



Effect of Previous Plant Extract Treatment on Sensory and Physical Properties of Frozen Bonito (*Sarda sarda*) Fillets

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Abstract

Green tea (*Camelia sinensis*), grape seed (*Vitis vinifera*) and pomegranate peel (*Punica granatum*) extracts were used to keep quality characteristics of bonito (*Sarda sarda*) fillets during frozen storage. Fillets were dipped into plant extract solutions before freezing, then glazed and stored at -18°C for 5 months. Preference of panellists was focused on green tea applied fillets, means of odour and texture at the end of the storage period. Also, brightness values were found to be high. Redness and yellowness of the fillets were affected from natural colour of grape seed and pomegranate peel extracts, respectively. More stiff and less adhesive product was obtained with control groups, followed by green tea extract applied fillets. Deformation of muscle structure was considerably decreased by dipping fillets into plant extracts before glazing.

Keywords: Plant extract, bonito, texture profile analysis, sensory, frozen storage.

Bitki Ekstraktları Uygulamasının Dondurulmuş Palamut (*Sarda sarda*) Balığı Filetolarının Duyusal ve Fiziksel Özellikleri Üzerine Etkisi

Özet

Palamut balığı (*Sarda sarda*) filetolarının dondurulmuş depolama süresince kalitesinin korunması amacıyla yeşil çay (*Camelia sinensis*), üzüm çekirdeği (*Vitis vinifera*) ve nar kabuğu (*Punica granatum*) ekstraktları uygulanmıştır. Dondurma işlemi öncesinde filetolar bitki ekstraktlarına daldırılmış, glaze edilmiş ve -18°C'de 5 ay depolanmıştır. Depolama süresinin sonunda koku ve tekstür bakımından yeşil çay ekstraktı uygulanmış filetoler panelistlerin beğenisini kazanmıştır. Aynı zamanda, parlaklık değerleri de yüksek belirlenmiştir. Filetoların kırmızılık değeri üzüm çekirdeği ekstraktının ve sarılık değeri ise nar kabuğu ekstraktının doğal renginden etkilenmiştir. Kontrol grubu sıkı yapılı ve az yapışkanlık göstermiş, yeşil çay ekstraktı uygulanan filetoler kontrol grubunu takip etmiştir. Filetoların glazing işlemi öncesi bitki ekstraktlarına daldırılması ile kas yapısında meydana gelen deformasyon oldukça azaltılmıştır.

Anahtar Kelimeler: Bitki Ekstraktı, Palamut balığı, tekstür profil analizi, dondurulmuş fileto.

Introduction

Harvested and captured fishery products are shipped to all over the world. Increasing distance in transporting needs stability of physical and sensory attributes as well as chemical aspects. Freezing is one of the most employed methods used for preserving fresh fish and other seafood products. However, deterioration of fish occurs depending on many factors like fish species, storage temperature, time and enzymatic degradation during frozen storage.

Mechanical endurance is weakened during freezing, thawing, storage and fish muscle turns out to be more sensible to spoilage (Sigurgisladottir *et al.*,

2000). The quality of frozen fish is affected by the loss of moisture during freezing, influencing ice crystal size and location. Fast freezing rate minimizes the migration of water into the extracellular spacing and forms smaller intracellular ice crystals. Otherwise, this may lead to denaturation of the muscle proteins as well as structural damage of membranes, which can result in increased drip loss, loss of water holding capacity and textural changes (Duun and Rustad, 2007). Freeze-thaw cycle ruins the stability of muscle structure and cause prooxidant scattering and accumulation of lipid oxidation (Schubring, 2002).

When lipids are exposed to environmental

factors such as air, light, heat undesirable taste, colour, rancid odour, loss of nutritional value, even toxic compounds are formed (Alonso *et al.*, 2004). In spite of lacking unfavourable attributes in primary oxidation products like hydroperoxides, decomposition products such as aldehydes, ketones and hydrocarbons have strong unpleasant taste and odour (Mendes *et al.*, 2009).

The lipids oxidize, but instead of forming carbonyls and other compounds associated with rancidity, they become bound up in lipid-protein complexes, which accounts for the toughened texture of over stored or poorly stored frozen fish (Castell, 1971). The changes in protein due to freezing mainly occur in myofibrillar proteins, which undergo denaturation and aggregation through protein-protein bonding. Also, these changes are associated with lipid oxidation occurring during frozen storage. However, texture and functionality alterations observed in the muscle of fatty and semi fatty species are smaller than in lean species due to the protective effect of lipids in proper concentration and location on protein (Tejada, 2001).

Since the shelf life of fillets are lower than for whole fish, it is necessary to develop methods to retain sensory and nutritional properties of fish. Natural antioxidants used to extend the shelf-life of marine products are receiving great attention (Lin and Lin, 2005; Pazos *et al.*, 2005; Alonso *et al.*, 2007; Lugasi *et al.*, 2007; Alghazeer *et al.*, 2008; Ibrahim and El-Sherif, 2008).

The most important characteristic of tea, *Camellia sinensis* is being totally natural with obtaining no artificial colorants, preservatives and odours. Tea has been consuming for 5000 years in China as medicine. Natural antioxidants of tea polyphenols, which has protective effect on active oxygen radicals, are extracted by using water, ethanol, methanol and acetone. The protective effect of grape seed (*Vitis vinifera*) against oxidation agents has been attributed to the high content of phenolic compounds (Negro *et al.*, 2003; Shaker, 2006). By-products of grape juice and wine plants are the sources of alternative natural antioxidants. It is reported that pomegranate fruit, juice, peel, seed oil and seed extracts have high antioxidative value and also different parts of the plant have antibacterial, antidiarrhetic, antifungal properties (Kaur *et al.*, 2006).

In this study, it was aimed to determine the effect of previous green tea, grape seed and pomegranate peel extract treatments on sensory and physical properties of frozen bonito fillets during the storage at -18°C.

Materials and Methods

Bonito (*Sarda sarda*) was purchased from the main fish market in Antalya, Turkey and transferred to the laboratory in polystyrene boxes with crushed

ice. The mean weight and length of fish were 181.4±18.91 g and 27.96±1.35 cm, respectively.

Extraction

Antique green tea leaves (*Camellia sinensis*) harvested in 2004 and 2005 shooting periods were purchased from the local market in Antalya and dried at 40°C for 12 h in an oven. All samples were then ground into a fine powder with a mill. The powders dissolved in ethanol (1:20 w/v) and then extracted in a water bath with shaker at 40°C for 4 h. The extracts were filtered and concentrated in a rotary evaporator to get crude extracts.

Grapes (*Vitis vinifera* sp., Calkarası) were purchased from the market in Antalya and seeds were manually separated. The seeds were dried at 50°C until a constant weight and ground to powder and extracted in a soxhlet extractor with petroleum ether for 6 h.

Pomegranate (*Punica granatum*) were also purchased from the market and peeled. Pomegranate peels were dried at 50°C until a constant weight and ground to powder. Defatted seed powder and pomegranate peel powder were dissolved in ethanol (1:20 w/v) and then extracted in a water bath with shaker at 40°C for 4 h. The extracts were filtered and concentrated in a rotary evaporator to get crude extracts.

All green tea, grape seed and pomegranate peel extracts were then stored under nitrogen at -20°C until use.

Treatments

Extract solution was prepared by dissolving 1.0 g plant extract in 100 ml distilled water. Fillets were separately dipped into three extract solutions and frozen in an air-blast freezer at -40°C. Another group fillets were dipped into water as control. Frozen bonito fillets were wrapped using stretch film (10 micron thickness) and aluminum folio (20 micron thickness), placed in carton boxes and stored at -18°C.

Analyses

Quality control analyses were performed during the storage on monthly intervals. Frozen fillets were thawed in a refrigerator (at 4°C) for each sampling time.

Total Phenolics in Plant Extracts

Total phenolic contents of the extracts were determined spectrophotometrically according to the Folin-Ciocalteu colorimetric method (Spanos and Wrolstad, 1990). Each extract of 0.1 ml was introduced to 5 ml Folin-Ciocalteu's reagent (0.2 N), 4 ml sodium carbonate (7.5 g L⁻¹) and 0.9ml distilled water. The mixture was allowed to stand for 2 hours

before absorbance measurement against blank at 765 nm (Shimadzu UV 160A, Tokyo, Japan). Results were expressed as mg gallic acid equivalents (GAE) in mg/100 g.

Antioxidant Capacity of Plant Extracts

The antioxidant capacity of plant extracts was determined by measuring the formation of the radical cation ABTS, according to the photometric method of Re *et al.* (1999). 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS⁺) radical cations were prepared by adding 7 mM ABTS reagent including 2.45 Mm potassium per sulphate and allowed to stand 12-16 hours at room temperature in darkness. The radical was adjusted to 0.700±0.02 at 734 nm on each spectrophotometric measurement with ethanol. Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), a water soluble analog of vitamin E, was used as an antioxidant standard. Serial concentrations of plant extracts were prepared and the standard calibration curve composed of concentration against inhibition rate was constructed for Trolox. Trolox equivalent antioxidant capacity values were calculated as proportioning the slope of the curves. The results were expressed in mmol Trolox equivalents (mmol TE) as means of three consecutive measurements of the same sample.

Color Measurements

Surface color measurements were determined using a CR-400 Chroma Meter (Minolta Co., Osaka,

Japan) which consisted of a measuring head (DP-400). The colorimeter was calibrated to a standard white tile in which 'L' (lightness), 'a' (redness) and 'b' (yellowness) values were determined as 97.02, 0.08, 1.75, respectively. Measurements were averaged over four zones of fillets.

Sensory Analysis

Ten experienced panelists, staff members of the department of food engineering, who had experience on fish and fish products were chosen to evaluate the quality of fillets. Before presentation to the panel, fish samples were steam cooked for 20 minutes and served warm to the panelists. The samples were coded using letters and randomly presented to the panelists. Panelists were asked to evaluate appearance, odor and taste of samples. Appearance, odor and taste were scored on a 10-point hedonic scale (Table 1). A score of zero was used as a rejection attribute point (Simeonidou *et al.*, 1997).

Texture Measurements

Instrumental texture profile analysis (TPA) was performed by using TA-XT2 (Stable Micro Systems, Godalming, Surrey, England) conducting a spherical probe (SMSP5) with the diameter of 5 mm and equipped with 5 kg load cell. Thawed fish fillets having average height of 2.86±0.97 cm were placed under the probe and at least 4 portions of each sample were used with test speed of 2 mm/sec and penetration distance of 5 mm. From the resulting force-time curve these parameters were determined:

Table 1. Sensory evaluation scale for frozen fish fillets¹

Score	Odour	Taste	Texture
10	Strong seaweed-like odours	Fresh sweet flavours characteristic of the speches	Very succulent and coherent
9	Some loss of seaweed-like odour	Some loss of sweetness	Succulent and coherent, flaky
8	Loss of odours, neutral odours	Loss of the characteristic flavour of the species	Succulent after chewing several times, coherent and less flaky
7	No sour or stale odours; boiled milk or potatoes, wood sap	Neutral flavour, no off-flavours, slightly meaty	Moderately-succulent after chewing several times, moderately coherent and flaky
6	Slightly sour, caramel-like odours	Slightly rancid, slightly insipid, slightly soapy	A little succulent or watery at first chewing, a little dry at later chewing
5	Lactic acid and sour milk	Trace of 'off-flavours', rancid, slightly bitter	Not succulent at all at first chewing, dry and tough at later chewing
4	Lower fatty acids, rancid butter, slightly cold storage odour	Bitter, sour (citric), cold storage flavor 'woody'	Dry and tough or fibrous from first chewing
3	Slight amines, slight ammonia	Strong bitter flavour, soapy, slight amine flavour	Very dry and tough or fibrous during chewing
2	Ammonia, very sour, cold storage odour (cardboard), some sulphide odours	Strong bitterness, strong sour, not nauseating	Strongly dry and tough, a little difficult to chew
1	Strong ammonia and amines, faecal	Strong bitterness, strong sour, a little nauseating	Strongly dry and tough, difficult to chew
0	Very strong ammonia and faecal, sulphid, acrid, putrid odours	Strong 'off-flavour' of amines, putrid, difficult to taste	Strongly dry and tough, very difficult to chew

¹ 10-8 "excellent", 7-8 "good", 5-6 "fair", 3-4 "poor", 1-2 "very poor" Scores <4 is rejected

hardness (N), resistance at the maximum compression; adhesiveness (Nmm), negative area at first compression; springiness (mm), ability of deformed fillets to recover its original form; cohesiveness, the area of second compression cycle relative to the area of the first compression cycle; gumminess (N), the force needed to disintegrate a semisolid sample to a steady state of swallowing. Three measurements were performed on each fillet, and the mean value was used for statistical analyses of the data.

Statistical Analysis

Two replications of the experiment were conducted at separate times and all analyses were performed in duplicates. Means and standard errors were calculated. Data were analyzed by a two factor factorial arrangement in a completely randomized design. The two factors were the four dipping solutions (grape seed, green tea, pomegranate peel, water), and six storage months (0, 1, 2, 3, 4, 5). Analysis was conducted using the SAS software (Statistical Analysis System, Cary, NC, USA). When main effects or interactions were significant, Duncan's Multiple Range test was used.

Results and Discussion

Total Phenolics and Antioxidant Activity of Plant Extracts

In this research, dry matter of green tea, grape seed and pomegranate peel extracts were determined as 53.83±0.65%, 52.93±0.67% and 52.28±0.82%, respectively.

The presence of phenolic compounds in green tea extract was 2.278±1.83%, where statistically higher ($P<0.01$) than the other plant extracts. Antioxidant activity was found as 1.772±0.071 mM trolox. Ivanova *et al.* (2005) reported that total phenolic material content of green tea, extracted in hot water was 317.62±3.76 µM quercetin equivalent and antioxidant activity 5.91±0.14 mM trolox. Chan *et al.* (2007) found these parameters as 20.55±0.21 g GAE/100 g for phenolic content in microwave applied green tea, extracted in methanol and 126±4.5 mg GAE/g FRAP and 3000±778 mg AA/100 g DPPH for antioxidant activity. Total phenolic content of Greek and China green tea contents were 88.1±0.41 and 1216±32 mg GAE/cup, respectively (Atoui *et al.*, 2005). Also, these researchers found antioxidant activities as 0.13 mg extract/mg DPPH for Greek tea and 0.57 mg extract/mg DPPH for China tea.

Total phenolic compound content and antioxidant activity of grape seed extract were found as 1.077±1.52% and 1.605±0.045 mM trolox. A positive relation was observed between phenolic content and antioxidant activity of plant extracts as reported by Rusak *et al.* (2008). Green tea, grape seed

and pomegranate peel have high antioxidative properties. Antioxidant activity of grape peel, pulp and seed was compared and found the highest activity in seed with the values of 55.54±1.62 mmol/100 g (Guo *et al.*, 2003), 25.12±0.53% (Negro *et al.*, 2003). Jayaprakasha, Selvi and Sakariah (2003) determined antioxidant activity of defatted grape seed extract solved in acetone:water:acetic acid as 215.6±18.2 µmol/g. The same researchers found phenolic content as 54.0±4.86% catechin equivalent in grape seed extract obtained by ethyl acetate:water (Jayaprakasha *et al.*, 2001). Total phenolic content of different grape seed species were ranged between 143±16 and 2228±26 mg GAE/100 g (Guendez *et al.*, 2005).

Existence of total phenolic compound in pomegranate peel extract was found as 1.065±0.97, and antioxidant activity was 1.784±0.033 mM trolox. In spite of containing less total phenolic content than green tea extract, pomegranate peel had similar antioxidant activity which was statistically insignificant. Singh *et al.* (2002) found the highest antioxidant activity in pomegranate peel and seed extracts obtained by methanol solvent. Li *et al.* (2006) used ethanol, methanol, acetone and their mixture as extraction solvent. Total phenolic content of pomegranate peel extract was found 249±17.2 mg/g tannic equivalent. The highest antioxidant activity was obtained with the mixture of solvents. Ferric reducing ability of plasma analysis was performed in order to determine antioxidant activity of peel, pulp and seed of many fruits. These values in pomegranate peel, pulp and seed were 82.11±4.01, 3.10±0.12 and 0.72±0.05 mmol/100 g, respectively. Trolox equivalent antioxidant capacity value of gallic catechin-(4-8)-catechin pure extract obtained from pomegranate peel was found as 3.56±0.11 (Plumb *et al.*, 2002).

Plant variety, moisture of material, used solvent, applied time and temperature differences excite disparity in findings. Moreover, antioxidant activity determination method was another factor makes it difficult to compare the results. It is not appropriate to compare these data with the results of our study directly due to the differences in the reaction mechanisms of antioxidant capacity assays.

Colour Analysis

First decision about food before consuming is up to its brightness. Bonito is a pelagic fish living close to sea surface. L values of samples did not differ in the first two months for all groups, however on the 4th and 5th months fillets treated with green tea extract showed the highest scores (Table 2). L values of fillets treated with pomegranate peel extract remained at low levels for all storage months and significantly ($P<0.05$) decreased, whereas the other groups leveled off their initial L values during frozen storage and found as insignificant in statistical evaluation.

Rigor strengthening and muscle structure

Table 2. Instrumental color scores of frozen bonito fillets previously treated with plant extracts¹

Colour Analysis		Storage Time (Months)				
Treatments		1	2	3	4	5
L Value	GT	48.3±1.2 ^{aY}	50.7±1.2 ^{aXY}	53.2±2.8 ^{aX}	50.7±1.1 ^{aXY}	49.2±0.7 ^{aXY}
	GS	47.0±0.8 ^{aX}	46.1±4.8 [*]	46.0±0.9 ^{bXY}	45.1±0.7 ^{bXY}	43.9±1.0 ^{bY}
	PP	51.8±2.2 ^{aX}	47.7±0.1 ^{bXY}	43.2±3.2 ^{bYZ}	41.1±1.5 ^{cZ}	40.9±1.4 ^{cZ}
	C	49.6±6.7 ^{aX}	47.0±0.3 ^{bX}	46.8±2.5 ^{abX}	47.2±0.8 ^{bX}	46.1±0.5 ^{bX}
a Value	GT	4.7±0.0 ^{bX}	5.9±0.4 ^{bX}	6.8±0.3 ^{abX}	5.7±1.1 ^{abX}	6.0±1.7 ^{aX}
	GS	9.1±0.0 ^{aX}	9.4±2.1 ^{aX}	8.5±0.1 ^{aX}	8.4±0.5 ^{aX}	7.9±0.9 ^{aX}
	PP	6.4±2.3 ^{abX}	5.5±0.6 ^{bX}	5.2±0.3 ^{bcX}	5.3±0.6 ^{abX}	5.7±1.7 ^{aX}
	C	6.7±1.7 ^{abX}	5.3±0.3 ^{bX}	4.8±1.3 ^{cX}	4.6±2.2 ^{bX}	4.9±0.1 ^{aX}
b Value	GT	7.2±0.4 ^{cY}	7.9±0.2 ^{bXY}	9.0±0.2 ^{abX}	9.0±0.5 ^{aX}	8.1±0.7 ^{aXY}
	GS	11.6±0.2 ^{aX}	11.3±0.8 ^{aX}	10.1±0.4 ^{abXY}	10.7±1.1 ^{aXY}	9.1±1.1 ^{aY}
	PP	9.6±0.4 ^{bX}	10.8±1.5 ^{aX}	10.2±0.9 ^{aX}	10.9±1.4 ^{aX}	10.1±1.5 ^{aX}
	C	8.5±0.6 ^{bX}	8.1±0.7 ^{bX}	8.2±0.8 ^{bX}	9.1±0.7 ^{aX}	8.9±1.0 ^{aX}

¹ Values are mean ± standard deviation

* not included statistically due to high standard deviation

Means within the same column (a,b,c) and the same row (X, Y, Z) with different letters are different (P<0.05).

GT: Green tea extract, GS: Grape seed extract, PP: Pomegranate peel extract, C: Control

changes with freezing process. Fillet appearance differs due to affected reflectance properties of muscle. Decrease in protein solubility and denaturation make muscle seem opaque (Einen *et al.*, 2002). Fish fillet is more sensitive to oxidation than bovine meat because of hemoglobin increase after harvest (Richards *et al.*, 2002). Potential oxidation was gradually protected by plant extracts.

Undesirable changes in appearance such as loss of color intensity, development of freezer burns, surface dehydration, muscle opacity are thought to be irreversible changes occur in proteins or protein-bound pigments or changes of myoglobin and oxymyoglobin into metmyoglobin. Oxidation of ferrous to ferric iron (Fe²⁺ to Fe³⁺) gives a brown appearance to the meat (Asghar and Pearson, 1959). High content of conjugated double bonds exposed to air oxygen causing color loss in carotenoids (Choubert and Baccaunaud, 2006). These alterations affected the frozen bonito fillets and resulted in gradual decrease on redness scores of pomegranate peel and grape seed extracts. The changes in a* value of all groups were statistically unimportant for all groups during frozen storage (Table 2). Redness scores of fillets were significantly affected by grape seed extracts. The lowest scores for a* value were found in the samples treated with pomegranate peel extract and control.

Shrimps were dipped into grape seed extract in different concentrations to hinder melanosis formation (Gokoglu and Yerlikaya, 2008). Application of extract extended original shrimp color and especially L* and a* values were positively affected. Sanchez-Alonso *et al.* (2008) reported that a* and b* values of horse mackerel mince with added white grape pomace did not change significantly during frozen storage. However, b* value increased in control sample which was thought to be related with brown oxidation pigments (met-heame proteins).

Yellow color is often associated with oxidation (Özoğul *et al.*, 2006). Yellowness was fluctuated for

each bonito fillet groups during frozen storage (Table 2). Fillets treated with grape seed extract had the highest b* values. Yellowness in the fillets treated with green tea extract significantly changed during the storage and the lowest b* value was found in this group at the end of storage. The natural plant color of pomegranate peel showed itself and kept the yellowness value at high levels for all months. Hamre *et al.* (2003) stated that lipid oxidation often causes yellow fluorescent pigments to accumulate in the fillet and determined a weak correlation between oxidation indicator parameters and b* value of herring. Oxidation is the cause of color deterioration. It was reported that L* and a* values showed high correlation with sensory scores (O'Sullivan *et al.*, 2003). However, in this study the natural colors of plant extracts affected the a* and b* values and lessen the brightness of control fillets except for green tea treatment.

Sensory Analysis

Frozen storage prevents food from undesired sensory and chemical aspects caused by microorganisms and spoilage reaction, however cannot be totally hindered. Reactions especially occur in proteins and lipids affect sensory properties and cause unpleasant odour, taste and texture changes.

Odour scores of fish samples were gradually decreased during the storage as can be seen on Table 3. Initially the best odor score were found in control samples, however the score significantly decreased at the end of the storage period. The samples treated with green tea extract were the most preferred fillets followed by the samples with grape seed extract.

Horse mackerel and Mediterranean hake fillets and minces were exposed to rosemary extract and frozen (Vareltzis *et al.*, 1997). Plant extract applied groups were more preferred than control groups like in our study.

According to taste scores preference of panelists

Table 3. Sensory analysis scores of frozen bonito fillets previously treated with plant extracts¹

Sensory Analysis	Treatments	Storage Time (Months)				
		1	2	3	4	5
Odour	GT	8.0±0.6 ^{abX}	7.8±0.3 ^{aXY}	6.9±0.2 ^{aYZ}	6.7±0.1 ^{aZ}	7.4±0.4 ^{aXYZ}
	GS	7.1±0.5 ^{bX}	7.4±1.2 ^{aX}	6.9±1.2 ^{aX}	6.4±0.0 ^{aX}	7.0±0.6 ^{aX}
	PP	8.0±0.0 ^{abX}	7.9±0.7 ^{aX}	7.8±0.5 ^{aX}	7.2±0.8 ^{aX}	6.8±0.1 ^{aX}
	C	8.9±0.4 ^{aX}	7.3±0.2 ^{aY}	6.8±0.3 ^{aY}	6.5±0.7 ^{aY}	6.9±0.1 ^{aY}
Taste	GT	6.5±0.1 ^{cX}	7.6±0.6 ^{aX}	7.0±1.0 ^{aX}	6.7±0.4 ^{aX}	7.1±0.4 ^{abX}
	GS	8.3±0.2 ^{abX}	7.2±0.7 ^{aX}	6.9±1.5 ^{aX}	6.7±0.7 ^{aX}	6.3±0.6 ^{bX}
	PP	7.1±0.4 ^{bcX}	7.7±0.4 ^{aX}	7.4±0.5 ^{aX}	6.9±0.1 ^{aX}	7.4±0.2 ^{aX}
	C	8.8±0.9 ^{aX}	7.1±0.6 ^{aY}	6.6±0.3 ^{aY}	6.4±0.2 ^{aY}	6.9±0.1 ^{abY}
Texture	GT	7.0±0.0 ^{aX}	7.2±0.7 ^{aX}	6.4±0.3 ^{aX}	6.3±0.1 ^{aX}	7.7±0.9 ^{aX}
	GS	8.0±0.6 ^{aX}	7.8±0.3 ^{aX}	6.9±0.2 ^{aX}	6.7±0.1 ^{aX}	7.4±0.4 ^{aX}
	PP	7.1±0.5 ^{aY}	7.4±1.2 ^{aXY}	6.9±1.2 ^{aX}	6.4±0.0 ^{aXY}	7.0±0.6 ^{aXY}
	C	8.0±0.0 ^{aX}	7.9±0.7 ^{aX}	7.8±0.5 ^{aX}	7.2±0.8 ^{aX}	6.8±0.1 ^{aX}

¹ Values are mean ± standard deviation

Means within the same column (a, b, c) and the same row (X, Y, Z) with different letters are different (P<0.05).

GT: Green tea extract, GS: Grape seed extract, PP: Pomegranate peel extract, C: Control

gradually decreased especially in control group (Table 3). Control samples had the highest (P<0.05) taste scores on the first month, however left its place to the treatments of pomegranate peel and green tea extract at the end of the storage. Decreasing odour and taste scores during storage were thought to be related to penetration of plant extracts into fish fillets. The difference in treatment groups was statistically insignificant.

Physical and chemical changes in proteins of fish during frozen storage cause texture deterioration. This problem also affects sensory aspects. Formaldehyde formation makes cross-links with protein, lessen protein solubility and decrease water holding capacity (Steen and Lambelet, 1997). These alterations also cause taste losses, odour changes. Turan and Erkoyuncu (2004) reported that control group had less preference compared with bonito applied plant extracts.

Thawed fish flesh are generally tough, dry and fibrous. Fillets had fluctuation in texture scores during frozen storage (Table 3). The fillets treated with green tea extract were the most preferred samples in terms of sensory characteristics, whereas control samples had low sensory scores. These findings were also supported by instrumental analysis. Refsgaard *et al.* (1999) reported similar results for salmon.

Texture Profile Analysis

Many researchers tend to use instrumental texture measurements because of the variations in sensory evaluation performed by human. In texture profile analysis, the response of a sample to a compressive or tensile force is measured by means of time. Basic mechanical variables that characterize texture of food are hardness, springiness and cohesion (Casas *et al.*, 2006). Texture profile analysis scores for bonito fillets were all given in Table 4.

Fish is mostly composed of water, and water

expands during freezing. This tears the flesh of the fish and makes it mushy. Hardness values of fillets treated with grape seed, pomegranate peel and control groups were gradually changed (P<0.05) during storage period, whereas alteration in the sample treated with green tea was statistically insignificant. Plant extracts soften the structure. The difference between treatments was statistically significant (P<0.05), where control samples had the highest hardness scores at the end of the frozen storage.

Fresh muscle structure alters due to replacement of water to intercellular spaces during freezing and frozen storage (Hurling and McArthur, 1996). Protein denaturation and loss of water holding capacity make fish flesh firmer (Mackie, 1993). It is also reported that codfish muscle structure deforms even stored in convenient conditions and thawing.

Adhesion is the ability to stick or bond to a substrate. Initial adherent value of fillets was low in the first month. Gradual decrease was observed for treatments of grape seed extract and control samples, whereas increase in adhesion values of fillets treated with pomegranate peel extract were statistically significant (P<0.05). The highest scores were determined for the treatment of green tea extract. Texture properties of frozen fish alter because of many reasons like degradation of compounds, regenerated reactions, and formation of ice-crystals, thawing, and water replacement.

Texture properties are also affected by differences in chemical composition and physical structure among the fillets. Therefore, different parts of fillets should be taken into consideration to perform accuracy (Sigurgisladdottir *et al.*, 1999; Johnsson *et al.*, 2001) as regarded in this research.

Cohesion is deformation occurs before break off while biting, another definition; is the force among molecules. Cohesion=1 means, muscle keep its stability and returns to its original structure during pause between the compressions. Cohesion<1 means,

Table 4. Texture profile analysis scores of frozen bonito fillets previously treated with plant extracts¹

Texture Profile		Storage Time (Months)				
Analysis	Treatments	1	2	3	4	5
Hardness	GT	139.8±4.6 ^{aX}	125.1±2.0 ^{aX}	130.0±20.0 ^{aX}	111.3±34.0 ^{aX}	105.6±5.8 ^{bX}
	GS	118.1±5.0 ^{aX}	125.6±4.0 ^{aX}	91.6±17.1 ^{abY}	76.0±4.8 ^{aY}	78.2±7.7 ^{cY}
	PP	102.4±5.0 ^{aXY}	132.8±26.0 ^{aX}	89.6±8.2 ^{bY}	68.5±5.8 ^{aY}	71.4±4.3 ^{cY}
	C	73.1±6.0 ^{aW}	95.5±2.8 ^{aZ}	98.5±1.4 ^{abZ}	111.4±3.0 ^{aY}	122.6±3.6 ^{aX}
Adhesiveness	GT	4.8±2.4 ^{aX}	2.9±1.7 ^{aX}	1.8±0.1 ^{aX}	1.7±0.4 ^{aX}	2.1±0.4 ^{aX}
	GS	7.3±7.8 ^{aX}	2.1±0.7 ^{aX}	1.7±0.01 ^{aX}	2.2±1.4 ^{aX}	1.8±0.1 ^{aX}
	PP	0.8±0.2 ^{aZ}	1.2±0.4 ^{aYZ}	3.0±0.8 ^{aX}	2.5±0.6 ^{aXY}	1.5±0.5 ^{abYZ}
	C	4.6±4.8 ^{aX}	3.4±0.5 ^{aX}	3.2±2.4 ^{aX}	1.7±0.02 ^{aX}	0.7±0.1 ^{bX}
Cohesiveness	GT	0.44±0.02 ^{aZ}	0.53±0.01 ^{aXY}	0.58±0.01 ^{aX}	0.53±0.04 ^{aXY}	0.51±0.02 ^{abY}
	GS	0.44±0.01 ^{aY}	0.55±0.05 ^{aX}	0.59±0.01 ^{aX}	0.49±0.04 ^{aXY}	0.43±0.04 ^{bY}
	PP	0.58±0.22 ^{aX}	0.61±0.01 ^{aX}	0.49±0.05 ^{aX}	0.51±0.01 ^{aX}	0.41±0.04 ^{bX}
	C	0.53±0.02 ^{aX}	0.54±0.01 ^{aX}	0.54±0.05 ^{aX}	0.55±0.01 ^{aX}	0.55±0.05 ^{aX}
Springiness	GT	0.96±0.00 ^{aX}	0.95±0.01 ^{aX}	0.95±0.03 ^{abX}	0.95±0.04 ^{aX}	1.00±0.02 ^{aX}
	GS	0.98±0.02 ^{aX}	0.94±0.01 ^{aXY}	0.92±0.01 ^{bXY}	0.91±0.03 ^{aY}	0.96±0.02 ^{abXY}
	PP	0.96±0.06 ^{aX}	0.95±0.02 ^{aX}	0.96±0.01 ^{abX}	0.96±0.04 ^{aX}	0.99±0.09 ^{aX}
	C	0.97±0.03 ^{aX}	0.99±0.02 ^{aX}	1.01±0.01 ^{aX}	0.99±0.05 ^{aX}	0.82±0.03 ^{bY}
Gumminess	GT	62.9±24.3 ^{aXY}	67.0±2.8 ^{aXY}	76.2±11.4 ^{aX}	58.7±13.4 ^{abXY}	37.9±6.3 ^{bY}
	GS	57.1±13.0 ^{aX}	62.2±2.5 ^{aX}	54.4±11.5 ^{abXY}	52.3±10.0 ^{abXY}	31.7±4.7 ^{bY}
	PP	38.7±16.9 ^{aXY}	61.0±16.5 ^{aX}	43.8±1.2 ^{bXY}	34.7±2.3 ^{bXY}	29.8±3.9 ^{bY}
	C	38.7±1.1 ^{aZ}	52.0±2.6 ^{aY}	52.4±7.0 ^{abY}	61.5±1.1 ^{aXY}	68.5±6.7 ^{aX}

¹ Values are mean ± standard deviation

Means within the same column (a, b, c) and the same row (X,Y,Z,W) with different letters are different (P<0.05).

GT: Green tea extract, GS: Grape seed extract, PP: Pomegranate peel extract, C: Control

the deformation of first compression is irreversible (Veland and Torrissen, 1999). The difference of cohesion values was insignificant among treatment groups. Storage period had no effect on cohesion scores of fillets treated with pomegranate peel extract and control samples. However, cohesion values of green tea and grape seed extract treatment groups raised on the 2nd and 3rd months.

Adhesion > Cohesion = Sticky and Adhesion < Cohesion = Not sticky; in this study all fillet samples were determined as sticky. Also, green tea applied fillets and control samples were the ones that mostly kept their stability.

Springiness is the ability of the product physically springs back after it has been deformed during the first compression. Differences in springiness values of fillets in all treatment groups were insignificant during frozen storage period. The highest scores were obtained by treatment of green tea extract, whereas control samples had the least values on the 5th month.

Storage period affected gumminess values of control samples (P<0.05), whereas all other treatment groups leveled off. The difference between treatment groups was statistically insignificant until the 5th month of storage, in which gumminess value of control samples were higher than of the other groups, followed by green tea extract applied fillets. It was clarify that, there was a strong relation between firm structure and gumminess.

Conclusion

Lipid oxidation and protein degradation cause

unpleasant physical and sensory alterations even during freezing process. In this study, it was introduced that use of plant extracts as natural antioxidants have positive effects on quality parameters during frozen storage. Plant extract applications met high scores than control groups. Green tea extract treatment was more preferred in terms of brightness, odour and texture aspects. Also, instrumental analysis of this group met successful results, whereas it is obvious that pomegranate peel extract did not possess agreeable determinations after all evaluations. The present findings will be useful in leading to further experiments on the identification and characterization of natural sources that are responsible for extending quality of food products.

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