

Effects of Sodium Salt Solutions (Sodium Acetate, Lactate and Citrate) on Physicochemical and Sensory Characteristics of Persian Sturgeon (*Acipenser persicus*) Fillets under Refrigerated Storage

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ABSTRACT

Effects of sodium salt solutions on physicochemical and sensory characteristics of refrigerated Persian sturgeon (*Acipenser persicus*) fillets during 12 days of storage were investigated. Fillets were dipped in solutions (2.5% w/v) of sodium acetate (SA), sodium lactate (SL), sodium citrate (SC) and distilled water (as control) for 10 minutes and then packaged. An assessment of TBARS, FFA, pH, heme iron and sensory attributes (flavor, color and odor) was carried out on 0,3,6,9 and 12 days past the storage time. Results indicated that TBARS values of each sample increased with the storage time ($P < 0.05$). Control showed the highest values of TBARS while sodium acetate sample had the significantly ($P < 0.05$) lowest figures among the treatments (1.04 for SA versus 2.34 for control). Lipid hydrolysis assessment revealed that the sodium salt treated samples, especially sodium acetate, acquired the lower FFA amounts ($P < 0.05$) as compared with control. No significant differences ($P > 0.05$) were observed among the pH values of the treatments. Heme iron assessment showed that the samples treated with sodium acetate contained more heme iron as compared with control. Sensory assessment revealed more desirable scores for the sodium acetate treated group as compared with others samples. The order for the sodium salt treated effects was: SA > SC > SL. As a consequence, sodium salts, in particular sodium acetate, might be considered as effective tools in preventing the quality degradation of the fillets, resulting in an extension of their shelf life.

Key words: Antioxidant, Persian sturgeon (*Acipenser persicus*), Sodium salts.

INTRODUCTION

No doubt, of all various food materials that have ever been valued for containing naturally high potential benefits, there is none that can be compared with fish and shellfish products. Marine lipids with their high level of polyunsaturated fatty acids (PUFA), in particular eicosapentaenoic (EPA, 20:5 ω 3) and docosahexaenoic acids (DHA, 22:6 ω 3) largely contribute to human diet (Ackman, 1999; Pigott and Tucker, 1987). On the other hand, they may act as

primary agents in autoxidative damage (Flick and Martin, 1992; Hsieh and Kinsella, 1989). Lipid oxidation is the most important problem for safeguarding seafood during either storage or processing (Tang *et al.*, 2001). The use of good manufacturing practices and hazard analysis of critical control point (HACCP) is crucial in production, storage, distribution and retailing of refrigerated foods, and because of consumer's demand for fresh refrigerated foods with extended shelf life, considerable researches have been directed toward using

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various preservation strategies to prolong the shelf life, while ensuring the safety of fresh foods including fishery products (Sallam, 2007). Sodium salts of such organic acids as acetic, lactic and citric acids, containing antimicrobial, antioxidative and flavor improving properties, have been proposed for prolonging the shelf life of meat (Maca et al., 1997) fish muscle (Zhuang et al., 1996) as well as poultry flesh (Williams and Philips, 1998).

Persian sturgeon is one of the Caspian Sea fishes with high commercial value and acceptability in various marketplaces. Therefore, it is necessary to pay the needed attention to the quality improvement as well as to extending the shelf life of this valuable commodity during storage. The aim of this study is to investigate the antioxidative properties of sodium salts (acetate, citrate and lactate) on prolonging the storage duration of Persian sturgeon.

MATERIALS AND METHODS

Preparation and Treatment of Fish Sample

Four fresh fish (*Acipenser persicus*) were purchased 3-4 hours after being caught, from a local market in Gorgan, North of Iran. The weight of the fish batch was about 11.0 ± 3.0 kg. The fish were headed, eviscerated, washed and immediately transported to the laboratory in boxes containing enough slurry ice. The fish were skinned and filleted manually. The muscular part of the truck was used for the analysis. Before preparing the treatments, the amounts of FFA, TBARS, heme iron and pH were assessed in the fresh sample (Table 1). Sodium salt solutions were prepared at the concentration of 2.5% w/v. Fillets were

dipped in pre-chilled (4.0°C) solutions of SA, SL, SC and distilled water (as control) up to 10 minutes. The ratio of the fillets to each solution was 1:2.5. The treatments were placed in Styrofoam boxes separately and packaged by over-wrapping with polyvinylidene film. The samples were then stored under refrigerated conditions (4.0°C) up to 12 days. For each group, triplicate samples were taken for chemical analyses on 0, 3, 6, 9 and 12 days of the storage time.

Preparation of Chemicals

All chemicals employed (sodium salts, solvents and reactants) were reagent grade (E, Merck, Darmstadt, Germany).

Physicochemical Analysis

Measurement of Proximate Pomposition

Fresh sturgeon fillets were analyzed in triplicate for moisture, lipid, protein and ash content according to the standard methods of AOAC (1990).

Measurement of TBARS

TBARS was determined according to the procedure of Tarladgis (1969). Commintuted fish fillet (10 gr) was thoroughly homogenized with 100 ml of distilled water and 2.5 ml of HCL (4M) along with 6-7 droplets of antifoaming. The mixture was subjected to the distillation process for 10 minutes. The obtained liquid (5 ml) was added to 4 ml of a solution containing 0.0288 gr thiobarbituric acid and 90% acetic acid. The mixture was heated in boiling water bath for 30 minutes and then cooled to ambient temperature ($\sim 22^{\circ}\text{C}$). TBARS was measured at 538 nm as follows, where D is the absorbance of the solution against the

Table 1. The values of TBARS, FFA, heme iron and pH in fresh sample.

	TBARS	FFA	pH	Heme iron
Fresh sample	0.11 ± 0.04	1.09 ± 0.1	5.96 ± 0.03	3.58 ± 16

blank sample prepared by adding 5 ml of distilled water and 5 ml of TBA solution.

TBARS (μM of malonaldehyde) = $7.8 \times D$

Measurement of FFA (Lipid ydrolisis)

Free fatty acid (FFA) content was measured as described by Egan (1997). Comminuted fish fillets (150 gr) were thoroughly mixed with 250 ml of chloroform for 10 minutes and filtered through Wattman filtr paper. Ten ml of the filtered dried liquid was used for sample oil content after being dried at 105°C . Free fatty acid content was expressed as oleic acid% through titration of filtered liquid (2 ml) containing 3 droplets of phenol phetalein with NaOH (0.1N) as follows:

(Oleic acid%) FFA = $(\text{ml of NaOH used in the titration} \times 28.2 \times 100) / (\text{Oil sample weight} \times 1000)$

Measurement of pH

For pH measurement, 5 gr of fish muscle was well mixed with 45 ml of distilled water and then the pH values recorded through a pH meter (Metrohm, 713 pH Meter-Herisau Switzerland) (Suvanich *et al.*, 2000).

Measurement of Heme Iron

The heme iron content was determined according the method of Clark *et al.* (1997). Nine ml of acetone acid was added to a 2 gr sample of fish. After being kept in a dark cabinet for 30 minutes, the total heme pigment content was assessed by direct spectrophotometric measurement at 640 nm:

Total pigment (ppm) = $A_{640} \times 680$

Heme iron (ppm) = $\text{Total pigment} \times 8.82 / 100$

Sensory Analysis

Sensory assessments included the evaluation of three parameters (flavor, color and odor). This was conducted by a panel (5 trained people) according to a hedonic scale

(ASTM, 1969) with slight modification. (Table 2)

Statistical Analysis

All the data were presented as means \pm standard error. The experimental design was a factorial $4 \times 5 \times 3$ (4 treatment including control, five storage times and three replicates). ANOVA was employed to find the interactions between values of different analyses and storage times. Data from pH, chemical and sensory results were subjected to ANOVA followed by Least Significant Difference test (LSD) using Statistical Analysis System (SAS Institute, Inc., 1996).

RESULTES AND DISCUSSION

Proximate Composition

The proximate composition of Persian sturgeon fillet averaged: 63.19% moisture, 20.69% crude protein, 1.96% ash, and 14.12% crude lipid.

TBARS Assessment

The levels of tissue malondyaldehyde, a degradation product of lipid, are often measured to determine the extent of lipid proxidation that has occurred in biological systems (Khayat and Schwall, 1983). Table 3 presents the changes of TBARS values in Persian sturgeon fillets. TBARS values of the control and sodium salts treated samples increased with the storage time ($P < 0.05$), the highest values being obtained on day 12 (2.34 for the control). However, a little decrease ($P > 0.05$) in TBARS value of

Table 2. Sensory assessment of Persian sturgeon fillets.

Score	Sensory attribute description
7	Natural flavor, color and odor of fillet
5	No sensible change in natural flavor, color and odor
3	Sensible decrease in natural flavor, color and odor
1	No natural flavor, color and odor
0	Completely off-flavor, white color and off-odor

Sourced by ASTM (1969).

**Table 3.** Changes in TBARS value during refrigerated storage under sodium acetate (SA), sodium citrate (SC), sodium lactate (SL), and control (distilled water).

Factors	Storage time (Days)				
	t ₁ = 0	t ₂ = 3	t ₃ = 6	t ₄ = 9	t ₅ = 12
SA	0.13±0.49 ^a _C	0.19±0.1 ^b _C	0.18±0.05 ^c _C	0.64±0.11 ^b _B	1.04±0.1 ^c _A
SC	0.12±0.03 ^a _C	0.19±0.01 ^b _C	0.60±0.1 ^b _B	1.40±0.2 ^a _A	1.73±0.2 ^b _A
SL	0.12±0.02 ^a _D	0.21±0.1 ^b _D	0.62±0.1 ^b _C	1.40±0.2 ^a _B	1.90±0.05 ^{ab} _A
Control	0.12±0.03 ^a _D	0.40±0.01 ^a _D	0.90±0.01 ^a _C	1.75±0.2 ^a _B	2.34±0.2 ^a _A

Different superscript letters characterize significant differences in each column (a-c) and in each row (A-D) for $P < 0.05$.

sodium acetate was observed on day 6. This may be the result of the interaction between malondyaldehyde and other such components of fish body as amines, nucleosides, nucleic acids, proteins, phospholipids and other aldehydes which are some of the end-products of lipid oxidation (Aubourg, 1993). In this study, the use of sodium salts reduced the rate of TBARS formation in sturgeon fillets as compared with control. A comparison of different samples revealed that TBARS levels of fillets treated with sodium acetate were significantly ($P < 0.05$) lower than those treated with sodium lactate and control on most of the storage dates (Figure1). On this basis, sodium citrate treatment attained lower TBARS values by analogy with control and sodium lactate samples whereas the differences between sodium citrate and sodium lactate treatments were not significant ($P > 0.05$) during the trial. No significant ($P > 0.05$) differences were observed between control and sodium lactate group on most of the storage trial times (Table3). Conversely, William *et al.* (1995) revealed the significantly lower TBARS levels in Catfish fillets treated with 2% sodium lactate through 8 days of storage at 1°C. However, our results are in agreement with the previous work by Sallam (2007) who demonstrated that dipping of sliced Salmon fillets in sodium lactate produced lower TBARS levels than those of the control with the differences between sodium lactate and the control not being significant ($P > 0.05$) during chilled storage. It was also demonstrated that sodium citrate treatment

ended up with better results in comparison with sodium acetate and lactate samples. In this study, TBARS values of fillets treated with sodium acetate were significantly ($P < 0.05$) lower than those of sodium citrate and lactate treated after 3 days of storage (1.04 for SA versus 1.73 and 1.90 for SC and SL, respectively). Different effects of these salts on lipid oxidation may be the result of various factors including the extent of microbial growth, packaging method as well as storage time (Sallam, 2007). However, the use of sodium salts, especially sodium acetate, reduced the rate of TBARS formation in the fillets, demonstrating the effectiveness of these salts in reducing the oxidation rate during chilled storage.

Lipid Hydrolysis (FFA)

An evaluation of lipid hydrolysis in Persian sturgeon is presented in Figure 2. In comparison with the initial material, the free fatty acid (FFA) values of the control and sodium salts treated samples significantly ($P < 0.05$) increased with the storage time and reached their maximum levels after 12 days in each sample. This may be due to the effect of lipid hydrolyzing enzymes (mainly lipase and phospholipase) in decomposing the fats in fish tissue (phospholipids and triglycerides) (Serdaroglu and Felekoglu, 2005). Sodium salts treated samples showed lower FFA values when compared with control ($P < 0.05$). Accordingly, inhibitory effects of sodium salts on lipid hydrolysis could be concluded. In this study, the release

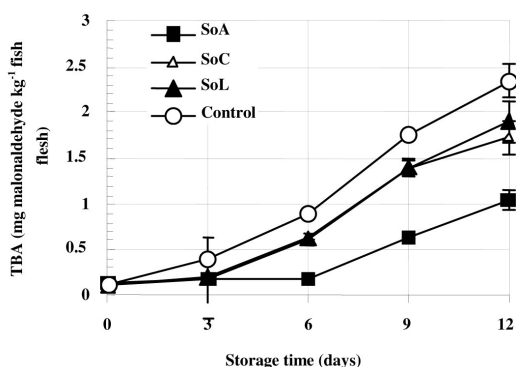


Figure 1. Comparison of TBARS values in persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

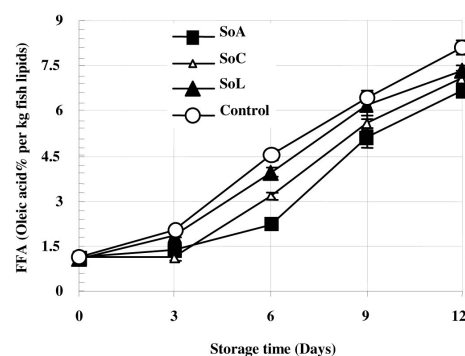


Figure 2. Comparison of FFA values in persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

of FFA in sodium acetate sample was significantly ($P < 0.05$) lower than that in sodium lactate group (6.65 for SA versus 7.38 for SL on day 12, Table 3). The order of the preventive effects of these salts on FFA formation was: sodium acetate > sodium citrate > sodium lactate. However, no significant ($P > 0.05$) differences have been observed in FFA levels either between sodium acetate and sodium citrate or between sodium citrate and sodium lactate treatments. Although the formation of FFA itself does not lead to nutritional losses, it has been proved that the accumulation of FFA is related to lipid oxidation (Serdaroglu and Felekoglu, 2005) and texture deterioration through interaction proteins (Mackie, 1993). In the current study, sodium salts, in particular sodium acetate conspicuously reduced the rate of lipid damage in Persian sturgeon fillet.

Changes in pH

The changes of pH values of the fillets during the 12 days are presented in Figure 3. The level of pH in living fish is close to the neutral (Massa *et al.*, 2005), however, it could be affected by some such factors as species, feeding and storage temperature (Pacheco-Aguilaret *et al.*, 2000). Acidity (pH values) of all samples slightly increased

with the storage time ($P > 0.05$). An increase in pH may be attributed to the increase in volatile bases caused by bacterial activity (Cann *et al.*, 1983). Benjakul *et al.* (2002) showed that the decomposition of nitrogenous compounds caused increase in pH of fish flesh. In this study, sodium salts did not have any significant ($P > 0.05$) effect on pH changes of sturgeon fillets as compared with the control and as well no significant ($P > 0.05$) differences were observed between different treatments (Table 5). Similar results have been reported by Al-Sheddy *et al.* (1999), who showed that there were not significant differences in sodium acetate (10% w/w) treatment in

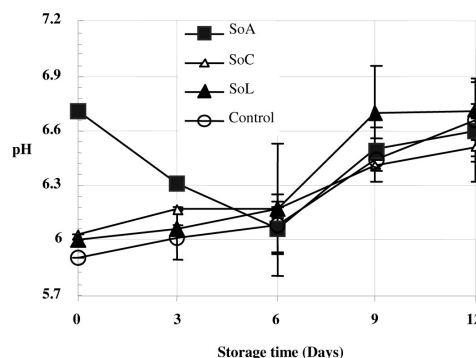


Figure 3. Comparison of pH values in persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

**Table 4.** Changes in FFA content during refrigerated storage under sodium acetate (SA), sodium citrate (SC), sodium lactate (SL), and control (distilled water).

Factors	Storage time (days)				
	t ₁ = 0	t ₂ = 3	t ₃ = 6	t ₄ = 9	t ₅ = 12
S.A	1.10±0.1 ^a _D	1.14±0.04 ^b _D	2.25±0.3 ^c _C	5.11±0.1 ^c _B	6.65±0.3 ^c _A
S.C	1.10±0.1 ^a _D	1.12±0.14 ^b _D	3.19±0.25 ^{bc} _C	5.60±0.3 ^{bc} _B	7.09±0.1 ^{bc} _A
S.L	1.10±0.04 ^a _D	1.18±0.14 ^b _D	3.94±0.5 ^{ab} _C	6.19±0.15 ^{ab} _B	7.38±0.24 ^b _A
Control	1.10±0.12 ^a _E	2.45±0.2 ^a _D	4.51±0.2 ^a _C	6.45±0.23 ^a _B	8.12±0.1 ^a _A

Different superscript letters characterize significant differences in each column (a-c) and in each row (A-D) for $P < 0.05$.

comparison with sodium lactate (5% v/v of 60% solution) and trisodium citrate (1.5% w/w) treatments in camel meat after 12 days of storage.

Changes in Heme Iron Content

Iron mainly exists in heme pigments, ferritin protein and as well in such active parts of enzymes as lipoxigenase (Dragoev *et al.*, 1998). This is the main active catalyst in meat rancidity (tang *et al.*, 2001). The determination of heme iron content as a quality degradation index indicates the relationship between the release of iron and increase of rancidity in fish (Dragoev *et al.*, 1998). Figure 4 shows the changes of heme iron contents in sturgeon fillets treated with sodium salts solutions and distilled water during refrigerated storage. During the first 3 days of storage, the heme iron content increased markedly ($P < 0.05$). Thereafter, the levels of heme iron decreased with increase in the storage time. This might be because of the higher soluble heme pigment in fresh meat caused by autolysis that might contribute to the greater extractability of

heme pigments. Also, the decrease observed in heme iron content may be presumably due to the release of free iron from heme. Benjakul and Bauer (2001) reported that the decrease in heme iron content was inversely related to non-heme iron content. The comparison of different treatments showed that the control group had lower heme iron content and reached its minimum value on day 9. Conversely, higher heme iron content was observed in sodium acetate treatment. No significant ($P > 0.05$) differences have been observed in heme iron contents between the control and sodium lactate as well as between sodium citrate and sodium lactate treated groups (except the day 9). However, in our study, sodium acetate showed higher amounts of heme iron as compared with other treatments, although the differences were not significant ($P > 0.05$) on most of the storage days.

Sensory Analysis

The results of sensory analysis of fillets are shown in Figures 5, 6 and 7. Fishes are particularly sensitive to oxidative rancidity

Table 5. Changes in pH value during refrigerated storage under sodium acetate (SA), sodium citrate (SC), sodium lactate (SL), and control (distilled water).

Factors	Storage time (days)				
	t ₁ =0	t ₂ =3	t ₃ =6	t ₄ =9	t ₅ =12
S.A	6.07±0.03 ^a _A	6.31±0.12 ^a _A	6.06±0.12 ^a _A	6.50±0.27 ^a _A	6.60±0.23 ^a _A
S.C	6.03±0.01 ^a _C	6.17±0.04 ^a _B	6.17±0.03 ^a _B	6.41±0.04 ^a _A	6.51±0.06 ^a _A
S.L	6.00±0.03 ^a _C	6.06±0.4 ^a _{BC}	6.17±0.25 ^a _{ABC}	6.70±0.04 ^a _{AB}	6.71±0.16 ^a _A
Control	5.91±0.12 ^a _A	6.02±0.17 ^a _A	6.10±0.12 ^a _A	6.44±0.23 ^a _A	6.66±0.32 ^a _A

Different superscript letters characterize significant differences in each column (a-c) and in each row (A-E) for $P < 0.05$.

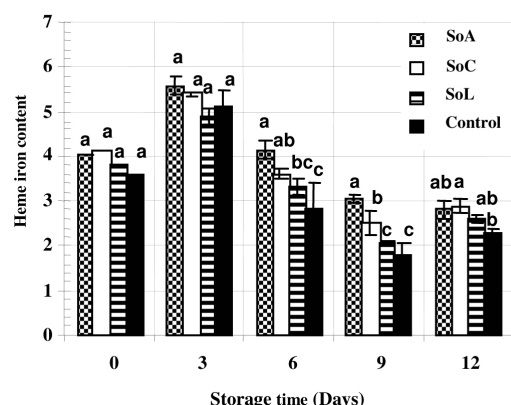


Figure 4. Comparison of heme iron values in persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

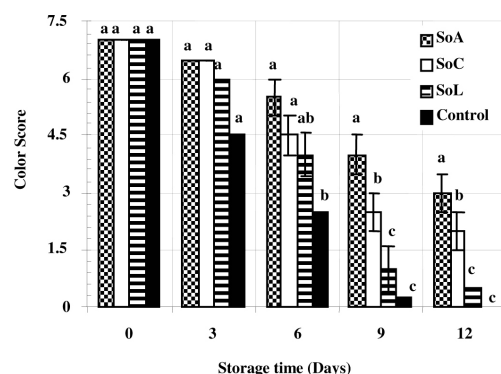


Figure 5. Comparison of color scores given to persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

(Khayat and Schwal, 1983) because of containing high levels of unsaturated fatty acids (Morris and Culkin, 1989). Rancidity development may produce some undesirable changes in flavor (Karahadian and Lindsay, 1989), color (Haard, 1992) and external qualities (Undeland and Lingnert, 1999). Sensory assessments led to the best score (score 7) on day 1 for all samples and then the quality of the fillets decreased with the storage time. Significant effect of the storage time on sensory attributes of the fillets varied in the different treatments. Sensory analysis indicated that the use of sodium

salts reduced the rate of fillets' deterioration in comparison with control which achieved the lowest score (score 0) on terminal days and was no longer acceptable, however, treated samples did not attain this score during the trial. Sodium citrate treatment, obtained better sensory scores as compared with sodium lactate, but their differences were just significant ($P < 0.05$) on days 9 and 12. In the current study, an important improvement was obtained in sensory attributes of fillets treated with 2.5% sodium acetate although there were no significant ($P > 0.05$) differences between sodium salts

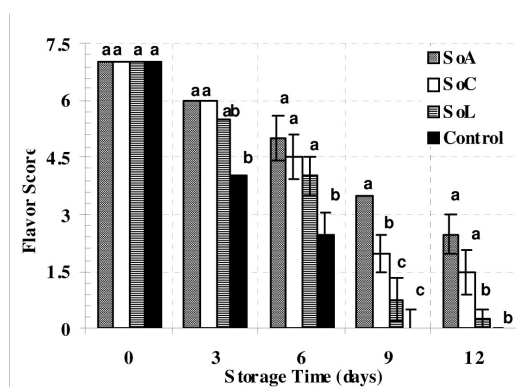


Figure 6. Comparison of flavor scores given to persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).

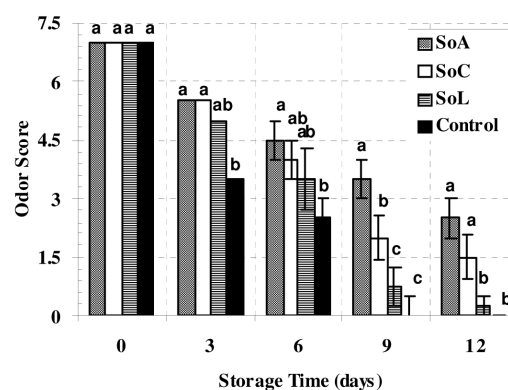


Figure 7. Comparison of odor scores given to Persian sturgeon fillets treated with SoA (sodium acetate), SoC (sodium citrate), SoL (sodium lactate), vs. control. Bars denote standard error of the mean (n= 3).



treatments up to day 6. In fact, the best scores were given to the sodium acetate treatment by panel group when compared with other treatments. In this study, the results of color evaluation, which showed better results for sodium acetate sample, confirmed those of Al-Sheddy (1999) who reported that dipping of camel meat in sodium acetate (10% w/w) produced a significant ($P < 0.05$) reduction in surface discoloration in comparison with control. Also, previous work by Manju *et al.* (2007) revealed that vacuum-packaging in conjunction with 2% sodium acetate can be safely employed to improve the sensory attributes and extend the shelf life of Pearl spot (*Etroplus suratensis*) samples up to 15 days at 2°C. Mendonca *et al.* (1989) indicated that using sodium acetate with acetic acid improved color quality of pork chops more than the use of acetic acid alone. However, among the different kinds of molecules produced as a result of lipid oxidation, secondary ones are considered the chief compounds responsible for oxidized flavor (Kurade and Baranowski, 1987). Accordingly, a close relationship was observed between the TBA assessment and the rancidity odor in the present study.

CONCLUSIONS

This study demonstrates that the sodium salts of organic acids (sodium citrate, sodium lactate and specially sodium acetate) can reduce the rate of lipid oxidation and improve the sensory attributes of fillets, so they can be employed as useful antioxidants in prolonging the shelf-life of refrigerated Sturgeon fillets.

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بررسی اثر محلول های نمکی سدیم (استات، لاکتات و سیترات سدیم) بر خواص
فیزیکی و شیمیایی وحسی فیله تاسماهی ایرانی (*Acipenser persicus*)
در شرایط نگهداری در یخچال

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چکیده

در این تحقیق، اثر محلول های نمکی سدیم بر خواص فیزیکی و شیمیایی وحسی فیله تاسماهی ایرانی (*Acipenser persicus*) طی ۱۲ روز نگهداری در یخچال بررسی گردید. بدین منظور، فیله ها در محلول های آبی (2.5 wt/vol) استات سدیم، لاکتات سدیم، سیترات سدیم و آب مقطر (به عنوان شاهد) به مدت ۱۰ دقیقه غوطه ور و سپس بسته بندی شدند. ارزیابی شاخص های TBA, FFA, pH، آهن هم و خواص حسی (طعم، رنگ و بو) در روزهای ۰، ۳، ۶، ۹ و ۱۲ نگهداری، صورت گرفت. نتایج حاکی از آن بود که میزان TBA تمام تیمارها با گذشت زمان افزایش یافت ($P < 0.05$). تیمار شاهد بالاترین میزان TBA را نشان داد در حالیکه نمونه حاوی استات سدیم پایین ترین میزان را در بین سایر تیمارها داشت ($P < 0.05$). در ارزیابی هیدرولیز چربی، تیمارهای حاوی نمک های سدیم، به خصوص استات سدیم، میزان FFA پایین تری در مقایسه با شاهد داشتند. بین میزان pH نمونه ها تفاوت معنی داری مشاهده نشد ($P < 0.05$). بررسی آهن هم نیز نشان داد که تیمارهای حاوی نمک های سدیم به ویژه استات سدیم، دارای مقادیر بالاتری از آهن هم نسبت به شاهد بودند. در ارزیابی وحسی فیله ها، بهترین امتیازات به تیمار استات سدیم داده شد. ترتیب اثر نمک های سدیم بدین صورت بود: $SA > SC > SL$. بنابراین، نمک های سدیم به ویژه استات سدیم را می توان به عنوان آنتی اکسیدان در برابر افت کیفی و همچنین افزایش مدت زمان ماندگاری فیله ماهی موثر دانست.