

Original Research Article

OPTIMIZING SALTING AND IMMERSION TIME IN LIQUID SMOKE OF FARMED GILTHEAD SEA BREAM (*SPARUS AURATA*) FILLETS USING RESPONSE SURFACE METHODOLOGY

ABSTRACT

Aims: To produce smoked gilthead sea bream fillets using the liquid smoke dipping method during brining and optimize the brining-smoking process using the response surface method.

Study Design: The experimental design was a two-level Central Composite Design that included 14 runs divided into two blocks.

Place and Duration of Study: Department of Fisheries and Aquaculture Technology, Technological Educational Institute of Western Greece, Messolonghi, Greece, between April 2015 and December 2015

Methodology: Second order polynomial models for yield, moisture content, salt content, water activity, pH, redness index and instrumental texture parameters of salted and smoked with liquid smoke gilthead sea bream filets were developed as a function of salt brine content (from 8.7 to 19.3 % (w/w)) and immersion time (from 30 to 139 minutes) in brining solutions that contained 10% w/w liquid smoke. A numerical optimization was used to find the optimum values for salt brine content and immersion time. For the confirmation of the models smoked gilthead sea breams were prepared using the optimal settings of the factors. The yield, salt content, water content, water activity, redness index and the textural parameters were determined. The results were statistically compared to the values predicted by the mathematical models. A consumer panel evaluated the smoked gilthead sea breams prepared using the optimal settings of the factors. The proximate composition of the smoked gilthead sea bream was also determined.

Conclusions: Within the experimental range, response surface methodology and the numerical optimization method proved useful in determining the optimum settings for salting and smoking with the liquid smoke of gilthead sea bream fillets. The optimal conditions for salt brine content and immersion time were 15.867 % (w/w) and 109.975 minutes, respectively. Under optimal conditions, yield, water and salt content, water activity, pH, redness index, maximum shear force and work of smoked gilthead sea bream were 79.73 %, 61.99%, 3.81% 0.94, 5.55, 0.41, 1269.1 (g*) and 3659.8(g* sec), respectively. The water, protein, lipid and ash content of the smoked product were 61.87%, 20.89%, 12.43% and 3.56%, respectively, and the consumer panel highly accepted it.

Keywords: Liquid smoke, gilthead sea bream, salting, optimizing

1. INTRODUCTION

Gilthead sea bream (*Sparus aurata*) is one of the most important fish species farmed in Greece. However, in recent years, the increased supply of gilthead sea bream caused prices to decline (1). In certain, also, periods of the year, there is a plentiful supply of fresh fish in the market, which causes a further decline in the prices (2). There is, therefore, need to look for the development of value-added products of gilthead sea bream for commercial or industrial use. Such products should fulfil consumers' demands and make fish farming industries more profitable. VASILIADOU *et al.*(2005) (3) suggested that smoked gilthead sea bream could be such a product.

Smoking is a traditional processing method that gives particular flavour, aroma, texture and colour to the smoked fish (4). Liquid smoke has several advantages over traditional smoking. Smoked fish with liquid smoke contain fewer quantities of the carcinogenic polynuclear aromatic hydrocarbons compared to those smoked with conventional methods (5). Also, several common food-borne pathogens have shown sensitivity to liquid smoke in vitro and food systems (6). Moreover, liquid smoke has the advantage over the traditional smoking methods of lowering costs and less environmental damage (7). Thus, liquid smoke has the potential for use as an alternative to the traditional smoking process. It has been tested in various sea foods, including salmon (8), trout (4, 7, 9, 10), mackerel (11), anchovy (12), swordfish (13) and various species of bivalve molluscs (14-16).

Immersion in a brine solution is the most common method of salting fish that are going to be smoked. Brine solutions can also be used to apply liquid smoke onto the fish tissue (11). Salt functions as a flavour enhancer; improves the texture and has a noticeable preservative effect to the smoked fish (17). In published studies, the salting process presents large variation regarding the salt brine content and immersion time (12, 18)

Response surface methodology (RSM) is a statistical method used for developing, improving, and optimizing processes when several factors influence the response of interest. The objective of RSM is to build a mathematical model that precisely describes the overall process (19). It has been used to optimize processes in several kinds of seafood such as sausages from minced mullet (20), surimi from mechanically recovered fish muscle (21), smoked mussels (16) and smoked catfish (22).

Although the effects of traditional smoking on quality parameters of gilthead sea bream have been investigated (3), no reference concerning the use of liquid smoke for the production of smoked gilthead sea bream has been found in the literature. Therefore, the aim of this work was to produce smoked gilthead sea bream fillets using the liquid smoke dipping method during brining and optimize the brining-smoking process using the RMS method.

2. MATERIALS AND METHODS

2.1 Fish

Gilthead sea breams (average weight and length 356 g and 251 mm, respectively) were offered by a fish farming company located in Western Greece (Sao SA, Patra, Greece). Fish were packed in an insulated polystyrene container with flaked ice and delivered to the Technological Educational Institute of Western Greece in Messolonghi on the next day of their harvesting. At the laboratory, the fish were washed, eviscerated and filleted by hand. Then, the fillets were wrapped individually in aluminium foil, placed in plastic bags and vacuumed. Afterwards, the fillets were stored frozen at -80°C until use. The mean weight of the skinned on fillets was 65.0 ± 3.8 g (average $\pm$ S.D.).

2.2 Processing of Gilthead Sea Bream fillets with liquid smoke

The vacuum packed fillets were allowed to thaw in a refrigerator chamber at 2°C overnight (12 hours). The thawed fillets were weighed and soaked in separate containers containing the pre-chilled at 2°C brining/smoking solution (named from now on immersion solution). The immersion solution contained sodium chloride, sugar at concentration 20% (w/w) of sodium chloride content, the colorant E-102 at concentration 0.0070% (w/w) and 10% (w/w) of liquid smoke. The salt brine content and immersion time for each run are shown in Table 1. The liquid smoke was a commercially available, water-soluble liquid smoke condensate (Papadimitriou SA, Sindos, Greece). The fish to brine ratio was 1:2, and brine temperature was 2°C. After brining and smoking, the fillets were dried in an oven with forced air circulation at 35°C for 60 min, smeared with sunflower and cooked at 90°C for 60 min, to an internal temperature of 70°C at the end of the process. The internal temperature of the smoked product was monitored using K-type thermocouples (0.5 mm, Comark Instruments, U.K) and a recording thermometer (Comark KM1242, Comark Instruments, UK). The smoked fillets were cooled at 2°C overnight and then they re-weighted for processing yield determinations. Then, the smoked fillets were sliced into two portions as follows: a) the anterior portion, which was used for the chemical and colour determinations, and b) the middle-tail portion that was used for the instrumental textural determinations.

2.3 Physicochemical analyses

Yield was calculated following the formula:

$$\text{Yield} = \{(\text{weight of raw fillet in gramme}) \times (\text{weight of cooked fillet in gramme})^{-1}\} \times 100$$

A portion of fish muscle (2g) was used for moisture determination according to the method of AOAC 1995 (23). Salt content in samples was determined by the Mohr method (24). Water activity (aw) measurements were made using the Novasina apparatus, model Lab master-aw (Novasina AG, Switzerland). The pH was measured according to the method of Varelztsis et al., 1988(25). Readings were taken using a Crison Basic 20 model pH-meter (Crison Instruments SA, Spain) at ambient temperature. Colour measurements were carried out using a Handerlab Miniscan EZ Meter (Hunter Associates Laboratory, Inc., USA). The instrument was standardized against a white and black tile before each measurement. Each measurement was repeated three times for each sample. a* and b* values were measured, and redness index (RI) was calculated using the equation $RI = a \times b^{-1}$ (4). The shear force and work of smoked sea breams were measured using a universal testing machine (Stable Micro System, Model TA-XT Plus, Texture Exponent, Surrey, UK) equipped with a modified 4-bladed Kramer shear cell. The cross-head speed was 3 mm sec⁻¹; the cutting distance was 48 mm, and the load cell was 50 kg. The proximate composition of the smoked sea bream fillets that were prepared using the optimal salt brine content and immersion time was determined. The residue from the moisture determination was heated at 550°C for 24 hours for ash determination. Crude protein of the sea bream burgers was analyzed by the Kjeldahl method (23). Total fat content (%) was determined from 4g freeze dried sample using petroleum ether and a Soxtherm S-360D extraction unit (Gerhardt, Germany).

127 Table 1. Design matrix for smoked sea bream

Blocks	Runs	Salt Brine Content g/100g		Immersion Time minutes	
		Coded values	Actual values	Coded values	Actual values
Block 2	1	0	12.9	0	75
Block 2	2	0	12.9	1.41	139
Block 2	3	0	12.9	0	75
Block 2	4	1.41	19.3	0	75
Block 2	5	0	12.9	0	75
Block 2	6	0	12.9	-1.41	11
Block 2	7	-1.41	6.5	0	75
Block 1	8	-1	8.4	1	120
Block 1	9	1	17.4	1	120
Block 1	10	-1	8.4	-1	30
Block 1	11	0	12.9	0	75
Block 1	12	0	12.9	0	75
Block 1	13	0	12.9	0	75
Block 1	14	1	17.4	-1	30

128

129 2.4 Experimental design and statistical analysis

130 All statistical analyses, generation of response surfaces, optimization, verification and
 131 contour plots were accomplished using the Expert Design version 9 (Stat-Ease Inc.,
 132 Minneapolis, USA) statistical software. The experimental design was a two-level Central
 133 Composite Design that included 14 runs divided into two blocks. The coded and actual
 134 values of each run are shown in Table 1. A response surface methodology was used to
 135 analyze the effect of the two factors (A and B) on the responses (Y) using the second-order
 136 polynomial response surface with the corresponding interactions:
 137 $Y = b_0 + b_1A + b_2B + b_{12}AB + b_{11}A^2 + b_{22}B^2$ (1)

138 Where Y is the corresponding response variable; A and B are the codified factors (A=salt
 139 brine content, and B= immersion time), and b_1 and b_2 are linear, b_{12} is interaction, and b_{11} and
 140 b_{22} are quadratic coefficients of the model. A stepwise methodology was followed to
 141 determine the significant terms in Equation (1). The significances of all coefficients in the
 142 equations were judged statistically at a probability (P) of 0.05. Adjusted and prediction R^2 ,
 143 prediction error sum of squares (PRESS), and adequacy precision were used to confirm the
 144 adequacy of the models (19). Prediction R^2 comparable to adjusted R^2 , a low PRESS value
 145 and adequacy precision higher than 4 suggest that the model as fitted is adequate to
 146 predicting. For these experiments, forty-two fish were used, that were obtained in two blocks
 147 of twenty-one fish. For each run, the physicochemical properties of six fillets were analyzed.

148 A numerical optimization was used to find the optimum values for salt brine content
 149 and immersion time. For the confirmation of the models, two batches of six smoked sea
 150 breams were prepared using the optimal settings of the factors. The yield, salt, and water
 151 content, aw, RI and the textural parameters were determined as described in Materials and
 152 Methods section. Finally, results were statistically compared to the values predicted by the
 153 mathematical models.

2.5 Sensory evaluations

A consumer panel of sixty-three university students and other people evaluated smoked sea breams prepared using the optimal settings of the factors. For this purpose, a total number of 42 fillets were processed as described above. Sensory evaluations of saltiness, texture, flavour and acceptability were performed following the sensory scales described by (3). According to these sensory scales, the limit of the acceptability was defined between 3 and 5 for saltiness, texture, flavour and acceptability.

3. RESULTS AND DISCUSSION

3.1 Statistical Analysis on Model Fitting

The results of the physicochemical parameters of gilthead sea bream fillets subjected to different levels of brine concentration and immersion time are presented in Table 2. The factors and the responses were fitted to the following coded equations:

$$\text{Yield} = +82.32 - 1.86 * A - 1.76 * B \quad (2)$$

$$\text{Water Content} = +64.67 - 1.51 * A - 1.34 * B - 1.25 * AB \quad (3)$$

$$\text{Salt Content} = +2.98 + 0.71 * A + 0.56 * B + 0.26 * AB - 0.34 * B^2 \quad (4)$$

$$\text{Water Activity (aw)} = +0.96 - 0.010 * A - 9.251E-003 * B - 5.333E-003 * AB \quad (5)$$

$$\text{pH} = +5.71 - 0.21 * B \quad (6)$$

$$\text{Redness Index (RI)} = +0.38 + 0.018 * A + 0.43 * B - 0.026 * B^2 \quad (7)$$

$$\text{Max Shear Force} = +1060.69 + 111.91 * A + 173.30 * B \quad (8)$$

$$\text{Shear Work} = +3063.31 + 240.67 * A + 563.44 * B \quad (9)$$

The analyses of variance showed that the models and the coefficients were significant ($P < 0.05$). The models as fitted explain 72.44, 83.04, 96.09, 99.49, 73.42, 86.98, 83.51 and 93.20% of the variability in the yield, water content, salt content, water, aw, pH, RI, maximum shear force, and work, respectively. All models possessed no significant ($P > 0.05$) lack of fit values, low PRESS, prediction R^2 comparable to fitted R^2 , and adequacy precision higher than 4 (Table 2). Therefore, the models as fitted provided an adequate approximation to the true system.

Table 2. Data of physical, chemical and textural properties of smoked sea bream processed with different levels of salt brine contents and immersion times in the brining salting solution

Std	Block	Run	A: Salt content % (w/w)	brine B: Time minutes	Immersion Yield %	Water Content % (w/w)	Salt Content %(w/w)	aw	pH	RI	Max Force g*	Shear g*x sec	Shear Work
12	Block 2	1		12.9	75	84 ¹	63.73	2.94	0.957	5.70	0.366	1065.25	3004.94
11	Block 2	2		12.9	139	78.29	63.49	2.97	0.945	5.52	0.386	1229.95	3594.55
14	Block 2	3		12.9	75	83	64.97	2.99	0.957	5.70	0.379	1149.66	3141.39
9	Block 2	4		19.3	75	79.36	61.69	3.85	0.942	5.62	0.405	1310.24	3396.89
13	Block 2	5		12.9	75	81	64.49	3.31	0.957	5.49	0.403	1069.6	3140.19
10	Block 2	6		12.9	11	84.60	67.78	1.56	0.971	6.12	0.249	759.336	2237.16
8	Block 2	7		6.5	75	85.05	66.21	1.79	0.970	5.65	0.327	829.8	2521.03
3	Block 1	8		8.4	120	82.20	65.99	2.32	0.962	5.55	0.400	1146.13	3547.57
4	Block 1	9		17.4	120	80.59	60.63	4.24	0.930	5.49	0.420	1343.67	3927.41
1	Block 1	10		8.4	30	86.6	65.80	1.62	0.970	5.93	0.327	875.334	2290.12
6	Block 1	11		12.9	75	81.59	64.61	3.00	0.956	5.82	0.395	1028.41	3267.61
5	Block 1	12		12.9	75	81.20	65.80	3.01	0.956	5.62	0.397	1057.31	3072.39
7	Block 1	13		12.9	75	83.64	64.72	2.88	0.955	5.81	0.376	1091.33	3148.06
2	Block 1	14		17.4	30	81.35	65.44	2.48	0.959	5.92	0.344	893.62	2597.01
	Lack of Fit					0.7132	0.2719	0.4280	0.2379	0.7672	0.4388	0.0731	0.1444
	Adj R-Squared					0.7244	0.8304	0.9609	0.9949	0.7342	0.8698	0.8351	0.9320
	Pred R-Squared					0.5570	0.6727	0.9223	0.9901	0.5573	0.7431	0.6628	0.8722
	PRESS					30.18	14.57	0.61	1.661E-005	0.20	6.456E-003	1.331E+005	4.069E+005
	Adeq Precision					10.832	12.975	25.580	81.944	12.623	17.144	14.493	23.910

¹Mean from six independent measurements

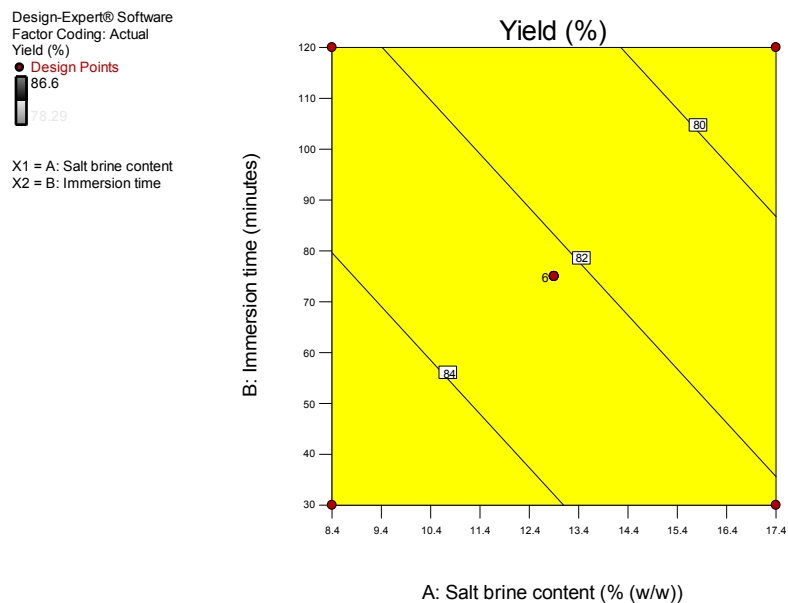
3.2 Effect of salt brine content and immersion time on physicochemical parameters of smoked gilthead sea bream fillets

The level of b_1 and b_2 values (Equation 2) indicates the higher linear negative effect of salt brine content followed by that of immersion time on the yield of smoked sea bream. This result suggests a decrease in yield with an increase of these two factors and can be attributed to the changes in water content of the smoked sea bream, as will be discussed later in this study. It appears that the yield reaches optimal conditions at approximately 13 % (w/w) salt brine content and 80 minutes immersion time (Fig. 1). In this study, the mean filleting yield was 36.6% of the weight of whole sea breams, and that of the smoked sea bream ranged from 86.7 to 79.4 % of the initial weight of the fillets. Thus, the overall processing yield of the smoked sea bream fillets ranged from 31.7 to 29.1%. By contrast to the results of the present study, VASILIADOU *et al.* (2005)(3) report an average overall processing yield of 37% for hot smoked gilthead sea breams that were processed without a head, guts, skin, and tail. However, the processing yield of fish after smoking depends on their size and biological condition, the form of the treated samples (e.g., whole fish or fillets) and the technology applied (26).

The level of b_1 and b_2 values (Equation 3) indicates the higher negative linear effect of salt brine content followed by that of immersion time on the water content of smoked sea bream. This result suggests a decrease in water content with an increase of these two factors. The interaction coefficient (b_{12}) of salt brine content and immersion time has, also, a negative effect. In the contour plot (Fig.2), it is observed that the water content of the smoked sea bream reaches its minimum at approximately 17% (w/w) salt brine content and 110 minutes immersion time.

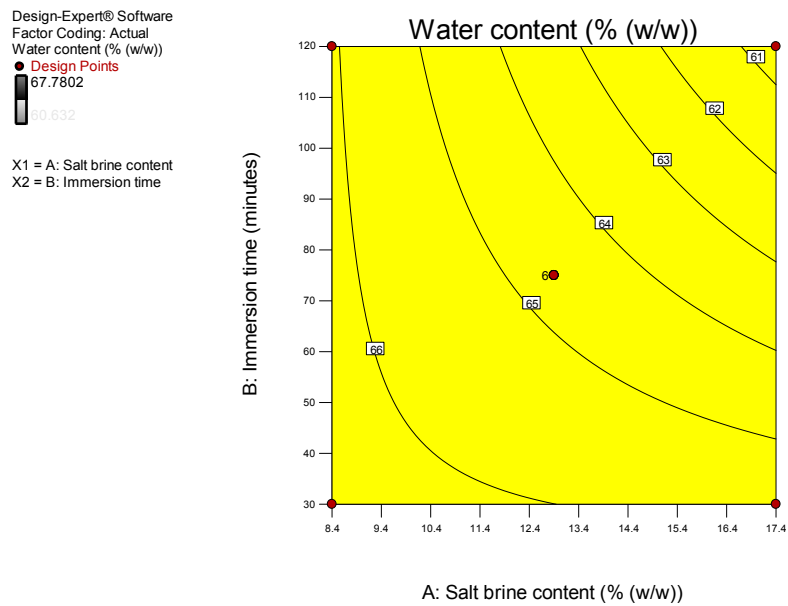
The level of b_1 and b_2 values (Equation 4) indicates the higher positive effect of salt brine content followed by that of immersion time on the salt content of the smoked sea bream. This result shows an increase of the salt content of smoked sea bream with an increase of these two factors. The interaction coefficient (b_{12}) of salt brine content and immersion time has, also, a positive effect, whereas the quadratic coefficient (b_{22}) indicates an adverse effect of immersion time. In the contour plot (Fig. 3), it is observed that the salt content of the smoked sea bream reaches its maximum at approximately 17% (w/w) salt brine content and 96 minutes immersion time. JITTINANDANA *et al.* (2002)(9) observed similar results for smoked trout and CORZO *et al.* (2015)(22) for smoked catfish with those found in the present study for water and salt content of smoked sea bream. When a fish muscle is brined, water diffuses out of muscle and salt diffuses from brine into muscle, due to the differences in concentration and osmotic pressures among inter-muscles and salting agent (27, 28). Thus, the overall result is the enrichment with sodium chloride and partial dehydration of fish muscle.

244 Figure 1. Contour plot of yield of smoked sea bream



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246 Figure 2 Contour plot of water content of smoked sea bream



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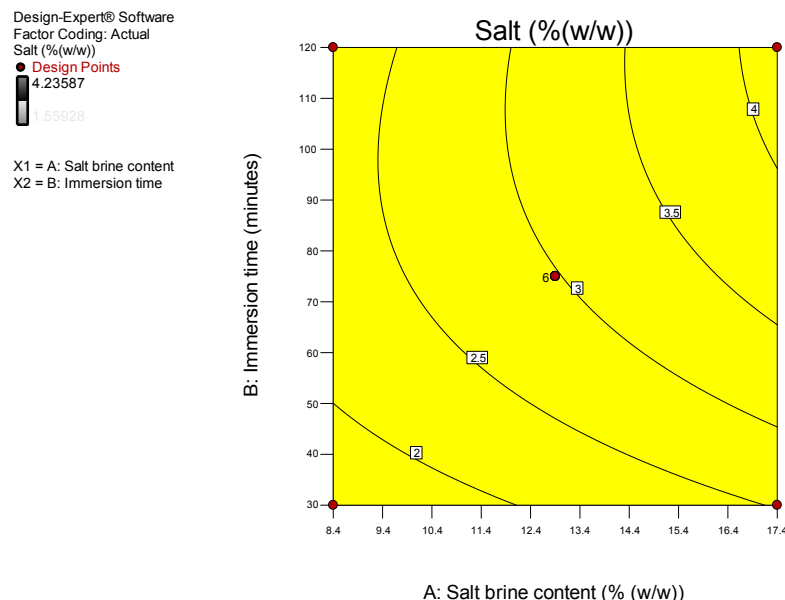
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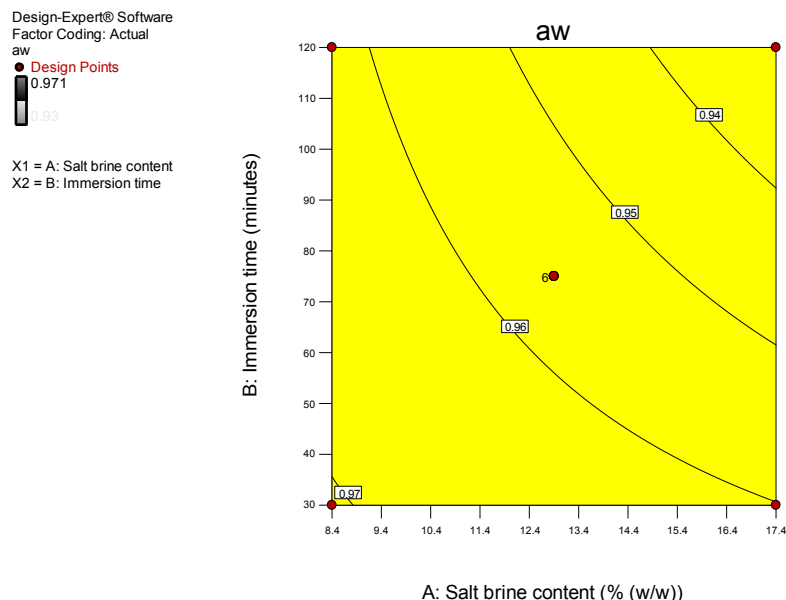
254 Figure 3. Contour plot of the salt content of smoked sea bream



255 The level of b_1 and b_2 values (Equation 5) indicates the negative linear effect of salt
 256 brine content and immersion time on a_w of smoked sea bream. The mentioned effect suggests
 257 that the water activity of the smoked sea bream decreases with the increase of salt brine
 258 content and immersion time. The interaction coefficient (b_{12}) of the salting parameters has a
 259 small negative effect. Bacterial growth is retarded by salt levels which reduce water activity
 260 (a_w) of fish muscle to about 0.97 or less. Smoked fish with salt contents greater than 3.5% in
 261 their water phases will usually have such a value of water activity although many factors can
 262 cause variation. A water activity of less than 0.85 is necessary to make products stable at
 263 room temperature, and a value of a_w of about 0.75 or less is needed to inhibit mold growth
 264 (29). In the contour plot (Fig.4), it is observed that the water activity of smoked sea bream
 265 reaches its minimum at approximately 14% (w/w) salt brine content and 94 minutes
 266 immersion time.
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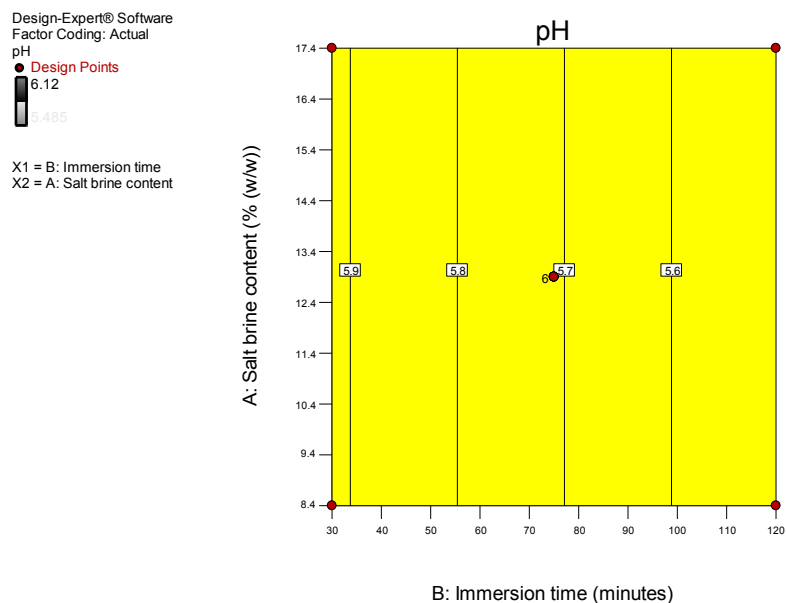
268 The level of b_2 value (Equation 6) indicates the negative linear effect of immersion
 269 time on pH. This result suggests that pH of smoked sea bream decreases with the increase of
 270 immersion time. The result is mainly due to the high acidity of the liquid smoke used in the
 271 present study (13). The mentioned effect is shown in Figure 5 by the shape of parallel lines.
 272 Minimal values of pH are approached at an immersion time of approximately 100 minutes.
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274 Figure 4 Contour plot of water activity (aw) of smoked sea bream



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276 Figure 5 Contour plot of pH of smoked sea bream

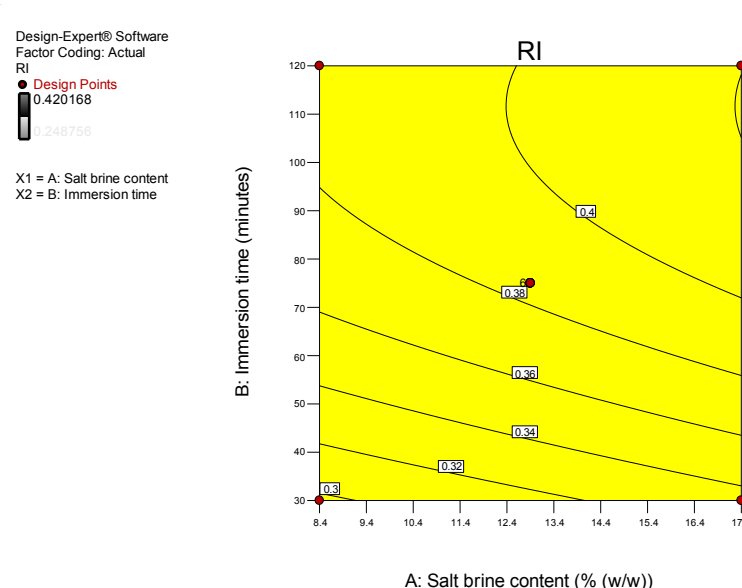


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278 The level of b_1 and b_2 values (Equation 7) indicates the higher linear positive effect of
 279 immersion time followed by salt brine content on RI of smoked sea bream. The result
 280 suggests that the red tone (higher a^* values) of the smoked sea bream becomes more intense
 281 with an increase of salt brine content and immersion time. As expected, the amount of the
 282 absorbed from the fish liquid smoke, which had a dark red colour, increases with immersion
 283 time. Also, as was mentioned earlier in this paper, the water content of sea bream muscle
 284 decreases with increases of both salt brine content and immersion time, which causes

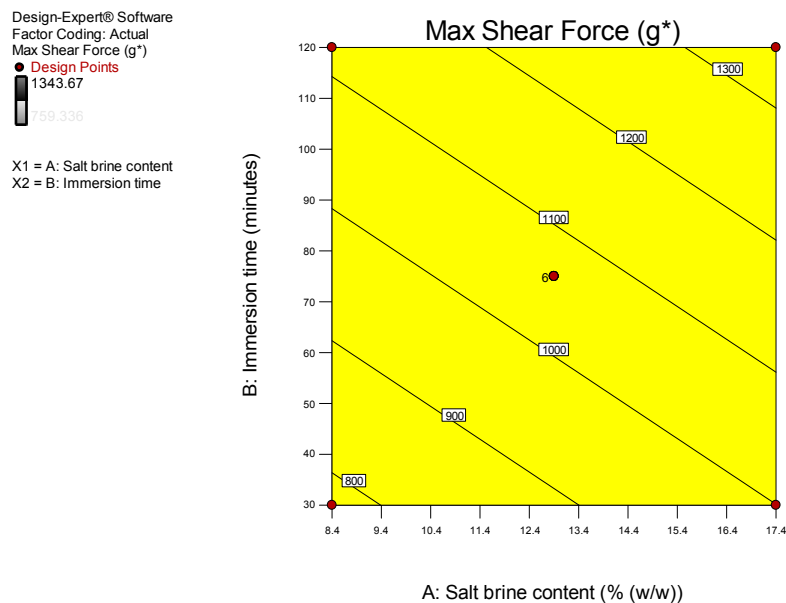
condensation of the absorbed liquid smoke in sea bream muscle giving a more intense reddish tone to the finished product. The quadratic coefficient (b_{22}) indicates a negative effect of immersion time on RI. Colour measurements of smoked fish can be of great interest for producers or retailers due to the impact of consumer purchase. Previous studies on smoked fish have shown that the colour of the products is related to the pigment received in the diet, the fat content of the fish and the smoking technology applied (30). In the contour plot (Fig.6), it is observed that smoked sea bream reaches maximum redness at approximately 13% (w/w) salt brine content and 75 minutes immersion time.

Figure 6. Contour plot of redness index (Ri) of smoked sea bream



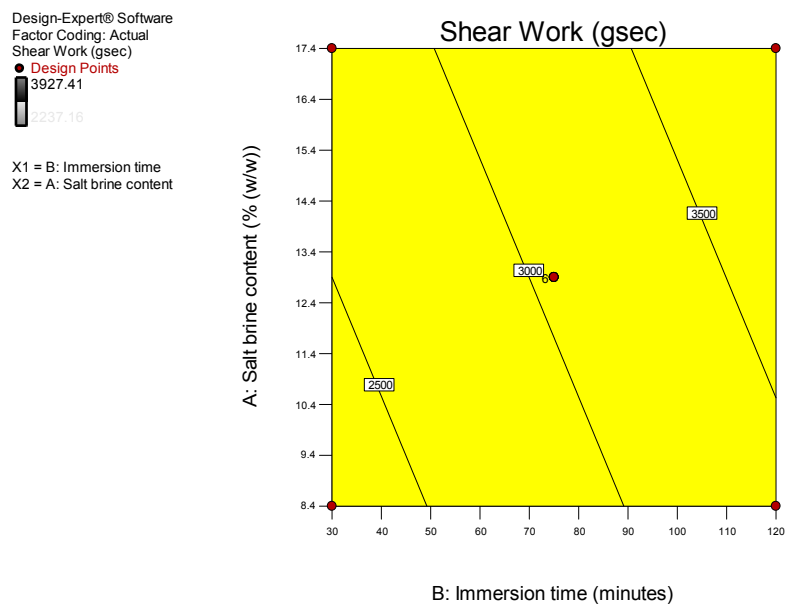
The level of b_1 and b_2 values in Equations 8 and 9 suggest a positive linear effect of brine concentration and immersion time on maximum shear force (i.e. hardness/softness values) and shear work (i.e. toughness/tenderness values) of smoked sea bream. Furthermore, the effect of immersion time is higher than that of salt brine content in both responses. These results suggest that the smoked sea bream fillets are harder and tougher as salt brine content and immersion time increase. These effects are shown in Figures 7 and eight by the shape of parallel lines which approach their maximal values above levels of salt brine content 14.4% and immersion time 100 minutes for maximum shear force, respectively, and of salt brine content 10.4% and immersion time 90 minutes for shear work, respectively. Salt brine content and duration of brining had a major effect on the texture of smoked rainbow trout (9). Changes in instrumental texture of salmon treated with liquid smoke flavourings are also reported by MARTINEZ *et al.* (2007)(8). Fish species with less water content are harder in texture than those with more water content (31). The texture of smoked fish is, mainly, determined from the state of myofibrillar proteins that is primarily related to the salt concentration in the water phase of fish (32). Interaction of fish myofibrillar protein with salt may cause protein denaturation, which causes changes in texture and reduced water holding capacity (33). Altogether, the changes in the texture of smoked sea bream of the present study can be attributed to the decreasing moisture and increasing salt content in the water phase of the smoked fillets as salt brine content and immersion time were increasing.

315 Figure 7. Contour plot of maximum shear force of smoked sea bream



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317 Figure 8. Contour plot of shear work of smoked sea bream



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319 3.3 Numerical Optimization

320 Table 3 shows the optimization criteria for each factor and response. The optimum
321 combination of salt brine content and immersion time were 15.867 % (w/w) and 109.975
322 minutes, respectively, with an overall desirability $D = 0.606$. Table 3 also shows the
323 predicted responses for this combination.

324 Table 3. Criteria and output for numerical optimization of smoked sea bream

Criteria	Goal	Lower Limit	Upper Limit	Output
A: Salt brine content	is in the range	8.4	17.4	15.864
B: Immersion time	is in range	30	120	109.975
Yield (%)	Maximize	78.29	86.6	79.73
Water content (%)	Minimize	60.632	67.7802	61.99
Salt (5)	Maximize	1.55928	4.23587	3.81
aw	Minimize	0.93	0.971	0.939
pH	Minimize	5.485	6.12	5.55
RI	is in range	0.248756	0.420168	0.414
Max Shear Force (g*)	is in range	759.336	1343.67	1269.10
Shear Work (g* sec)	is in range	2237.16	3927.41	3659.76
Desirability				0.606

325 3.4 Verification of the model

326 Once the optimum combination of the factors was determined, it was used to prepare
 327 smoked sea breams, and all the response variables of this product were determined.
 328 Experimental values of each response were compared with those predicted by the models,
 329 and both sets of values are presented in Table 4. The average values from the confirmation
 330 experiments were within the confirmation prediction interval thus the models were
 331 confirmed.

332 Table 4 predicted and experimental physical chemical and textural properties of smoked sea
 333 bream processed using salt brine content 15.86% and immersion time 109.97 minutes

Response	Predicted	Experimental mean	95% PI ² low	95% PI high
Yield (%)	79.73	80.28	77.39	82.06
Water content (¹ n=2, %)	61.99	61.87	60.42	63.56
Salt (n=2, %)	3.81	3.90	3.48	4.13
aw (n=2)	0.939	0.94	0.94	0.94
pH (n=2)	5.55	5.39	5.37	5.73
RI	0.414	0.392	0.38	0.45
Max Shear Force (n=2, g*)	1269.10	1331.21	1131.72	1406.48
Shear Work (n=2, g* sec)	3659.76	3706.86	3409.07	3910.45

334 ¹ n= number of replicates (each replicate contained 6 fillets), ²PI= Prediction interval

3.5 Proximate composition and sensory assessment of smoked gilthead sea bream

The results for the proximate composition of the raw fish are similar to those reported by KYRANA *et al.* (1997)(34) for the farmed gilthead sea bream. Salting, drying and cooking reduced the moisture content and increased the protein, fat and ash contents of the smoked product (Table 5).

The mean scores of the sensory attributes of the smoked product were close to the “best” scores of every one of the sensory properties (Table 5): Thus, the smoked sea bream was highly accepted by the consumer panel.

Table 5. Proximate composition and sensory attributes of smoked sea bream processed using salt brine content 15.86% and immersion time 109.97 minutes

Proximate Composition	Raw Gilthead Sea Bream	Smoked Product
Water Content	73.52±0.86 ¹	61.87±2.75
Protein Content	19.98±1.05	20.89±1.05
Fat Content	3.93±0.83	12.43±0.79
Ash	1.37±0.07	3.56± 0.21
Sensory Attributes		
Saltiness		4.38±0.65 ²
Texture		4.25±0.62
Flavour		4.32±0.90
Acceptability		4.15±0.71

¹ Mean value ± standard deviation (n= 3), ²Mean value ± standard deviation (n= 63)

Conclusion

Second order polynomial models for yield, moisture content, salt content, aw, pH, redness index and instrumental texture parameters of salted and smoked with liquid smoke gilthead sea bream filets were developed as a function of salt brine content and immersion time in brining solutions that contained 10% w/w liquid smoke. Within the experimental range, response surface methodology and the numerical optimization method proved useful in determining the optimum settings for salting and smoking with the liquid smoke of gilthead sea bream fillets. The optimal conditions for salt brine content and immersion time were 15.867 % (w/w) and 109.975 minutes, respectively. Under optimal conditions, yield, water and salt content, water activity (aw), pH, redness index, maximum shear force and work of smoked gilthead sea bream were 79.73 %, 61.99%, 3.81% 0.94, 5.55, 0.41, 1269.1 (g*) and 3659.8(g* sec), respectively. The water, protein, lipid and ash content of the smoked product were 61.87%, 20.89%, 12.43% and 3.56%, respectively, and the consumer panel highly accepted it.

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