

Evaluation of Green Tea (*Camellia sinenses*) Extract and Onion (*Allium cepa* L.) Juice Effects on Lipid Degradation and Sensory Acceptance of Persian Sturgeon (*Acipenser persicus*) Fillets: A Comparative Study

S. Haghparast^{1*}, H. Kashiri¹, Gh. Alipour¹, and B. Shabanpour¹

ABSTRACT

This study was carried out to evaluate the effects of red onion (*Allium cepa* L.) juice and green tea (*Camellia sinenses*) extract on lipid oxidation and sensory characteristics of refrigerated (4°C) sturgeon fillets (*Acipenser persicus*). Fresh fillets were tumbled in 1%, 2.5%, and 5% (v/v) aqueous solutions of onion Juice (OJ) and tea extract (TE), and then stored for up to 8 days at 4°C in a refrigerator. Chemical indices of lipid oxidation as assayed by heme iron, thiobarbituric acid and free fatty acid contents indicated much more reduction in 2.5%TE, 5%TE and 5%OJ-treated samples relative to the other samples (P<0.05). Significant (P<0.05) differences in pH values of treatments and the control were observed during the first 2 days of storage time. For 5%OJ treatment, the pH remained constant during refrigerated storage (P>0.05) whereas gradual changes were detected in pH values of the others. Generally, the order of effectiveness for inhibiting oxidation in sturgeon fillets was found to be: 5% TE or OJ = 2.5% TE > 2.5 % OJ > 1% TE = 1% OJ. Based on sensory scores, higher amounts of onion juice (>1%) were more effective to improve attributive characteristics of the fillets.

Keywords: Green tea, Lipid degradation onion, Sturgeon fillets.

INTRODUCTION

Lipid oxidation is a major cause of muscle food deterioration (Ladikose and Lougovoise, 1990), subsequent off-flavors, unpleasant odors, toughening and muscle discoloration and nutritious value decrease (Frankel, 1998). It may be augmented by light, heat, metal ions and salt in fish flesh (Morrissey and Kerry, 2004). Addition of antioxidant with synthetic or natural origin is one of the strategies to reduce or retard oxidation and prevent the loss of quality and sensory attributes (Serdarglu and Felekoglu, 2005). Recently, the most commonly used synthetic antioxidants such as BHT, BHA and TBHQ (USDA, 2005) have been under attack due to

their potential action in carcinogenesis (Imaida *et al.*, 1983) which led to the rejection of these imperfect additives by consumer preferences (Sherwin, 1990). In addition, there is a growing interest in the use of natural antioxidants because of their considerable role as functional and biochemical inhibitors of oxidative damage induced by free radicals. Many plant tissues are good sources of phytochemicals, notably phenolic and flavonoids (Gorinstein *et al.*, 2005) that can act as the best alternatives to these mutagenic food additives. However, there might be a variety of differences in composition, concentrations and antioxidative properties in bioactive compounds of fruits and vegetables (Yang *et al.*, 2004).

¹ Department of Seafood Processing, Faculty of Fisheries, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Islamic Republic of Iran.

* Corresponding author, e-mail: sarah_haghparast@yahoo.com



Because of the current interest in healthy food, antioxidative properties of red onion (*Allium cepa* L.) as a ubiquitous aromatic plant and herbal green tea (*Camellia sinensis*) have been demonstrated in various studies (Helen *et al.*, 2000; Kawamoto, 2004). Phenolic compounds in red onions including anthocyanins, flavonoids, quercetin and volatile sulfuric components show chelating and free radical scavenging activities (Zielinska *et al.*, 2008), resulting in shelf life extension of fat-containing food systems. According to data published by Hertog *et al.* (1992), red onions exhibited the highest quercetin level (284-486 mg kg⁻¹) in a survey of 28 vegetables and 9 fruits. Also, green tea leaves (*Camellia sinensis* L.) contain other strong well known antioxidant components, in that, catechins have been shown to minimize the oxidation ability of fatty acids by chelating iron and copper which cause the disruption of metal-catalyzed free radical formation (Chander *et al.*, 2005). It has been detected that trolox and other natural antioxidant compounds such as caffeic acid, chrogenic acid, quercetin, rutin and catechins are stronger scavengers as compared to vitamins C and E (Qu *et al.*, 2001). Remarkably, an increasing volume of research during the past decade has provided the evidence that flavonoids in these two widely consumed food plants exert many beneficial properties on human health like hypochlosterolic, hypolemidemic, hypoglycaemic, thrombotic, potent anticancer and cardiovascular effects (Higdon and Frei, 2003; Yang *et al.*, 2001; Campos *et al.*, 2003). Persian sturgeon (*Acipenser persicus*) is a commercially best-sold fish in retail outlets. However, the presence of high concentrations of labile phospholipids in fillets makes it more susceptible to oxidative deterioration. To date, few reports on sensory and chemical changes of sturgeon fillets have been available to determine the acceptable time for storage under different conditions. The overall objective of the present study was to elucidate the feasibility of red onion (*Allium cepa* L.) and green tea (*Camellia sinensis* L.) for lipid peroxidation and quality

control of sturgeon fillets during refrigerated storage (4°C) with the preservation and enhancement of sensory properties. The specific objectives of this work were: (a) to compare the effectiveness of these two plants with naturally occurring antioxidative components, and (b) to optimize the level of green tea extract and onion juice addition to the fish fillet.

MATERIAL AND METHODS

Chemicals and Reagents

Sodium sulphate, acetone, TBA-i reagent, phenolphthalein, acetic acid, sodium carbonate, tannic acid, Folin-ciocalteu's phenol reagent and hydrochloric acid were supplied by Merck (KGaA, 64271 Darmstadt, Germany). Sodium hydroxide, ethanol and chloroform were purchased from Sigma (St. Louis, MO).

Plant Sample

Fresh red onions (*Allium cepa* L.) were purchased from a local grocery and the flesh part was used in the trial after peeling. Fresh leaves of *Camelia sinensis* were obtained from a retail market. To prepare ground dry green tea, the leaves were steamed (at 90±5°C) for 30 seconds, immediately cooled in iced-bowls and then reduced in size to give 80-mesh size powder. The crushed leaves were dried at 60 °C for 2 hours, and re-powdered in a burr mill.

Extraction of Green Tea

Ten g of ground dry green tea was added to 100 ml of distilled water and heated at 30-40°C for 45 min with a magnetic stirrer (DELTA Model HM-101). The mixture was then filtered with a Whatman filter paper No.42 and the filtered solution with soluble solid content was applied as green tea extract (TE) in the experiment.

Extraction of Onion Juice

Fifty g of finely chopped red onions (*Allium cepa* L.) were thoroughly agitated with 500 ml of preheated water (90°C) for 60 min in order to extract maximum content of phenolic compounds. The mixture was cooled to room temperature and was homogenized in a blender (Pars Khazar, 320, Iran). The homogenate was centrifuged at 10,000 rpm for 20 min in a high speed centrifuge (Sigma 3K30, Germany) and the resulting supernatant was used as onion juice (OJ).

Determination of Total Phenolics

Total phenolics were determined by a colorimetric method of Folin-Ciocalteu reagent (Singh et al., 2002). The green tea extract and onion juice at each prepared concentration were dissolved in 80% aqueous methanol (2: 1 v/v). 0.5 ml of the solution was well mixed with 1ml of diluted Folin-Ciocalteu reagent (1:10 with distilled water) and 0.8 ml of 7.5% Na₂CO₃. The mixture was allowed to stand for 30 min at room temperature and absorbance was measured at 765 nm with a spectrophotometer (Lightwave S2000 UV/VIS diode array spectrophotometer). The standard curve was prepared using 10, 20, 30, 40, 50, 60, 70, and 80 mg L⁻¹ solutions of tannic acid in methanol: water (80:20, v/v). Total phenol values were expressed in terms of tannic acid equivalent (TAE) (mg g⁻¹ of dry mass for green tea and fresh weight for red onion). The estimation of phenolic content was replicated three times and the results were averaged.

Sample Preparation

Fresh sturgeon fish were caught from the southeast part of the Caspian Sea (Iran) and transported to the laboratory in boxes containing enough ice within 2 hours *post mortem*. The average weight and total length of the fish were 12±2 kg and 80±5 cm. They were beheaded, eviscerated, skinned and

filleted by using common household methods in medium length of 15±1 (cm) and weight of 350±5 (g). The mean compositional contents of moisture, protein, lipid and ash (AOAC, 1995) in fresh fillets of sturgeon are presented in Figure 1. Preparation of the antioxidant solutions was performed freshly prior to fish catching in the laboratory. Different aqueous solutions of each plant extract were provided by dilution in distilled water at concentrations of 1%, 2.5%, and 5% (v/v). Fillets were tumbled in prepared dipping solutions in a ratio 1:1 (w/v) for 10 min. After packaging with polyvinylidene film, the samples were placed in Styrofoam trays and stored at 4 °C for up to 8 days. The analysis of lipid oxidation indices and sensory characteristics was conducted at 5 intervals (0, 2, 4, 6 and 8 days).

Chemical Analysis

The pH value was recorded using a pH meter (Metrohm, 713ph Meter-Herisau Switzerland), according to the method of Benjakul *et al.* (1997). Fish sample (0.5g) was centrifuged thoroughly with 4.5 ml of distilled water for 1 min at 1800 rpm and the homogenate was used for pH determination.

The lipid was extracted by the method of Bligh and Dyer (1959).

Peroxide value (PV) was calculated according to the method of Egan et al. (1997) and the results were expressed as meq oxygen kg⁻¹ lipids.

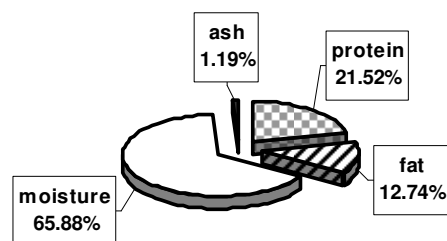


Figure1. Pie diagram showing the proximal composition of Persian sturgeon fillets.



The thiobarbituric acid index (TBA-i) was determined using the method of Tarladgis (1969) and expressed as mg malonaldehyde kg^{-1} of fish flesh. To do this, 10 g of fish sample was homogenized for each group, applied for TBARS analysis, and the color development was measured at 538 nm using a lightwave-S2000 UV/VIS diode array spectrophotometer.

Free fatty acid (FFA) content was measured as described by Egan *et al.* (1997). Results were expressed as %oleic acid (i.e. the cm^3 0.1 N NaOH used in the titration corresponding to % oleic acid).

Determination of heme iron content was conducted as described by Clark *et al.* (1997). Two g of fish sample was added to 9 ml of acetone solution containing 90% acetone, 8% distilled water and 2% HCl with the normality of 12.7. After keeping in a small dark chamber for 30 min, the heme iron content was calculated using the following equations where A is the absorbance of the sample against a blank of acetone acid at 640nm.

$$[1] \text{ Total pigment (ppm)} = A \times 680$$

$$[2] \text{ Heme iron (ppm)} = \text{total pigment} \times 8.82/100$$

Sensory Analysis

Sensory analysis (taste, color and odor) were assessed according to the descriptive sensory method of Hedonic (ASTM, 1969) with slight modification. A panel of 10 assessors was trained to evaluate each attribute and score them on a falling scale consisting of five points according to the guidelines in Table 1 at each sampling occasion. Fillets of fish were cooked in hot water steam for 20 min and the panelists were asked to wash their mouths with fresh water for several times before taste scoring.

Statistical Analysis

The experimental design was a factorial $7 \times 5 \times 3$ (7 treatments including the control, 5 sampling occasions, and three replicates).

Table1. Descriptive sensory evaluation definitions

Sensory attributes description	score
Natural flavor, color and odor of fillet	5
No sensible change in natural flavor, color and odor	4
Sensible discoloration, Slightly sour odor and incipient rancidity in flavor	3
No natural color, moderately off-odor and off-flavor	2
Sharply sour and extremely rancid flavor/odor, extremely discolored	1

Sourced by ASTM, 1969

The chemical and sensory data were subjected to one way ANOVA with $\alpha=0.05$. Comparisons within each analysis day and within a treatment at different sampling times were performed by least significant difference (LSD) test with statistical analysis system (SAS) program ($\alpha=0.05$).

RESULTS AND DISCUSSION

The mean compositional contents of moisture, protein, lipid and ash in fresh fillets of sturgeon were assessed by the method of AOAC (1995). As shown in Figure 1. The moisture, protein, lipid and ash contents of fresh sturgeon fillets were estimated to be 65.88%, 1.19%, 21.52% and 12.74%, respectively.

Phenolic Content of Plant Extract

Total phenolic content of extracts at different concentrations in terms of tannic acid equivalent (the standard curve equation: $y=0.00075x + 0.0091$, $r^2 = 0.9945$) is indicated in Table 2. In the present trial, the lowest (3.13 mg TAE g^{-1} dw) and highest (538.2 mg TAE g^{-1} dw) content were found in aqueous solutions of 1% OJ and 5% TE, respectively. According to the results in Table 2, green tea extract has much more content of phenolic compounds as compared to onion juice at the same concentration.

Table 2. Phenolic content of green tea extract (TE) and onion juice (OJ) at different concentrations.

Phenolic content (mg TAE/gdw) ^a			
TE 1%	145.6±0.037	OJ 1%	15.40±0.031
TE 2.5%	243.6±0.012	OJ 2.5%	37.39±0.022
TE 5%	538.2±0.055	OJ 5%	85.76±0.04

^a mg Tannic acid equivalents per gram dry weight of sample.

Benkeblia (2000) reported the total phenolics of red onion var. *Rouge Amposta* from 18 to 20 mg 100g⁻¹ fresh weight. Phenolic content in onions varies considerably particularly with cultivar (Bajaj et al., 1980). As evidenced by Samman et al (2001), the total phenolic compounds of green tea extracts were 117.3 mg gallic acid equivalents/g. It has been concluded that brewing conditions such as extraction temperature, period of extraction, ratio of tea leaves to extracting water, and stirring are important factors for determining the total phenolic content in green tea extract (Liebert et al, 1999).

Changes in pH Content

Changes in pH value of sturgeon fillets dipped in antioxidant solutions and the control during 8 days of storage are shown in Table 3. The pH value assessed as a crucial factor for determination of meat quality (Nam *et al*, 2001), might interfere with solubility activities of antioxidants by changing their electrical charges (Decker *et al*, 2005). In the present study, a gradual

increase in the pH value of fish fillets was observed during 8 days of storage (p<0.05) and reached to the maximum of 7.19 at the end of sampling time (day 8). The increase in pH was postulated to be due to an increase in volatile basic compounds produced by either endogenous or microbial enzymes (Cann et al, 1983), and decomposition of nitrogenous components (Benjakul *et al.*, 2002). It has been emphasized that the pH value can be raised to pH 7 or 8 during storage period (Poulter and Nicolaides, 1985). Additionally, Sikorski *et al.* (1990) reported that the enzymatic degradation of ATP caused the liberation of inorganic phosphate and ammonia, leading to the changes in pH value. The most significant (P<0.05) differences among various treatments and within each analysis day were observed through the first 2 days of storage time whereas the pH content of treated fillets and the control exhibited no significant (P>0.05) difference on days 4, 6 and 8. As storage time increased, the pH values of 5% OJ-treated samples showed no change while gradually irregular increases were detected in other samples, suggesting a degree of stability in pH by an increase in the amount of green tea extract added (Table 1).

Changes in PV Content

The effects of green tea extract and onion juice dipping solutions on the changes in PV content of the sturgeon fillets during 8 days of storage at 4°C are shown in Table 4. Peroxide value (PV) which is a measure of

Table 3. Changes in pH value of sturgeon fillets tumbled in tea extract (TE) and onion juice (OJ).

	Storage time (days)				
	0	2	4	6	8
1%TE	6.62±1.40 ^{BCD}	6.63±1.41 ^{CD}	6.54±1.38 ^{AB}	6.85±1.46 ^A	6.84±1.46 ^{ab}
2.5%TE	6.51±1.38 ^D	6.60±1.40 ^{bc}	6.58±1.40 ^{AB}	6.84±1.46 ^a	6.81±1.45 ^{ab}
5%TE	6.72±1.43 ^{ABC}	6.74±1.43 ^b	6.55±1.39 ^{AB}	6.83±1.46 ^{ab}	7.19±1.56 ^A
1%OJ	6.59±1.40 ^{CD}	6.73±1.43 ^{ABCD}	6.60±1.40 ^A	6.81±1.45 ^a	6.92±1.48 ^A
2.5%OJ	6.84±1.46 ^{ab}	6.79±1.45 ^{ab}	6.60±1.40 ^b	6.50±1.38 ^B	7.11±1.53 ^A
5%OJ	6.80±1.45 ^a	6.83±1.46 ^a	6.60±1.40 ^a	6.82±1.46 ^a	7.01±1.55 ^a
Control	6.63±1.41 ^{BCD}	6.69±1.42 ^{BCD}	6.37±1.35 ^B	6.65±1.41 ^{ab}	7.02±1.51 ^a

**Table 4.** Comparison of Peroxide value in sturgeon fillets tumbled in green tea extract (TE) and onion juice (OJ).

	Storage time (days)				
	0	2	4	6	8
1%TE	2.40±0.10 ^{eB}	3.26±0.10 ^{dB}	4.68±0.24 ^{cB}	6.28±0.09 ^{bB}	7.81±0.14 ^{aB}
2.5%TE	2.32±0.07 ^{dBC}	2.50±0.20 ^{dC}	3.26±0.12 ^{cD}	4.48±0.09 ^{bD}	4.81±0.21 ^{aD}
5%TE	2.21±0.09 ^{cC}	2.51±0.12 ^{dC}	3.06±0.11 ^{cD}	3.64±0.14 ^{bE}	4.28±0.11 ^{aE}
1%OJ	2.71±0.09 ^{eA}	3.89±0.10 ^{dA}	4.84±0.21 ^{cB}	6.42±0.18 ^{bB}	8.06±0.27 ^{aB}
2.5%OJ	2.70±0.10 ^{eA}	3.34±0.27 ^{dB}	3.96±0.12 ^{cC}	4.83±0.07 ^{bC}	6.12±0.08 ^{aC}
5%OJ	2.30±0.09 ^{eBC}	2.75±0.05 ^{dC}	3.32±0.08 ^{cD}	3.80±0.14 ^{bE}	4.75±0.21 ^{aD}
Control	2.68±0.06 ^{eA}	4.18±0.20 ^{dA}	5.68±0.08 ^{cA}	7.64±0.10 ^{bA}	10.01±0.11 ^{aA}

Results are expressed as means with standard error (n=3). Different superscript and subscript letters characterize significant difference in each column (A-D) and each row (a-e), respectively.

the concentration of peroxides and hydroperoxides formed in the initial stages of lipid oxidation is widely used for the estimation of oxidative rancidity in fats (Olafsdóttir et al., 1997). The PV scores rose sharply during storage time in all samples with significant differences being recorded among all treatments and the control at each sampling time. A slower increase in PV values was obtained in samples treated with onion juice and green tea extract, whereas a faster increase in PV of the control sample was evidenced after 4 days of storage time, demonstrating the oxidative stability of fish lipids by green tea extract and onion juice. The PV values of samples dipped in 5% OJ (4.75 meq oxygen kg⁻¹ lipids) or 5% TE (4.28 meq oxygen kg⁻¹ lipids) were approximately 1.7 and 1.8 times less than those of samples treated with 1% OJ (8.06 meq oxygen kg⁻¹ lipids) and 1% TE (7.81

meq oxygen kg⁻¹ lipids), respectively, at the end of storage time, which indicated that a high concentration of green tea and onion juice was more effective in controlling oxidation. Previous results by Alghazeer et al (2008) had confirmed that addition of instant green tea (250 and 500ppm) to Atlantic mackerel (*Scomber scombrus*) fillets decreased the rate of peroxides and hydroperoxides formation during 8 weeks of storage at -10°C, as compared to samples without green tea. However, it has been reported that a higher concentration of green tea may act as a pro-oxidant (Honglian and Etsuo, 2001).

Changes in TBA Content

Table 5 depicts the formation of malonaldehydes in sturgeon fillets during 8

Table 5. Comparison of thiobarbituric reactive substances (TBARS) content in sturgeon fillets tumbled in green tea extract (TE) and onion juice (OJ).

	Storage time (days)				
	0	2	4	6	8
1%TE	0.14±0.01 ^{dA}	0.27±0.08 ^{dAB}	0.60±0.06 ^{cBC}	1.16±0.17 ^{bBC}	1.84±0.09 ^{aABC}
2.5%TE	0.13±0.01 ^{cA}	0.24±0.03 ^{bB}	0.30±0.09 ^{cD}	0.61±0.13 ^{bD}	0.97±0.09 ^{aDE}
5%TE	0.11±0.02 ^{bA}	0.20±0.00 ^{bB}	0.28±0.07 ^{bD}	0.51±0.12 ^{abD}	0.80±0.25 ^{aE}
1%OJ	0.14±0.01 ^{dA}	0.31±0.01 ^{cdAB}	0.70±0.06 ^{cB}	1.34±0.27 ^{bAB}	2.05±0.19 ^{aAB}
2.5%OJ	0.12±0.01 ^{cA}	0.29±0.03 ^{cdAB}	0.48±0.06 ^{bcBCD}	0.79±0.14 ^{bBCD}	1.50±0.29 ^{aBCD}
5%OJ	0.12±0.00 ^{cA}	0.25±0.02 ^{bcAB}	0.37±0.11 ^{bcCD}	0.70±0.11 ^{bCD}	1.29±0.30 ^{aCDE}
Control	0.16±0.02 ^{dA}	0.37±0.03 ^{dA}	1±0.08 ^{cA}	1.87±0.23 ^{bA}	2.50±0.23 ^{aA}

Results are expressed as means with standard error (n=3). Different superscript and subscript letters characterize significant difference in each column (A-D) and each row (a-e), respectively.

days of refrigerated storage. The levels of tissue malonaldehyde, a degradation product of lipid, are often assessed in order to evaluate the rate of lipid peroxidation occurring in biological systems (Khayat and Schwall, 1983). As shown in Table 3, the formation of malonaldehydes was significantly retarded in samples treated with plant extracts. Up to the sixth day of storage, significant ($P<0.05$) differences were observed in the TBA content of the treatments at all sampling times except for samples dipped in 5% TE. There were no significant ($P<0.05$) differences among treatments of 2.5% OJ, 5% OJ, 2.5% TE and 5% TE at all analysis times while a significant difference ($P>0.05$) was verified between samples treated by 2.5% OJ and 5% TE at the end of the storage time. As shown in Table 5, samples treated by 1% OJ indicated no difference ($P>0.05$) in contrast to the control all over the time except day 4. Conversely, according to the results demonstrated by Serdaroglu and Felekoglu (2005), adding onion juice to frozen sardine mince (*Sardina pilchardus*) (1ml: 100 g) delayed lipid oxidation for 3 months, although the TBA values in both OJ and control treatments were out of consumable limits at the end of 5 month storage. As deduced by the comparison of thiobarbituric acid content, antioxidant efficiencies of onion juice and tea extract at the same concentration were similar ($P>0.05$) in sturgeon fillets on every sampling occasion. Sturgeon fish used in this study is a

medium-fat fish with nearly 13% lipid content and prone to lipid oxidation. On the 8th day of storage, the lowest amount of malonaldehyde was observed in refrigerated fillets tumbled in 5% TE. The minimum TBA value for fillets dipped in higher concentrations of green tea extract well suggests the positive correlation between phenolic content of green tea extract and antioxidant properties of these compounds to prevent or retard malonaldehydes formation. In an investigation by Tang *et al.* (2001), tea catechins added at concentrations greater than 300 mg kg⁻¹ were necessary to reduce lipid oxidation in mackerel patties as indicated by significant decrease ($P<0.05$) in TBA content. However, previous report by Van Het Hof (1997) revealed that green tea extract applications have no prohibition influence on lipid deterioration by using TBA and MDA analysis.

Changes in FFA Content

Variations in free fatty acid content of sturgeon fillets are depicted in Table 6. Progressive oxidation and enzymatic hydrolysis of unsaturated fatty acids is the main cause of lipid deterioration in fatty fish which is accompanied by the formation of free fatty acids (Srikar and Hiremath, 1972). It has been proved that FFA formation in fish products, is not the only agent of nutritional decrease, but its accumulation relating to lipid oxidation exerts a strong

Table 6. Comparison of free fatty acids (FFA) in sturgeon fillets tumbled in green tea extract (TE) and onion juice (OJ).

	Storage time (days)				
	0	2	4	6	8
1%TE	1.06±0.19 _d ^A	1.41±0.19 _d ^B	2.94±0.18 _c ^{AB}	3.98±0.16 _b ^{BC}	4.78±0.12 _a ^B
2.5%TE	1.10±0.10 _d ^A	1.29±0.44 _d ^B	2.23±0.22 _c ^{BC}	3±0.13 _b ^D	3.80±0.08 _a ^{CD}
5%TE	1.11±0.06 _c ^A	1.21±0.33 _c ^B	1.86±0.07 _{bc} ^C	2.85±0.54 _{ab} ^D	3.18±0.31 _a ^D
1%OJ	1.19±0.12 _c ^A	1.68±0.37 _c ^{AB}	2.97±0.15 _b ^{AB}	4.22±0.42 _a ^{AB}	4.77±0.29 _a ^B
2.5%OJ	1.23±0.13 _c ^A	1.73±0.26 _c ^{AB}	2.64±0.44 _b ^B	3.20±0.31 _{ab} ^{DC}	4.03±0.06 _a ^C
5%OJ	1.10±0.22 _c ^A	1.58±0.29 _c ^{AB}	2.59±0.26 _b ^{BC}	3.01±0.12 _{ab} ^D	3.62±0.41 _a ^{DC}
Control	1.15±0.17 _e ^A	2.47±0.29 _d ^A	3.56±0.22 _c ^A	4.95±0.09 _b ^A	5.94±0.10 _a ^A

Results are expressed as means with standard error (n=3). Different superscript and subscript letters characterize significant difference in each column (A-D) and each row (a-e), respectively.



influence on sensory attributes (Roldan *et al.*, 2005). In the present trial, lipid hydrolysis as measured by FFA indicated a general increase in all treatments including the control during refrigerated storage. No significant ($P>0.05$) differences in the FFA content of various treatments were observed during the first 2 days of storage. For the latter refrigerated storage time, significant ($P<0.05$) differences were detected among different samples treated by plant extracts; therefore different inhibitory effects of phenolic compounds on lipid hydrolysis of refrigerated sturgeon fillets could be implied, depending on the amount of plant extract added. As shown by marked increase in free fatty acids, significant degradation of n-3 PUFA was observed in control samples after 8 days of refrigerated storage (Table 6). By the end of the storage time, FFA content of the samples including 2.5% TE, 5% TE and 5% OJ were % 2.13, % 2.75 and % 2.31 lower than that of control, respectively. These data demonstrate that tumbling of fish fillets in green tea extract and onion juice at concentrations mentioned above was more effective for preventing free fatty acids formation as compared with the others. Kumudavally *et al.* (2008) perceived that the application of sprayed-green tea extract (10ml kg⁻¹ of meat weight) could extend the shelf life of fresh mutton for up to 4 days at 25°C when registered nearly a 25% increase in FFA content of samples treated with green tea extract at day 4 against a 83% increase in the control sample at the end of 1

day storage. Serdarglu and Felekoglu (2005) noted that rosemary extract (100ppm) showed better inhibition of lipid degradation than onion juice (1ml 100g⁻¹) in sardine (*Sardina pilchardus*) mince as assayed by more increases in FFA content of samples with 1%OJ (v/w).

Changes in Heme Iron Content

The changes in heme iron content of sturgeon fillets during 8 days are also presented in Table 7. Fish flesh is a source of iron-containing complexes including, myoglobin, haemoglobin and iron-bound proteins such as ferritin and transferrin (Hazell, 1982). Processing techniques such as filleting and mincing as well as storage can lead to the release of this iron from the complexes, and then catalyze lipid autoxidation in fish muscle (St. Angelo, 1996). As indicated in Table 7, heme iron content increased in the control and samples treated by 2.5% OJ, 2.5%TE and 5% TE after storage for 2 days. Afterwards, a significant ($P<0.05$) decrease was recorded which was especially sharp at day 6 for all treatments including the control. This could be due to the release of iron from heme to change into non-heme iron. In addition, measurement of heme iron content detected a significant ($P<0.05$) increase in the control and 2.5% OJ, 2.5% TE-treated samples during the last 2 days of storage time (days 6 and 8). Markedly, treatments with 2.5% TE,

Table 7. Comparison of heme iron content in sturgeon fillets tumbled in green tea extract (TE) and onion juice (OJ)

	Storage time (days)				
	0	2	4	6	8
1%TE	5.15±0.361 ^{abBC}	5.5±0.098 ^C	4.91±0.006 ^{bCB}	2.56±0.058 ^{CD}	3.02±0.157 ^{BC}
2.5%TE	5.66±0.04 ^{bA}	6.03±0.053 ^A	5.26±0.032 ^B	3.03±0.07 ^B	3.31±0.095 ^{dAB}
5%TE	5.52±0.174 ^{AB}	6.21±0.058 ^A	5.53±0.07 ^A	3.5±0.058 ^A	3.26±0.083 ^{cAB}
1%OJ	5.01±0.062 ^{aC}	5.26±0.078 ^{aCD}	4.46±0.235 ^C	2.69±0.167 ^{cCB}	2.86±0.058 ^{cCD}
2.5%OJ	4.94±0.117 ^{bC}	5.44±0.062 ^{aC}	4.93±0.162 ^{bCB}	2.31±0.058 ^{dD}	2.69±0.121 ^{dD}
5%OJ	5.11±0.115 ^{aBC}	5.1±0.115 ^D	5.1±0.362 ^{aB}	2.98±0.147 ^{bB}	3.34±0.058 ^{bA}
Control	5.21±0.061 ^{bABC}	5.76±0.072 ^{aB}	4.47±0.036 ^{cC}	2.32±0.164 ^{eD}	2.72±0.058 ^{dD}

Results are expressed as means with standard error (n=3). Different superscript ad subscript letters characterize significant difference in each column (A-D) and each row (a-e), respectively.

5% OJ, and 5% TE had the highest contents of heme iron at the end of storage time being 3.31, 3.34 and 3.12, respectively. This can be implicated in the positive effect of phenolic compounds presenting in considerable content in green tea extract added at concentrations higher than 1% (v/v) where even onion juice with a low content of phenolic components inhibited iron release in sturgeon fillets. Upon extended refrigerated storage, a slower rate of decrease in heme iron content of fillets was observed with increasing the concentration of green tea extract. However, fillets tumbled in 5% TE showed the highest heme iron content on most days of storage time as compared to other samples. Many in vitro studies have shown that metal-chelating properties of flavonoids in green tea have protective effects on cells and tissues against damages caused by free oxygen radicals (Rietveld and Wiseman, 2003; Kashima, 1999).

Changes in Sensory Attributes

Results of sensory difference testing in treated sturgeon fillets during 8 days of storage at 4°C are presented in Figure 2. All treatments developed off-odor with increased storage time, with the lowest and the highest off-odor detected on samples treated with 5% TE and 1% OJ, respectively. Meanwhile, the early signs of off-odor appeared in 1% OJ-treated fillets and the control on day 4 but developed in other treatments after 6 days of storage time. Younathan *et al.*, (2006) revealed that hot water extracts of yellow onion peels controlled rancid odor of cooked ground turkey even though TBA numbers were high. Sensory properties of food products are the key factors in consumer attraction (Gray *et al.*, 1996) and the implication of lipid peroxidation in flavor and aroma deterioration, as well as diminished food wholesomeness and food safety has been confirmed (Kanner and Rosenthal, 1992). Up to 4 days of storage, no significant

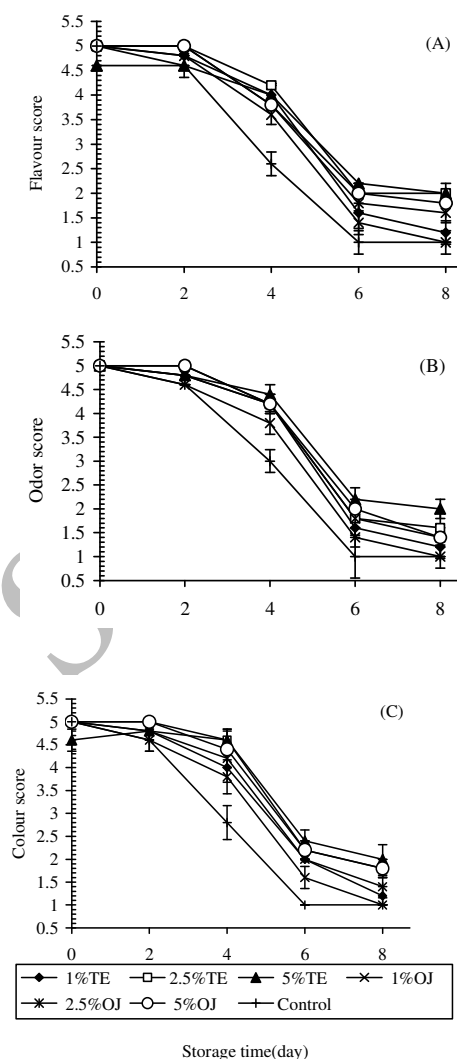


Figure 2. Comparative evolution of flavor (A), odor (B) and color (C) scores in sturgeon fillets treated by tea extract (TE) and onion juice (OJ) at three concentrations (1%, 2.5%, 5%) during refrigerated storage. Bars denote standard error of the mean (n=3).

($P > 0.05$) differences were observed among treated samples with the discovery of off-flavor in the control (Figure 2). Most differences among various treatments including the control were recognized on the 6th day when a sharp increase of off-flavor was detectable in 1% OJ and control samples. By the end of the storage time, samples tumbled in 1% OJ, 1% TE and the control had the lowest flavor scores while



treatments with upper concentrations of plant extracts were scored highly in flavor analysis. Tang and Cronin (2007) noted that the incorporation of onion juice during the preparation of encased turkey breast rolls led to the production of premium quality product processing and improved flavor quality and stability in sliced form. As stated by Scideman *et al.*, (1984) meat color is considered to be one of the most important quality attributes in the acceptance or rejection of the product. Refrigerated fillets treated by 2.5% TE, 5% TE and 5% OJ showed a much superior color quality after storage for 8 days at 4 °C (Figure 2). Although increase in the concentration of green tea extract had beneficial effects on chemical changes in surgeon fillets particularly at the end of the storage time, panelists characterized the new slightly bitter and grassy flavor with greenish-yellowish color for 5% and 2.5% TE treatments at all sampling times. The discoloration changes of fish fillets dipped in 2.5% and 5% TE steadily increased with time in storage. This might be implicated in the possible penetration of chlorophyll pigments and their subsequent interference with other biochemically active compounds in sturgeon fillets, which caused an undesirable change in meat color. Persian sturgeon is a white-muscled fish with pale pink flesh coloration after cooking by steam. This finding provides the evidence that tea extracts, particularly at higher concentrations, are good inhibitors to lipid damage progression but incapable of retaining and/or promoting the natural flavor and color of sturgeon fillets. In this sense, Liu (1997) stated that clove had an antioxidative impact on lipid degradation of marinated catfish with a significant decrease in the sweet taste of the meat. Also, polyphenols of green tea as a natural antioxidant had a similar effect on sensory attributes of (*Clupeonella cultriventris*) fillets during iced storage as compared to Ascorbic acid as synergist (, 2004). This is addressed as "Ojagh, S. M., Sahari, M. A., and Rezaei, M. 2004. Effects of natural

antioxidants (β -carotene and green tea poly phenols) on quality of *Clupeonella cultriventris* fillets during iced storage, *Iran. J. Mari. Sci. Tech.* 3 (4).

CONCLUSION

It might be concluded that green tea extracts and onion juice in concentrations higher than 1% (v/v) and 2.5% (v/v), respectively, had more antioxidant characteristics for oxidative stability of lipids and shelf life enhancement in refrigerated sturgeon fillets which was more detectable with increased storage time. The present study does not reflect a clear dose-relationship between the total phenolic content of plant extracts and the results obtained from the lipid oxidation indices assessed in refrigerated sturgeon fillets. However, color and flavor of samples with 5% TE were unfavorable and might not be appealing to consumers. For further evaluation, comparison of green tea extract and onion juice from various varieties, by different extraction methods, and at other concentrations is proposed with the employment of microbiological analysis which has not been investigated in the present trial.

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ارزیابی اثرات عصاره چای سبز و پیاز بر فساد چربی و خواص حسی فیله های تاسماهی ایرانی: مطالعه مقایسه ای

س. حق پرست، ح. کشیری، غ. علیپور و ب. شعبان پور

چکیده

این مطالعه به منظور مقایسه میزان اثر بخشی عصاره پیاز قرمز (*Allium cepa* L.) و چای سبز (*Camellia sinenses*) بر نرخ اکسیداسیون چربی و خواص حسی در فیله های تاسماهی (*Acipenser persicus*) طی نگهداری در یخچال صورت گرفت. بدین منظور، فیله های تازه در محلول های حاوی عصاره پیاز (OJ) و چای (TE) در سه غلظت (v/v) ۱٪، ۲٫۵٪ و ۵٪ غوطه ور شده و سپس به مدت ۸ روز در یخچال نگهداری شدند. شاخص های شیمیایی مرتبط با اکسیداسیون چربی طی ارزیابی توسط میزان آهن هم، TBARS، FFA، کاهش بسیاری را در نمونه های تیمار شده با ۲٫۵٪ TE، ۵٪ TE و ۵٪ OJ در مقایسه با سایر تیمارها نشان دادند ($P < 0.05$). بیشترین تفاوت معنی دار ($P < 0.05$) در مقادیر pH تیمارها و شاهد طی ۲ روز اول نگهداری مشاهده شد. میزان pH در تیمار



OJ ۵٪ در طول دوره نگهداری ثابت بود ($P > 0.05$) درحالیکه تغییرات تدریجی در مقادیر pH سایر تیمارها محرز گردید. ترتیب اثر بخشی در جلوگیری از اکسیداسیون فیله های تاسماهی بصورت ۵٪ TE = OJ ۵٪ < TE ۲٫۵٪ < OJ ۲٫۵٪ < TE ۱٪ = OJ ۱٪ مشخص شد. بر اساس امتیازات حسی، عصاره پیاز در مقادیر بالاتر (۱٪ >) در بهبود خواص حسی فیله ها موثرتر بود.

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