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Rizo Parraga, AM.; Fuentes López, A.; Fernández Segovia, I.; Masot Peris, R.; Alcañiz Fillol, M.; Barat Baviera, JM. (2013). Development of a new salmon salting smoking method and process monitoring by impedance spectroscopy. *LWT - Food Science and Technology*. 51:218-224. doi:10.1016/j.lwt.2012.09.025.



The final publication is available at

<https://dx.doi.org/10.1016/j.lwt.2012.09.025>

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Additional Information

# Accepted Manuscript

Development of a new salmon salting-smoking method and process monitoring by impedance spectroscopy

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PII: S0023-6438(12)00405-7

DOI: [10.1016/j.lwt.2012.09.025](https://doi.org/10.1016/j.lwt.2012.09.025)

Reference: YFSTL 3233

To appear in: *LWT - Food Science and Technology*

Received Date: 12 March 2012

Revised Date: 18 September 2012

Accepted Date: 25 September 2012

Please cite this article as: Rizo, A., Fuentes, A., Fernández-Segovia, I., Masot, R., Alcañiz, M., Barat, J.M., Development of a new salmon salting-smoking method and process monitoring by impedance spectroscopy, *LWT - Food Science and Technology* (2012), doi: 10.1016/j.lwt.2012.09.025.

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12 **Abstract**

13

14 In this work two objectives were proposed: (i) to optimize a new salmon salting-  
15 smoking method using vacuum packaging and (ii) to evaluate the application of  
16 impedance spectroscopy (IS) to the on-line monitoring of the process. Different  
17 processing conditions were evaluated (4 smoke flavoring (SF) salt concentrations, 3  
18 salting times, salting in vacuum or in air). Physico-chemical analyses and IS  
19 measurements were performed with three different sensors during the process. Salting  
20 with 16 g SF salt/100 g fish in vacuum packaging provided smoked salmon similar to  
21 products currently available on the market. This new method has the advantages of  
22 reducing processing times and waste. IS measurements were carried out by three  
23 different electrodes. The most appropriate sensor for process monitoring was a needle  
24 electrode, with which robust prediction models for NaCl content, moisture and  $a_w$   
25 during the salting-smoking process were obtained. The results showed the potential of  
26 IS as a rapid on-line monitoring method of the salmon salting-smoking process.

27

28 *Keywords:* Salmon; vacuum salting-smoking; impedance spectroscopy; process monitoring.

## 29 1. Introduction

30

31 Smoking is one of the oldest methods of fish preservation. The preservative effect of  
32 smoking is due to a combination of different factors, including the addition of salt,  
33 partial dehydration of the tissues that occurs throughout the different stages of the  
34 process, as well as the preservative action of the smoke components. The smoking  
35 process slows down the biological processes and oxidative damage and gives the final  
36 product sensory characteristics highly appreciated by consumers.

37 Improvements in the smoking process, including the reduction in processing times  
38 and the amount of brine wastes or the improvement of the hygienic quality would be of  
39 interest to this sector. To obtain high-quality smoked salmon with a long shelf life,  
40 optimization of the various stages that constitute the smoking process is essential. The  
41 salting step is especially critical. A salting process in which the exact amount of salt to  
42 be absorbed by the fish would be directly dosed, combined with vacuum packaging,  
43 could be an alternative to these techniques. With this new method both brine wastes and  
44 contamination would be reduced, since the lack of oxygen in vacuum packaging would  
45 delay microbial growth and lipid oxidation. The main disadvantage that could present  
46 this method is the growth of anaerobic microorganisms, such as *Clostridium botulinum*.  
47 Smoke flavoring salt could also be used and would provide salt and a smoky flavor to  
48 the product in a single stage, so that the total processing time would be significantly  
49 shortened.

50 It is well-known that certain physico-chemical parameters, such as  $a_w$  or salt content,  
51 directly affect the shelf life of smoked salmon. However, some studies have found high  
52 variability of these parameters within the same fish product (Cornu et al., 2006; Espe,  
53 Kiessling, Lunestad, Torrissen, & Røra, 2004; Fuentes, Fernández-Segovia, Barat &

54 Serra, 2010a), which have implications for consumers' safety and also for the sensory  
55 characteristics of the product. This is due to the fact that smoking processes are  
56 standardized for a certain fish species, without taking into account the effects of the  
57 initial characteristics (fat, moisture, fish size, freshness, etc.) of the raw material (Barat,  
58 Gallart-Jornet, Andrés, Akse, Carlehög, & Skjerdal, 2006). In this regard, the  
59 development of rapid non-destructive methods for on-line monitoring of the process, in  
60 order to detect when the product has reached optimum moisture, salt and/or  $a_w$  values  
61 would be of interest to producers.

62 Electronic sensors based on impedance spectroscopy (IS) could help to meet this  
63 objective. The relationship between sodium chloride content and impedance  
64 measurements has already been demonstrated (Guerrero et al. 2004; Karásková,  
65 Fuentes, Fernández-Segovia, Alcañiz, Masot, & Barat, 2011). In the IS technique an  
66 electrical sinusoidal stimulus is applied to the electrodes in order to measure the  
67 impedance of the sample at different frequencies. The module and phase of the  
68 impedance can vary significantly according to the charges present (free ions), types of  
69 microstructure and electrolytes, as well as texture, geometry and the electrodes used  
70 (Masot, 2010).

71 In this work two objectives were proposed. The first was to optimize a new salting-  
72 smoking method for salmon using vacuum packaging. The second was to evaluate the  
73 application of impedance spectroscopy in the on-line monitoring of the salting-smoking  
74 process.

75

## 76 **2. Materials and methods**

77

### 78 *2.1. Sample preparation*

79

80 Fillets of Atlantic salmon from a Norwegian farm (Hallvard Leroy AS) of  
81 commercial size 1.4-1.8 kg was used as raw material. The fillets were purchased in a  
82 local supermarket and transported to the laboratory under refrigeration.

83 Fourteen salmon fillets were employed for the complete test (8 for the first phase  
84 and 6 for the second). The fillets were cut transversally into 4 cm portions, obtaining 6  
85 or 7 samples per fillet. Each sample was weighed.

86 Smoke flavoring (SF) salt (Salinera Española, SA) was used in the salting-smoking  
87 stage. Its composition included 50% refined salt, white sugar, baking soda, smoke  
88 flavoring and anti-caking agent (E-536).

89

## 90 *2.2 Experimental design*

91

### 92 *2.2.1. Phase I: Optimization of salting-smoking process and selection of impedance* 93 *spectroscopy electrode.*

94 This phase of the study had two different aims. The first was to establish the  
95 appropriate salting conditions (correct amount of smoke flavoring salt, processing time  
96 and type of packaging) to obtain smoked salmon with similar characteristics to currently  
97 marketed products (60-63 g H<sub>2</sub>O/100 g, 3.5-3.8 g NaCl/100 g,  $a_w=0.963-0.965$ )  
98 (Cardinal, Gunnlaugsdottir, Bjoernevik, Ouisse, Vallet, & Leroi, 2004; Fuentes et al.,  
99 2010a) while generating the minimum of brine waste. The smoked salmon obtained by  
100 this new process is intended to be distributed vacuum packaged under refrigeration. The  
101 second objective was to select the most suitable electrode to monitor the salting-  
102 smoking process.

103 A total of 48 portions of fresh salmon obtained as described above were randomly  
104 divided into 4 batches. Each batch was submitted to a salting-smoking process under  
105 different conditions (Fig. 1). Four concentrations of smoke flavoring salt were studied:  
106 4, 6, 8 and 16 g SF salt/100 g fresh salmon. These concentrations were selected from  
107 previous studies (Fuentes, Pérez, Fernández-Segovia, & Barat, 2011). The weight of SF  
108 salt was spread over the fish muscle surface and the samples were individually placed  
109 inside plastic bags. Each batch was subdivided into 2 further groups, one was packaged  
110 in air and the other in vacuum (Fig. 1). Three processing times were also studied (12, 18  
111 and 24 h). The salting-smoking process was carried out at 4 °C. At the end of the  
112 processing time, the samples were placed in saturated brine under constant stirring for  
113 30 s to remove any traces of SF salt attached to the surface. Finally, the samples were  
114 dried with absorbent paper and re-weighed.

115 Two samples were used for each condition (n=2).

116 Analysis of moisture, pH, NaCl content and  $a_w$  were carried out on the fresh salmon  
117 and the smoked samples at different times during the study. Impedance spectroscopy  
118 measurements were also carried out using the 3 different sensors (double electrode,  
119 arrow electrode and a coaxial needle) described below.

120

121  Fig. 1

122

### 123 2.2.2. Phase II: Monitoring of salting-smoking process using impedance spectroscopy

124 The objective of this second phase of the study was to evaluate the application of  
125 impedance spectroscopy to monitoring the salmon salting-smoking process.

126 The salting-smoking conditions that provided smoked salmon similar to currently  
127 available products were selected from the results obtained in the previous phase. This



128 part of the study was repeated with 3 batches of fish consisting of 12 samples per batch.  
129 Each batch was purchased at intervals of 1 week.

130 The samples were salted-smoked with 16 g SF salt /100 g fresh salmon in vacuum  
131 packaging for 25 h at 4 °C. Analyses were carried out at 5 h-intervals, after rinsing the  
132 samples in brine and drying as described above. Physico-chemical analyses were carried  
133 out (moisture content, pH, NaCl content and  $a_w$ ) as well as impedance spectroscopy  
134 measurements with the electrode selected in Phase I (needle electrode). Two samples  
135 were used ( $n=2$  in each batch) for each salting time, including time 0, corresponding to  
136 fresh salmon.

137

### 138 *2.3. Analytical determinations*

139

140 The physico-chemical analysis and impedance spectroscopy measurements were  
141 performed in the centre of each fillet. The analyses were done in triplicate on each  
142 sample, except for pH, which was measured in quintuplicate.

143

#### 144 *2.3.1 Physico-chemical analyses*

145 Moisture content was determined according to the AOAC method 950.46 (1997).  
146 The pH measurements were carried out using a digital pH-meter micropH 2001 (Crison  
147 Instruments, S.A., Barcelona, Spain) with puncture electrode (Crison 5231) in five  
148 different locations on the sample. Water activity ( $a_w$ ) was measured in minced samples  
149 with a fast water activity-meter (GBX scientific FA-st/1, Cédex, France). Sodium  
150 chloride content was determined according to the procedure described by Fuentes,  
151 Fernández-Segovia, Barat, and Serra (2010b) using an automatic Sherwood Chloride  
152 Analyzer Model 926 (Sherwood Scientific Ltd., Cambridge, UK). Changes in total

153 mass, water and sodium chloride during the salting process were estimated by Eqs. (1),  
 154 (2) and (3):

$$155 \quad \Delta M_t^0 = \left( \frac{M_t^0 - M_0^0}{M_0^0} \right) \quad (1)$$

$$156 \quad \Delta M_t^W = \left( \frac{M_t^0 \cdot x_t^W - M_0^0 \cdot x_0^W}{M_0^0} \right) \quad (2)$$

$$157 \quad \Delta M_t^{NaCl} = \left( \frac{M_t^0 \cdot x_t^{NaCl} - M_0^0 \cdot x_0^{NaCl}}{M_0^0} \right) \quad (3)$$

158

159 ( $M_t^0$  and  $M_0^0$  are the salmon weight,  $x_t^W$  and  $x_0^W$  are the water weight fractions in the  
 160 salmon, and  $x_t^{NaCl}$  and  $x_0^{NaCl}$  are the NaCl weight fraction in the salmon, at sampling  
 161 times t and 0, respectively).

162 Sodium chloride concentration referred to the fish liquid phase ( $Z^{NaCl}$ ) was estimated  
 163 from the determinations of weight fractions of water ( $x^W$ ) and sodium chloride ( $x^{NaCl}$ )  
 164 according to Eq. (4), thus considering that nearly all the sodium chloride and water were  
 165 free in the salmon muscle.

$$166 \quad Z^{NaCl} = \left( \frac{x^{NaCl}}{x^W + x^{NaCl}} \right) \quad (4)$$

167

### 168 2.3.2. Impedance spectroscopy measurements

169 A low-cost, flexible, light, non-destructive measurement system was developed by  
 170 the Instituto de Reconocimiento Molecular y Desarrollo Tecnológico (IDM) at the  
 171 Universitat Politècnica de València (UPV) (Masot et al. 2010). This impedance  
 172 spectroscopy measurement system applies an electric signal to food products and  
 173 measures the response in a frequency sweep between 1Hz and 1MHz.

174 Since the electrical response depends on the type of electrode used, three different  
 175 electrodes were tested in this study. One was a double electrode (DE) composed of two  
 176 stainless steel needles 1.5 cm long and 1 mm in diameter, separated by a distance of 1

177 cm in a non-conductive frame. This design keeps the separation between both needles  
178 constant during measurements.

179 The second electrode, known as the Arrowhead (AH), was designed using thick-film  
180 technology, which uses high-resolution screen-printing methods to deposit pastes or  
181 inks of different electrical characteristics (conductive, resistive and dielectric) on an  
182 insulating substrate, in order to form an electronic circuit. This electrode is designed  
183 with a pointed end to help it to penetrate through the sample.

184 The third sensor (needle electrode) consisted of a hollow needle with an internal  
185 isolated wire, so that a two-electrode system is configured. The external part of the  
186 needle is made of stainless steel and acts as the outer electrode. The internal wire is also  
187 made of stainless steel and acts as the inner electrode. Both electrodes are separated by  
188 dielectric material (epoxy resin). The hollow needle (TECAN 53156, Oxford-  
189 FEDELEC) has an outer diameter of 0.46 mm.

190 The impedance measurements were taken by inserting the sensors into the sample  
191 perpendicular to the muscle fibers of the fish. The penetration depth of the electrodes  
192 was constant in all the analyses (1.5 cm). All measurements were carried out at room  
193 temperature.

194

#### 195 *2.4. Statistical analysis*

196

197 Data are reported as mean  $\pm$  standard deviation. One-way ANOVA was conducted  
198 for each physico-chemical parameter evaluated in Phase II, to determine whether there  
199 were significant differences between the salting-smoking times. Statistical treatment of  
200 the data was performed using the Statgraphics Centurion XVI (Manugistics Inc.,  
201 Rockville, MD, USA).

202 In order to assess the feasibility of the impedance spectroscopy technique to  
203 discriminate between different moisture, NaCl contents, and/or  $a_w$  levels, three Principal  
204 Component Analyses (PCAs) were carried out with data obtained from the DE, AH and  
205 needle electrodes. PCAs were performed using impedance module and phase data  
206 obtained by the equipment in the frequency range established for each sensor. A PCA  
207 was conducted in the same way for the needle electrode with data obtained in Phase II.  
208 Partial Least Squares (PLS) were also carried out to create predictive models of each  
209 physico-chemical parameter evaluated from the IS measurements. PLS prediction  
210 models were created using a set of experimental data (calibration set). The model was  
211 then validated with a new set of experimental data (validation set). All multivariate  
212 analyses were performed using MATLAB ® PLS Tool-box (Eigenvector Research,  
213 Inc.).

214

### 215 **3. Results and discussion**

216

217 *3.1. Phase I: Optimization of the salting-smoking process and selection of the*  
218 *impedance spectroscopy electrode.*

219

220 *3.1.1. Physico-chemical analyses.*

221 Moisture, pH,  $a_w$  and sodium chloride content values of raw material (t=0) and  
222 smoked salmon are shown in Table 1. The values obtained for the raw material are  
223 similar to those reported by other authors for fresh salmon (Fuentes, Fernández-  
224 Segovia, Masot, Alcañiz, & Barat, 2010c; Gallart-Jornet, Barat, Rustad, Erikson,  
225 Escriche, & Fito, 2007).

226 In all the experimental conditions the salting-smoking process caused a significant  
227 reduction in the water content and  $a_w$  values, as well as an increase in the NaCl  
228 concentration, as compared with fresh salmon. Reducing the  $a_w$  values lengthens  
229 smoked salmon shelf-life. These changes are due to dehydration and NaCl absorption  
230 into the muscle. It should be noted that the samples with higher sodium chloride levels  
231 showed a slight decrease in pH values, due to the higher ionic strength of the internal  
232 solution in fish muscle cells, as described by Leroi and Joffraud (2000).

233

234

Table 1

235

236 In both types of packaging, the highest SF salt dosages (8 and 16 g SF salt/100 g  
237 fish) caused the largest increase in NaCl content, with the consequent reduction of  $a_w$   
238 values as processing time advanced. However, for the 4 and 6% SF salt doses, the  
239 magnitude of these changes was smaller in samples packaged in air, being practically  
240 negligible in salmon processed in vacuum packaging (Table 1). This could be explained  
241 by the fact that in these last cases at 12 h almost all the SF salt dose had been absorbed,  
242 so that the changes during the rest of the processing time were minimal.

243 Regarding the type of packaging, vacuum packaging caused faster sodium chloride  
244 absorption and dehydration of the salmon than air packaging. This effect was only  
245 observed for the highest SF salt dose (16 g/100 g fish), with minimal differences  
246 between the two types of packaging for the rest of the studied dosages, since these low  
247 amounts of salt are easily dissolved and absorbed in the first hours of processing in both  
248 types of packaging.

249 Of all the conditions studied, only those samples salted-smoked with 16 g SF  
250 salt/100 g of fresh salmon for 24 h in vacuum reached the levels of moisture, NaCl and

251  $a_w$ , previously established (60-63 g H<sub>2</sub>O/100 g, 3.5-3.8 g NaCl/100 g,  $a_w = 0.963$ -  
252 0.965). These were consequently the salting-smoking conditions selected for Phase II.

253

### 254 3.1.2. Impedance spectroscopy

255 Impedance spectroscopy was used to detect changes in the salmon muscle during the  
256 salting-smoking process. The impedance measurements of fresh salmon were compared  
257 with those of samples submitted to salting-smoking under the different conditions  
258 described above. In this phase, 3 different electrodes (DE, AH and needle) were studied  
259 as described in Section 2.3.2 on Materials and methods.

260 Impedance spectroscopy equipment generate 100 values for each measurement,  
261 corresponding to the modules and phases of the 50 frequencies analyzed. A Principal  
262 Component Analysis (PCA) was conducted for each electrode to determine whether  
263 impedance spectroscopy could discriminate between the different samples. The  
264 impedance data used in this analysis were from the samples processed for 24 h, since  
265 the highest differences in moisture, NaCl content and  $a_w$ , from the 4 SF salt levels were  
266 obtained for this time.

267 The results of the PCAs carried out on the DE, AH and needle electrodes are shown  
268 in Figs. 2.a, 2.b and 2.c, respectively.

269

270  Fig. 2

271

272 In all cases, a clear separation of the raw material (0%) from the rest of the samples  
273 was observed. The ED electrode could also discriminate samples processed with 16 g  
274 SF salt/100 g fish, while no discrimination was observed for the rest of the samples  
275 (Fig. 2.a). For the AH electrode, all samples subjected to the salting-smoking process

276 were overlapped (Fig. 2.b). The needle electrode showed 4 clusters: fresh salmon (0%),  
277 samples with 16% SF salt, samples with 8% SF salt and a fourth group with samples  
278 with 6% and 4% SF salt (Fig. 2.c). The best sensor in discriminating the different levels  
279 of moisture, NaCl contents and/or  $a_w$  was therefore the needle electrode and was  
280 consequently selected for the next phase of the study. These results confirm the  
281 importance of the measuring sensor design (electrode geometry and characteristics).

282

### 283 *3.2. Phase II: Monitoring of salting-smoking process using impedance spectroscopy*

284

#### 285 *3.2.1. Physico-chemical analyses.*

286 Values of moisture, pH and  $a_w$  of salmon submitted to the salting-smoking process  
287 (16% SF salt dosage, vacuum packaging) for 25 h are shown in Table 2. Moisture  
288 content progressively decreased during the salting process, with a higher rate at the  
289 beginning of the process, mainly due to the presence of salt crystals on the surface. The  
290 use of solid salt causes greater dehydration in the product at the beginning of the  
291 process. The water exiting from the muscle is needed to dissolve the salt on the surface  
292 and form brine at the interface, which enables the salt to later penetrate into the fish  
293 muscle.

294 The pH values showed a slight drop with processing time, although the differences  
295 were not significant. The  $a_w$  values decreased throughout the salting-smoking time, due  
296 to muscle dehydration and salt penetration.

297

298 

Table 2
---------

299

300 Fig. 3 shows the evolution of sodium chloride content ( $x^{\text{NaCl}}$ ,  $X^{\text{NaCl}}$  and  $Z^{\text{NaCl}}$ ) during  
301 the salting-smoking process. The NaCl content of the samples increased progressively  
302 with processing time. The highest increase was observed in the case of sodium chloride  
303 concentration expressed on a dry basis ( $X^{\text{NaCl}}$ ), because of the solute incorporation in  
304 fish muscle and osmotic dehydration that occurs during the process.

305

306 Fig. 3

307

308 Total weight, water weight and sodium chloride weight changes are shown in Fig. 4.  
309 Moisture and sodium chloride variations showed opposing behavior throughout the  
310 salting, as mentioned above (Fig. 4).

311

312 Fig. 4

313

314 Total weight changes could be considered a combination of both weight changes  
315 (water and NaCl). However, protein denaturation due to salt action would also  
316 contribute to the weight loss, as different authors have shown (Barat, Rodriguez-  
317 Barona, Andrés, & Fito, 2003; Ismail & Wootton, 1982). It can be assumed that there is  
318 pseudo-diffusional transport due to the strong dependence between the weight changes  
319 and the square root of time, as has been pointed out by other authors (Barat et al., 2006;  
320 Fuentes, Barat, Fernández-Segovia, & Serra, 2008; Gallart-Jornet et al., 2007).

321 It should be noted that samples reached the selected NaCl and  $a_w$  levels after 15 h of  
322 processing, a shorter time than in the preliminary study (Phase I). This is due to the  
323 salting process depending directly on the composition and initial quality of the raw  
324 material (Barat et al., 2006). Different batches of fresh salmon were used in Phases I



325 and II, so that the differences in the raw material could have been the cause of the  
326 differences found in the process kinetics.

327 The use of vacuum during processing and distribution of salmon could permit the  
328 growth of *Clostridium botulinum* type A, B and E, as well as toxin production. *C.*  
329 *botulinum* type E could grow in smoked salmon under vacuum at temperatures as low  
330 as 3.3 °C, if  $a_w$  is higher than 0.966. To minimize the risk of *C. botulinum* growth, the  
331 exact control of  $a_w$  is of outmost importance. This confirms the need for rapid  
332 monitoring methods that can be used on-line to determine the end of the salting process.

333

### 334 3.2.2 Impedance spectroscopy

335 A PCA was used to assess the feasibility of impedance spectroscopy for monitoring  
336 the salmon salting-smoking process, with the impedance spectroscopy values obtained  
337 at 5 h-intervals during the 25 h process.

338 Fig. 5 shows the results of the PCA performed with data obtained from the needle  
339 electrode according to processing time. The samples can be seen to be clearly separated  
340 according to processing time, except for 20 and 25 h, which are overlapped in the same  
341 graphic area. These two samples showed similar values for all the physico-chemical  
342 parameters studied, which justifies the behaviour observed in the PCA.

343

344  Fig. 5

345

346 Since the PCA analysis showed that impedance spectroscopy with the needle  
347 electrode could discriminate between different levels of moisture, NaCl and/or  $a_w$ , a  
348 statistical tool (PLS) was used to predict the values of these parameters from the  
349 measurements of the impedance device. In this way, statistical models were established

350 for all the parameters except for pH, whose evolution was not significant throughout the  
351 processing time.

352 Sodium chloride,  $a_w$  and moisture experimental values *versus* values predicted by  
353 the PLS statistical models are shown in Figs. 6.a, 6.b and 6.c, respectively.

354 In all cases, the predicted values successfully fitted to the experimental values  
355 (RMSEP values of 0.685 for NaCl, 0.006 for  $a_w$  and 3.579 for moisture), especially in  
356 the case of  $a_w$ , for which the intercept was near to 0 and the slope near to 1. These  
357 results agree with other studies on the use of impedance spectroscopy in the  
358 characterization of commercial smoked salmon and cod, in which the best predictions  
359 were obtained for  $a_w$  (Karásková et al., 2011).

360

361  Fig. 6

362

363 The results obtained from the PLS confirm the potential of impedance spectroscopy  
364 with needle electrode for monitoring the salmon salting-smoking process.

365

#### 366 4. Conclusions

367

368 The results obtained from the physico-chemical analyses showed that packaging  
369 under vacuum speeded up the process of NaCl absorption and dehydration in salmon,  
370 although this effect was observed only for the highest dosage of smoke flavoring salt  
371 (16 g/100 g). The optimum processing conditions to obtain a similar product to the  
372 currently available smoked salmon on the market were 16 g SF salt/100 g salmon in  
373 vacuum packaging. This new method has the advantages of reducing processing times  
374 and waste. Further sensory evaluation and shelf-life studies should now be carried out to

375 determine whether the sensory characteristics of smoked salmon obtained by the new  
376 method are comparable to currently marketed products and whether it has a similar or  
377 longer shelf-life.

378 Of the three electrodes used (double, arrowhead and needle electrode) in the IS  
379 measurements, the needle electrode was found to be the most appropriate for process  
380 monitoring. The increase in NaCl content and the reduction in moisture and  $a_w$  values  
381 with 16 g SF salt/100 g fish for 25 h in vacuum were detected by the EI technique using  
382 the needle electrode. This sensor was able to obtain robust NaCl content, moisture and  
383  $a_w$  prediction models during the process. The best of these was the  $a_w$  prediction model,  
384 which is particularly interesting because of the relationship between this parameter and  
385 the shelf-life of smoked products. The results therefore showed that IS is a rapid on-line  
386 monitoring method for the salmon salting-smoking process and could provide an  
387 important tool to obtain products of uniform quality.

388

### 389 **Acknowledgements**

390

391 The authors gratefully acknowledge the financial support for the work reported here  
392 received from the Generalitat Valenciana (GV/2011/098) and the Universitat  
393 Politècnica de València (UPV) (PAID-06-09-2940). A. Fuentes would like to thank the  
394 Campus de Excelencia Internacional at the UPV for its support. The proof-reading of  
395 this paper was funded by the UPV, Spain.

396

### 397 **References**

398

- 399 AOAC. 1997. Official Methods of analysis. *16th Ed. Association of Official Analytical*  
400 *chemists*. Arlington, Virginia.
- 401 Barat, J. M., Gallart-Jornet, L., Andrés, A., Akse, L., Carlehög M., & Skjerdal, O. T.  
402 (2006). Influence of cod freshness on the salting drying and desalting stages.  
403 *Journal of Food Engineering*, *73*, 9-19.
- 404 Barat, J. M., Rodriguez-Barona, S., Andrés, A., & Fito, P. (2003). Cod salting  
405 manufacturing analysis. *Food Research International*, *36*, 447-453.
- 406 Cardinal, M., Gunnlaugsdottir, H., Bjoernevik, M., Ouisse, A., Vallet, J. L., & Leroi, F.  
407 (2004). Sensory characteristics of cold-smoked Atlantic salmon (*Salmo salar*) from  
408 European market and relationships with chemical physical and microbiological  
409 measurements. *Food Research International*, *37*, 181–193
- 410 Chirife, J., & Resnik, S. L. (1984). Unsaturated solutions of sodium chloride as  
411 reference sources of water activity at various temperatures. *Journal of Food Science*,  
412 *49*, 1486–1488.
- 413 Cornu, M., Beaufort, A., Rudelle, S., Laloux, L., Bergis, H., Miconnet, N., Serot, T. &  
414 Delignette-Muller, M. L. (2006). Effect of temperature, water-phase salt and  
415 phenolic contents on *Listeria monocytogenes* growth rates on cold-smoked salmon  
416 and evaluation of secondary models. *International Journal of Food Microbiology*,  
417 *106*, 159-168.
- 418 Espe, M., Kiessling, A., Lunestad, B., Torrissen, O. J., & Røra, A. M. B. (2004).  
419 Quality of cold smoked collected in one French hypermarket during a period of 1  
420 year. *LWT-Food Science and Technology* *37*, 627-638.
- 421 Fuentes, A. (2007). Desarrollo de productos ahumados a partir de lubina.  
422 (*Dicentrarchus labrax* L.) Tesis doctoral. *Universidad Politécnica de Valencia*

- 423 Fuentes, A., Barat, J. M., Fernández-Segovia, I., & Serra, J. A. (2008). Study of sea bass  
424 (*Dicentrarchus labrax* L.) salting process: Kinetic and thermodynamic control. *Food*  
425 *Control*, *19*, 757-763.
- 426 Fuentes, A., Fernández-Segovia, I., Barat J. M., & Serra J. A. (2010a). Physicochemical  
427 characterization of some smoked and marinated fish products. *Journal of Food*  
428 *Processing and Preservation*, *34*, 83-103
- 429 Fuentes, A., Fernández-Segovia, I., Barat, J. M., & Serra, J. A. (2010b). Influence of  
430 sodium replacement and packaging on quality and shelf life of smoked sea bass  
431 (*Dicentrarchus labrax* L.). *LWT-Food Science and Technology*, *43*, 1426-1433
- 432 Fuentes, A., Fernández-Segovia, I., Masot, R., Alcañiz, M., & Barat, J. M. (2010c).  
433 Electronic sensor application in the detection of frozen-unfrozen fish. *International*  
434 *Conference on Food Innovation. Food Innova 2010*. Valencia, Spain.
- 435 Fuentes, A., Pérez, M. I., Fernández-Segovia, I., & Barat, J. M. (2011). Mejora en el  
436 proceso de ahumado de salmón. *VI Congreso Nacional de Ciencia y Tecnología de*  
437 *los Alimentos*. Valencia, Spain
- 438 Gallart-Jornet, L., Barat, J. M., Rustad, T., Erikson, U., Escriche, I., & Fito, P. (2007).  
439 Influence of brine concentration on Atlantic salmon fillet salting. *Journal of Food*  
440 *Engineering*, *80*(1), 267-275.
- 441 Guerrero, L., Gobantes, I., Oliver, I., Arnau, J., Guardia, M. D., Elvira, J., Riu, P.,  
442 Grèbol, N., & Monfort, J. M. (2004). Green hams electrical impedance spectroscopy  
443 (EIS) measures and pastiness prediction of dry cured hams. *Meat Science*, *66*, 289–  
444 294.
- 445 Ismail, N., & Wootton, M. (1982). Fish salting and drying: a review. *ASEAN Food*  
446 *Journal*, *7*(4), 175-183.

- 447 Karásková, P., Fuentes, A., Fernández-Segovia, I., Alcañiz, M., Masot, R., & Barat, J.  
448 M. (2011). Development of a low-cost non-destructive system for measuring  
449 moisture and salt content in smoked fish products. ICEF 2011. *Procedia – Food*  
450 *Science. I*: 1195-1201
- 451 Leroi, F., & Joffraud, J. J. (2000). Salt and smoke simultaneously affect chemical and  
452 sensory quality of cold-smoked salmon during 5 °C storage predicted using factorial  
453 design. *Journal of Food Protection*, 63(9), 1222-1227.
- 454 Masot, R. (2010). Desarrollo de un sistema de medida basado en espectroscopía de  
455 impedancia para la determinación de parámetros fisicoquímicos en alimentos. Tesis  
456 doctoral. *Universidad Politécnica de Valencia*.
- 457 Masot, R., Alcañiz, M., Fuentes, A., Schmidt, F. C., Barat, J. M., Gil, L., Baigts, D.,  
458 Martínez-Máñez, R., & Soto, J. (2010). Design of a low-cost non-destructive system  
459 for punctual measurements of salt levels in food products using impedance  
460 spectroscopy. *Sensors and Actuators A*, 158, 217–223.
- 461

462 **Figure Captions (black and white)**

463

464 **Fig. 1.** Experimental design of Phase I.

465

466 **Fig. 2:** Principal component analysis (PCA) performed on the impedance spectroscopy  
 467 measurements of samples in air and in vacuum with different smoke flavoring salt  
 468 dosages (0 (□), 4 (▼), 6 (●), 8 (★) and 16 (■) g SF /100 g) for 24 h. (a) Double  
 469 electrode, (b) Arrowhead electrode and (c) Needle electrode.

470

471 **Fig. 3.** NaCl weight fraction ( $x^{\text{NaCl}}$  (◆),  $y = 0.005x + 0.014$ ,  $R^2 = 0.933$ ), NaCl  
 472 concentration on a dry basis ( $X^{\text{NaCl}}$  (■),  $y = 0.012x + 0.053$ ,  $R^2 = 0.875$ ) and NaCl  
 473 weight fraction in the liquid phase of salmon ( $Z^{\text{NaCl}}$  (▲),  $y = 0.009x + 0.019$ ,  $R^2 =$   
 474  $0.951$ ) *versus* square root of the processing time ( $t^{0.5}$ ).

475

476 **Fig. 4.** Total weight changes ( $\Delta M_t^0$  (◆),  $y = -0.014x - 0.038$ ,  $R^2 = 0.974$ ), water weight  
 477 changes ( $\Delta M_t^w$  (■),  $y = -0.021x - 0.046$ ) ( $R^2 = 0.987$ ) and sodium chloride weight  
 478 changes ( $\Delta M_t^{\text{NaCl}}$  (▲),  $y = 0.005x + 0.012$ ) ( $R^2 = 0.915$ ) *versus* the square root of the  
 479 processing time ( $t^{0.5}$ ).

480

481 **Fig. 5.** Principal components analysis (PCA) performed with the impedance  
 482 spectroscopy measurements (needle electrode) of salmon submitted to the salting-  
 483 smoking process (16% smoke flavoring salt dosage, vacuum packaging) for different  
 484 processing times (0 (□), 5 (▼), 10 (●), 15 (★), 20 (■) and 25 (Δ) h).

485

486 **Fig. 6.** Predicted *versus* experimental values by the PLS statistical model (—) and ideal  
487 behaviour (---). (a) NaCl, (b)  $a_w$  and (c) moisture.

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488 **Figure Captions (color)**

489

490 **Fig. 1.** Experimental design of Phase I.

491

492 **Fig. 2:** Principal component analysis (PCA) performed on the impedance spectroscopy  
 493 measurements of samples in air and in vacuum with different smoke flavoring salt  
 494 dosages (0 ( $\square$ ), 4 ( $\blacktriangledown$ ), 6 ( $\bullet$ ), 8 ( $\star$ ) and 16 ( $\blacksquare$ ) g SF /100 g) for 24 h. (a) Double  
 495 electrode, (b) Arrowhead electrode and (c) Needle electrode.

496

497 **Fig. 3.** NaCl weight fraction ( $x^{\text{NaCl}}$  ( $\diamond$ ),  $y = 0.005x + 0.014$ ,  $R^2 = 0.933$ ), NaCl  
 498 concentration on a dry basis ( $X^{\text{NaCl}}$  ( $\blacksquare$ ),  $y = 0.012x + 0.053$ ,  $R^2 = 0.875$ ) and NaCl  
 499 weight fraction in the liquid phase of salmon ( $Z^{\text{NaCl}}$  ( $\blacktriangle$ ),  $y = 0.009x + 0.019$ ,  $R^2 =$   
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501

502 **Fig. 4.** Total weight changes ( $\Delta M_t^0$  ( $\diamond$ ),  $y = -0.014x - 0.038$ ,  $R^2 = 0.974$ ), water weight  
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506

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 510 processing times (0 ( $\square$ ), 5 ( $\blacktriangledown$ ), 10 ( $\bullet$ ), 15 ( $\star$ ), 20 ( $\blacksquare$ ) and 25 ( $\Delta$ ) h).

511

512 **Fig. 6.** Predicted *versus* experimental values by the PLS statistical model (—) and ideal  
513 behaviour (---). (a) NaCl (b)  $a_w$  and (c) moisture.

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514 **Table Captions**

515

516 **Table 1**

517 Physico-chemical parameters of raw material (S=0, t=0) and salmon submitted to  
518 salting-smoking with different smoke flavoring (SF) salt doses (S) (g SF salt/100 g fresh  
519 salmon), types of packaging (P) and processing times (t). Mean values  $\pm$  SD (n=2).

520

521 **Table 2**

522 Physico-chemical parameters of raw material (t=0) and salmon submitted to salting-  
523 smoking process (16% smoke flavoring salt dosage, vacuum packaging) for different  
524 processing times. Mean values  $\pm$  SD (n=6).

525

- A new salting-smoking method for salmon using vacuum packaging was developed
- Impedance spectroscopy was studied in the monitoring of the smoking process
- Vacuum packaging speeded up the process of NaCl absorption and salmon dehydration
- The needle electrode was the most appropriate for smoking process monitoring
- Impedance spectroscopy was a rapid monitoring method for the salmon smoking process

1 **Table 1**

2 Physico-chemical parameters of raw material (S=0, t=0) and salmon submitted to  
 3 salting-smoking with different smoke flavoring (SF) salt doses (S) (g SF salt/100 g fresh  
 4 salmon), types of packaging (P) and processing times (t). Mean values  $\pm$  SD (n=2).

5

<b>S</b>	<b>P</b>	<b>t (h)</b>	<b>Moisture (g H<sub>2</sub>O/100g)</b>	<b>pH</b>	<b>a<sub>w</sub></b>	<b>NaCl (g NaCl/100g)</b>
<b>0</b>		<b>0</b>	70.39 $\pm$ 1.47	6.13 $\pm$ 0.02	0.992 $\pm$ 0.002	0.00
		<b>12</b>	67.34 $\pm$ 0.13	6.10 $\pm$ 0.01	0.980 $\pm$ 0.000	1.84 $\pm$ 0.09
		<b>18</b>	65.91 $\pm$ 0.18	6.08 $\pm$ 0.08	0.978 $\pm$ 0.000	1.71 $\pm$ 0.07
<b>4</b>	<b>Air</b>	<b>24</b>	67.81 $\pm$ 0.24	6.07 $\pm$ 0.06	0.972 $\pm$ 0.002	2.03 $\pm$ 0.04
		<b>12</b>	67.03 $\pm$ 0.12	6.09 $\pm$ 0.04	0.979 $\pm$ 0.001	1.78 $\pm$ 0.05
		<b>18</b>	67.13 $\pm$ 0.29	6.09 $\pm$ 0.02	0.980 $\pm$ 0.001	1.79 $\pm$ 0.01
	<b>Vacuum</b>	<b>24</b>	68.03 $\pm$ 0.01	6.09 $\pm$ 0.03	0.980 $\pm$ 0.000	1.68 $\pm$ 0.01
		<b>12</b>	66.11 $\pm$ 0.21	6.12 $\pm$ 0.02	0.978 $\pm$ 0.001	1.77 $\pm$ 0.06
		<b>18</b>	65.13 $\pm$ 0.30	6.10 $\pm$ 0.03	0.977 $\pm$ 0.000	2.04 $\pm$ 0.01
<b>6</b>	<b>Air</b>	<b>24</b>	65.70 $\pm$ 0.17	5.99 $\pm$ 0.02	0.978 $\pm$ 0.001	2.54 $\pm$ 0.08
		<b>12</b>	65.52 $\pm$ 0.02	6.08 $\pm$ 0.04	0.978 $\pm$ 0.001	2.02 $\pm$ 0.02
		<b>18</b>	66.95 $\pm$ 0.18	6.15 $\pm$ 0.05	0.982 $\pm$ 0.001	1.67 $\pm$ 0.00
	<b>Vacuum</b>	<b>24</b>	67.23 $\pm$ 0.80	6.06 $\pm$ 0.05	0.976 $\pm$ 0.000	2.01 $\pm$ 0.02
		<b>12</b>	65.65 $\pm$ 0.20	6.14 $\pm$ 0.04	0.977 $\pm$ 0.000	2.16 $\pm$ 0.10
		<b>18</b>	65.52 $\pm$ 1.22	6.08 $\pm$ 0.02	0.976 $\pm$ 0.000	2.14 $\pm$ 0.05
<b>8</b>	<b>Air</b>	<b>24</b>	64.69 $\pm$ 0.35	6.10 $\pm$ 0.04	0.971 $\pm$ 0.000	2.58 $\pm$ 0.03
		<b>12</b>	65.15 $\pm$ 0.28	6.13 $\pm$ 0.03	0.978 $\pm$ 0.001	1.98 $\pm$ 0.09
		<b>18</b>	65.57 $\pm$ 0.17	6.16 $\pm$ 0.05	0.977 $\pm$ 0.000	2.13 $\pm$ 0.16
	<b>Vacuum</b>	<b>24</b>	64.20 $\pm$ 0.03	6.05 $\pm$ 0.02	0.965 $\pm$ 0.001	3.01 $\pm$ 0.00
		<b>12</b>	62.68 $\pm$ 0.28	6.15 $\pm$ 0.06	0.976 $\pm$ 0.002	1.79 $\pm$ 0.05
		<b>18</b>	62.34 $\pm$ 0.88	6.17 $\pm$ 0.02	0.978 $\pm$ 0.000	1.90 $\pm$ 0.14
<b>16</b>	<b>Air</b>	<b>24</b>	59.91 $\pm$ 0.06	5.99 $\pm$ 0.05	0.968 $\pm$ 0.001	3.40 $\pm$ 0.10
		<b>12</b>	60.53 $\pm$ 0.67	6.12 $\pm$ 0.06	0.969 $\pm$ 0.002	2.35 $\pm$ 0.22
		<b>18</b>	60.99 $\pm$ 0.26	6.01 $\pm$ 0.03	0.967 $\pm$ 0.000	3.12 $\pm$ 0.25
	<b>Vacuum</b>	<b>24</b>	62.45 $\pm$ 0.39	5.96 $\pm$ 0.01	0.963 $\pm$ 0.002	3.62 $\pm$ 0.02

6

1 **Table 2**

2 Physico-chemical parameters of raw material (t=0) and salmon submitted to salting-  
 3 smoking process (16% smoke flavoring salt dosage, vacuum packaging) for different  
 4 processing times. Mean values  $\pm$  SD (n=6).

5

<b>t (h)</b>	<b>Moisture (g H<sub>2</sub>O/100g)</b>	<b>pH</b>	<b>a<sub>w</sub></b>
<b>0</b>	70.97 $\pm$ 2.64 <sup>a</sup>	6.08 $\pm$ 0.04 <sup>ab</sup>	0.991 $\pm$ 0.002 <sup>a</sup>
<b>5</b>	66.45 $\pm$ 2.56 <sup>b</sup>	6.13 $\pm$ 0.04 <sup>b</sup>	0.972 $\pm$ 0.002 <sup>b</sup>
<b>10</b>	64.90 $\pm$ 1.94 <sup>bc</sup>	6.11 $\pm$ 0.07 <sup>b</sup>	0.968 $\pm$ 0.001 <sup>b</sup>
<b>15</b>	63.91 $\pm$ 2.32 <sup>bc</sup>	6.10 $\pm$ 0.07 <sup>b</sup>	0.963 $\pm$ 0.004 <sup>c</sup>
<b>20</b>	63.85 $\pm$ 2.84 <sup>cb</sup>	6.08 $\pm$ 0.02 <sup>ab</sup>	0.957 $\pm$ 0.008 <sup>d</sup>
<b>25</b>	62.42 $\pm$ 2.30 <sup>c</sup>	6.03 $\pm$ 0.06 <sup>a</sup>	0.957 $\pm$ 0.002 <sup>d</sup>
<b><math>\alpha</math></b>	***	ns	***

6 Same letters in the same column indicate homogeneous group membership.

7 Significance level ( $\alpha$ ): ns no significant difference; \*\*\* p < 0.001

8

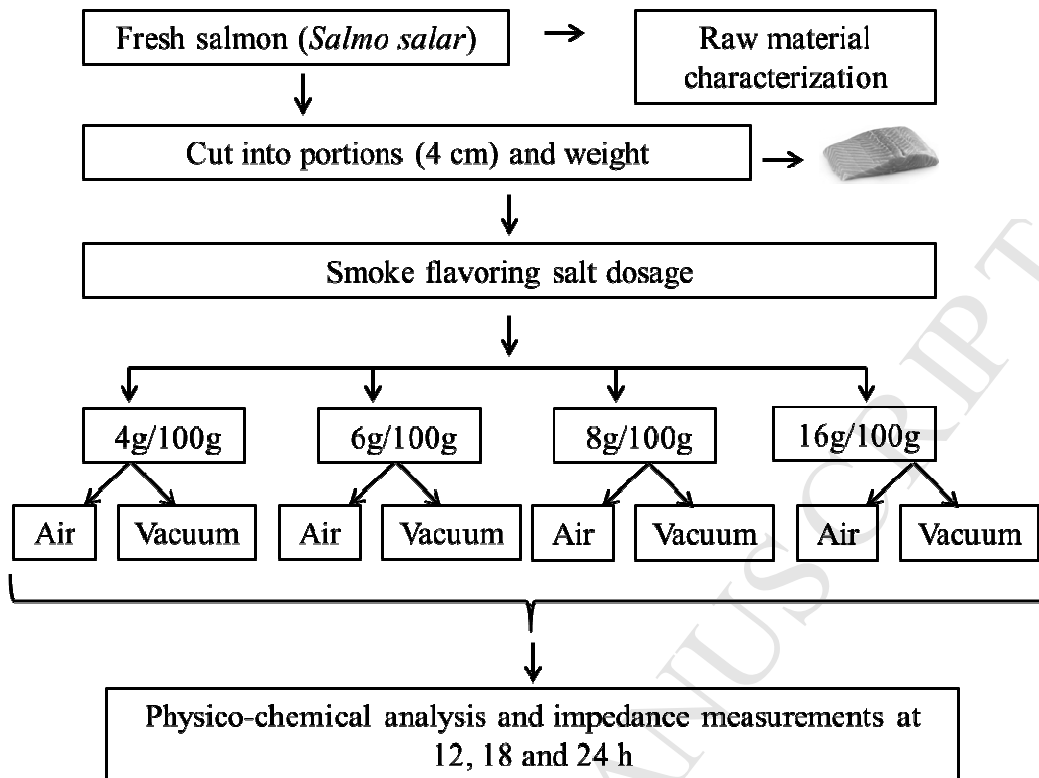


Fig. 1

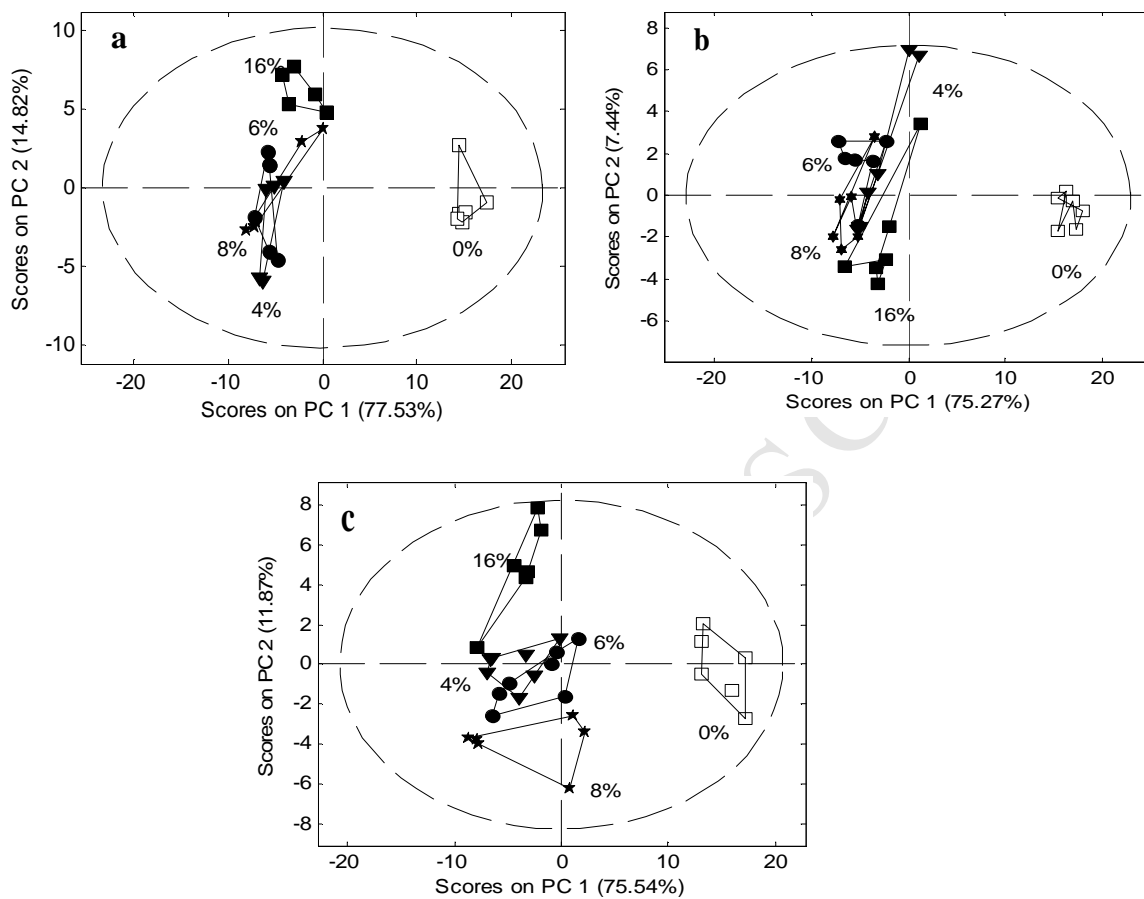


Fig. 2



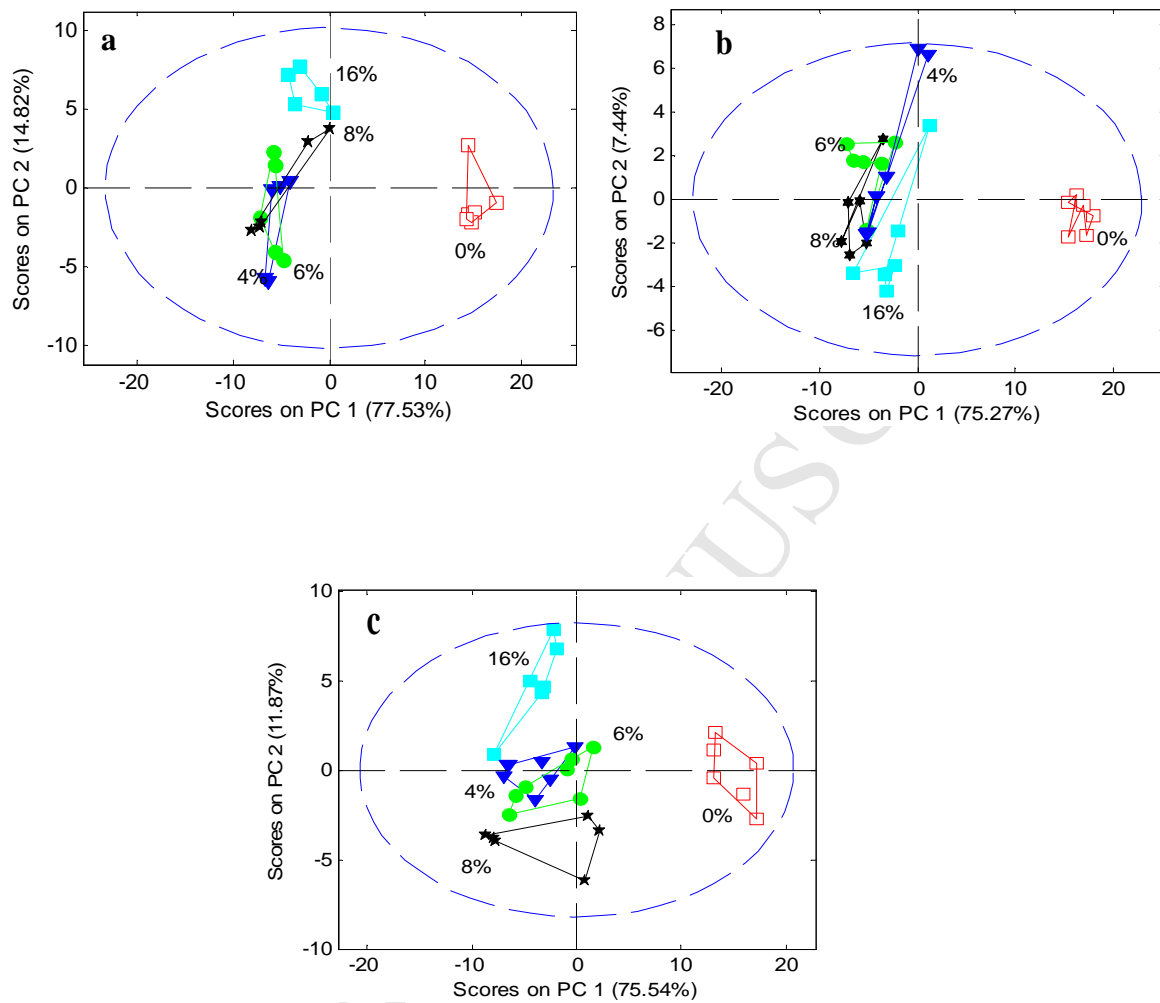
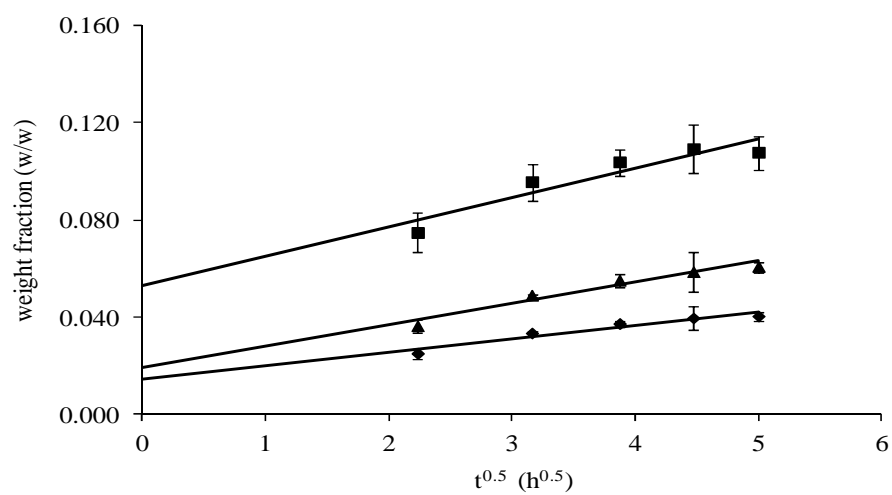
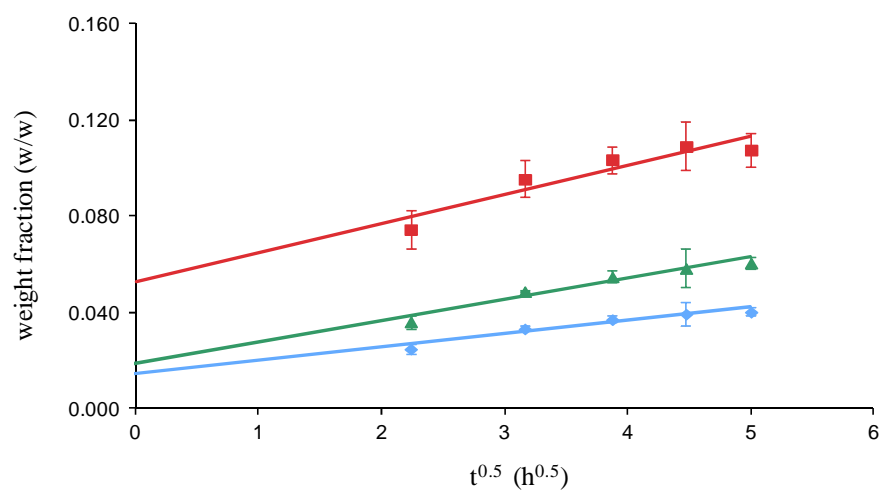


Fig. 2

**Fig. 3**

**Fig. 3**

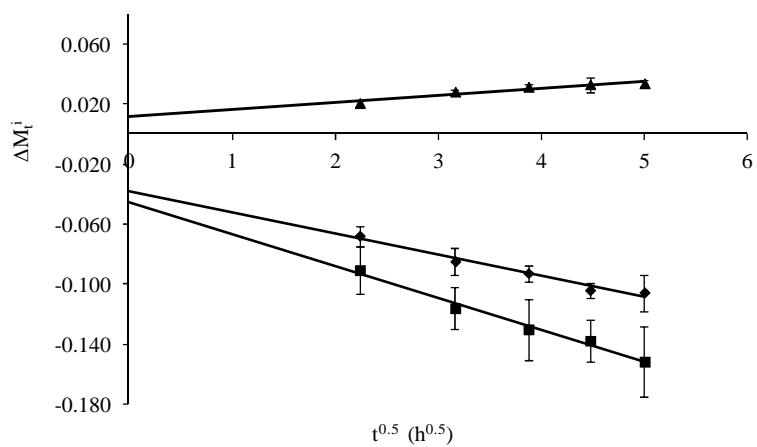
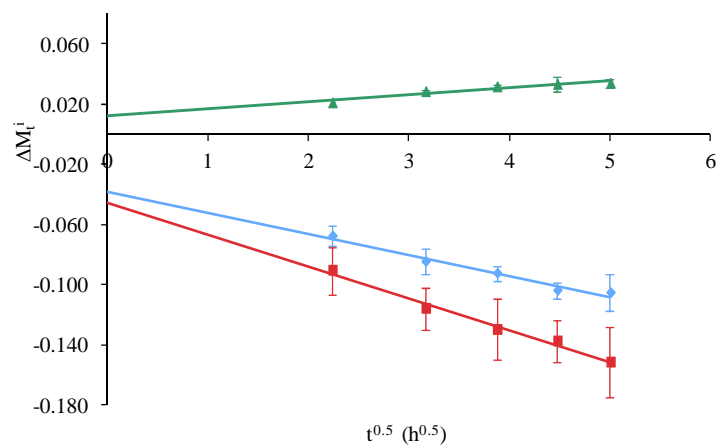
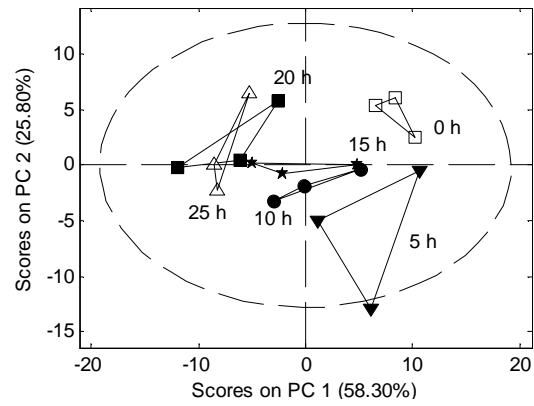


Fig. 4

**Fig. 4**

**Fig. 5**

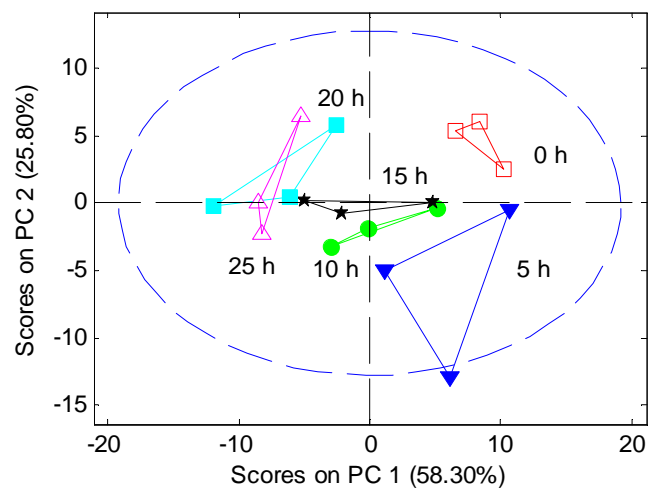
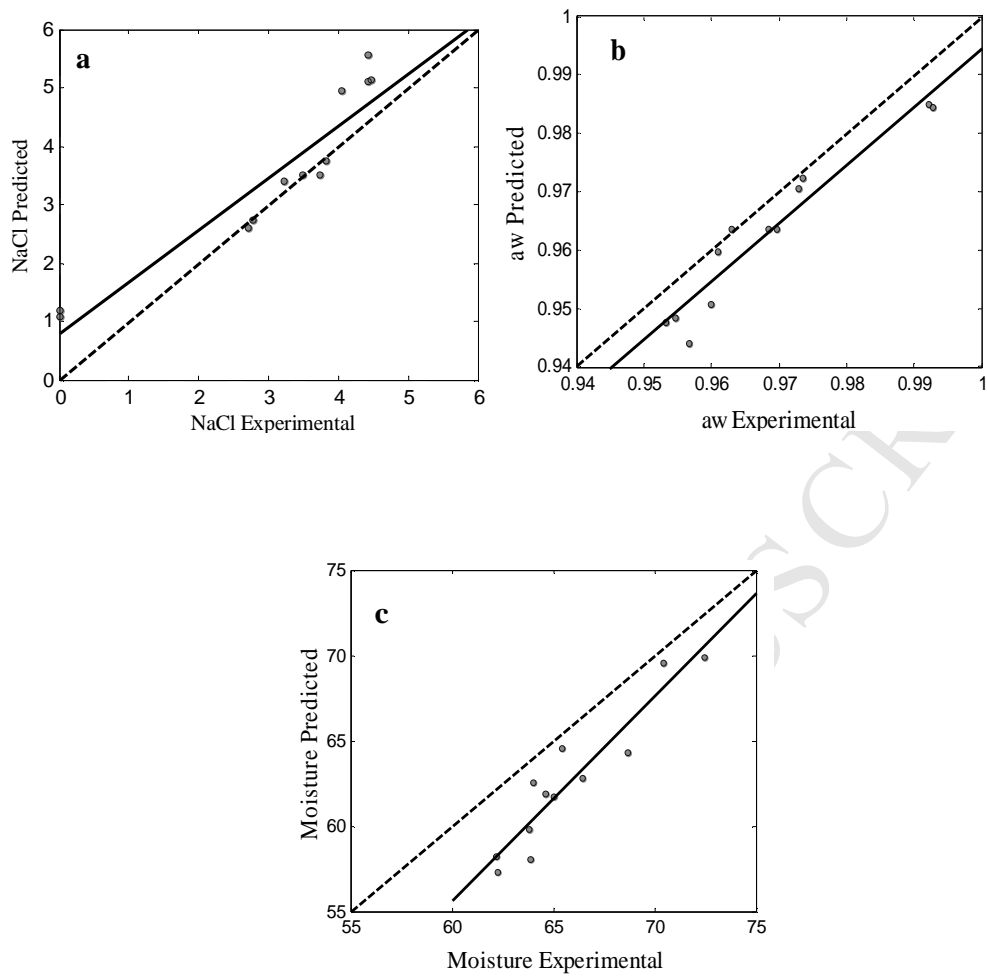
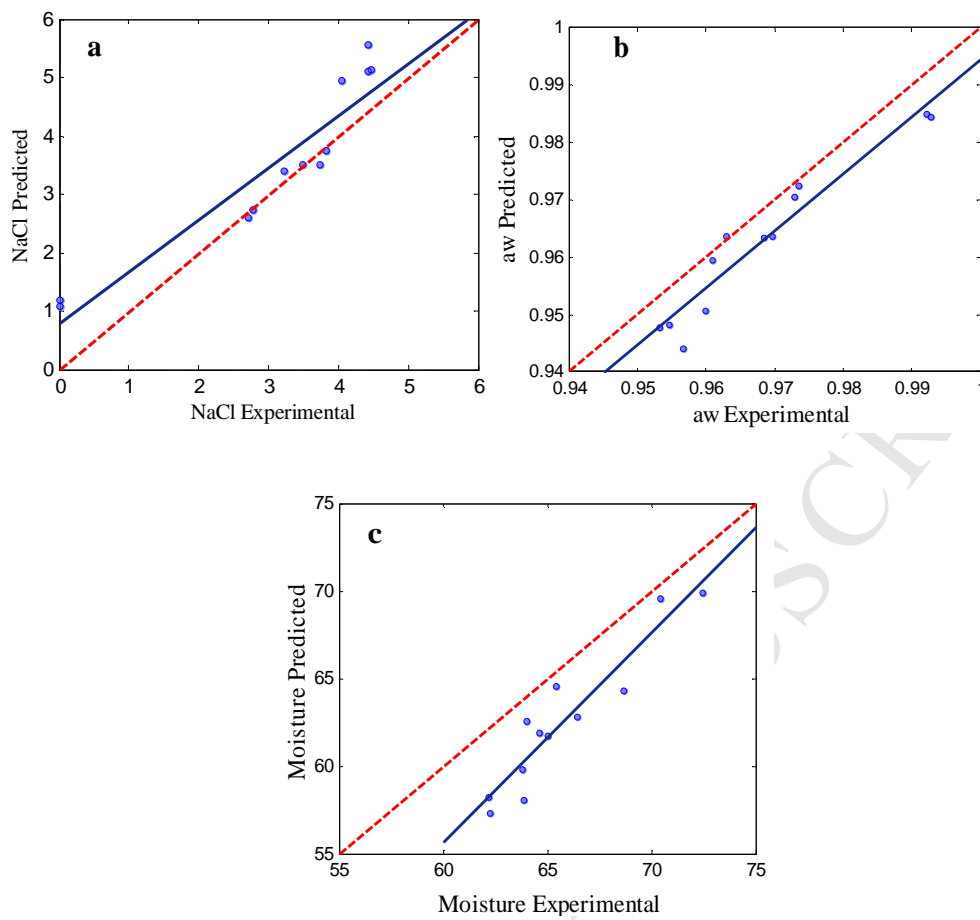


Fig. 5

**Fig. 6**



**Fig. 6**