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Efficacy of electrolyzed oxidizing water as a pretreatment method for reducing *Listeria monocytogenes* contamination in cold-smoked Atlantic salmon (*Salmo salar*)

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ABSTRACT

Listeria monocytogenes contamination in ready-to-eat (RTE) fish products, in particular in cold-smoked salmon is an important food safety concern. This study evaluated the antimicrobial activity of electrolyzed oxidizing (EO) water as a pretreatment method during the process of cold-smoked salmon to inactivate *L. monocytogenes*. In addition, the effect of EO water treatment on the sensory and textural quality of the final product was also evaluated. Raw Atlantic salmon (*Salmo salar*) fillets were inoculated with *L. monocytogenes* (with an approximately cell number of 6×10^5 CFU/g *L. monocytogenes* ATCC 19114) and treated with EO water at three different temperatures (20, 30, and 40 °C) and at three different exposure time of 2, 6, and 10 min before the cold-smoking process. A combination of EO water and a mild temperature (40 °C) had reduced *L. monocytogenes* populations by 2.85 $\log_{10}$ CFU/g. The sensory as evaluated by a consumer panel (N = 71) and texture, which was measured by texture analysis showed no significant changes between EO and mild temperature treated samples and the control.

1. Introduction

Listeria monocytogenes, a psychotropic, Gram-positive and facultative anaerobe, is a food-borne pathogen capable of causing serious illness in vulnerable groups such as elderly, newborns, and individuals with weakened immune systems (Gombas, Chen, Clavero, & Scott, 2003; McCarthy & Burkhardt, 2012). The organism is well adapted to different environmental conditions including high salt content (10–20%), low temperature (less than 1 °C), low oxygen, and pH values below 6 (Farber & Peterkin, 1991) and it is a major concern to producers of RTE foods. Listeriosis has the third highest mortality rate among foodborne infections; approximately 2500 illnesses and 500 deaths are attributed to listeriosis in the United States annually (Levine, Rose, Green, Ransom, & Hill, 2001).

Despite all the efforts to eradicate *L. monocytogenes* from RTE foods, its contamination continues to occur. One class of the RTE products recognized as a potential risk for *L. monocytogenes* is cold-smoked salmon, because the temperature applied during the process (20–30 °C) is not sufficient to inactivate the organism, and if conditions are not properly controlled could encourage its growth. These foods are consumed without further cooking (Gudmundsdóttir et al., 2005; Rørvik, 2000).

The United States has set a zero tolerance level for the *L. monocytogenes* in RTE foods, including RTE seafood (Porsby, Vogel, Mohr, & Gram, 2008). Among all of the U.S. Food and Drug Administration regulated RTE foods recalled due to the *L. monocytogenes* risk from 1990 to 2006, seafood had the highest percentage of recalls (CSPI., 2008; Kang et al., 2012; Ozer & Demirci, 2006) and according to the Rapid Alert System for Food and Feed (RASFF) in EU, compared with other food product, seafood is second only to vegetables in the number of alerts activated between 2009 and 2012 (RASFF, 2013).

To date, several RTE products, such as smoked salmon, smoked mussel, smoked trout, cooked crawfish, and seafood salad, have been found to be contaminated with *L. monocytogenes* (McCarthy & Burkhardt, 2012). Therefore, reducing the occurrence of *L. monocytogenes* in RTE seafood is an important food safety goal.

So far application of different types of antibacterial compounds, such as nisin, hypochlorite, chlorine, chlorine dioxide, trisodium

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phosphate solution, acidified sodium chlorite, potassium lactate in combination with sodium acetate or sodium diacetate have been suggested to eliminate or minimize the occurrence of pathogens from raw and minimally processed seafood including cold-smoked salmon (Bremer & Osborne, 1998; Kim, Huang, Marshall, & Wei, 1999; Lakshmanan & Dalgaard, 2004; Lin, Huang, Cornell, Lin, & Wei, 1996; Mu, Huang, Gates, & WU, 1997; Nykänen, Lapveteläinen, Hietanen, & Kallio, 1999; Park, Rua, & Acker, 1991; Su & Morrissey, 2003; Vogel, Yin Ng, Hyldig, Mohr, & Gram, 2006; Yoon, Burnette, Abou-Zeid, & Whiting, 2004).

One of the disinfection methods which is becoming more common in seafood processing and sanitation is the use of electrolyzed oxidizing water (EO water), which its antimicrobial activity against a variety of microorganisms and food-borne pathogens has reported (Fabrizio & Cutter, 2003; Hricova, Stephan, & Zweifel, 2008; Huang, Hung, Hsu, Huang, & Hwang, 2008; McCarthy & Burkhardt, 2012; Ozer & Demirci, 2006; Rasco & Ovissipour, 2015).

EO water is a novel antimicrobial agent developed in Japan, and produced when dilute salt water is put through an electric current in a sealed chamber. Two different types of acidic and alkaline water, are produced from the process. The former is capable of killing harmful microorganisms, while the latter can be used to remove dirt and grease from items such as cutting boards and other kitchen utensils (Huang et al., 2006).

The disinfecting effect of EO water on fish pathogenic bacteria (Kasai, Ishikawa, Hori, Watanabe, & Yoshimizu, 2000; Ovissipour et al., 2015) and in the surface sanitization of seafood products (McCarthy & Burkhardt, 2012) have been reported. Several studies have been conducted using EO water to control the microbial contamination in different types of seafood products; soaking whole carp in EO water reduced the numbers of aerobic bacteria (Mahmoud et al., 2004); using ice prepared with EO water during the refrigerated storage, hindered the growth of aerobic and psychrotrophic bacteria (Kim et al., 2006); treatment of raw salmon and yellowfish tuna with EO water and/or ice, reduced the number of *Escherichia coli* O157:H7, *L. monocytogenes*, Enterobacter aerogenes and *Morganella morganii* (Ozer & Demirci, 2006; Phuvasate & Su, 2010). Soaking inoculated tilapia (with *E. coli* O157:H7 and *Vibrio parahaemolyticus*) into EO water resulted in 2.6 log CFU/cm^2 reduction of *V. parahaemolyticus* after 10 min, and an additional 0.7 log CFU/cm^2 reduction compared to tap water on *E. coli* O157:H7 after 1 min treatment (Huang et al., 2006). Raw salmon treated with EO water (pH of 2.6, ORP of 1150 mV and free chlorine of 90 mg/L) at 35 °C for 64 min resulted in a 1.07 log CFU/g (91.1%) and 1.12 log CFU/g (92.3%) reduction in *E. coli* O157:H7 and *L. monocytogenes*, respectively (Ozer & Demirci, 2006).

Compared to traditional disinfectant solutions, EO water has the potential to be more cost effective, less expensive, and environment friendly (Hricova et al., 2008; Ozer & Demirci, 2006; Rasco & Ovissipour, 2015). Moreover, using EO water on different food products did not negatively affect the organoleptic properties of color, scent, flavor, or texture in different food products (Hara, Watanuki, & Arai, 2003; Kim, Hung, Brackett, & Lin, 2003; Kobayashi, Tosa, Hara, & Horie, 1996; Mahmoud, 2007).

To date, there is no study of the effectiveness of EO water against pathogenic bacteria during the process of cold-smoked salmon production. Studying the efficacy of this disinfecting agent to eliminate or reduce the growth of *L. monocytogenes* in a high-risk seafood product, such as cold-smoked salmon, can provide useful information for seafood producers to prevent food-borne illnesses associated with *L. monocytogenes*.

The objective of this study was to evaluate the efficacy of the EO water treatment to inactive *L. monocytogenes* in cold-smoked Atlantic salmon (*Salmo salar*). Investigating the effect of EO water treatment on sensory and textural properties of cold-smoked salmon at different exposure times and temperature treatments was the second goal of this study.

2. Material and methods

Fresh Atlantic salmon (*S. salar*) fillets were purchased from a local grocery store in Pullman, WA. Fillets were stored in freezer at −20 °C for 24 h before conducting the experiments, followed by thawing at refrigerator for 6 h before the experiments. For the microbiology, fillets were cut into 50 g pieces (5 × 3.5 × 2 cm) prior to inoculation and EO water treatments.

2.1. Microbial analysis

2.1.1. Inoculum preparation, and inoculation of salmon fillets

L. monocytogenes ATCC 19114 was obtained from Microbiologics®, Inc. (St. Cloud, MN). *L. monocytogenes* was cultured on 50 mL of tryptic soy broth TSB (BactoTM) with 1% yeast extract for 18 h at 37 °C. After appropriate incubation of bacterial cultures, 10 mL broth of the *L. monocytogenes* was transferred under aseptic conditions to a sterile centrifuge tube, and then centrifuged for 15 min at 5000 rpm (3380×*g*) to harvest bacterial cells (AccuSpin™ model 400 bench top centrifuge, Fisher Thermo Scientific, Pittsburgh, PA). To eliminate any effect of broth components and bacterial metabolites, the resultant pellets were resuspended in 10 mL of sterile saline solution (0.85% (w/v) NaCl). After the second centrifugation, the supernatant was decanted, and the resulting washed pellets were then resuspended in sterile 10 mL aliquots as before. These were then used as pure cell suspensions to inoculate salmon fillets. The approximate initial cell number of (3 × 10^8 CFU/mL) *L. monocytogenes* was used for inoculation of salmon fillets. Samples were inoculated by spreading 0.1 mL of the cell suspension on the surface of the 50 g fillets. Inoculated fillets were kept in laminar flow hood at room temperature for 1.5 h before treated with EO water. The population of *L. monocytogenes* on the inoculated salmon fillets were about 6 × 10^5 CFU/g of fish fillets. Each experiment was conducted in triplicate.

2.1.2. Preparation of EO water

Electrolyzed water (pH: 2.7; ORP:1150 mV; free chlorine: 60 ppm) was generated at 9–12 V direct current (dc) for 15 min using a two-compartment batch scale electrolysis apparatus (Super Oxseed Labo, Electrolyzed Water Generator, Aoi Electronic Corp., Kannami, Shizuoka, Japan), with the anode and cathode sides of the chamber divided by an ion exchange diaphragm. The ORP and pH were measured with a pocket-sized redox meter (HI 98201, HANNA® Instruments, Ann Arbor, Michigan, USA) and a pH meter (FE20, Mettler-Toledo, Columbus, OH, USA), respectively. The free chlorine concentration of the EO water was measured with a DPD assay (Colorimeter™ Analysis System, Hach Co., Loveland, CO, USA) according to the manufacturer instructions. The acidic EO water was collected for the treatment of salmon fillets.

2.1.3. Treatment of salmon fillets with EO water

Inoculated samples were subjected to EO water with different temperatures (20, 30, and 40 °C) at different times (2, 6, and 10 min). Clean plastic beakers with EO water were placed into the water bath at given temperatures. All samples were immersed into the EO water at different temperatures with different exposure times. The sterile deionized water was used to study the influence of temperatures and times on *L. monocytogenes* reduction. However, since no reduction was observed, the data not shown here.

2.1.4. Preparation of cold-smoked salmon

Salmon fillets (50 g) including control and EO water treated samples, where processed with a dry-curing method (three parts brown sugar and 1 part crystalline additive salt by weight) for 18 h at 4 °C, followed by rinsing the fillets with sterile deionized water to remove the excess sugar and salt. Then hickory liquid smoke (B&G Foods, Inc., Parsippany, NJ) was spread on both sides of the fillets and they were held on the drying rack for 2 h under a laminar hood at room temperature to allow for pellicle formation. The water phase salt and water activity of the cold-smoked salmon samples were in the range of 3.7–4.2%, and 0.91–0.94 (at 20 °C), respectively.

2.1.5. Recovery of Listeria monocytogenes and culture media

To recover *L. monocytogenes*, at the end of each experiment, 50 g salmon fillets were removed from beakers and placed in sterile strainer-filter bags and homogenized with 100 mL of sterile 0.1% peptone water (Bacto, BD, Sparks, MD) using a Stomacher 400 (Seward Ltd., London, UK). One mL of the homogenized suspension was then serially diluted (dilution range, 10^0 to 10^{-5}) in 9 mL of sterile 0.1% peptone (Bacto, BD, Sparks, MD). A 1-mL aliquot of each dilution was divided into 5 aliquots of 0.2 mL and cultured using spread plate technique on Listeria Palcam agar (CM0877, Oxoid Ltd., Basingstoke, United Kingdom) (Al-Qadiri et al., 2015). Listeria agar plates were aerobically incubated at 37 °C for 24–48 h, and the numbers of viable cells were determined as CFU per gram of salmon fillet (N = 3).

2.2. Physical and chemical analyses

It should be noted that for physicochemical analysis of cold-smoked salmon, un-inoculated fish fillets were used. The same aforementioned methods were used for all steps, including EO water preparation, treatment of samples with EO water, and cold-smoked salmon preparation. Physicochemical properties of samples were compared with the control sample, which was untreated EO water cold-smoked salmon.

2.2.1. Color measurement

Color was measured with a tristimulus spectrophotometer (model CM-5, Konica Minolta Sensing Inc, Japan) recording: L^* (lightness), a^* (redness), and b^* (yellowness). Each sample was measured in triplicate.

2.2.2. Expressible liquid

Expressible liquid was measured as described by Das, Anjaneyulu, Gadekar, Singh, and Pragati (2008), with slight modification. Approximately 5.0 g of smoked salmon was weighed and wrapped with a filter paper (Whatman, grade 541), then placed into a centrifuge tube. The tubes were centrifuged for 10 min at (500 × *g*) at 10 °C (AccuSpin™ model 400 bench top centrifuge, Fisher Thermo Scientific, Pittsburgh, PA). The amount of expressible liquid (EL) was measured by the following equation:

$$\mathrm{El} = \frac{W_a - W_b}{W_a} \times 100$$

where W_a is the initial weight and W_b is the centrifuged weight of the sample. Triplicate measurements were performed on each sample.

2.2.3. Texture

The TA.XT2 texture analyzer with a load cell of 25 kg was used (Stable Micro System Ltd., Surrey, UK). In Warner–Bratzler shearing blade with a thickness of 1 mm, length of 9 cm and width of 7 cm was used with a test speed of 2.5 mm/s. The results were reported as the maximum peak force (shear force in N) required to shear through the sample (Gudbjörnsdóttir, Jonsson, Hafsteinsson, & Heinz, 2010).

Three samples were tested from each treatment (N = 3) with measurements taken at three different locations on each sample (7 cm × 2 cm × 2 cm, 32 g).

2.2.4. Water phase salt and water activity

Water phase salt was calculated from the values of salt content and moisture content (Hilderbrand, 1992). Salt content was determined by silver nitrate titration for the chloride ion (Shin & Rasco, 2007) based upon AOAC method 937.09.

Water activity of the samples was measured using an AquaLab water activity meter (Model 3 TE, Decagon Devices, Pullman, WA) at 20 °C.

2.2.5. Sensory evaluation

Consumer panelists were recruited from the Washington State University community to serve on a sensory panel. Panelists (N = 71) were from a diverse ethnic background and ranged in age from 18 to 71. Approximately 45% of the test subjects were males, and 55% of the test subjects females. All participants signed an Informed Consent Form and the project was approved by the Washington State University Institutional Review Board. Sensory testing was conducted during a single day in the WSU School of Food Science sensory laboratory using a directional paired preference test. Three pairs of cold-smoked salmon (samples pretreated with EO water at three different temperatures (20, 30, and 40 °C; for 10 min) vs control were presented to the panelists and pairs were presented in separate flights. Panelists were presented with the 20 °C vs control, 30 °C vs control and 40 °C vs control. For each pair presented, consumers were asked which sample was preferred based upon appearance, aroma, texture, taste/flavor and overall preference. Samples were presented in random order under standard incandescent lighting.

Panelists were provided with filtered water, unsalted saltine crackers and grapes for cleansing the palate. A cuspidor was provided for expectoration of samples. The results were analyzed by Compusense-at-Hand software (Compusense Inc., Guelph, ON). Statistical significance was determined according to (Roessler, Pangborn, Sidel, & Stone, 1978) at a 5% probability level.

2.2.6. Statistical analysis

One-way analysis of variance using SPSS statistical software, (IBM SPSS Statistics 21) was carried out to compare the effect of different treatment on microbial levels, color, and texture between treatments. Comparison of means were performed using Tukey's HSD multiple comparison test at the 5% probability level.

3. Results and discussion

3.1. Effect of EO water treatment on inactivation of L. monocytogenes as a function of time and temperature in cold-smoked salmon

Acidic EO water treatment reduced *L. monocytogenes* on cold-smoked salmon (Table 1). Inoculated salmon fillets were treated with acidic EO water at three different temperature of 20, 30, and 40 °C with different exposure time of 2, 6, and 10 min prior to the brining and treatment with liquid smoke. Treating inoculated samples (5.77 $\log_{10}$ CFU/g) with EO water at 40 °C for 6 and 10 min reduced *L. monocytogenes* levels by 2.4 $\log_{10}$ CFU/g (40%) and 2.9 $\log_{10}$ CFU/g (50%), respectively. Significant microbial reduction was observed when the temperature of EO water and the treatment

Table 1
Mean counts of *Listeria* (log_{10} CFU/g) treated with EO water in cold-smoked salmon fillet.[a,b,c,d]

Temperature	20 °C			30 °C			40 °C		
Time (min)	2	6	10	2	6	10	2	6	10
Bacteria log (CFU/g)	4.36 ± 0.09a*Aa* (1.41)	4.22 ± 0.03a*Aa* (1.55)	4.19 ± 0.15a*Aa* (1.58)	4.25 ± 0.15a*Aab* (1.52)	3.95 ± 0.06ab*Aa* (1.82)	3.51 ± 0.29b*Aab* (2.26)	3.83 ± 0.02ab*Ab* (1.94)	3.41 ± 0.09bc*Bb* (2.36)	2.92 ± 0.07c*Cb* (2.85)

[a] Reduction in counts (expressed in CFU/g) shown in parentheses for each treatment, Initial count (control) = 5.77 log_{10} CFU/g.
[b] Mean values with different letters differed significantly ($P < 0.05$).
[c] Upper-case letters are showing effect of time in a constant temperature, differed significantly ($P < 0.05$).
[d] Italic letters are showing the effect of temperature in a constant time, differed significantly ($P < 0.05$).

time increased to 30 °C and 10 min ($P < 0.05$); treating fillets at 20 °C did not reduce bacterial levels ($P > 0.05$). Increasing the exposure time at 20 and 30 °C, did not show any significant increase on the reduction of *L. monocytogenes* ($P > 0.05$). Increasing the exposure time for the EO treatment at 40 °C, reduced the number of the *L. monocytogenes* from 5.77 log_{10} CFU/g to 3.83 log_{10} CFU/g (at 2 min) and to 2.92 log_{10} CFU/g (at 10 min), a 24% reduction. Depending upon the food product and treatment conditions, increasing exposure time for EO water treatment may increase treatment efficacy. At the exposure times of 2, 6 and 10 min, increasing the temperature of EO water from 20 °C to 30 and 40 °C significantly increased the bacterial reduction ($P < 0.05$).

Huang et al. (2006) used EO water to inactivate *E. coli* O157:H7 and *V. parahaemolyticus* on tilapia, and reported significant microbial reduction as treatment time increased from 1 to 10 min. In another study, Issa-Zacharia, Kamitani, Morita, and Iwasaki (2009) showed that increasing exposure time (from 1 to 5 min) did not significantly reduce *E. coli* in RTD Japanese mustard green. Furthermore, Fabrizio and Cutter (2004) studied the effect of EO water to reduce pathogens on fresh pork, and suggested that an increased contact time (>10 min) was needed to adequately reduce the microbial contaminants. Ovissipour et al. (2015) observed higher bacterial reduction by increasing EO exposure time in a study with bacterial cell suspensions finding that only 2 min was required for complete inactivation of *L. monocytogenes* in cell suspension, while for *E. coli* O104:H4, 4 min was required to achieve complete inactivation. Ovissipour et al. (2015) noted that the presence of organic matter can significantly decreased the efficacy of EO water, which can be compensated for by increasing the exposure time, temperature and the available chlorine concentration.

Raising the temperature to 40 °C can increase the efficacy of the EO water treatments as observed here at 40 °C for *L. monocytogenes* in salmon fillets. Others have found that mild temperature significantly enhanced the effectiveness of the acidic electrolyzed water against *V. parahaemolyticus* on shrimp (Xie, Sun, Pan, & Zhao, 2012) over a range of 4–20 °C showed no significant changes in the reduction of *V. parahaemolyticus*, but when the treatment temperature reached 50 °C 1.5 to 2 log reductions were observed. Ozer and Demirci (2006) reported a reduction of *L. monocytogenes* Scott A population in the range of 0.40 log_{10} CFU/g (60%) at 22 °C to 1.12 log_{10} CFU/g (92.3%) at 35 °C. Venkitanarayanan, Ezeike, Hung, and Doyle (1999) reported no differences in the inactivation rates of *E. coli* O157:H7, *Salmonella enteritidis*, and *L. monocytogenes* which were treated by EO water at 4 and 23 °C. However at 35 and 45 °C, much higher rates of reductions were observed for all three pathogens. Increasing the temperature may change the bacterial cell wall physico-chemical properties which can accelerate the EO water entrance into the cell (Fabrizio & Cutter, 2003).

3.2. Texture analysis

Forces required to shear cold-smoked salmon treated with EO water were compared to untreated controls (Fig. 1). However, the samples treated with EO water at 40 °C for 10 min, showed slightly softer texture, no significant differences were observed in the firmness of the samples between the control and any EO water treated smoked salmon ($P > 0.05$). It has been shown by other researchers that if the cold-smoking processing conditions including brining and curing method and smoking temperature can be carefully controlled, will result in safer products with similar firmness (Gudbjörnsdóttir et al., 2010; Sigurgisladottir, Sigurdardottir, Torrissen, Vallet, & Hafsteinsson, 2000).

Since, salmon fillets were cured after EO water treatment, the brining procedure may counteract the effect of higher temperature EO water treatments by increasing flesh firmness. Salt penetration

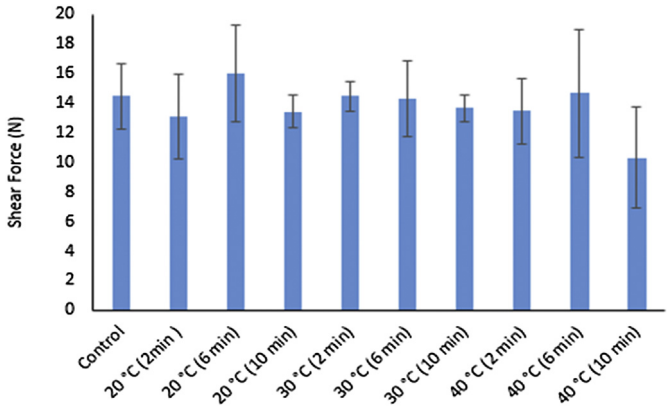

Fig. 1. Effect of EO water treatment on firmness of the cold-smoked salmon samples (N = 3).

into the muscle tissue, causes myosin to swell. At a high amount of salt, protein denatures, reduces protein gelation and increases water loss and results in firm texture (Duerr & Dyer, 1952).

3.3. Color measurements

Lightness (L^*) and redness (a^*) of the smoked salmon are shown in Figs. 2 and 3, respectively. No significant differences were observed in the lightness and redness ($P > 0.05$), indicating that EO treatment at different temperature and exposure time had no effect on the color of the final product.

One of the main quality aspects of fish products in terms of consumer acceptance is color. Previous research has been shown that high pressure processing (HPP) as a technique to inactivate *L. monocytogenes* in cold-smoked salmon, can negatively impact color and texture limiting the application o of high pressure processing as a pasteurization method for salmonids (Amanatidou et al., 2000; Basaran-Akgul et al. 2010; Gudbjörnsdóttir et al., 2010). A significant advantage of EO water treatment is that the color of the final product did not change as shown by both instrumental and sensory testing (Table 2) even at 40 °C showing the promise for this technique for cold smoked and potentially other ready to eat seafood and meat products.

3.4. Expressible liquid

Expressible liquid is an important indication of the water holding capacity and important quality factor from both

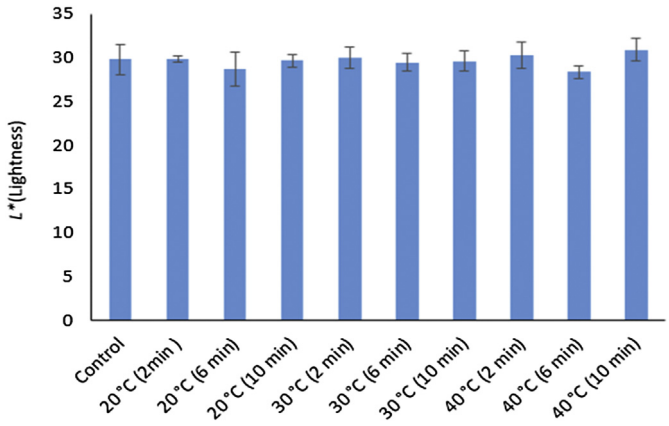

Fig. 2. The lightness (L^*) of salmon fillets treated with EO water at three different temperatures and exposure time (N = 3).

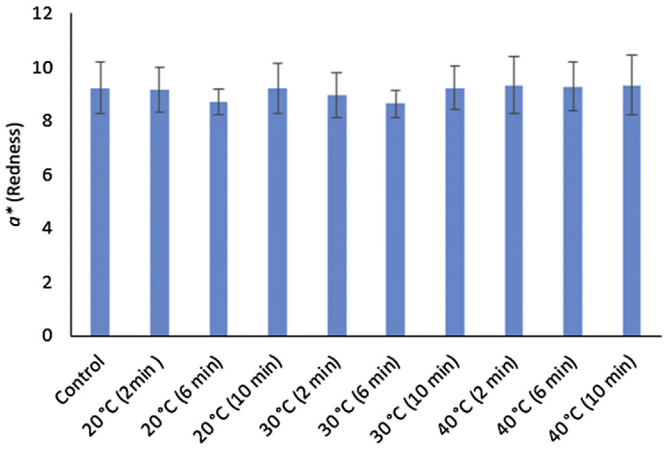

Fig. 3. The redness (a^*) of salmon fillets treated with EO water at three different temperatures and exposure time (N = 3).

commercial and consumer acceptance. No correlation between treatment temperature and/or exposure time of EO and the amount of expressible water was found in different treated samples (Fig. 4), indicating that EO treatment did not negatively influenced the water holding capacity of cold-smoked salmon. The expressible liquid in current study was ranged from 5.5 to 8% which is in agreement with other researchers findings about cold-smoked salmon. It has been mentioned that the expressible liquid in cold-smoked salmon is due to the muscle integrity structure because of the different salting methods (Cardinal et al., 2004; Schubring, 2006).

Since, in current study the same method of brining was used for all samples, no difference was observed among the treatments.

3.5. Sensory evaluation

Results of the paired comparison preference test are shown in Table 2. No significant differences for preference were found between the control and experimental treatments for any of the attributes, including appearance, aroma, texture and taste/flavor. No significant difference in overall liking was found.

EO water as a pretreatment method for cold-smoked salmon did not negatively influence the consumer acceptance or preference, indicating the promise this method has for microbial control for RTE seafood. The influence of electrolyzed water on the sensory properties of RTE foods has not been studied. The influence of EO water on some of the sensory attributes such as color, odor, firmness, and texture on raw chicken (Rahman, Park, Song, Al-Harbi, & Oh, 2012), pork (Rahman, Wang, & Oh, 2013) and shrimp (Lin et al., 2013) was studied. Rahman et al. (2012), reported that the fresh chicken breast which was treated by EO water, showed better sensory score after 7 days storage at 4 °C compared to the sample which was not treated by EO water.

Table 2
Panelist scores indicating the preferred sample in a pair comparing: 20 °C, 30 °C, or 40 °C to Control. Data represents 71 responses.

Attributes	Pairs compared*					
	20 °C	Control	30 °C	Control	40 °C	Control
Appearance	15	26	26	23	19	20
Aroma	17	24	20	29	23	18
Texture	22	19	30	19	22	19
Taste/Flavor	23	18	29	20	24	17
Overall	21	20	29	20	23	18

*P = ≤0.05.

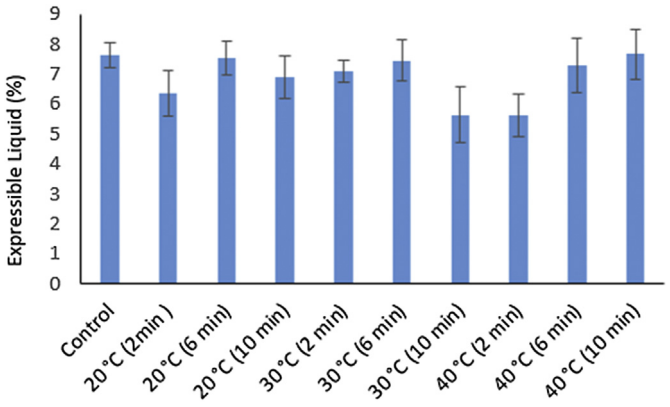

Fig. 4. Expressible liquid of different samples treated with EO water at three different temperatures and exposure time (N = 3).

4. Conclusion

Acidic EO water treatment of raw salmon fillets (*S. salar*) prior to curing of cold-smoked salmon at 40 °C for 10 min can reduce the level of *L. monocytogenes* by approximately 3 log without causing any significant changes in their sensory and textural properties. EO treatments at lower temperatures (20 or 30 °C) or for shorter times (2–6 min) resulted in a 1.5 to 2.0 log reduction.

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