Assessing proteomic changes in dry-cured ham under different salt concentration and pastiness

López-Pedrouso, M.¹, Pérez-Santaescolástica, C.², García-Pérez, José V.³, Fulladosa, E.⁴, Benedito, J.³, Coll-Brasas, E⁴, Zapata, C.¹, Franco D.² and Lorenzo, J.M.²

¹Department of Zoology, Genetics and Physical Anthropology, University of Santiago de Compostela, Santiago de Compostela -15872, Spain; ²Centro Tecnológico de la Carne de Galicia, Rúa Galicia Nº4, Parque Tecnológico de Galicia, San Cibrao das Viñas, 32900 Ourense, Spain; ³ASPA Group, Department of Food Technology, Universitat Politècnica de València, Camí de Vera s/n, E-46022 València, Spain; ⁴IRTA, XaRTA, Food Technology, Finca Camps i Armet, E-17121 Monells, Girona, Spain

*Corresponding author email: jmlorenzo@ceteca.net

Abstract –The reduction of salt content in dry-cured meat products is a long-standing challenge but it is well known that reducing salt drastically increases the proteolysis. Specifically, excessive protein degradation during ripening process contributes to the development of defective textures such as pastiness that devalues the final quality in dry-cured ham. The aim of this study was to evaluate the degree of proteolysis in dry-cured ham processed under different salt contents using a proteomic analysis. The 2-DE proteome profiles of dry-cured hams were examined in relation to its salt content and pastiness level in order to find proteins markers associated with the process. A positive relationship was found between the total and unique number of spots and dry-cured ham pastiness. Thus, the number of spots was significantly (P<0.05) higher in high pastiness than in low pastiness dry-cured hams.

Key Words - Defective textures, food proteomics, protein degradation

I. INTRODUCTION

Dry-cured ham is a traditional product with a great popularity among European consumers. The processing of drycured ham requires salt as preserving agent. It is also responsible for texture, flavour and final quality of the product. However, according to World Health Organization, reducing salt intake has been identified as one of the most important measures to improve population health. On the other hand, it is well known that a change in salt concentration affects protein degradation depending on many factors such as rearing conditions (e.g. feeding, sex and slaughter age), pig line and the ripening process [1]. In addition, muscle proteins undergo an intense proteolysis during the curing process that contributes to singular dry-cured ham quality. However, excessive proteolysis may lead to higher pastiness, which consists of excessive softness, mushy texture and unpleasant flavours [2]. Therefore, a rigorous control of salt content in the dry-cured ham process is necessary. The aim of the present study was to determine the degree of proteolysis in dry-cured ham in relation to its salt concentration and pastiness level using a proteomic analysis.

II. MATERIALS AND METHODS

Eight dry-cured hams were selected from an initial set of two hundred raw hams obtained from a commercial slaughterhouse resulting from Large White x Landrace breed crosses (medium fat content). Dry-cured hams were classified in two groups (low and high pastiness) according to sensorial panel of IRTA. The protein extraction and the 2-DE were performed according to Franco *et al.* [3]. A total of 250 µg of total protein was loaded into immobilized pH gradient (IPG) strips (24 cm, pH 4-7 linear). The second dimension was performed using 12% SDS-PAGE gels and stained with SYPRO Ruby fluorescent stain. After 2-DE, gels were visualized and digitalized using Gel Doc XR+ system. The detection and quantification of spots were performed using PDQuest software. Statistical differences in the number of spots in sample groups were tested by the Mann-Whitney U test using the SPSS Statistic software.

III. RESULTS AND DISCUSSION

Proteolysis is one of the most important biochemical process related with salt concentration, which generates differences in the end product quality. In the present work, a proteomic approach has been used to evaluate significant differences between low and high pastiness samples. Figure 1 shows a representative gel image from low

and high pastiness dry-cured hams. High-quality 2-DE gels were obtained and biological replicates showed a high reproducibility within sample groups. We found that the average number of spots for low and high pastiness samples was 92 and 123, respectively. It is noteworthy that 5 and 37 spots were unique spots represented only in low and high pastiness, respectively. The total and unique number of spots showed statistically significant differences (p-value <0.05) between groups. Overall, our observations show that number of spots was remarkably higher in high pastiness samples than in low pastiness dry-cured hams. This suggests that differential protein degradation by proteolytic enzymes had a marked effect on the ham texture (pastiness).



Figure 1. 2-DE gel of dry-cured ham relative to low pastiness and high pastiness.

Previous studies have shown that proteolysis differences were correlated not only with moisture content and salt content, but also with enzymatic activity [4]. According to that study, water content, salinity and their interaction affect structural protein and hence the final dry-cured ham texture characteristcs.

IV. CONCLUSIONS

The use of proteomic has proved to be a powerful tool to assess the proteolysis degree in dry-cured ham. It was found a relationship between the total and unique number of spots and dry-cured ham pastiness variations.

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VI. REFERENCES

[1] Mora, L., Calvo, L., Escudero, E., Toldrá, F. (2016). Differences in pig genotypes influence the generation of peptides in dry-cured ham processing. Food Research International 86: 74-82.

[2] Škrlep, M., Čandek-Potokar, M., Mandelc, S., Javornik, B., Gou, P., Chambon, C., Santé-Lhoutellier, V. (2011). Proteomic profile of dry-cured ham relative to *PRKAG3* or *CAST* genotype, level of salt and pastiness. Meat Science 88: 657-667.

[3] Franco, D., Mato, A., Salgado, F.J., López-Pedrouso, M., Carrera, M., Bravo, S., Parrado M., Gallardo J.M., Zapata C. (2015). Tackling proteome changes in the *longissimus thoracis* bovine muscle in response to pre-slaughter stress. Journal Proteomics 122: 73-85.

[4] Harkouss, R., Astruc, T., Lebert, A., Gatellier, P., Loison, O., Safa, H., Portanguen, S., Parafita, E., Mirade, P.-S. (2015). Quantitative study of the relationships among proteolysis, lipid oxidation, structure and texture throughout the dry-cured ham process. Food Chemistry 166: 522-530.