NJ) and bovine skin gelatin (Sigma, St. Louis, MO, USA) were used in film preparation. Film forming solution (FFS) was formulated to contain 6.75% gelatin, and tannic acid (Sigma, St. Louis, MO, USA) at concentrations of 0, 5, 10 and 15% (wt/wt of gelatin). Initially, gelatin and tannic acid solutions were prepared separately by dissolving in warm deionized water. Tannic acid solution was added, in 8-10 increments, to the gelatin solution while being stirred at approximately 70 °C for 30 min. The FFS was denatured in 90 °C water bath for 30 min and cooled to room temperature. Glycerol (Sigma, St. Louis, MO, USA) was added to the FFS at a concentration of 25% (wt/wt of gelatin) followed by degassing. Amount of dry solids in each film preparation was kept constant (3 g) to maintain uniform film thickness. The FFS was poured

into Teflon plates and left to dry for 24 h at 25 °C, 15% RH. Dried films were stored at 50% RH until use.

Tensile properties of films: Tensile properties including tensile strength (TS), elastic modulus (EM) and percent elongation at break (%E) were measured using standard method D 882-01 (ASTM, 2001). To facilitate mounting on the clamps of a texture analyzer (TA-XT2, Stable Micro System Ltd., Surrey, UK), the prepared films were cut into 50 mm (long) x 8 mm (wide) strips and conditioned in a 50% RH chamber for 2 days at 22±2 °C before testing. A 5-kg load cell and crosshead speed of 50 mm/min was used for determining the tensile properties.

Water vapor permeability of films: Modified Gravimetric Cup method based on ASTM standard E 96-92 (McHugh *et al*., 1993) was used to determine the water vapor transmission rate (WVTR) of films. Desiccation chambers fitted with fans to attain air velocity of 152 m/min were maintained at 0% RH and 22±2 °C. Polymethylmethacrylate circular test cups with lid and screws were filled with deionized water (6 ml). Circular discs were cut from the films and placed in between the circular test cup lids. They were screwed tight to form a seal. Reduction in weight due to loss of water was recorded every 2 hours for 8 hours and the last reading was at 24th hour. Calculated water loss with time was divided by cup area (m^2) to give water vapor transfer rate (g/h-m^2). Permeance (g/kPa-h-m^2) was obtained by dividing water vapor transfer rate with partial pressure at the inner surface of the film. Permeance was multiplied by average film thickness to yield water vapor permeability (WVP) (g-mm/kPa-h-m^2).

Water solubility of films: Film soluble matter was determined by the modified method of Zhang *et al*. (2010b). Film (1 g) was taken in a centrifuge tube and 50 ml water at 90 °C was added to it. The centrifuge tubes were immersed in water bath at 90 °C for 30 min. The tubes were then cooled and centrifuged (Sorvall Legend Mach 1.6, Thermo Scientific, Waltham, MA) for 15 min at 2000 *g*. The supernatant was discarded and the pellet was dried for 6 h at 106 °C. The % soluble matter was the difference in the weight of the film taken and the dried pellet times one hundred.

Glass transition temperature (T_g) of films: Glass transition temperatures of films were measured using a Modulated Differential Scanning Calorimeter (MDSC) (DSC Q200, TA Instruments, New Castle, DE). Sample (10±1 mg) was taken in standard DSC pan, placed in the furnace with nitrogen flow rate of 20 ml/min. Sample was subjected to heating rate of 10 °C/min between temperature range of -50 to 150 °C. Heat flow data was collected and analyzed using instrument software (Universal Analysis 2000, v. 4.3, TA Instruments, New Castle, DE) to calculate the T_g.

Antioxidant activity of films: Film antioxidant activity was determined following DPPH (2,2-diphenyl-1-picrylhydrazyl) assay (Bao *et al*., 2009) using 0.072 mM/L DPPH reagent in ethanol. Films were stored in a 50% RH chamber at 23±1 °C and sampled on day 0, 10 and 20. Percent DPPH quenched was calculated by [1-(AS-AO)/AC)]*100); where AS, AC, AO are the absorbance values of sample, control, and solution of 5.5 ml ethanol and 500 µl sample, respectively.

Effect of tannin incorporated gelatins on the oxidative stability of salmon fillets: Atlantic salmon fillets (*Salmo salar*) were obtained from a local retailer (Pullman, WA). Fillets were sliced using a sterile knife into pieces of approximately 15±0.5 g and stored at 3°C for one day until use. Pieces of same shape and size were sliced so that all pieces would have similar surface area (approx. 25 cm^2). Fillet pieces were dipped in fish and bovine gelatin solution with tannic acid. Samples were dried under a fan for 15 min. After drying, samples were placed in foam meat trays (Genpack, Gens Falls, NY) and covered with a Saran® wrap to mimic packaging in retail display. Samples were stored at 4 and 10 °C for 12 days and analyzed for TBARS every third day. Treatments included uncoated fillets (positive control), fillets dipped in control FFS (negative control) and fillets dipped in FFS solutions with 5, 10 and 15% tannic acid.

Modified protocol for TBARS assay was adopted from Kim *et al*. (2012). Briefly, 15±0.3 g fish fillet was blended (Waring Blender, Model HGBTAC30, Warring Commercial, Torrington, CT) with 40 ml of 1.5% trichloroacetic acid (Sigma, St. Louis, MO) for 15 seconds. Blended mixture (20 ml) was taken and centrifuged (Sorvall Legend Mach 1.6, Thermo Scientific, Waltham, MA) for 15 min at 3300 *g*. Supernatant (2ml) was added to 2 ml of 20 mM thiobarbituric acid (Sigma, St. Louis, MO) solution. The mixture was heated in a 95 °C water bath for 30 min and cooled to room temperature. Solution was filtered through 0.45 µm Whatman (Maidstone, Kent, UK) GD/X syringe filters. The absorbance of samples was taken at

532 nm and TBARS values (mg MDA/Kg fish) were calculated using the extinction coefficient.

Statistical analysis: Pooled ANOVA assuming randomized complete block design was used for salmon oxidation studies. Data analysis was done using statistical analysis software version 9.2 (SAS Institute, Cary, NC). For determining TBARS, each treatment consisted of triplicate samples at every time point. Pair wise comparisons equivalent to Fishers LSD were used for physical properties of the films. Each mean ± standard error was the average of 2, 5, 6, 8 and 10 replicates for glass transition temperature, DPPH radical scavenging activity, water vapor permeability, tensile properties and water solubility of films, respectively.

RESULTS AND DISCUSSION

Tensile properties: Incorporation of tannic acid to gelatin films altered tensile properties. Tensile strength increased ($P > 0.05$) and % elongation decreased ($P > 0.05$) with increasing concentration of tannic acid in bovine gelatin films (Fig 1, 2). In case of fish gelatin films, there was a slight decrease ($P < 0.05$) in tensile strength compared to control. Percent elongation of fish gelatin samples with intermediate concentrations of tannic acid (5% and 10%) increased ($P > 0.05$) compared to control and decreased ($P < 0.05$) for films with 15% tannic acid. Increase in the TS and decrease in the % elongation of crosslinker-incorporated gelatin films has been reported by other authors (Cao *et al*., 2006; Kim *et al*., 2005; Rivero *et al*., 2010). Increase in tensile strength can be due to the crosslinkers stabilizing the film matrix, which in turn decreases film elongation. Incorporation of tannic acid increased ($P > 0.05$) film stiffness (Fig 3), which is evident by an increase in the elastic modulus of bovine and fish gelatin samples.

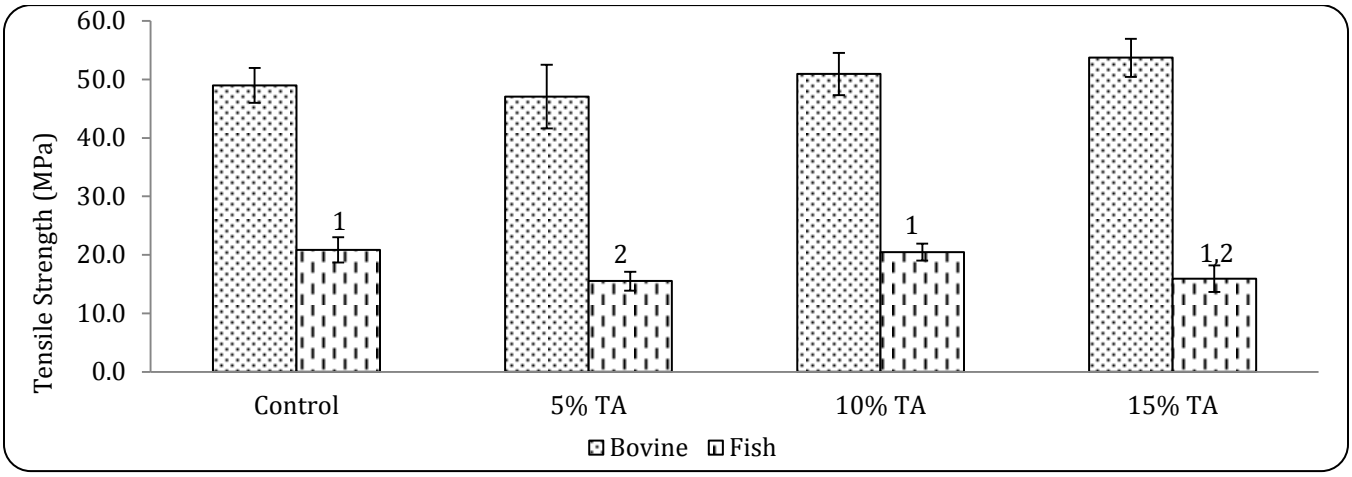

Figure 1. Tensile strength (MPa) of tannic acid incorporated bovine and fish gelatin samples. Values represent means ± standard error of means (n=8). Means with same numbers and without numbers are not significantly different ($P > 0.05$).

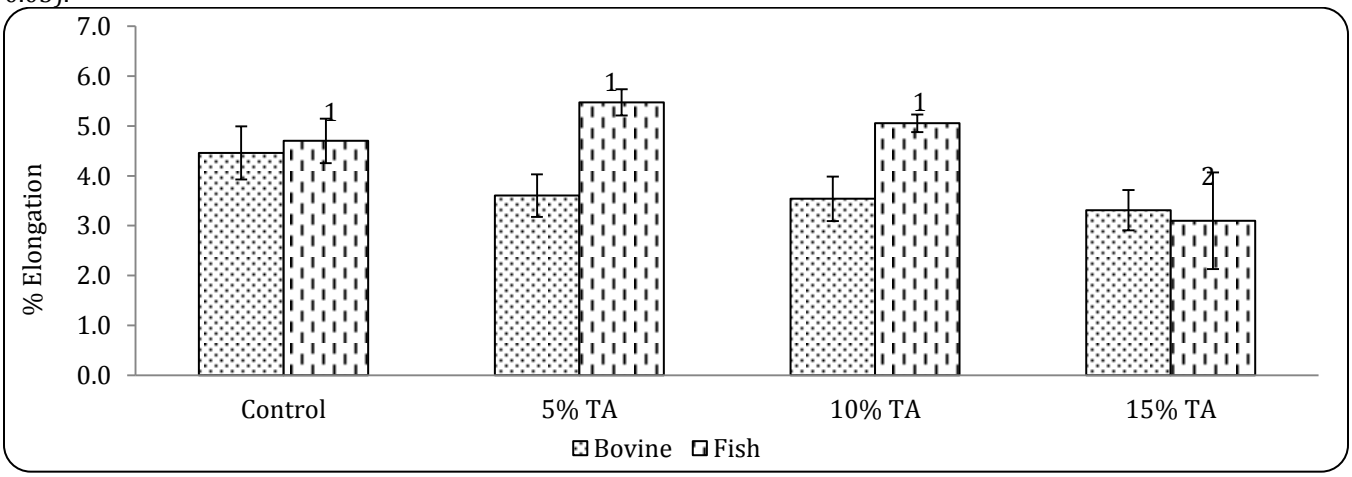

Figure 2. Percent elongation of tannic acid incorporated bovine and fish gelatin samples. Values represent means ± standard error of means (n=8). Means with same numbers and without numbers are not significantly different ($P > 0.05$).

Tensile strength increased from 84 to 88 MPa as the concentration of tannic acid increased from 0 to 40 mg/g gelatin in bovine gelatin films (Cao *et al*., 2006). The same study showed a reduction of approximately 2 MPa in the elastic modulus of the samples with increase in tannin concentration. Kim *et al*. (2005) reported an increase of tensile strength from 44 to 68 MPa and no significant change in the elastic modulus of the samples with the incorporation of condensed tannins in gelatin chitosan matrix. Rivero *et al*. (2010) reported an increase in tensile strength of chitosan films incorporated with tannic acid. Crosslinkers change physical properties to varying degrees. However, direct comparison of data among different gelatin samples is not reliable due to different formulations, test conditions and gelatin composition.

Tensile strength and elastic modulus of bovine gelatin samples are significantly higher compared to fish gelatin samples (Fig 1, Fig 3). This can be explained by the difference in the amino acid composition. Proline and hydroxylproline content is higher in bovine gelatin compared to fish gelatin (Avena-Bustillos *et al*., 2006, 2011; Gomez-Gullien *et al*., 2007; Gomez-Estaca *et al*., 2009). These amino acids stabilize the triple helix structure resulting in increased tensile strength and elastic modulus. Hydroxyproline has major role in stabilizing the triple helix structure due to H-bonding to its hydroxyl group. High content of proline and hydroxyproline is believed to be the reason for highly viscous properties for mammalian gelatins (Aveena-Bustillos *et al*., 2006).

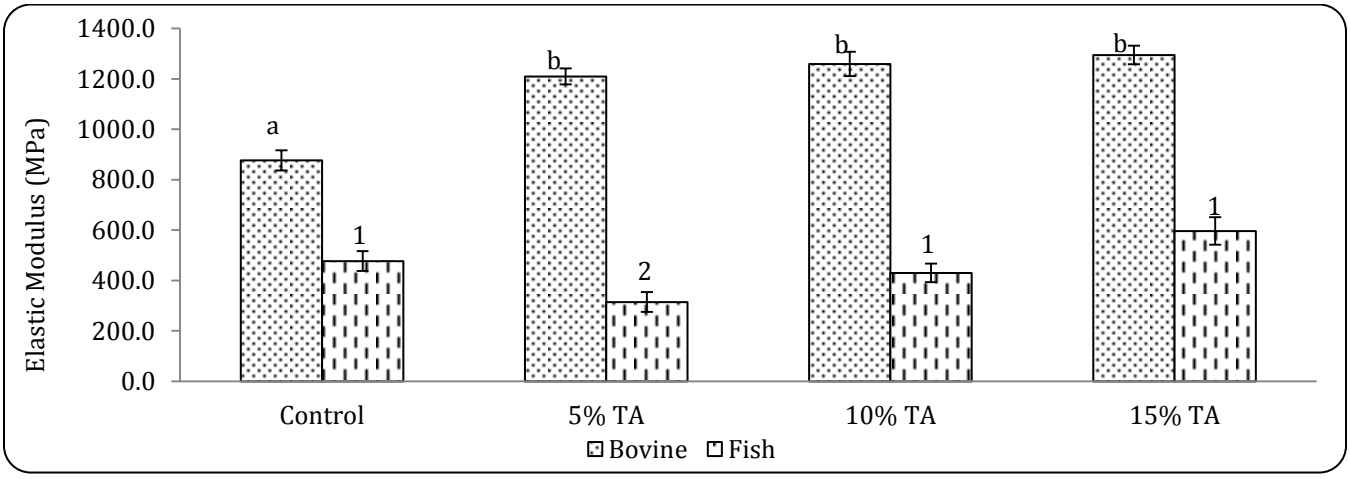

Figure 3. Elastic modulus (MPa) of tannic acid incorporated bovine and fish gelatin samples. Values represent means ± standard error of means (n=8). Letters and numbers represent significant difference in the means of fish and bovine gelatin samples, respectively. Means with same letters or numbers and without letters or numbers are not significantly different ($P > 0.05$).

Water vapor permeability: Incorporation of tannic acid did not make a significant ($P > 0.05$) difference in the WVP of the bovine and fish gelatin films. However, samples with 15% tannic acid have lower WVP compared to controls. Difference was significant in case of bovine gelatin films. Tannic acid has numerous hydroxyl groups which can interact with water (Cao *et al*., 2006). As a result, the WVP of samples did not change significantly. No significant change in the WVP of tannic acid crosslinked bovine gelatins (Cao *et al*., 2006) and transglutaminase crosslinked fish gelatins (Piotrowska *et al*., 2008) was reported.

Water solubility: Solubility of bovine gelatin films reduced significantly ($P < 0.05$) with incorporation of tannic acid. The percent soluble matter was reduced from 55.4±3.9 to 12.9±1.7 with incorporation of 15% tannic acid in bovine gelatin films. In case of fish gelatin, solubility did not change significantly ($P > 0.05$) with incorporation of tannic acid. Film solubility increased for intermediate concentrations of tannic acids and reached value close to the control for fish gelatin films with 15% tannic acid. Increased degree of crosslinking can result in decreased combination of gelatin with water (Cao *et al*., 2006).

Approximately 8% decrease in solubility of tannin crosslinked chitosan was reported by Rivera *et al*. (2010). Solubility of bovine gelatin films crosslinked with tannic acid (3 wt %) decreased significantly (Zhang *et al*., 2010b). Formation of covalent crosslinks in the film matrix was suggested as possible reason for reduction in film solubility. Reduction in solubility from 99 to 27% was reported

for cod fish gelatin films crosslinked with 0.3 mg/ml transglutaminase (Piotrowska *et al*., 2008). Kolodziejska *et al*. (2006) reported reduction in solubility of chitosan-fish gelatin films crosslinked with transglutaminase. It is likely that the film solubility depends on the extent and type of crosslinking and the method used to determine solubility (boiling time and temperature).

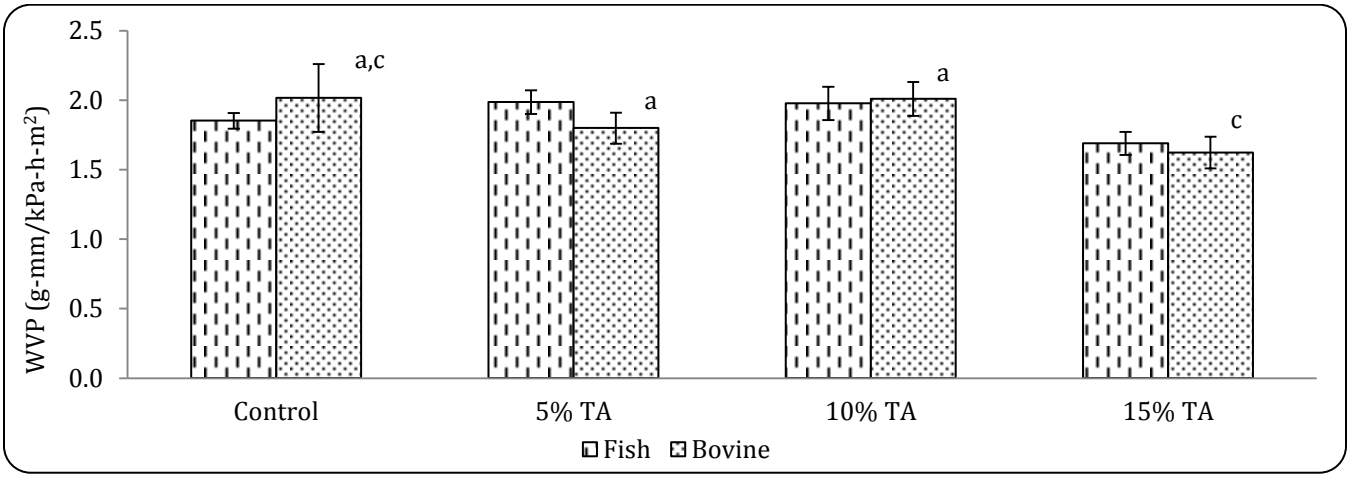

Figure 4. Water vapor permeability of tannic acid incorporated bovine and fish gelatin samples. Values represent means ± standard error of means (n=6). Means with same letters and without letters are not significantly different ($P > 0.05$).

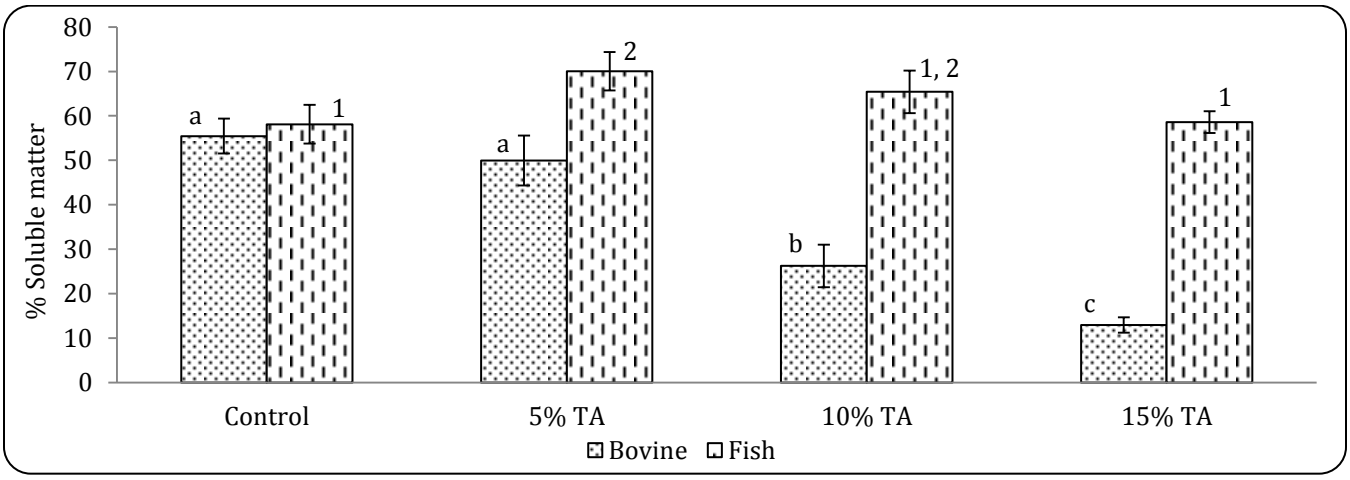

Figure 5. Water solubility of tannic acid incorporated bovine and fish gelatin samples. Values represent means ± standard error of means (n=10). Letters and numbers represent significant difference in the means of bovine and fish gelatin samples, respectively. Means with same letters or numbers and without letters or numbers are not significantly different ($P > 0.05$).

Glass transition temperature: Material transition from a glassy or brittle state into a rubbery state over a temperature range is described by glass transition. Incorporation of tannic acid increased the glass transition temperature by 12 and 6 °C for bovine and fish gelatin films, respectively. When tannins are incorporated into the films, the film matrix becomes more rigid, which in turn increases the energy needed for transition into a rubbery state.

Antioxidant activity of films: Bovine and fish gelatin controls showed 10% DPPH radical scavenging activity.

Table 1. Glass transition temperature (T_g) of bovine and fish gelatin films with tannic acid.

Treatments	Bovine gelatin films	Fish gelatin films
	Glass Transition temperature (°C)	
Control	26.5±0.39	31.6±1.4
5% TA	33.68±1.05	28.7±1.0
10% TA	33.42±1.09	34.4±2.7
15% TA	38.75±0.77	37.8±0.7

Values represent means ± standard error of means (n=2).

Bovine gelatin controls retained this level of activity throughout the 20 day storage while the fish gelatin

controls did not. Samples with tannic acid possess 70 to 90% radical scavenging activity. Level of tannic acid in the films had no significant effect ($P > 0.05$) on the radical scavenging activity of the films. Cao *et al*. (2006) reported an improvement in the properties of tannic acid crosslinked bovine gelatin films during 90-day storage. It was proposed that tannic acid interacts with gelatin in a step-by-step manner, resulting in improved properties (Cao *et al*., 2006; Frazier *et al*., 2003). Similar phenomenon might be responsible for changes observed in this study to retain radical scavenging activity during storage. However, it should be noted that effect of storage on other film properties was not investigated in this study.

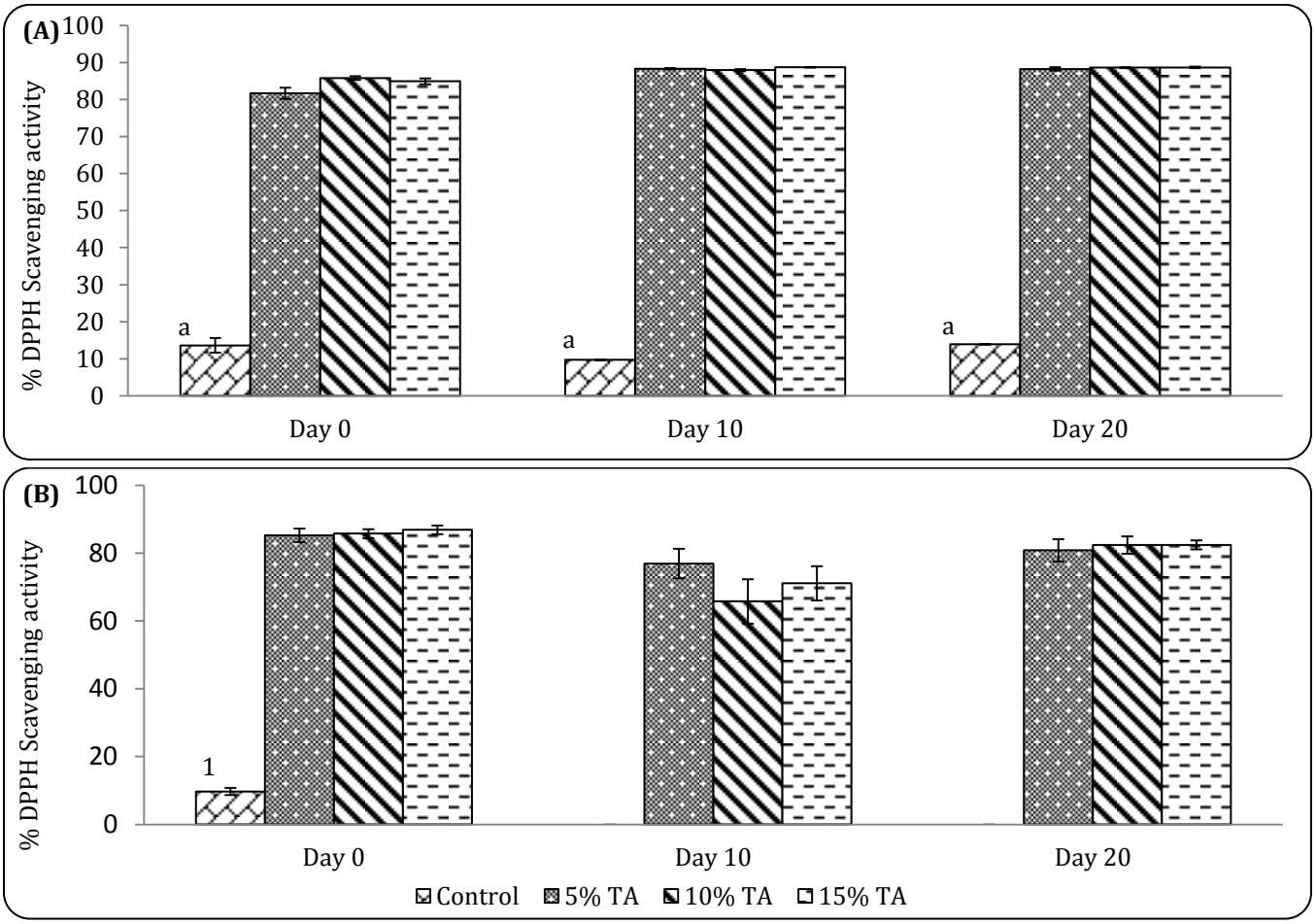

Figure 6. DPPH radical scavenging activity of tannic acid (0, 5, 10, and 15%) incorporated bovine (A) and fish (B) gelatin films. Letters and numbers represent significant difference in the means of bovine and fish gelatin samples, respectively. Means (n=5) with same letters or numbers and without letters or numbers are not significantly different ($P > 0.05$).

Effect of tannin incorporated gelatins on the oxidative stability of salmon fillets: There was a tendency for salmon fillets treated with bovine and fish gelatin solutions to show lower TBARS values compared to untreated controls (Table 2), but the trend was not strong and the results were similar after day 9. At 4 °C storage, bovine and fish gelatin treatments resulted in lower TBARS values than untreated samples and controls at day 3, 6 and 9. However, the differences were not significant ($P > 0.05$). Similar results were reported for samples stored at 10 °C.

It is likely that the treatment effect is being masked by the variability in the assay. Variability in TBARS can be due to several reasons. Degradation of fatty acids results in the formation of malondialdehyde (MDA). It is a minor component of fatty acids with double bonds. The MDA is used as indicator of oxidative rancidity in biological samples. Monoenolic form of MDA reacts with methylene groups of TBA and forms a complex (Mendens *et al*., 2009), which has absorption maxima at 532 nm. However, TBA also reacts with other compounds like alkenals,

alkadienes, pyridines, pyrimidines, sucrose and urea (Shahidi and Zhong, 2005). The generic term TBARS is used to describe these substances. Also, TBA reacts with other food compounds including carbohydrates, amino acids and pigments (Mendens *et al*., 2009). The TBARS assay can lack accuracy in certain food systems due to these factors. Underestimation of oxidation products in the sample can occur when MDA reacts with amino acids, proteins and glycogen in the food (Jo and Ahn, 1998). However, this method is widely used to assess oxidative spoilage in foods due to its simplicity.

Table 2. TBARS (mg MDA/Kg fish) values of salmon fillets stored at 4 and 10 °C treated with bovine and fish gelatin solutions incorporated with 5, 10 and 15% (wt/wt of gelatin) tannic acid.

		4 °C		10 °C	
Storage time	Treatment	Bovine gelatin	Fish gelatin	Bovine gelatin	Fish gelatin
		mg MDA/Kg fish			
Day 0		1.16±0.15		1.29±0.09	
Day 3	Untreated	1.33±0.55	1.24±0.29	0.77±0.34	0.95±0.43
	Control	1.07±0.18	1.38±0.18	0.75±0.22	1.03±0.49
	5% TA	0.41±0.16	0.39±0.19	0.42±0.23	0.30±0.12
	10% TA	0.51±0.20	0.27±0.12	0.34±0.07	0.22±0.10
	15% TA	0.2±0.06	0.24±0.13	0.36±0.13	0.18±0.05
Day 6	Untreated	0.55±0.09	0.66±0.36	0.39±0.12	0.73±0.27
	Control	1.02±0.24	0.61±0.11	0.59±0.01	0.23±0.06
	5% TA	0.30±0.03	0.31±0.15	0.05±0.00	0.18±0.11
	10% TA	0.13±0.04	0.09±0.03	0.15±0.05	0.04±0.00
	15% TA	0.08±0.03	0.06±0.01	0.12±0.05	0.04±0.01
Day 9	Untreated	1.01±0.66	0.82±0.31	0.64±0.29	0.19±0.12
	Control	0.86±0.20	0.80±0.28	0.56±0.30	0.20±0.14
	5% TA	0.26±0.03	0.19±0.02	0.38±0.13	0.30±0.13
	10% TA	0.46±0.16	0.40±0.31	0.04±0.00	0.07±0.03
	15% TA	0.78±0.69	0.53±0.45	0.61±0.13	0.12±0.06
Day 12	Untreated	0.21±0.02	0.56±0.16	0.19±0.06	0.10±0.03
	Control	0.86±0.19	1.17±0.53	0.32±0.13	0.16±0.09
	5% TA	0.29±0.15	0.84±0.64	0.17±0.09	0.09±0.02
	10% TA	0.58±0.26	0.21±0.07	0.22±0.16	0.12±0.05
	15% TA	0.57±0.25	0.14±0.04	0.07±0.05	0.15±0.02

Values represent means ± standard error of means (n=3).

CONCLUSION

Incorporation of tannic acid resulted in reduction in solubility of bovine gelatin films and other minor changes to fish and bovine gelatin film properties. Treatments reduced the TBARS values of salmon fillets at best during the first 9 days of storage. However, general trend in data was not evident due to variability in the assay. Perhaps a more sophisticated assay such as HPLC would be more suitable to study treatment effects of the added antioxidant.

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