Study of the moulds and yeasts counts throughout the manufacture of dry-cured "lacón". Effect of salt levels

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Abstract- The influence of salt levels on moulds and yeasts counts during the manufacture of dry-cured "lacón" was studied. In this work, three batches with different salt levels were used in salting stage: 0.75, 1 and 1.25 days/kg. Process time influenced significantly ($P<0.05$) on moulds and yeasts counts in the three batches studied. However, content of salt was not significantly affected on these microbial groups counts. The counts of these microbial groups increased throughout the manufacturing process, particularly after the post-salting stage.

Keywords: **Dry-cured "lacón", Moulds and yeasts, Salt levels**

I. INTRODUCTION

Traditional cured meat products, made from whole meat pieces of pork or beef, are commonly produced and consumed in different countries throughout the world. Dry-cured lacón, a product made in the North-West of Spain (Galicia), which has a wide acceptance by consumers, is among these traditional cured meat products.

Its elaboration is begun by cutting the fore extremity of the pig at the shoulder blade–humerus joint and the stages of the process are very similar to those followed in the production of dry-cured ham.

During ripening stage of dry-cured meat enzymatic and chemical reactions are involved in the development of flavour. The typical microflora of these meat products is made up of *Micrococcaceae*, lactic acid bacteria and moulds and yeasts, with the involvement of the *Micrococcaceae* being very important. Proteolysis and lipolysis constitute the main biochemical reactions in the generation of flavour precursors, where the endogenous enzymes play the most important role (Toldrá, 1998). The contribution of the fungal population and their enzymes to proteolysis in dry-cured meat products, such as dry-cured sausages, (Bruma, Hierro, de la Hoz, Mottram, Fernández, & Ordoñez, 2003, Sunesen, & Stahnke, 2003) and dry-cured hams (Monte, Villanueva, & Domínguez, 1986; Nuñez, Rodríguez, Córdoba, Bermúdez, & Asensio, 1996; Martín, Córdoba, Nuñez, Benito, & Asensio, 2004) is widely known.

The objective of this research was study the effect of salt levels on moulds and yeasts counts throughout the manufactured of dry-cured "lacón".

II. MATERIAL AND METHODS

II.1 Samples

In order to carry out this study three batches of "lacón" were manufactured in our centre. Each batch consisted of four "lacón" pieces that in the fresh stage weighted 4 kg each. The first batch was salted during 3 days (0.75 days/kg), second during 4 days (1 day/kg) and the last during 5 days (1.25 days/kg) being the temperature of the salting room between 2 and 5 ºC and the relative humidity (RH) between 80 and 90 %. After the salting stage, the pieces were taken from the pile, brushed, washed and transferred to a post-salting room where they stayed for 14 days at a 2-5 ºC and 85-90% RH. After the post-salting stage, the pieces were transferred to a room at 12 ºC and 74-78% RH where a drying ripening process took place for 84 days. For microbial analysis, in each batch, samples were taken from after 7 and 14 days of post-salting and after 7, 14, 28, 56 and 84 days of drying ripening.

II.2. Microbial analysis

Surface samples, approximately 2 mm thickness, were aseptically removed from different areas of the lean surface, and collected together in a sterile bag. The samples were weighed and diluted 1:3 (w/w) with peptone physiological solution (PPS) composed of 8.5 g NaCl (Merck); 1 g trytone (Oxoid) and 1000 mL distilled water. They were homogenized using a IUL-INSTRUMENT mod. Masticator for 2 min. For each sample, serial dilutions were made in PPS and 0.1 mL of the appropriate dilution was spread-plate inoculated onto Oxytetracycline Glucose Yeast Extract agar plates (OGYEA, Merck). Yeast counts were obtained after incubation at 25ºC for 4-5 days and plates with 30-300 colonies were counted.

II.3. Statistical analysis

For the statistical analysis of the results, data were analyzed using the SPSS 15.0 for Windows (SPSS, Chicago, IL, USA) software package. One way analysis of variance was used to analyze the effects of time of process and salt levels.

III. RESULTS AND DISCUSSION

Table 1 shows the evolution of moulds and yeast during the manufactured of dry-cured "lacón". Time of process influenced significantly ($P<0.05$) on moulds and yeasts counts in the three batches studied. However, salt levels were not significantly affected on these microbial groups counts. After 7 days of post-salting the counts of moulds and yeasts were higher in the batches manufactured with less content of salt. This tendency can be observed in almost all points of "lacón" process studied. This salting effect could be indicated that moulds and yeasts are slightly sensitive to salt levels.

Table 1. *Evolution of moulds and yeast counts (log cfu/g) in the surface of pieces during the manufactured of dry-cured lacón. Effect of salt levels*

Salt content (days/kg)	Post-salting (days)		Dry-ripening (days)				
	7	**14**	**7**	**14**	**28**	**56**	**84**
0.75	2.68 ± 0.30^{a}	3.58 ± 0.32^{b}	6.82 ± 0.34^{c}	6.29 ± 0.06^{cd}	6.36 ± 0.25^{cd}	6.46 ± 0.05^{cd}	6.57 ± 0.24^{d}
1.00	2.43 ± 0.23^{a}	3.68 ± 0.49^{b}	6.03 ± 0.89^{c}	6.88 ± 0.20^{cd}	6.58 ± 0.33^{cd}	6.48 ± 0.21^{cd}	6.46 ± 0.16^{d}
1.25	2.31 ± 0.26^{a}	3.47 ± 0.36^{b}	5.60 ± 0.38^{c}	6.24 ± 0.07^{d}	6.61 ± 0.20^{d}	6.41 ± 0.05^{d}	6.26 ± 0.10^{d}

[a-d] Different letters in the same row show significant differences ($p<0.05$, Duncant test)

Our results observed after 7 days of post-salting were lower than showed in a previous study 4.19 log cfu/g (Lorenzo, García-Fontán, Franco, & Carballo, 2005) and those found by Vilar, García-Fontán, Prieto, Tornadijo, & Carballo (2000) who reported values of 7.12 log cfu/g in surface "lacón" samples. At the end of post-salting stage we observed an increased on content of moulds and yeasts in three batches studied. These results are in agreement with those showed in other raw-cured meat products made from whole pieces, such as ham (Huerta, Hernández, Guamis, & Hernández, 1988; Nuñez et al., 1996) and Spanish beef "cecina" (García, Zumalacárregui, & Díez, 1995).

During the drying-ripening stage moulds and yeasts counts remained constant until the end of the process (84 days of drying-ripening). These results observed at the end of process were slightly lower than those determinated in a previous work 7.29 log cfu/g (Lorenzo et al., 2005) and those higher than showed by Vilar et al. (2000) who found final values of 4.22 log cfu/g. Nuñez et al. (1996) also showed final values in Iberian ham samples similar those found by us (around 6.2 log CFU/g).

IV. CONCLUSIONS

The salt contents did not significantly affect on moulds and yeasts counts However, process time influenced significantly ($P<0.05$) on these microbial groups counts in the three batches studied. The batches manufactured with less content of salt showed higher moulds and yeasts counts. At the end of the process of dry-cured "lacón" we found mean values of 6.43 log cfu/g.

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