

Textural and physicochemical changes in salmon (*Salmo salar*) treated with commercial liquid smoke flavourings

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Abstract

The textural and physicochemical properties of fillets of farmed Atlantic salmon (*Salmo salar*) treated with two commercial liquid smoke flavourings (LS1 and LS2) were examined after 15, 30 and 45 days of storage at 2 °C in oxygen impermeable bags. Salmon flesh treated with LS1 was characterized by high water-soluble protein, fat and moisture contents, plus low hardness, fracturability, gumminess and chewiness, and a low alkali-insoluble protein content. These characteristics were similar to those of control salmon (not treated with liquid smoke flavouring). Storage time changed these properties similarly in both. Salmon flesh treated with LS2 was characterized by high hardness, fracturability, gumminess, chewiness and alkali-insoluble protein levels, plus low water-soluble protein, fat and moisture contents. Storage time appeared to have a far less important effect on salmon flesh treated with LS2.

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1. Introduction

Salmon (*Salmo salar*) is an important food product, both from a nutritional and economic point of view. The raising of Atlantic salmon in aquaculture has contributed greatly to its market presence (Cardinal et al., 2001). About 15% of cultured and wild-caught salmon is converted into cured and smoked products (Huang, Cavinato, Mayes, Bledsoe, & Rasco, 2002).

Smoking is a centuries-old food preservation technique. However, the conventional smoking process is now being substituted by the use of smoke flavourings (Guillén & Ibargoitia, 1998). Smoke flavourings have been used for some 30 years as preservatives and aromatisers of meat and fish (Morais et al., 1996; Schindler, 1997). These have several advantages over traditional smoking procedures. For example, such treatment is much cheaper and less

taxing on the environment (Pszczola, 1995). Many smoke flavourings are also free of harmful compounds such as polycyclic aromatic hydrocarbons (PAHs) (Gomaa, Gary, Rabie, Lopez-Bote, & Booren, 1993). Finally, smoke flavourings of different composition are available and can be combined to obtain products with very different organoleptic qualities (Guillén, Manzanos, & Ibargoitia, 1996a). This is confirmed by the number of patents on smoke flavourings that have appeared in recent years (Guillén, Manzanos, & Ibargoitia, 1996b).

Several authors have studied the antimicrobial activity (MacRae, Hudson, & Towers, 1989), antioxidant effects (Barclay, Xi, & Norris, 1997; Guillén & Ibargoitia, 1998) and influence on organoleptic properties (Guillén, Manzanos, Ibargoitia, Cabo, & Sopelana, 1998; Kim, Kurata, & Fujimaki, 1974) of smoke flavourings. However, limited information is available on their effects on the physicochemical and textural characteristics of fish products (Goncalvez & Prentice-Hernandez, 1998). The aim of the present study was to investigate the effects of two commercial liquid smoke flavourings on these

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variables in fillets of Atlantic salmon. The final goal was to determine whether these flavourings might be able to replace traditional salmon smoking methods.

2. Materials and methods

2.1. Liquid smoke flavourings

Two commercial liquid smoke flavourings (LS1 and LS2) used in the Spanish food industry were used to smoke fillets of Atlantic salmon (*Salmo salar*). LS2 contained all the components of traditional smoke in similar proportions, while LS1 was composed mainly of phenol derivatives.

2.2. Samples and treatments

Forty farmed Atlantic salmon each weighing 2.7 ± 0.3 kg were obtained from a commercial Spanish fish processing plant. After slaughter (gill cut and bleeding), the fish were eviscerated, cleaned and placed on ice for transport to the laboratory. Within 2 days of slaughter, the fish were filleted and trimmed by hand to remove skin, bones, fins and visible adipose tissue.

These fillets were then dry-salted by hand with coarse grain salt to dry the flesh, leaving them for 16 h at 4 °C. The salt was then cleaned-off. Twenty right fillets were treated by immersion in LS1 for 30 s (SLS1). The other 20 right fillets were treated with LS2 (SLS2). All left fillets were used as control samples (CS); these underwent no smoking treatment. Each fillet was then vacuum-packed in an oxygen impermeable bag and stored at 2 °C until analysis at 15, 30 or 45 days.

2.3. Analyses

Fig. 1 shows the parts of the salmon fillets used for analysis. For the determination of pH, 10 g of flesh were blended with 10 ml of distilled water. The pH of the homogenized sample was then measured using a Crison Basic 20 pH meter (Crison Instruments, Barcelona, Spain).

The moisture content (%) was determined by drying 2 g samples of salmon flesh at 105 °C to a constant weight (ISO, 1999).

The total fat content (%) of 2.5 g of fish was extracted with petroleum ether in a Soxtherm S-306.D extraction unit (Gerhardt, Bonn).

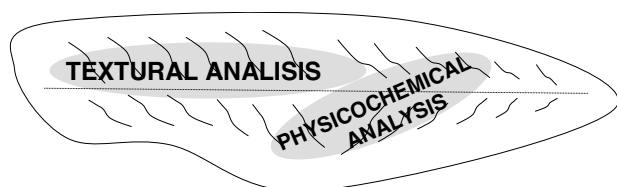


Fig. 1. Illustration showing which parts of the salmon fillets were used for analyses.

Protein was fractionated by the method of Hashimoto, Watanabe, Kono, and Shiro (1979), as modified by Vissanguan, Benjakul, Riebroy, and Thepkasikul (2004). The nitrogen content (%) of all fractions (non-protein nitrogen, NPN; water-soluble fraction, WSP; salt-soluble fraction, SSP; alkali-soluble fraction, ASP; alkali-insoluble fraction, AIP) was determined by the Kjeldahl method (ISO, 1997).

Fillet texture was examined using a TA XT2 Texture Analyser (Stable Micro Systems, Godalming, England) equipped with a load cell of 25 kg and a 20 mm diameter cylindrical probe. The test conditions involved two consecutive cycles of 30% compression with 5 s between cycles. The cross-head moved at a constant speed of 2 mm/s. Texture variables (hardness (H) (N/cm²), fracturability (F) (N/cm²), cohesiveness (C), springiness (S) (cm), gumminess (G) (N/cm²) and chewiness (CH) (N/cm)) were calculated as described by Bourne (1978).

2.4. Statistical analysis

Four determinations were made on each fillet for each physicochemical characteristic, and another ten determinations for each texture variable. Data were subjected to ANOVA. Means were compared by the least squares difference (LSD) method. Significance was set at 5%.

Pearson correlation analysis was performed to determine the relationships between variables. Principal component analysis (PCA) was used to reduce the number of variables. All calculations were performed using Statgraphics STSC Inc. software (version 5.0).

3. Results and discussion

Table 1 shows the mean values for the texture variables of the CS, SLS1 and SLS2 fillets after 15, 30 and 45 days of storage. The results for the SLS1 fish were quite similar to those of the CS fish, except in terms of cohesiveness and springiness. However, SLS2 fish showed significant differences to the controls with respect to all texture variables. Further, the hardness, fracturability, cohesiveness, gumminess and chewiness values were higher than in either the CS or SLS1 groups of fish at all times. Gómez-Guillen, Montero, Hurtado, and Bordenas (2000) indicate cold-smoking to produce a considerable increase in shear strength, while Sigurgisladottir, Sigurdardottir, Torrissen, Vallet, and Hafsteinsson (2000) report that the force required to shear smoked fillet samples is significantly greater than that required to shear unprocessed fillets. Similar results have been reported by Birkeland, Rora, Skara, and Bjerkgeng (2004) who showed the mean firmness (force) of smoked salmon fillets to be approximately 2–3 fold greater than that of the raw material.

With respect to changes in texture variables during storage, the CS fillets showed a reduction in the values of all variables, except for hardness and fracturability which

Table 1
Means and standard deviations for texture variables in the different salmon groups

Variable	Storage time (days)	Salmon groups		
		CS	SLS1	SLS2
Hardness (N/cm ²)	15	5.26 ± 0.26 ^{cb}	6.38 ± 0.43 ^{bx}	19.42 ± 1.52 ^{ax}
	30	5.88 ± 0.41 ^{bx}	5.56 ± 0.17 ^{bβ}	18.06 ± 1.92 ^{ax}
	45	5.71 ± 0.72 ^{bx}	4.49 ± 0.47 ^{bβ}	15.83 ± 0.85 ^{aβ}
Fracturability (N/cm ²)	15	2.62 ± 0.16 ^{cx}	3.19 ± 0.21 ^{bx}	9.71 ± 0.76 ^{ax}
	30	2.75 ± 0.31 ^{bx}	2.90 ± 0.45 ^{bx}	9.05 ± 1.01 ^{ax}
	45	2.73 ± 0.35 ^{bx}	2.49 ± 0.23 ^{bβ}	7.91 ± 0.42 ^{aβ}
Cohesiveness (ratio)	15	0.50 ± 0.07 ^{ax}	0.39 ± 0.06 ^{bx}	0.43 ± 0.03 ^{aβ}
	30	0.37 ± 0.04 ^{bβ}	0.37 ± 0.01 ^{bx}	0.43 ± 0.01 ^{aβ}
	45	0.30 ± 0.01 ^{cγ}	0.35 ± 0.02 ^{bx}	0.47 ± 0.03 ^{ax}
Springiness (cm)	15	0.87 ± 0.07 ^{ax}	0.92 ± 0.08 ^{ax}	0.67 ± 0.05 ^{bx}
	30	0.64 ± 0.04 ^{aβ}	0.55 ± 0.01 ^{bβ}	0.58 ± 0.02 ^{bβ}
	45	0.44 ± 0.01 ^{cγ}	0.52 ± 0.03 ^{bβ}	0.56 ± 0.02 ^{aβ}
Gumminess (N/cm ²)	15	2.60 ± 0.43 ^{bx}	2.78 ± 0.49 ^{bx}	8.87 ± 1.49 ^{ax}
	30	1.98 ± 0.45 ^{bβ}	1.88 ± 0.27 ^{bβ}	8.75 ± 1.58 ^{ax}
	45	1.73 ± 0.16 ^{bβ}	1.67 ± 0.30 ^{bβ}	6.94 ± 0.35 ^{aβ}
Chewiness (N/cm)	15	2.47 ± 0.43 ^{bx}	2.60 ± 0.35 ^{bx}	5.70 ± 0.60 ^{ax}
	30	1.15 ± 0.33 ^{bβ}	1.05 ± 0.17 ^{bβ}	4.07 ± 0.31 ^{aβ}
	45	0.78 ± 0.09 ^{bγ}	0.95 ± 0.20 ^{bβ}	4.47 ± 1.71 ^{ax}

CS, control salmon; SLS1, salmon treated with smoke flavouring LS1; SLS2, salmon treated with smoke flavouring LS2.

Different letters (a, b, c) in the same row indicate significant differences between means ($P < 0.05$).

Different letters (α, β, γ) in the same column for each variable indicate significant differences between means ($P < 0.05$).

showed a small increase by day 30 of storage. Cohesiveness was the only variable that did not change over time in the SLS1 fish; the values for all other variables decreased.

Finally, all texture values for the SLS2 fish decreased over storage time, except for cohesiveness which increased. It should be noted that the SLS1 and SLS2 fish showed the lowest hardness and fracturability values at 45 days of storage. Hattula, Elfving, Mroueh, and Luoma (2001) reported spoilage to be due to the activity of autolytic enzymes in fish tissue; these appear to have a major effect on texture deterioration. Hultmann, Rora, Steinsland, Skara, and Rustad (2004) reported smoking to increase the activity of endogenous proteases; these enzymes (e.g. collagenase) hydrolyse different proteins in muscle and break down the connective tissue. They therefore cause undesirable textural changes. Laksmanan, Piggott, and Paterson (2003) reported reduction in hardness to be a consequence of weakened connective tissue; these authors reported the Z lines in salt-cured salmon to be completely extinguished by day 21 of storage.

Table 2 shows the mean values for the physicochemical variables of the CS, SLS1 and SLS2 fish after 15, 30 and 45 days of storage. Fish treated with either liquid smoke flavouring showed higher NPN values and lower SSP, ASP, moisture and pH values than the control. Hassan (1988); Montero, Gómez-Guillen, and Borderias (2003) showed that the NPN increased with cold-smoking and suggest this may be due to protein breakdown by proteolytic enzymes. In addition, Lund and Nielsen (2001) reported that the smoking process enhances the formation of large peptide fragments.

Hultmann et al. (2004) and Gómez-Guillen et al. (2000) showed that the amount of salt-soluble proteins was reduced as a result of smoking due to the denaturation/aggregation of myofibrillar proteins. Further, these investigators, and Montero et al. (2003), showed the water content of salmon fillets to be reduced after cold-smoking. Finally, Hassan (1988) reported that smoking causes a reduction in flesh pH, perhaps due to smoke acid absorption, a loss of moisture and the reaction of phenols, polyphenols and carbonyl compounds with protein SH and amino groups, respectively.

No differences were seen between CS and SLS1 fish at any time with respect to WSP, ASP and fat values. However SLS2 fish showed lower WSP and fat values and higher AIP than the other two groups. Moreover, the AIP levels of SLS2 fish were approximately 6-fold higher, and the WSP levels were 9-fold lower than those recorded for CS and SLS1 fish at all times. Gómez-Guillen et al. (2003) showed that smoking leads to the 'insolubilisation' of the connective tissue, while Hultmann et al. (2004) reported that the extractability of water-soluble proteins was significantly reduced in smoked samples.

With respect to changes in the values of physicochemical variables during storage, the NPN, SSP, ASP, fat and pH values of SLS1 and SLS2 fish varied in line with the changes observed in CS fish. However, the AIP and moisture contents of CS fish remained stable, as in SLS1, both increased; however in SLS2 fish while AIP decreased and moisture increased. Hultmann et al. (2004) reported that after continued storage, the water content of smoked

Table 2

Means and standard deviations of physicochemical variables in the different salmon groups

Variable	Storage time (days)	Salmon group		
		CS	SLS1	SLS2
NPN (%)	15	1.63 ± 0.05 ^{bx}	2.00 ± 0.04 ^{ax}	1.99 ± 0.05 ^{ax}
	30	1.54 ± 0.04 ^{bβ}	1.81 ± 0.07 ^{aβ}	1.89 ± 0.02 ^{aβ}
	45	1.36 ± 0.03 ^{by}	1.73 ± 0.04 ^{aβ}	1.84 ± 0.01 ^{aβ}
WSP (%)	15	9.28 ± 0.05 ^{bβ}	9.74 ± 0.24 ^{ax}	0.93 ± 0.19 ^{cβ}
	30	9.16 ± 0.10 ^{aβ}	9.28 ± 0.14 ^{aβ}	1.53 ± 0.22 ^{bx}
	45	9.47 ± 0.05 ^{ax}	9.16 ± 0.08 ^{bβ}	1.34 ± 0.15 ^{cx}
SSP (%)	15	7.16 ± 0.07 ^{ax}	6.09 ± 0.09 ^{cx}	6.44 ± 0.05 ^{bx}
	30	6.92 ± 0.18 ^{aβ}	5.89 ± 0.10 ^{cβ}	6.34 ± 0.06 ^{bβ}
	45	6.44 ± 0.08 ^{aγ}	5.67 ± 0.05 ^{cγ}	6.15 ± 0.04 ^{bγ}
ASP (%)	15	2.82 ± 0.09 ^{ax}	1.91 ± 0.05 ^{bx}	1.67 ± 0.13 ^{cx}
	30	1.90 ± 0.17 ^{aβ}	2.10 ± 0.26 ^{ax}	1.04 ± 0.30 ^{bβ}
	45	1.12 ± 0.07 ^{aγ}	0.52 ± 0.1 ^{bβ}	13.33 ± 0.08 ^{bγ}
AIP (%)	15	2.30 ± 0.14 ^{bx}	2.57 ± 0.29 ^{bβ}	13.90 ± 0.08 ^{ax}
	30	2.28 ± 0.07 ^{bx}	2.49 ± 0.26 ^{bβ}	13.91 ± 0.10 ^{ax}
	45	2.34 ± 0.08 ^{cx}	2.97 ± 0.12 ^{bx}	13.33 ± 0.08 ^{aβ}
Fat (%)	15	15.33 ± 0.27 ^{by}	15.82 ± 0.13 ^{aγ}	15.32 ± 0.20 ^{bβ}
	30	16.07 ± 0.12 ^{bβ}	16.15 ± 0.06 ^{aβ}	15.12 ± 0.08 ^{bβ}
	45	16.63 ± 0.10 ^{ax}	16.58 ± 0.08 ^{ax}	15.50 ± 0.10 ^{bx}
Moisture (%)	15	64.43 ± 0.33 ^{ax}	61.98 ± 0.41 ^{bβ}	61.73 ± 0.37 ^{bβ}
	30	64.31 ± 0.33 ^{ax}	62.63 ± 0.21 ^{bx}	61.27 ± 0.22 ^{cβ}
	45	64.70 ± 0.45 ^{ax}	63.15 ± 0.34 ^{bx}	62.38 ± 0.24 ^{cx}
pH	15	6.14 ± 0.00 ^{ax}	6.12 ± 0.00 ^{bx}	5.96 ± 0.00 ^{cx}
	30	5.93 ± 0.00 ^{aβ}	5.87 ± 0.00 ^{bγ}	5.76 ± 0.00 ^{cβ}
	45	5.93 ± 0.00 ^{aβ}	6.09 ± 0.02 ^{aβ}	5.64 ± 0.03 ^{cγ}

CS, control salmon; SLS1, salmon treated with smoke flavouring LS1; SLS2, salmon treated with smoke flavouring LS2; NPN, non-protein nitrogen; WSP, water-soluble protein; SSP, salt-soluble protein; ASP, alkali-soluble protein; AIP, alkali-insoluble protein.

Different letters (a,b,c) in the same row indicate significant differences between means ($P < 0.05$).

Different letters (α, β, γ) in the same column for each variable indicate significant differences between means ($P < 0.05$).

salmon increased slightly, but was still lower than in fresh salmon.

Rora and Einen (2003) showed that storage leads to progressive changes in the solubility and denaturation of muscle collagen in Atlantic salmon. The reduction in insoluble collagen that occurred was probably a result of environmental changes in the salmon muscle, where enzymes such as collagenases, neutral proteinases and acid proteinases had cleared parts of the collagen triple helix (Eckhoff, Aidos, Hemre, & Oyvind, 1998).

PCA was used to identify similarities and differences among samples on the basis of the textural and physicochemical data obtained. Three principal components (PC1, PC2 and PC3) were obtained which accounted for 90.01% of the total variation. PC1 explained 57.80% of the variance and was mainly related to hardness, fracturability, gumminess, chewiness, WSA, AIP and fat content. PC2, which accounted for 21.34% of the variance, was mainly related to cohesiveness, springiness, ASP and fat

content. PC3 represented 10.86% of the total variance and was mainly related to NPN, SSP, moisture content and pH.

Fig. 2 shows the distribution of the samples and the respective changes experienced over storage time as a function of the three principal components mentioned above. The control salmon groups (CS-15, CS-30 and CS-45) fell into two subgroups. CS-15 fish showed positive values with respect to PC2, and were therefore characterised by high cohesiveness, springiness, ASP and pH values. CS-30 and CS-45 fish showed negative values with respect to PC1, and were therefore characterized by high WSP, high fat and moisture contents, and low hardness, fracturability, gumminess, chewiness and AIP values.

The fish treated with LS1 (SLS1-15, SLS1-30 and SLS1-45) showed a distribution similar to the CS groups. SLS1-15 showed negative values with respect to PC3, with high NPN and pH values and low SSP and moisture content values. SLS1-30 and SLS1-45 showed negative values with respect to PC1, and were characterized by having similar characteristics to CS-30 and CS-45 fish. The effect of storage time on both the CS and SLS1 fish was evident, their dominant PCA axes changing over time.

Finally, the fish treated with LS2 (SLS2-15, SLS2-30 and SLS2-45) differed from all other samples in that they showed positive values with respect to PC1. They were therefore characterized by high hardness, fracturability, gumminess, chewiness and AIP, but low WSP, and moisture and fat contents. Storage time does not, therefore, appear to have a dramatic effect on Atlantic salmon treated in this way.

These results agree with those obtained by other authors. Gómez-Guillen et al. (2003) showed that smoking leads to the insolubility of connective tissue and caused water loss, resulting in the hardening of the flesh. Firmness and break strength are therefore inversely

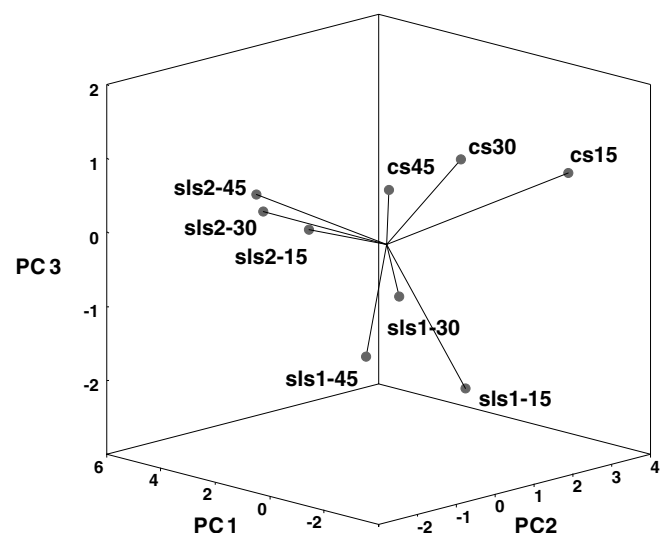


Fig. 2. PCA plot showing the distribution of salmon groups over storage time, based on the physicochemical and textural data.

Table 3
Correlation matrix between textural and physicochemical variables

	H	F	C	S	G	CH
NPN	0.57 ^c	0.59 ^c	0.56 ^c	0.33 ^a	0.60 ^c	0.67 ^c
WSP	0.97 ^c	0.97 ^c	0.51 ^b	0.12	0.95 ^c	0.90 ^c
SSP	0.07	0.01	0.18	0.31 ^a	0.03	0.07
ASP	0.31 ^a	0.31 ^a	0.08	0.71 ^c	0.27	0.17
AIP	0.98 ^c	0.98 ^c	0.51 ^b	0.12	0.96 ^c	0.90 ^c
Fat	0.40 ^c	0.41 ^c	0.74 ^c	0.52 ^c	0.74 ^c	0.73 ^c
Moisture	0.71 ^c	0.71 ^c	0.39 ^a	0.04	0.72 ^c	0.65 ^c
pH	0.58 ^c	0.58 ^c	0.13	0.54 ^c	0.56 ^c	0.45 ^b

H, hardness; F, fracturability; C, cohesiveness; S, springiness; G, gumminess; CH, chewiness; NPN, non-protein nitrogen; WSP, water-soluble protein; SSP, salt-soluble protein; ASP, alkali-soluble protein; AIP, alkali-insoluble protein.

^a $P < 0.05$

^b $P < 0.01$.

^c $P < 0.001$.

related to the water content of smoked salmon fillets (Indrasena, Hansen, & Gill, 2000; Jittinandana, Kenney, Slider, & Kiser, 2002; Rora et al., 1998). The present results (Table 3) showed a positive correlation ($r = 0.981$) to exist between hardness and AIP, and a negative correlation ($r = -0.715$) between hardness and moisture. Rora et al. (1998), however, reported a negative correlation between fat content and shear force in smoked salmon fillets, as reported here ($r = -0.404$). Finally, Hultmann et al. (2004), reported no linear relationship to exist between the F_{\max} and the salt-soluble protein content; this also agrees with the present results ($r = 0.071$).

Clearly, the two liquid smoke flavourings used have different effects on salmon flesh, perhaps due to their different compositions. Toth and Potthast (1984) indicate that during the surface treatment of fish and meat products with traditional smoke, reactions between carbonyls and proteins occur which would give more consistency to the final product. However, phenolic derivatives can form hydrogen bonds with water (Maga, 1988), and hardness decreases with increasing water content (Rongrong, Carpenter, & Cheney, 1998).

In conclusion, the two commercial smoke flavourings studied led to changes in the physicochemical and texture characteristics of Atlantic salmon flesh. These changes were more accentuated with SL2, perhaps because of its particular composition. Studies investigating microbiological changes, shelf-life and consumer attitudes are still needed, but with textural and physicochemical characteristics exclusively in mind, these smoke flavourings could be used to cold-smoke Atlantic salmon.

Acknowledgements

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