



## Research article

Effect of salting procedures on quality of hake (*Merluccius hubbsi*) filletsMarion Daniela Marchetti<sup>a,b,\*</sup>, Paula Luisina Gomez<sup>b,c</sup>, María Isabel Yeannes<sup>a,b</sup>,  
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## ABSTRACT

The influence of salting procedures on the proximate analysis, mechanical parameters, and color of hake (*Merluccius hubbsi*) was investigated. Three procedures were comparatively evaluated: dry salting (DS), mixed salting (MS) and brining (BS). MS samples had the highest fat content, a considerable protein content and an equilibrium salt content similar to BS. MS samples had a great water loss, as DS method, but hardness and other mechanical parameters were similar to that obtained with BS, i.e. significantly lower than DS. All samples showed color parameters significantly different as compared to fresh hake, turning more red-orange as the salting time increased. Lightness diminished,  $a^*$  values increased and  $b^*$  values did not show a clear trend throughout the salting time. Principal component analysis (PCA) described the relationship between some variables ( $Z_{NaCl}$ , color, and mechanical parameters) with salting time. High Pearson's correlation coefficients were found between  $Z_{NaCl}$  and hardness, springiness, cohesiveness and  $a^*$  parameter ( $r = 0.76$ ,  $p < 0.001$ ;  $r = 0.93$ ,  $p < 0.0001$ ;  $r = 0.95$ ,  $p < 0.001$  and  $r = 0.93$ ,  $p < 0.0001$ , respectively). Luminosity was negatively correlated with  $Z_{NaCl}$  ( $r = -0.87$ ,  $p = 0.0001$ ). The correlation curves showed nonlinear relationships ( $R^2_{adj}$  between 83.7 % and 97.4 %), which could be used to predict quality attributes of hake fillets as a function of salting time. This work contributed to know the effect of different salting procedures on the quality attributes of a species widely available in the Southwest Atlantic Ocean.

## 1. Introduction

Species of *Merluccius* (*Merlucciidae*), known as hake and/or whiting, inhabit the Atlantic continental platform, southwestern Indian, eastern Pacific and New Zealand (Nelson et al., 2016). They are caught with bottom trawls and constitute one of the most important exploited demersal fish. *Merluccius hubbsi* is a dominant specie in Argentina and two populations have been identified: a northern population between 22 °S and 41 °S, and a southern or Patagonian population between 41 °S and 55 °S (Belleggia et al., 2019). Although Argentina is a pioneer country in the exploitation of *M. hubbsi*, it exports the 90 % of its fishery production in value (INDEC, 2020).

Since 10 years ago, seafood is accepted as an essential food for humans (FAO, 2010) due to its high biological value proteins, polyunsaturated fatty acids (PUFA), principally omega-3 (i.e. docosahexaenoic and eicosapentaenoic acids), and other nutrients, such as minerals,

trace elements, and vitamins (FAO, 2010). These nutrients are essential for human body functions and the development of the brain and nervous system (Hamed et al., 2015). They also have anticancer properties (Hamed et al., 2015), anti-inflammatory action (Calder, 2017), reduce risk of cardiovascular disease (Tomdino et al., 2019), play an essential role in prevention of neurodegenerative diseases (Kerdiles et al., 2017), among others. Myotome is one of most important constituent of fish muscle structure; it is formed by a package of individual fibers linked by intramuscular connective tissue called myocommata (Bao et al., 2019). Collagen is the most important connective tissue in fish muscle, representing between 3 and 10 % of the protein. This plays a vital role in maintaining fillet integrity and muscle cohesiveness (Varghese and Mathew, 2017).

Salting is an ancient practice used by many cultures to preserve food, in particular meat and fish (Gagaoua and Boudechicha, 2018). Nowadays, despite the emergence of other preservation methods, as

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refrigeration or freezing, salting fish remains popular because this process generate good sensory properties (color, taste, aroma and texture) (Crotova et al., 2021; Fan et al., 2014; Martínez-Alvarez and Gómez-Guillén, 2013). There are three salting procedures: brining, dry salting and a combination of both procedures (Marchetti et al., 2020).

The salting process causes changes on the seafood chemical composition associated with physicochemical and structural modifications (Jiang et al., 2019a). Oxidation of unsaturated highly lipids is accelerated (Jónsdóttir et al., 2011) and mineral content is modified because salting involves osmotic concentration. Structure is altered since proteins suffer denaturation, being myosin heavy chain more susceptible to denaturing by strong salting. Salting causes a series of changes in the tissue components and protein properties which are influenced by salting condition, that is, salting in and salting out effects (Jiang et al., 2019a, 2019b).

The evaluation of different quality parameters, such as texture and color, are frequently employed to examine and evaluate fish quality; these attributes are key to the new products development or process variables optimization. Currently, the evaluation of texture and color can be performed by different sensory and instrumental tests. Sensory analyses is the best method to evaluate all parameters involving in texture but this is difficult to apply on large sample series. For this reason, instrumental measurements have been used for a long time to evaluate texture and color. Double-compression test allows to obtain the mechanical parameters (hardness, springiness, cohesiveness, adhesiveness, gumminess and chewiness) applying the texture profile analysis (TPA) (Bourne, 2002). In fish, hardness, springiness, and cohesiveness are the most important mechanical parameters (Cheng et al., 2014).

Many studies in the literature have been focused on the salting process in cod (*Gadus morhua*) (Heredia et al., 2007; Oliveira et al., 2012; Nguyen et al., 2012; Martínez-Alvarez and Gómez-Guillén, 2013; Bjørkevoll et al., 2014). However, until our knowledge, only a small number of articles discuss the salting process in a species similar to cod available in the Southwest Atlantic Ocean (Marchetti et al., 2020). Practically, there are not reported studies on the physicochemical, rheological and color changes caused by salting in *M. hubbsi*. In this context, this study was aimed to investigate the effect of salting on Proximate Chemical Composition, mechanical parameters and color of hake using three salting procedures: brining, dry salting and mixed salting. In addition, this study assessed and validated the relationships between salt concentration (referred to liquid phase) and some fish quality attributes.

## 2. Materials and methods

### 2.1. Sample preparation and salting procedures

The analyzed hake (*M. hubbsi*) specimens were collected in the Southwestern Atlantic Ocean (41–50 °S), in February 2018. Whole hake specimens were kept on ice from capture to arrival at the processing plant (Mar del Plata, Argentina). Two fillets were obtained from each sample by longitudinal cuts and stored at  $2 \pm 1$  °C for 2–3 h until processing. Skinless fillets were manually cut into  $100 \times 50 \times 10$  mm pieces to perform salting procedures. The thickness was obtained by direct measurement with a Teclock dial micrometer model SM-124 ( $\pm 0.0001$  m, Japan). Brining (BS), mixed (MS) and dry salting (DS) methods were evaluated. The experiments were carried out at  $4 \pm 1$  °C (Barat et al., 2003). Fish brining was performed by immersing the samples into a NaCl saturated solution 26 % w/w (CELUSAL, Buenos Aires, Argentina) using a 1:10 fish-to-brine ratio. The brine/hake ratio was high enough to avoid any significant change in brine concentration during the salting experiments (Barat et al., 2003). DS was carried out by placing alternate layers of fish and salt (0.6 kg salt/kg fish) (Boeri et al., 1982). The liquid released from the tissue was drained out of the container. The MS fillets were also stacked between layers of salt (0.3 kg salt/kg fish), but the liquid extracted from the muscle due to the strong salting was retained in the container to form a saturated brine (Boeri et al., 1982). For all

treatments, hake fillets samples were taken at different times until equilibrium was reached (0, 2, 4, 6, 8, 10, 14, 24, 28, 34 and 48 h). DS and MS samples were immersed in saturated brine for 30 s to remove the excess of salt. Then, all samples were superficially dried with absorbent paper and preserved at  $4 \pm 1$  °C for further analysis. The whole experience was replicated once.

### 2.2. Physicochemical analysis

Samples were homogenized using a mixer (MOULINEX, Buenos Aires, Argentina) to determine different physicochemical analysis. Moisture content ( $x_w$ ) was measured by the gravimetric method at 100 °C up to constant weight (24 h) (AOAC, 1990 Sec 984.25). Ash content was analyzed by using a muffle furnace heated to 550 °C during 8 h (AOAC, 1993 Sec. 945.46). Chloride content ( $x_{NaCl}$ ) was determined in the ashes using the Mohr's method (Kirk et al., 1996). Salt concentration referred to liquid phase ( $z_{NaCl}$ ) was estimated from the water and sodium chloride content ( $x_w$  and  $x_{NaCl}$ , respectively) as shown in Eq. (1) (Barat et al., 2002).

$$z_{NaCl} = \frac{x_{NaCl}}{x_w + x_{NaCl}} \quad (1)$$

Total lipids content were evaluated by the acid hydrolysis method (AOAC, 1993 Sec 922.06). Total protein content was analyzed with the Kjeldahl method (AOAC, 1993 Sec. 920.152). Water activity ( $a_w$ ) was measured with an Aqua-Lab CX-2 (Decagon Devices Inc., Pullman, WA) water activity meter at 20 °C. Measurements of pH were carried out in the fish samples previously homogenized with water (ratio 1:1) using a pH-meter (ALTRONIX, model EZDO-PC, Saen SRL, Argentina). Each analysis was performed in triplicate, except the salt content that was determined in quadruplicate.

### 2.3. Texture Profile Analysis

The Texture Profile Analysis was performed using a TMS-Pro texturometer (Food Technology Corporation, Virginia, USA) using a 500 N load cell. A double compression at 70 % deformation was performed using a 25 mm diameter cylindrical probe with a crosshead speed of 0.001 m/s on hake cylinders (9 mm thickness and 16.5 mm diameter) (Tomac et al., 2020). The following mechanical parameters were obtained from the force vs. time curves using the Texture Expert software (Version 1.0, Stable Micro Systems Ltd.): Hardness (H), Springiness (S), Cohesiveness (C), Adhesiveness (A), Gumminess (G) and Chewiness (Ch) according to the definitions of Bourne (2002). The test was replicated at least 8 times for each sample and mean values for each parameter were calculated.

### 2.4. Color measurement

Color of fresh and salted hake was measured with a portable colorimeter (Lovibond, SP60, United Kingdom). Values were obtained for D65 illuminant and 10° observer (Tomac et al., 2020). Prior to testing, the instrument was calibrated with a black and white calibration standard. The  $L^*$ ,  $a^*$ ,  $b^*$  components of CIELAB space were recorded, where  $L^*$  indicates lightness or luminance,  $a^*$  indicates chromaticity on green (-) to red (+) axis, and  $b^*$  chromaticity on blue (-) to yellow (+) axis. These numerical values were converted into "chroma" (C) and "hue angle" (h) color functions using Eqs. (2) and (3), respectively (Vásquez-Mazo et al., 2019):

$$C = (a^{*2} + b^{*2})^{1/2} \quad (2)$$

$$h = \arctg \left( \frac{b^*}{a^*} \right) \quad (3)$$

Hue is an angle in a color wheel of 360°, and can be distributed in the four quadrants of the  $a^* b^*$  plane. Values are defined as followed: red-

**Table 1.** Proximate chemical composition, NaCl content, water activity and pH of fresh and salted hake fillets. FH: fresh hake, BS: brining, MS: mixed salting, DS: dry salting.

	FH	BS	MS	DS
Water g/g	0.803 ± 0.006 <sup>a</sup>	0.627 ± 0.001 <sup>b</sup>	0.555 ± 0.006 <sup>c</sup>	0.566 ± 0.019 <sup>c</sup>
Proteins g/g	0.163 ± 0.008 <sup>a,b</sup>	0.140 ± 0.008 <sup>a</sup>	0.178 ± 0.008 <sup>b,c</sup>	0.209 ± 0.027 <sup>c</sup>
Fat g/g	0.022 ± 0.002 <sup>a</sup>	0.051 ± 0.002 <sup>b</sup>	0.074 ± 0.015 <sup>c</sup>	0.023 ± 0.003 <sup>a</sup>
Ashes g/g	0.0105 ± 0.0008 <sup>a</sup>	0.180 ± 0.007 <sup>b</sup>	0.190 ± 0.001 <sup>c</sup>	0.195 ± 0.004 <sup>c</sup>
NaCl g/g	0.0018 ± 0.0002 <sup>a</sup>	0.175 ± 0.002 <sup>b</sup>	0.176 ± 0.003 <sup>b</sup>	0.187 ± 0.002 <sup>c</sup>
a <sub>w</sub>	0.996 ± 0.002 <sup>a</sup>	0.791 ± 0.009 <sup>b</sup>	0.754 ± 0.006 <sup>c</sup>	0.756 ± 0.005 <sup>c</sup>
pH	6.83 ± 0.05 <sup>a</sup>	6.11 ± 0.01 <sup>b</sup>	6.32 ± 0.01 <sup>c</sup>	6.17 ± 0.01 <sup>d</sup>

Different letters in the same row indicate that there were significant differences ( $p < 0.05$ ).

purple: 0, yellow: 90°, bluish-green: 180° and blue: 270°. Chroma is the attribute that represents the intensity or saturation of the hue, and it is neutral in the center and increases towards the outer edge of the color wheel. The measurements were replicated 5 times for each sample and mean values for each parameter were calculated.

### 2.5. Statistical analysis

Two-way analysis of variance (ANOVA) was used to establish the presence or absence of significant differences in proximate chemical composition and  $z_{\text{NaCl}}$  values of hake fillets, according to the factors “salting” and “time”. Significance level was set at  $p < 0.05$ . The main effects (i.e. individual effects of each factor separately) were examined in case of significant interactions between factors and multiple comparisons were performed using the Tukey test. Two-way multivariate analysis of variance (MANOVA) was used to detect significant differences in mechanical and color parameters, according to the factors “salting” and “time” ( $p < 0.05$ ). In case of finding significant differences, Hotelling corrected for Bonferroni test was performed. Principal analysis component (PCA) was used to illustrate the relationship among variables ( $z_{\text{NaCl}}$  values and the mechanical and color parameters) and salted samples at different times.

Pearson's correlation coefficients were calculated among all the studied variables and nonlinear regression models were analyzed to assess the instrumental parameter- $z_{\text{NaCl}}$  relationships. Models were internally validated to determine if could adequately describe the experimental data by means of ANOVA, the adjusted determination coefficient ( $R^2_{\text{adj}}$ ) and the Fisher test. In addition, they were externally validated with other  $z_{\text{NaCl}}$  values. Predicted instrumental values were compared to the instrumental measured ones by using the Student t-test. The Infostat v 2019 software (National University of Córdoba, Argentina) was used.

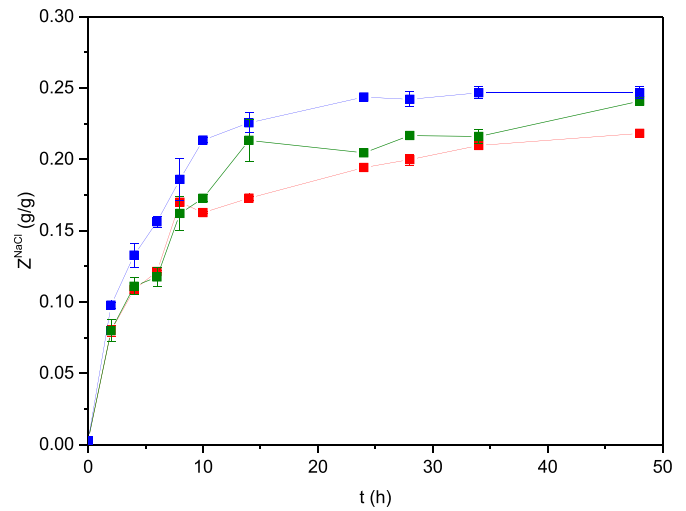
### 2.6. Ethical approval

Ethical approval is not applicable to this work, as dead fish were collected from the local processing plant and no endangered or protected fish species were involved. Specimen collection and maintenance were performed in strict accordance with the [United Kingdom Animals \(Scientific Procedures\) Act, \(1986\)](#).

## 3. Results and discussion

### 3.1. Characterization fresh and salted hake fillets

Proximate chemical composition, NaCl content, water activity and pH values of fresh and salted hake are shown in [Table 1](#). The fresh hake values are in accordance with the data previously obtained for fresh *M. hubbsi* fillets ([Marchetti et al., 2018, 2020](#)). The final  $x_w$  was significantly lower in DS and MS samples than in BS fillets. However, values of the equilibrium  $x_{\text{NaCl}}$  did not present significant differences



**Figure 1.** Changes in  $z_{\text{NaCl}}$  content during hake fillet salting. BS (Red), MS (Green) and DS (Blue).

between BS and MS salting procedures. In DS,  $x_{\text{NaCl}}$  was slightly higher than others salting procedures. The direct contact of salt crystals with hake tissue produces an important concentration difference between the outside and inside of the muscle, causing an increase in the dehydration and the salt penetration rate. Other authors reported the same behavior in cod ([Barat et al., 2002, 2004](#); [Andrés et al., 2002](#)) and chub ([Binici and Kaya, 2017](#)).

To analyze and compare the fat and protein content during salting procedures, these values were corrected by subtracting the  $x_w$  and  $x_{\text{NaCl}}$  from the total mass (i.e. only the protein and fat content were considered for the calculation basis, data not shown). Analyzing the protein content values, BS and MS samples presented a decrease compared to FH samples, whereas DS samples practically showed no changes. BS and MS cause a loss of proteins soluble in salt and water and non-protein nitrogen compounds towards the surrounding medium ([Szymczak, 2011](#)). Regarding the fat content, BS and MS samples showed an increase compared to FH samples, whereas DS samples remained practically without significant changes. Protein loss in MS and BS samples produces a concentration effect on the lipid content. On the other hand, the brine prevents contact of fillets with the air avoiding the lipid oxidation ([Collignan and Raoult-Wack, 1994](#)).

$A_w$  values decreased due to the effect of salting. DS and MS samples presented significantly lower  $a_w$  values compared to BS samples. Similarly, [Oliveira et al. \(2012\)](#) reported  $a_w$  values in dry salted cod in the range 0.70–0.75 and slightly higher values for brining procedure. Regardless of the applied procedure, the salting caused a slight decrease in pH values in all samples (BS, MS and DS). Similar values have been reported in other studies of salted cod, due to protein aggregation and muscle dehydration ([Lauritzen et al., 2004a, 2004b](#); [Martínez-Alvarez](#)

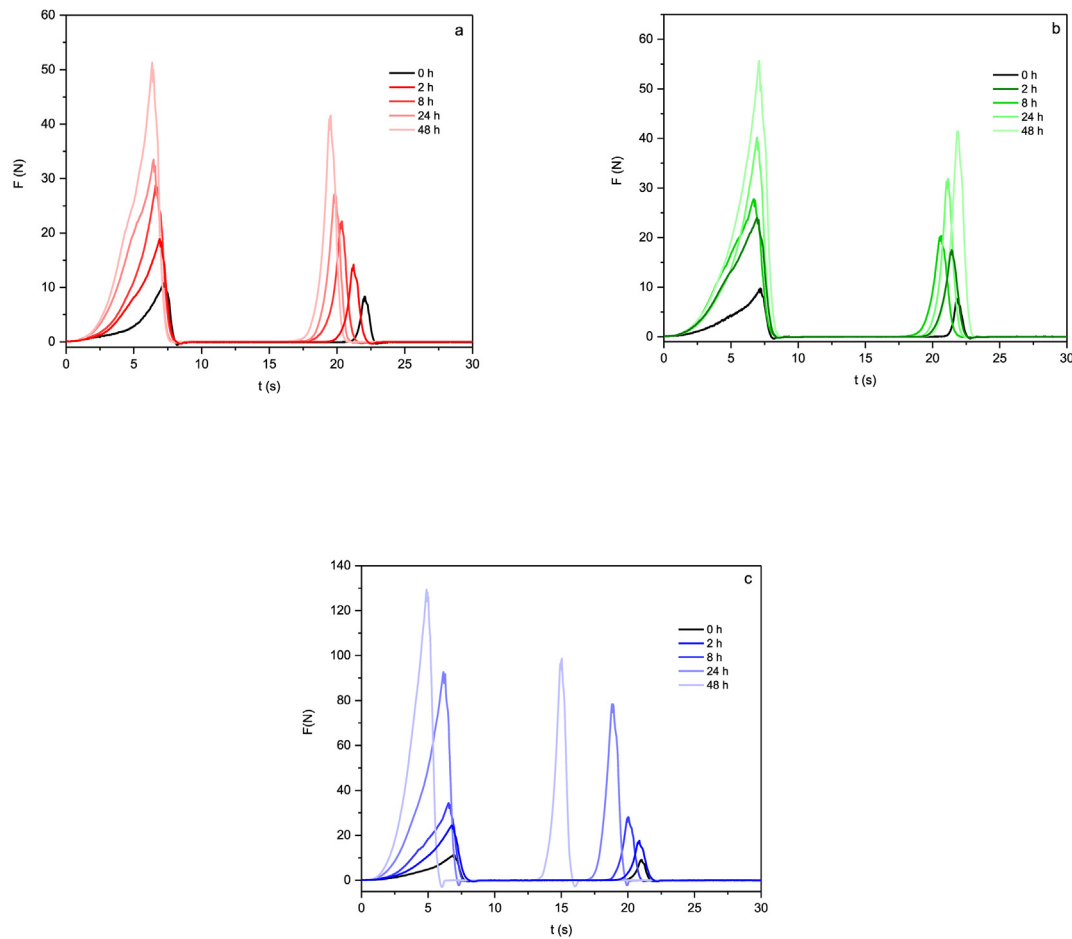


Figure 2. Typical double compression curves for salted hake fillets at 0, 2, 8, 24 and 48 h, a) BS, b) MS and c) DS.

and Gomez-Guillen, 2005). At pH < 6.5, the release of the heavy chains of actin and myosin are favored. On the other hand, the isoelectric point of proteins in fish muscle is approximately 5 (Martínez-Alvarez and Gomez-Guillen, 2005).

### 3.2. z<sub>NaCl</sub> changes throughout the salting procedures

The evolution of z<sub>NaCl</sub> for salted hake fillets with brining, mixed salting and dry salting are illustrated in Figure 1. For all salting

procedures, z<sub>NaCl</sub> variation showed an increase with salting time. The two-way variance analyses showed significant difference in the interaction between time and salting (p < 0.05). After 2 h, z<sub>NaCl</sub> in the DS samples was significantly higher compared to MS and BS samples. Non-significant differences were observed between MS and BS until 14 h of salting. There were not significant differences in z<sub>NaCl</sub> values between DS samples salted by 14 h and MS samples salted by 24 h; and between DS samples salted by 14 h and BS samples salted by 48 h. Analyzing the effect of salting over time, significant differences

Table 2. Average mechanical parameters corresponding to double compression curves of salted hake fillets at 0, 2, 8, 24 and 48 h. FH: Fresh hake, BS: brining, MS: mixed salting, DS: dry salting.

Salting	t (h)	H (N)	H <sub>2</sub> (N)	C (-)	S (-)	G (N)	Ch (J)	A (J)	
FH	0	10.60 ± 0.84	8.00 ± 0.78	0.112 ± 0.026	0.142 ± 0.016	1.18 ± 0.25	0.168 ± 0.041	0.43 ± 0.18	a
BS	2	17.94 ± 2.38	13.80 ± 1.85	0.181 ± 0.036	0.314 ± 0.056	3.28 ± 1.02	1.07 ± 0.52	0.22 ± 0.14	b
	8	27.12 ± 2.62	20.93 ± 1.99	0.216 ± 0.025	0.398 ± 0.040	5.86 ± 0.99	2.36 ± 0.61	0.153 ± 0.050	c,d
	24	36.29 ± 6.96	28.82 ± 5.72	0.219 ± 0.017	0.361 ± 0.055	7.98 ± 1.81	2.86 ± 0.71	0.171 ± 0.053	d,e,f
	48	50.31 ± 3.70	40.58 ± 2.90	0.280 ± 0.023	0.454 ± 0.033	14.09 ± 1.18	6.38 ± 0.49	0.070 ± 0.075	g
MS	2	23.16 ± 1.53	16.71 ± 0.91	0.150 ± 0.021	0.291 ± 0.033	3.48 ± 0.54	1.02 ± 0.23	0.213 ± 0.15	b,h
	8	28.94 ± 2.60	21.77 ± 1.82	0.189 ± 0.015	0.401 ± 0.038	5.48 ± 0.87	2.21 ± 0.47	0.155 ± 0.099	c
	24	40.03 ± 5.11	31.73 ± 4.48	0.215 ± 0.023	0.387 ± 0.089	8.66 ± 1.75	3.44 ± 1.30	0.32 ± 0.12	e
	48	56.03 ± 7.32	44.89 ± 7.51	0.263 ± 0.019	0.406 ± 0.051	13.79 ± 2.67	5.60 ± 1.30	0.082 ± 0.058	g
DS	2	22.51 ± 2.11	16.83 ± 0.98	0.140 ± 0.018	0.242 ± 0.030	3.16 ± 0.64	0.77 ± 0.22	0.47 ± 0.14	h
	8	31.95 ± 2.64	25.34 ± 2.51	0.194 ± 0.018	0.304 ± 0.024	6.20 ± 0.87	1.89 ± 0.37	0.257 ± 0.095	f
	24	89.18 ± 4.49	74.56 ± 3.52	0.259 ± 0.014	0.372 ± 0.035	23.14 ± 2.26	8.57 ± 0.87	0.49 ± 0.17	i
	48	121.23 ± 9.05	96.01 ± 7.04	0.285 ± 0.035	0.434 ± 0.032	34.64 ± 5.43	15.10 ± 3.00	0.51 ± 0.17	j

Rows identified with different letter showing differences with a 95 % confidence level.

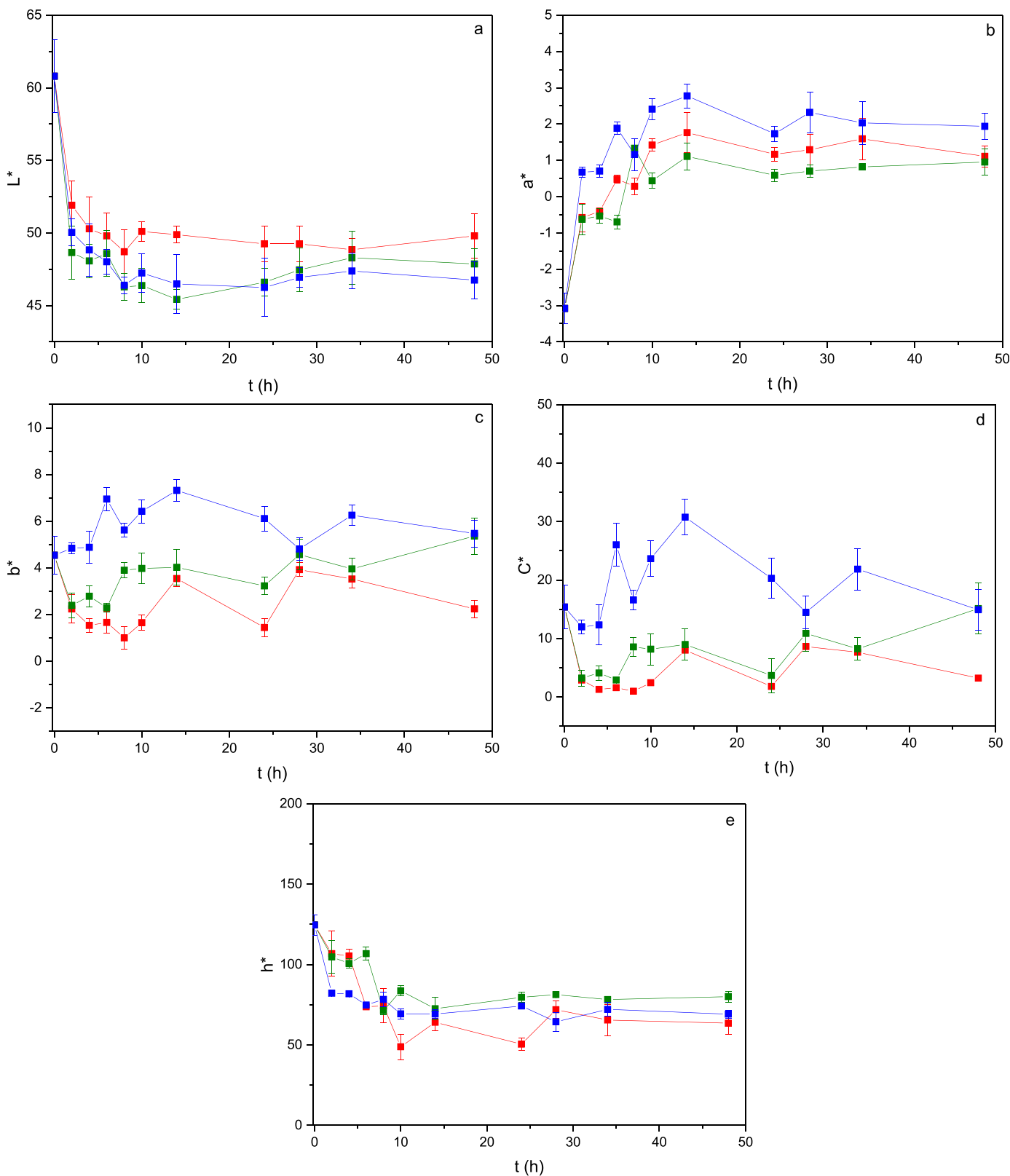
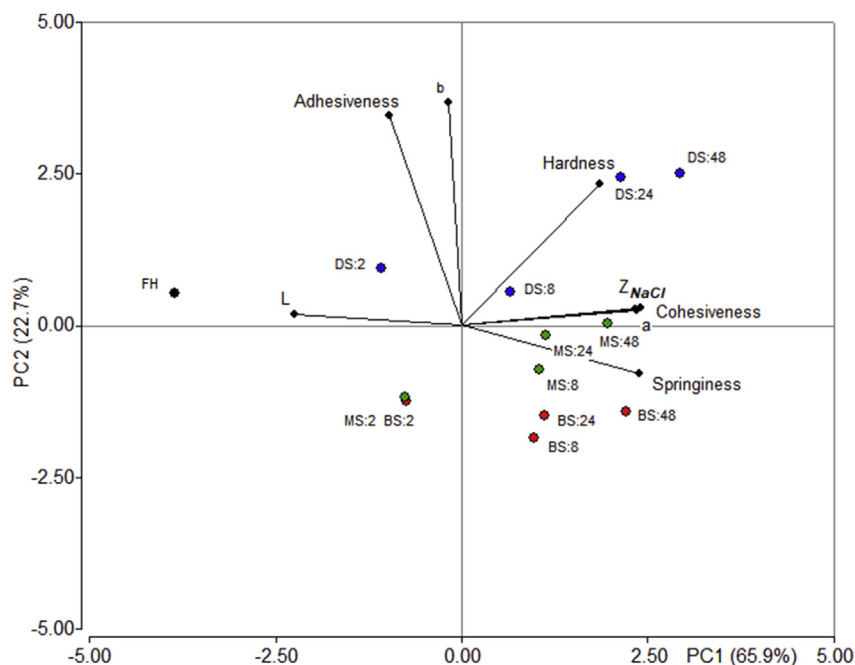


Figure 3. Changes in  $L^*$  (a),  $a^*$  (b),  $b^*$  (c),  $C^*$  (d) and  $h^*$  (e) parameters and functions during hake fillet salting. BS (Red), MS (Green) and DS (Blue).

were observed at  $t = 2$  h and 8 h for all samples. For short salting times ( $t < 14$  h), MS behavior was quite similar to that observed for BS samples. For  $t = 48$  h, non-significant differences in  $z_{NaCl}$  values were found between DS and MS samples; DS showed the highest  $z_{NaCl}$  values

and BS presented the least ones. The differences observed in  $z_{NaCl}$  values between the different salting could be attributed to the differences in the driving forces involved in the process. Barat et al. (2002 and 2003)



**Figure 4.** Principal Component Analysis (PCA) bi-plot of  $z_{NaCl}$  content, mechanical parameters and color corresponding to salted hake samples at different times. BS (Red), MS (Green) and DS (Blue). Salted times in hours: 0 (fresh hake), 2, 8, 24 and 48.

reported similar results at different brine concentrations during cod salting.

### 3.3. Texture Profile Analysis and color

Figure 2 shows the evolution of texture profiles with time for each salting procedure. At the beginning of the test, all samples showed a maximum peak associated with hardness of tissue, and a second peak associated with second compression. For longer salting times, texture profiles increased on the y axis, that is, showed an increase in the force and the area under the curve of both compressions. The slope of the curves of the first seconds of the first compression gradually increased with the salting time, reflecting an increase in hardness due to processing and a decrease in deformability. These changes were more abrupt in DS fillets (Figure 2c) than in BS and MS samples (Figure 2a and b, respectively).

Analyzing the effect of salting over time, for  $t = 2$  h, slight differences were observed in BS profiles compared to the DS and MS profiles (data not shown). In general, there were no differences between BS and MS profiles for  $t = 8, 24$  and  $48$  h. However, DS samples exhibited a slight increase in the compression force at  $t = 8$  h. This increase was more abrupt after 24 and 48 h of processing. Sigurgisladottir et al. (2000) and Birkeland et al. (2004) reported similar results on the texture of dry salted salmon. These changes were associated to the loss of water caused by the fast salt absorption at the beginning of the process. Proteins were rapidly denatured and aggregate due to extreme changes in salt and water content, losing water-holding capacity.

Table 2 shows the mechanical parameters obtained from fresh and salted hake fillets. MANOVA analysis found significant differences in the interaction between time and salting, indicating that the evolution of the mechanical parameters over time were dependent on salting procedures. Salting induced significant changes in the mechanical properties of hake fillets. All salted samples showed an increase in hardness, springiness, cohesiveness, gumminess and chewiness compared to fresh fillets as salting time increased. In addition, BS and MS samples did not show significant differences in the mechanical parameters throughout the salting process. In contrast, DS samples exhibited significant differences compared to the other salting methods for times 8, 24 and 48 h, which

were associated with more abrupt modifications in the mechanical properties. According to the results reported by other authors, the salting method influences the structural and mechanical properties of fish muscle (Andrés et al., 2002; Barat et al., 2002, 2003; Thorarinsdóttir et al., 2004).

Figure 3 shows the evolution of colorimetric parameters,  $L^*$ ,  $a^*$ ,  $b^*$ , and functions,  $C^*$  and  $h^*$ , during salting. All treated samples were significantly different compared to fresh hake samples. The two-way multivariate variance analyses showed significant difference in the interaction between time and salting ( $p < 0.05$ ), indicating that the evolution of color over time was dependent on salting procedures. In general, for the three procedures (BS, MS and DS), the lightness of hake slightly diminished during the first 8 h of salting, then it did not present larger variations for higher salting times (Figure 3a).  $a^*$  values presented the opposite trend over time, that is, they increased during the first hours of salting ( $\approx 8$  h) (Figure 3b). Instead,  $b^*$  values presented slight changes in relation to the others parameters ( $L^*$  and  $a^*$ ) and did not show a clear trend throughout salting.  $b^*$  increased slightly in the DS samples in some times and decreased slightly for the MS and BS samples (Figure 3c). At the end of salting, DS samples presented  $b^*$  values similar to the fresh hake, while MS and BS samples showed a decrease in  $b^*$  values, being more important the change amplitude in BS samples. Chroma values showed nearly the same pattern that was reported for  $b^*$  values (Figure 3d). In accordance with the pattern observed for  $a^*$  and  $b^*$  parameters,  $h^*$  values decreased after eight hours of salting and then remained relatively constant, indicating that hake color became more red-orange (Figure 3e). Analyzing the effect of time on salting methods, BS samples did not show significant differences in color parameters after 8 h of treatment. In contrast, DS y MS samples showed significant differences between times 8 and 48 h, and 24 and 48 h. Evaluating the salting methods at different times, for  $t = 2$  h, DS samples showed significant differences in color parameters compared to BS and MS samples; there were no significant differences between BS and MS samples. On the other hand, MS samples with 48 h of processing did not show significant differences in color parameters with DS samples processed 8 and 24 h.

The decrease in  $L^*$  observed in salted fillets could be due to the dehydration and protein denaturation during salting, causing opacity in

the tissue. This behavior has been observed previously in hake (Marchetti et al., 2018; Tomac et al., 2020), Atlantic salmon (Birkeland et al., 2004), and mackerel (Agustinelli, 2014). Moreover, an increase in  $b^*$  values due to the effect of brining was also reported in sea bass (Fuentes et al., 2012), cod (Oliveira et al., 2012) and hake (Marchetti et al., 2018), which was associated with the dehydration and lipid oxidation, causing the appearance of yellowish tones in fish tissue. In contrast, other authors (Corzo and Bracho, 2006; Agustinelli, 2014; Jiménez Lugo, 2017) reported a decrease in  $b^*$  values after BS using low salt concentrations and it was related to the diffusion from the tissue towards the solution of the remaining blood and other pigments. In addition, Marchetti et al. (2018) reported that BS at high salt concentrations caused an increase in  $a^*$  values in hake, while Agustinelli (2014) and Jiménez Lugo (2017) reported a decrease in  $a^*$  values during BS at low salt concentrations of mackerel and liza, respectively.

Figure 4 shows the Principal Component Analysis performed to explain the relationship between some variables ( $z_{\text{NaCl}}$ , color and mechanical parameters) and salted samples at different times (0, 2, 8, 24 and 48 h). The analysis indicated that principal component 1 (PC 1) explained 65.9 % of the variability of the data, and including principal component 2 (PC 2) was possible to explain 88.6 % of total variation. PC 1 was positively associated with  $x_{\text{NaCl}}$ , hardness, springiness, cohesiveness and  $a^*$  parameter and negatively correlated with  $L^*$ . PC 2 was positively represented by adhesiveness and  $b^*$  parameter. The variables hardness 2, gumminess and chewiness were not included in the PCA because they were strongly correlated with hardness. As shown in Figure 4, FH sample were very close to the  $L^*$  and adhesiveness (dominant properties) and placed to the left of the graph. Samples salted by 2 h were placed in the left quadrant slightly shifted to the right. This behavior indicated a loss of luminosity and an increase in hardness,  $a^*$  parameter, cohesiveness and springiness. This effect was more pronounced as the salting time increased (samples placed in the right quadrant). In general, there were no differences between BS and MS throughout salting time. On the other hand, DS always was located in the positive quadrant (moving from left to right), indicating that these samples presented the highest values in adhesiveness,  $L^*$  and hardness.

### 3.4. Correlation between quality attributes and $z_{\text{NaCl}}$

Pearson's correlation coefficients between the quality parameters and  $z_{\text{NaCl}}$  values were calculated. Hardness, springiness, cohesiveness and  $a^*$  parameter were highly correlated with  $z_{\text{NaCl}}$  variable ( $r = 0.76$ ,  $p < 0.001$ ;  $r = 0.93$ ,  $p < 0.0001$ ;  $r = 0.95$ ,  $p < 0.001$  and  $r = 0.93$ ,  $p < 0.0001$ , respectively).  $L^*$  was negatively correlated with  $z_{\text{NaCl}}$  values ( $r = -0.87$ ,  $p = 0.0001$ ). Figure 5 shows the correlations between hardness, cohesiveness and  $L^*$  parameter with  $z_{\text{NaCl}}$  values in hake fillets salted by the different procedures. As it can be observed, they were well correlated. The correlation curves obtained showed nonlinear relationships with an upward concavity. Table 3 summarizes the parameters corresponding to the models for the variables: hardness, cohesiveness and luminosity. The adjust determination coefficients indicated that between 83.7 % and 97.4 % of the variability could be explained by the models. In the case of springiness and  $a^*$  parameter, the curves could not be adequately adjusted by the models. Table 4 lists the additional  $z_{\text{NaCl}}$  values used to externally validate the obtained models for hardness; cohesiveness and luminosity. Additionally, Table 4 shows the instrumental measurements and the predicted values obtained according to the models. Student-t test comparisons between observed and predicted values showed that the proposed models used in this study adequately predict the quality attributes of salted samples at different times. All p-values were not statistically significant at  $\alpha = 0.05$ .

An exponential increase in hardness and cohesiveness was observed during the process due to the increase in  $z_{\text{NaCl}}$  content (Figure 5a and b). In contrast, an exponential decrease in  $L^*$  parameter was seen (Figure 5 c). In general, there were no differences between the hardness of BS and MS samples for the range of  $z_{\text{NaCl}}$  studied; however, DS samples exhibited a

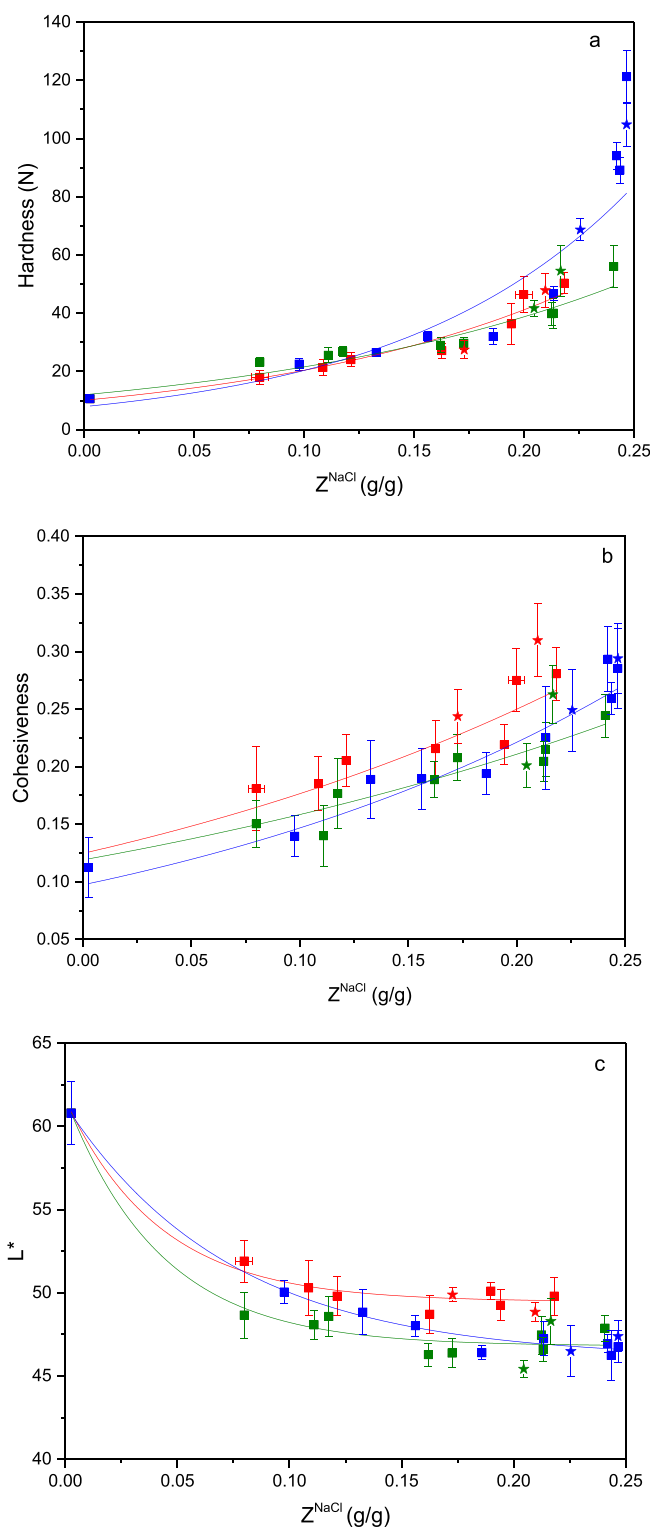


Figure 5. Correlations between  $z_{\text{NaCl}}$  content and some quality attributes during salted hake fillet. a) Hardness, b) Cohesiveness and c) Luminosity. BS (Red), MS (Green) and DS (Blue) experimental data; (—) model prediction; (I) error bar; (star) external observed value.

more abrupt increase with the concentration of  $z_{\text{NaCl}}$ . The dry salted samples showed a very large increase in hardness when approaching to the equilibrium value of  $z_{\text{NaCl}}$ , associated with the highest values of  $z_{\text{NaCl}}$  reached ( $\approx 0.25$ ). These results were in agreement with Barat et al. (2002), who previously reported an exponential correlation between hardness and  $z_{\text{NaCl}}$  values during salting of cod. They attributed the

**Table 3.** Estimated model coefficients corresponding to correlation between hardness, cohesiveness and luminosity with  $z_{NaCl}$  values during hake fillet salting. BS: brining, MS: mixed salting, DS: dry salting.

	Hardness 1 (N)		Model <sup>a</sup> : $y = a \cdot e^{b \cdot x}$		$R^2_{adj}$
	a		b		
BS	10.09 ± 0.67		7.03 ± 0.44		0.969
MS	11.98 ± 1.13		5.87 ± 0.65		0.914
DS	7.91 ± 1.07		9.44 ± 0.27		0.901
	Cohesiveness (-)		Model <sup>a</sup> : $y = a \cdot e^{b \cdot x}$		$R^2_{adj}$
	a		b		
BS	0.125 ± 0.014		3.48 ± 0.65		0.837
MS	0.120 ± 0.007		2.86 ± 0.32		0.931
DS	0.097 ± 0.009		4.10 ± 0.43		0.939
	L*		Model <sup>a</sup> : $y = y_0 + a \cdot e^{(-x/b)}$		$R^2_{adj}$
	$y_0$		a	b	
BS	49.46 ± 0.32		12.15 ± 1.34	0.042 ± 0.010	0.922
MS	46.80 ± 0.49		14.91 ± 1.95	0.042 ± 0.012	0.879
DS	46.06 ± 0.28		15.22 ± 0.94	0.074 ± 0.007	0.974

<sup>a</sup> y = Instrumental measure; x:  $z_{NaCl}$  score.

**Table 4.** Means and standard deviations of observed and predicted instrumental parameter score of new  $z_{NaCl}$  values. BS: brining, MS: mixed salting, DS: dry salting.

$z_{NaCl}$ (g/g)	Hardness 1 (N)			Cohesiveness (-)			L*			
	Observed value	Predicted value	P value <sup>§</sup>	Observed value	Predicted value	P value <sup>§</sup>	Observed value	Predicted value	P value <sup>§</sup>	
BS	0.173 ± 0.001	27.44 ± 3.18	34.00 ± 4.85	0.122 <sup>*</sup>	0.244 ± 0.024	0.227 ± 0.052	0.634 <sup>*</sup>	49.88 ± 0.43	49.66 ± 0.54	0.481 <sup>*</sup>
	0.210 ± 0.004	47.86 ± 5.88	44.06 ± 7.01	0.512 <sup>*</sup>	0.310 ± 0.032	0.258 ± 0.065	0.282 <sup>*</sup>	48.84 ± 0.60	49.55 ± 0.43	0.171 <sup>*</sup>
MS	0.204 ± 0.001	41.68 ± 2.77	39.81 ± 9.09	0.750 <sup>*</sup>	0.201 ± 0.019	0.214 ± 0.026	0.523 <sup>*</sup>	45.42 ± 1.10	46.92 ± 0.47	0.096 <sup>*</sup>
	0.217 ± 0.003	54.52 ± 8.62	42.75 ± 10.10	0.200 <sup>*</sup>	0.262 ± 0.025	0.221 ± 0.029	0.137 <sup>*</sup>	48.29 ± 1.38	46.89 ± 0.06	0.154 <sup>*</sup>
DS	0.226 ± 0.004	68.66 ± 3.73	66.60 ± 4.96	0.596 <sup>*</sup>	0.249 ± 0.036	0.246 ± 0.047	0.934 <sup>*</sup>	46.48 ± 1.52	46.78 ± 0.54	0.764 <sup>*</sup>
	0.247 ± 0.004	104.78 ± 13.53	81.24 ± 12.60	0.092 <sup>*</sup>	0.294 ± 0.030	0.268 ± 0.054	0.506 <sup>*</sup>	47.38 ± 0.93	46.60 ± 0.49	0.268 <sup>*</sup>

<sup>\*</sup> Value non statistically significant at  $\alpha = 0.05$ .

<sup>§</sup> P-value corresponding to the comparison between observed and predicted value using Student's t-test.

differences found to variations in the state of the proteins. Cohesiveness increased proportionally with the concentration of  $z_{NaCl}$ . However, the final cohesiveness values did not show great differences, suggesting that the integrity of the fillets was kept in all the salting procedures (Yashoda and Suryanarayana, 1998). Lightness diminished abruptly in the  $z_{NaCl}$  range from 0 to 0.15 with slight differences among samples (BS, MS, DS). Variations in L\* between procedures would be mainly associated with variations in the  $x_w$  of the fillets, since dehydration causes opacity, as was previously mentioned.

#### 4. Conclusions

Quality of hake fillets has been affected by the different salting procedures and the changes were associated with physicochemical and structural modifications. The main modifications occurred during DS, where the direct contact of salt crystals produces an important concentration difference between the outside and inside of the muscle. The hake fillets subjected to MS had the highest fat content, a considerable protein content, and an equilibrium salt content similar to BS. In addition, although MS produced a loss of water similar to DS, the hardness and other mechanical parameters were similar to those obtained with BS. Regardless of the method, all samples showed color parameters significantly different compared to fresh hake, turning more red-orange as the salting time increased. Lightness diminished, a\* values increased and b\* values did not show a clear trend throughout the salting time. Therefore, according to these results, MS would be the recommended salting method in the production of salted hake, since it combines advantages of the other two methods, resulting in a product strongly salted with better mechanical properties and optical properties and nutritional

composition. On other hand, useful nonlinear correlations were found between  $z_{NaCl}$  values and hardness, cohesiveness and L\*, which could be used to predict the quality attributes of hake as a function of salting time. Thus, salted hake could be an alternative to increase the added value of fish products and diversify the seafood offer.

#### Declarations

##### Author contribution statement

Marchetti, Marion Daniela: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Gomez, Paula Luisina: Analyzed and interpreted the data; Wrote the paper.

Yeannes, María Isabel: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

García Loredó, Analia Belen: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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**Data availability statement**

The data that has been used is confidential.

**Declaration of interests statement**

The authors declare no conflict of interest.

**Additional information**

No additional information is available for this paper.

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