Instrumental Texture Analysis on the Surface of Dry-cured Ham to Define the End of the Process

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Abstract.- The end of the elaboration process of dry-cured ham is currently decided by product weight loss and/or by an expert who carries out an evaluation of the tactile texture on the surface. The objective of this study was to define the optimal measurement conditions of an instrumental texture analysis on the surface of the dry-cured ham (ITAS), to define the end of process. 120 dry-cured hams were classified by experts into Hard (appropriate) or Soft (non-appropriate) texture groups and used to perform compression tests using different probes on three anatomical positions. Results showed that the small probe in position 2 gave the most discriminant conditions, providing representative information of the internal texture. Although classification using only weight loss was possible with an accuracy rate of 80.4 % or 66.7% depending on the weight loss, the maximum classification accuracy was obtained when using ITAS in combination with weight loss. Further studies at industrial level are needed.

Keywords: dry-cured ham, texture, instrumental evaluation, non-destructive technologies, weight loss

1. Introduction

In the dry-cured ham industry, the end of the process is decided by the product weight loss and/or by the evaluation of the texture of the whole hams made by experts on the surface. Weight loss criterion is also used in the Traditional Speciality Guaranteed (TSG) "Jamón Serrano" (Jamón Serrano Foundation, 2007) as a quality standard, requiring a minimum weight loss of 33%. However, weight loss is not always well-correlated to the internal texture because other factors such as salt content, drying temperature and elaboration conditions or raw meat pH can also influence texture development (Harkouss et al., 2015; Morales, Arnau, Serra, & Gou, 2007; Morales, Serra, Guerrero, & Gou, 2007; Tomažin et al., 2020) and therefore, its use alone to decide the end of the process may lead to dry-cured hams with non-appropriate texture characteristics being sent to the market. For this reason, not only a sensory analysis of aspect and flavour, but also of the textural attributes of a small part of the dry-cured ham production is also required in the Jamon Serrano TSG seal. Although some studies to develop faster instrumental methods to evaluate the sensory perception of texture are being investigated, both sensory and instrumental tests are tedious, destructive and can only be performed for a small part of the production. Currently, non-invasive technologies able to determine internal textural characteristics of the product are not available on the market, even though some authors have pointed out the possibility of applying them in the future by using "*in-line*" multi energy X-ray (Fulladosa et al., 2018) or magnetic resonance imaging (García-García, Fernández-Valle, Castejón, Escudero & Cambero, 2019) devices.

Meanwhile, different strategies could be used to more efficiently decide the end of the process according to texture. Information on the fat content of the raw material and acquired salt content using non-destructive technologies implemented in-line (De Prados et al., 2015; Fulladosa, Munoz, Serra, Arnau, & Gou, 2015; Fulladosa, Santos-Garces, Picouet, & Gou, 2010; Santos-Garcés, Gou, Garcia-Gil, Arnau, & Fulladosa, 2010; Santos-Garcés, Muñoz, Gou, Garcia-Gil, & Fulladosa, 2014; Schivazappa et al., 2017) could provide relevant information for the estimation of the optimal duration of the elaboration process since there is a well-known relationship between the drying time and fat and salt content of the dry-cured ham.. Besides, texture development is influenced by salt content (Andrés, Cava, Ventanas, Thovar, & Ruiz, 2004; Benedini, Parolari & Toscani, 2012), giving an idea of the final texture of the product. Information on raw meat pH or impedance measured "*in-line*" could also provide further information to predict the final texture of the product (Guerrero et al., 2004; Guerrero, Gou, & Arnau, 1999; Schivazappa et al., 2002). However, because dry-cured ham texture can also be influenced by other processing variables more difficult to control, it would be of interest to measure texture "*in-line*" at the end of the process for classifying hams into Hard (appropriate) or Soft (non-appropriate) texture groups before sending them to the market. An instrumental analysis of the texture from the surface of the ham could reduce workload, subjectivity and also improve the quality of the product marketed.

The objective of this study was to define the optimal measurement conditions (probe size and anatomical location) of a non-destructive instrumental texture analysis on the surface of the dry-cured ham (ITAS), to evaluate whether the product had the appropriate texture to be sent to the market or not. The usefulness of including information about weight loss and subcutaneous fat thickness to improve the prediction of product texture was also evaluated. The representativeness of texture analysis on the surface to define the internal textural characteristics of the ham were also evaluated.

2. Material and methods

2.1 Experimental design

Experiment 1: Setting up the ITAS method

One hundred and twenty dry-cured hams from Duroc, Large White and Landrace breeds crosses skinned with the typical V-shape (Fig.1A), and with an elaboration process time of at least 24 months, were selected according to experts texture evaluation in a dry-cured ham facility owned by Monte Nevado (Segovia, Carbonero el Mayor, Spain). The weight loss of the sampled dry-cured hams ranged between 29% and 42%. Sixty hams were selected for having an appropriate texture for sending to the market after evaluation of the texture on the surface by experts (Hard texture). The rest of the hams (n=60) were selected for having a non-appropriate texture showing too much softness (Soft texture). Classification of hams into Hard or Soft texture groups was carried out by three different experts who routinely perform this activity in the company. They evaluated the texture of the hams in triplicate by pressing with the fingers on the external surface close to *Semimembranosus* (SM) and *Biceps femoris* (BF) as well as on the knuckle muscles. This is the standard procedure in many dry-cured ham companies to estimate the texture of the internal muscles from the surface of the ham. The most common score of the three experts (appropriate/hard texture or not appropriate/soft texture) was considered as the reference value. Triplicates were performed by evaluating each ham on three different days.

These hams were used to define the measurement conditions of an instrumental compression test. In order to minimise any possible damage to the ham, the instrumental texture was only evaluated in the part covered with subcutaneous fat. Three different anatomical positions were selected (1, 2 and 3, as shown in Figure 1) and three different size spherical probes (large, medium and small, with diameters of 5.5, 3.5 and 1.9 cm respectively, see Figure 2) were evaluated. Position 1 and 2 were chose because are the areas evaluated by experts at the industry. Position 3 was chosen because it would be the most convenient area of measurement in the case of industrial implementation. Spherical probes were chosen in order to simulate the shape of the human fingers. Dry-cured hams were distributed according to the experimental design defined in Table 1. Subcutaneous fat thickness in each anatomical position and volumetric content of subcutaneous, intermuscular and intramuscular fat (%), the perimeter of the slice covered with subcutaneous fat (%) and the lean slice thickness (%) were determined using computed tomography image analysis (section 2.3). Salt content of the hams was also estimated using previously developed predictive models based on computed tomography data (Santos-Garces et al., 2010; Santos-Garcés, Munoz, Gou, Sala, & Fulladosa, 2012).

In order to study the relationship between the textural parameters measured on the surface and those measured in the BF muscle, five hams from the soft texture group and 4 hams from the Hard texture group were randomly selected. A Stress relaxation (SR) and a Texture profile analysis (TPA) test were performed on the BF muscle of these hams as described in section 2.4.

Experiment 2. The effectiveness of the ITAS method to classify hams into different texture groups

Twenty-seven dry-cured hams with an elaboration process of 24 months and similar weight losses (36.5%±2.52) were selected according to their texture from the same dry-cured ham producer as in Experiment 1. This is the mean weight loss of the company production before being sent to market and the weight loss range in which the internal texture prediction is more difficult to assess by experts. Fifteen hams had a hard texture and twelve had a soft texture according to the most common score of the three different experts (reference value). The most discriminant measurement conditions defined in experiment 1 were used to perform the analysis. The discriminant ability of ITAS was assessed using the measured texture parameters on the surface of the ham in combination with the subcutaneous fat thickness and/or weight loss.

2.2 Description of ITAS method

Uniaxial compression tests (samples compressed in one direction and unrestrained in the other two) were performed using the different spherical probes shown in Figure 2 on the three anatomical positions described in Figure 1. All the measurements were carried out by using a Universal Texture Analyser TA.TX2 (Stable Micro Systems Ltd., Surrey, England) using a load cell of 30 Kg at a constant crosshead speed of 40 mm/s. Crosshead displacement was 10 mm when using the large probe (Ø= 5.5 mm), and 15 mm when using the medium (Ø= 3.5 mm) and the small (Ø= 1.9 mm) probes. These distances were previously calculated to simulate the pressure exerted by a human finger at a maximum force. The displacement was measured automatically by the equipment itself after contacting the sample surface (minimum contact force of 10 g). Anatomical positions 1 and 2 were selected because they fell within the area evaluated by the experts. Anatomical position 3 was centrally located and avoided movement of the ham during ITAS analysis, facilitating in-line evaluation of the hams at the industry without any mounting system. Dry-cured hams were fixed with wedges to prevent any movement during compression. Figure 2A shows the instrumental texture analysis performed at the ham surface.

The force/time curves were recorded and evaluated by Exponent stable micro Systems software (version 6.1.16.0). The parameters were calculated by macros written using the same software. The following parameters were determined (Figure 2C): Maximum compression force (F_{max}, N); area under the curve until maximum compression force (A_1, N.mm) representing the total work performed to compress the ham; area under the curve after the maximum compression force (A_2, N.mm) representing sample recovery to its initial state after being compressed (to some extent related to the elasticity of the product), and the slope of the curve between 20% and 80% of the maximum compression force (Slope, N/mm) representing Young's modulus or the modulus of elasticity.

2.3 Computed tomography image analysis and salt content prediction

All the dry-cured hams were scanned at IRTA centre of Monells, using a scanner model HiSpeed Zx/i from General Electric Healthcare (GE Healthcare, Barcelona, Spain). An axial protocol was used with settings 80 kV, 250 mA and rotation time of 2 s. Image size was 512 x 512 voxels and the Displayed Field of View (DFOV) was 461 mm (resolution=1.1 pixels/mm). Before scanning, the hams were properly aligned in the CT equipment. Then, a 10 mm thick slice was scanned 10 cm from the aitch bone in the distal direction. This was the area where the ITAS was performed. The acquired matrixes of values were analysed using Centricity Radiology RA600 v.7.0 (GE Medical Systems, Barcelona, Spain). From each image, the subcutaneous fat thickness (mm) at each anatomical position (Figure 1B) was manually measured perpendicular to the tray.

Salt content was estimated in BF muscle by using previously developed predictive models (Santos-Garces et al., 2010) that used tomograms acquired at 80 kV (RMSE=0.263%). To do so, a central part of BF muscle was automatically selected, and the predictive model applied with an in-house script written in Matlab. This script used a combination of thresholding and gradient detection algorithms to segment intramuscular fat (Munoz, Rubio-Celorio, Garcia-Gil, Guardia, & Fulladosa, 2015). Subcutaneous fat and intermuscular fat were segmented applying thresholding after segmentation of the intermuscular fat. Depending on the position of the segmented fat patches, pixels were classified as subcutaneous fat if the fat patch was close to the ham contour, otherwise pixels were classified as intermuscular fat. The content of subcutaneous, intermuscular and intramuscular fat (%) was estimated as the number of pixels corresponding to each class of fat over the total number of pixels of the ham slice (See Figure 1B). The lean slice thickness (%) was calculated as the percentage of pixels containing lean in a line perpendicular to the tray and at 3 cm from the edge of the bone over the number of pixels containing lean and fat. The perimeter covered with subcutaneous fat (%) was calculated as the percentage of number of pixels of the perimeter of the slice segmented as subcutaneous fat into the total number of pixels of the perimeter (see Figure 1B).

2.4 Instrumental texture analysis of internal muscles

From each ham selected for internal texture analysis (n=10), a minimum of twelve parallelepipeds from BF muscle were accurately carved with a scalpel (20 mm x 20 mm x 15 mm, length x width x height) for SR and TPA tests. Before the texture analysis the pieces were stored at 4ºC for 24h wrapped in plastic film to prevent them from drying out. A SR test was performed using a Universal Texture Analyser TA.TX2 (Stable Micro Systems Ltd., Surrey, England), provided with a 30 kg load cell and a 60 mm compression plate. Samples were compressed to 25% of their original height, perpendicular to the muscle fibre bundle direction, at a crosshead speed of 1mm/s and at a temperature of 4ºC ± 2°C. The force decay or relaxation versus time $Y_{(t)}$ was calculated as follows:

$$Y_{(t)} = \frac{F_0 - F_{(t)}}{F_0}$$

where F_0 (N) is the initial force and $F_{(t)}$ is the force recorded after t seconds of relaxation. The force decay at 2s (Y_2) and 90s (Y_{90}) was calculated (Morales, Guerrero, Serra, & Gou, 2007).

The Texture Profile Analysis was performed using a universal Texture Analyser TA.HD.plus (Stable Micro Systems Ltd., Surrey, England) provided with a 250 kg load cell and a 75mm compression plate. Samples were compressed twice to 75% of their original height (time = 0s between the two compression cycles), perpendicular to the muscle fibre bundle direction, at a crosshead speed of 1mm/s and at a temperature of 4ºC ± 2°C. Force versus time was recorded and the following parameters were calculated: hardness (N), cohesiveness (dimensionless) and springiness (dimensionless). Hardness is defined as the maximum peak force during the first compression cycle; cohesiveness as the ratio between the positive force area during the second compression cycle and that found during the first compression cycle; and springiness as 'the height that the food recovers during the time that elapses between the end of the first bite and the start of the second bite (Bourne, 1978). In both SR and TPA tests, for each parameter, the average of the minimum six pieces per ham was used for the statistical analysis.

2.5 Statistical analysis

The reproducibility of each expert was determined by analysing their frequency of successes and failures in the three repetitions performed for each ham compared to the most frequent classification (hard or soft texture) given by the three experts in the three replicates (n=9). The expert was considered to be reproducible when the result of the three repetitions of the same expert was the same and agreed with the most frequent classification given by them all.

Differences of internal texture between Soft and Hard texture groups were analysed using a one-way ANOVA procedure including texture group as fixed effect. Textural parameters measured at the ham's surface (F_{max}, A_1, A_2 and slope) were also analysed by using a one-way ANOVA procedure for each combination of the anatomical position (1, 2 or 3) and each probe size (large, medium and small). The model included the Texture Group defined by the experts (Hard/Soft) as fixed effect. Weight loss and/or subcutaneous fat thickness were also included as covariates in additional models. Differences between mean values were tested by means of Tukey's test ($p<0.05$).

A discriminant analysis (DA) and a Partial Least Square discriminant analysis (PLS-DA) were applied to classify the dry-cured hams into the different, predefined texture groups (Hard and Soft) on the basis of the textural parameters measured on the surface (Force/time curve or F_{max}, A_1, A_2 and slope), weight loss and subcutaneous fat thickness. The percentage of samples correctly classified according to cross-validation analysis was calculated. Linear correlations between surface and internal texture parameters were calculated in those hams where both measurements (on the surface and on BF muscle) were performed (n=9). All the analyses were performed using the XLSTAT v19.7 (2017) (Addinsoft, Paris).

3. Results and discussion

3.1 Experiment 1: Definition of probe size and anatomical measurement position for ITAS

Soft and Hard texture groups showed significant differences in internal texture (F_0 and Y_{90} from the SR test and hardness and cohesiveness from the TPA test) ($p<0.05$) (Table 2). This analysis was performed to corroborate the ability of experts to predict the internal texture of the ham from the surface when evaluating hams with significantly different weight losses. Besides, the mean reproducibility of experts when classifying the hams into the two different texture groups was 89.1%. This means that experts would classify 89.1% of production into the same group (soft or hard) after evaluating the hams three times, producing an error in classification in 10.9% of the production.

Table 3 shows physicochemical characteristics of the hams used in the experiments performed with the different probe sizes. Hams assigned to the different experiments using large, medium and small probes had similar characteristics ($p>0.05$). Hard hams showed a significantly ($P<0.05$) higher salt content, weight losses, lower subcutaneous fat thickness (except in the small probe batch) and lower percentage of perimeter covered with subcutaneous fat than the soft hams. However, no significant differences were found in lean slice thickness, intermuscular fat and intramuscular fat percentage. The higher salt content of hard hams contributed to reduce the activity of Ca-dependent proteases and cathepsin D (Sárraga, Gil, Arnau, & Monfort, 1989) although during the process m-calpain could increase its activity when salt content is up to 0.5M (Rosell &Toldrà, 1996). An increase of hardness is observed as the water content is reduced during drying (Ruíz-Ramírez, Arnau, Serra, & Gou, 2006).

The higher percentage of the perimeter covered with subcutaneous fat and the higher subcutaneous fat thickness in soft hams than in hard hams slowed down the dehydration process and facilitated proteolysis during the process. Moreover, weight losses had a significant correlation ($P<0.05$) with the percentage of the perimeter covered with subcutaneous fat ($r=-0.61$) and with the percentage of lean surface ($r=0.54$), but the correlation with the percentage of intermuscular and intramuscular fat was not significant ($P>0.05$) ($r= -0.23$ and $+0.14$, respectively).

Table 4 shows F values and significance (p-values) of the texture group effect for each of the textural parameters (F_{max}, A_1, A_2 and Slope) when using different probe sizes on the different anatomical positions. All the parameters provided relevant information to classify hams into the two texture groups defined by experts ($p<0.05$). The highest F values for F_{max} and A_1 were obtained with the small probe in position 2, whereas slightly higher F values for A_2 and slope were obtained with the large probe in position 1. The use of the small probe in position 2 was considered the most discriminant condition and was used for further analysis. The reason could be that the small probe had a smaller surface of contact with the ham. The deformation of the ham surface in the areas surrounding the point of contact between the probe and the ham was lower. Therefore, the small probe was less affected by the areas adjacent to the measurement point, being the more representative measurement of the internal texture of the ham.

For anatomical position 2, the reason could be that, among the three studied, it was the part of the ham with the thinnest subcutaneous fat layer, and beneath this position there are the knuckle muscles and BF, and normally not thick intermuscular fat streaks. However, at anatomical position 2, movements are more prone to occur during the compression because of the shape of the ham. Therefore, a mounting system needs to be designed for deploying this technology in the factory. Anatomical positions 1 and 3 seem less convenient. Beneath position 1 there are the *Gracilis,* SM and *Semitendinosus* muscles*,* with the thickest subcutaneous fat layer which can produce changes on the results. Beneath position 3, the BF, SM and *Gracilis* muscles are located and intermuscular fat layers can be found.

It must be noted that there was a significant correlation between the textural parameters obtained with the small sensor in position 2 (F_{max} and A_2) and the internal texture measured in the BF muscle (F_0 and hardness), showing significant correlation coefficients between A_2 and F_0 (r=0.72) and between A_2 and hardness (r=0.67) (p<0.05). No significant correlations were found between the other parameters. High correlations found between internal texture and F_{max} and A_2 values measured on the surface of the ham can be explained because both parameters provide relevant textural information on the whole hams. F_{max} represents the maximum force needed to compress the samples. Obviously, the harder the ham, the more force is needed, although variability in subcutaneous fat thickness and the percentage of the perimeter covered with fat can interfere on this correlation. A_2 represents the energy that the ham returns when the probe moves back from the ham surface. If the ham is soft inside, the returned energy will be lower, since the sample becomes deformed for longer and does not return to its original position immediately after removing the compression force. These results suggest that textural measurements on the surface of the ham using the small sensor in position 2 could be used to estimate the texture in the BF muscle.

When weight loss (%) and subcutaneous fat thickness (mm) were included in the analysis of variance as covariates (results not shown), a positive significant weight loss effect was observed in all cases ($p \leq 0.05$). Therefore, weight loss was also an important parameter for the classification of hams into different textural groups. This result was expected since hardness increased with weight loss (Serra, Ruíz-Ramírez, Arnau, & Gou, 2005). Weight loss was also positively correlated to F_{max} A_1, A_2 and slope, which were useful for the classification of the hams. In contrast, subcutaneous fat thickness only had a significant effect if weight loss was dropped from the model. The reason for this is the significant negative correlation between these two covariates. An increase in subcutaneous fat thickness slows down the drying process and decreases the weight loss. In the case of anatomical positions 1 and 3, correlations between weight loss and subcutaneous fat thickness were around -0.60 whereas a lower correlation (r=-0.46) was found in position 2. The reason is the lower representativeness of position 2 with respect to the overall fat content of the ham. Internal differences in texture were also related to other parameters such as salt and fat content (Table 3).

The obtained results drive the development of a multisensor method to more accurately and less destructively identify dry-cured hams with an appropriate texture before being sent to the market. To do so, the use of non-destructive technologies that provide information about the product during the process (De Prados et al., 2015; Pérez-Santaescolástica et al., 2019) and the application of data mining techniques (Peromingo, Caballero, Rodríguez, Caro, & Rodríguez, 2020) in combination with ITAS data could be useful to estimate the remaining processing time to achieve the target texture.

3.2 Classification performance of dry-cured ham production using different data

Results from discriminant analysis showed that classification performance of dry-cured ham production, with weight loss between 28% and 42% (whole weight loss range), into Hard and Soft texture groups using only weight loss was possible with a correctness of 80.4 % (Table 5). However, there was still an important part of hams, mainly in a range of weight loss between 34.7 and 37.5% (medium weight loss range), that were misclassified. The rate of feasibility for the classification of hams with a medium weight loss was just 66.7%. This means that, in a company such as the one which provided the hams for this study, in which the main part of the production (54%) had a weight loss between 34.5 and 37.5% before being sent to the market, 36% of the production would be misclassified if only weight loss was used to decide the end of process.

Textural parameters from ITAS provided relevant information for classification, showing similar classification scores to those of using weight loss when hams had a wide range of weight losses and increasing the rate of precision from 66.7% to 70.4% when only hams with a medium weight loss were used. The use of the whole force/time curve for the classification did not provide extra information showing similar classification scores than the textural parameters extracted from the curve. When combining textural parameters and subcutaneous fat thickness information, the maximum classification accuracy was obtained, both when using the whole weight loss range (82.1%) or only the medium weight loss range (74.1%). Therefore, the use of non-destructive measurements (such as individual weight loss, ITAS parameters and subcutaneous fat thickness using computed tomography) provided relevant information for dry-cured ham classification before being sent to the market. Further studies at industrial level with a larger amount of dry-cured hams with different intramuscular and intermuscular fat contents, the ageing process and salt contents are needed to generalise these results.

4. Conclusions

Texture parameters measured on the surface of the ham can discriminate dry-cured hams with appropriate texture from those not ready to be sent to the market. These texture parameters, together with subcutaneous fat thickness, could help to improve classification especially in those hams with a similar weight loss. However, a process validation using different types of hams (with different degrees of proteolysis, intramuscular and intermuscular fat content and ageing conditions) and including relevant information of the product (such as fat content in raw material or salt content after salting) obtained in-line during the process is needed.

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6. References

Andrés, A. I., Cava, R., Ventanas, J., Thovar, V., & Ruiz, J. (2004). Sensory characteristics of Iberian ham: Influence of salt content and processing conditions. *Meat Science, 68*(1), 45-51. doi:http://dx.doi.org/10.1016/j.meatsci.2003.08.019

Benedini R., Parolari G., Toscani T., & R., V. (2012). Sensory and texture properties of Italian typical dry-cured hams as related to maturation time and salt content. *Meat Science, 90*, 431-437.

Bourne, M. C. (1978). Texture profile analysis. *Food Tecnology, 32*(7), 62-66.

De Prados, M., Fulladosa, E., Gou, P., Munoz, I., Garcia-Perez, J. V., & Benedito, J. (2015). Non-destructive determination of fat content in green hams using ultrasound and X-rays. *Meat Science, 104*, 37-43. doi:10.1016/j.meatsci.2015.01.015

Fulladosa, E., Austrich, A., Muñoz, I., Guerrero, L., Benedito, J., Lorenzo, J. M., & Gou, P. (2018). Texture characterization of dry-cured ham using multi energy X-ray analysis. *Food Control, 89*, 46-53. doi:https://doi.org/10.1016/j.foodcont.2018.01.020

Fulladosa, E., Munoz, I., Serra, X., Arnau, J., & Gou, P. (2015). X-ray absorptiometry for non-destructive monitoring of the salt uptake in bone-in raw hams during salting. *Food Control, 47*, 37-42. doi:10.1016/j.foodcont.2014.06.023

Fulladosa, E., Santos-Garces, E., Picouet, P., & Gou, P. (2010). Prediction of salt and water content in dry-cured hams by computed tomography. *Journal of Food Engineering, 96*(1), 80-85. doi:10.1016/j.jfoodeng.2009.06.044

Fundación Jamón Serrano (2007). Pliego de Condiciones para la elaboracón del Jamón Serrano (Available on 28/07/07 at URL: http://www.fundacionserrano.org/fjamones/es/public/biblioteca/Pliego%20de%20Condiciones.pdf).

García-García, A. B., Fernández-Valle, M. E., Castejón, D., Escudero, R., & Cambero, M. I. (2019). Use of MRI as a predictive tool for physicochemical and rheologycal features during cured ham manufacturing. *Meat Science, 148*, 171-180. doi:https://doi.org/10.1016/j.meatsci.2018.10.015

Guerrero, L., Gobantes, I., Oliver, M. À., Arnau, J., Dolors Guàrdia, M., Elvira, J., . . . Monfort, J. M. (2004). Green hams electrical impedance spectroscopy (EIS) measures and pastiness prediction of dry cured hams. *Meat Science, 66*(2), 289-294. doi:https://doi.org/10.1016/S0309-1740(03)00101-3

Guerrero, L., Gou, P., & Arnau, J. (1999). The influence of meat pH on mechanical and sensory textural properties of dry-cured ham. *Meat Science, 52*, 267-273.

Harkouss, R., Astruc, T., Lebert, A., Gatellier, P., Loison, O., Safa, H., . . . 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. doi:http://dx.doi.org/10.1016/j.foodchem.2014.06.013

Morales, R., Arnau, J., Serra, X., & Gou, P. (2007). Meat pH influence on texture of dry-cured ham with reduced salt content and submitted to different storage temperatures. *6th International Symposium on the Mediterranean Pig*, 267-273.

Morales, R., Guerrero, L., Serra, X., & Gou, P. (2007). Instrumental evaluation of defective texture in dry-cured hams. *Meat Science, 76*, 536-542.

Morales, R., Serra, X., Guerrero, L., & Gou, P. (2007). Softness in dry-cured porcine biceps femoris muscles in relation to meat quality characteristics and processing conditions. *Meat Science, 77*, 662-669.

Munoz, I., Rubio-Celorio, M., Garcia-Gil, N., Dolors Guardia, M., & Fulladosa, E. (2015). Computer image analysis as a tool for classifying marbling: A case study in dry-cured ham. *Journal of Food Engineering, 166*, 148-155. doi:10.1016/j.jfoodeng.2015.06.004

Pérez-Santaescolástica, C., Fraeye, I., Barba, F. J., Gómez, B., Tomasevic, I., Romero, A., . . . Lorenzo, J. M. (2019). Application of non-invasive technologies in dry-cured ham: An overview. *Trends in Food Science & Technology, 86*, 360-374. doi:https://doi.org/10.1016/j.tifs.2019.02.011

Peromingo, B., Caballero, D., Rodríguez, A., Caro, A., & Rodríguez, M. (2020). Application of data mining techniques to predict the production of aflatoxin B1 in dry-cured ham. *Food Control, 108*, 106884. doi:https://doi.org/10.1016/j.foodcont.2019.106884

Rosell, C. M., & Toldrá, F. (1996). Effect of curing agents on m-calpain activity throughout the curing process. Zeitschrift für Lebensmittel-Untersuchung und Forschung, 203(4), 320-325. doi:10.1007/BF01231069

Ruiz-Ramírez, J., Arnau, J., Serra, X., & Gou, P. (2006). Effect of pH24, NaCl content and proteolysis index on the relationship between water content and texture parameters in biceps femoris and semimembranosus muscles in dry-cured ham. *Meat Science, 72*, 185-194.

Santos-Garcés, E., Gou, P., Garcia-Gil, N., Arnau, J., & Fulladosa, E. (2010). Non-destructive analysis of a(w), salt and water in dry-cured hams during drying process by means of computed tomography. *Journal of Food Engineering, 101*(2), 187-192. doi:10.1016/j.jfoodeng.2010.06.027

Santos-Garcés, E., Munoz, I., Gou, P., Sala, X., & Fulladosa, E. (2012). Tools for Studying Dry-Cured Ham Processing by Using Computed Tomography. *Journal of Agricultural and Food Chemistry, 60*(1), 241-249. doi:10.1021/jf203213q

Santos-Garcés, E., Muñoz, I., Gou, P., Garcia-Gil, N., & Fulladosa, E. (2014). Including estimated intramuscular fat content from computed tomography images improves prediction accuracy of dry-cured ham composition. Meat Science, 96(2, Part A), 943-947. doi:http://dx.doi.org/10.1016/j.meatsci.2013.09.018

Sárraga, C., Gil, M., Arnau, J., & Monfort, J. M. (1989). Effect of curing salt and phoshate on the activity of porcine muscle proteases. *Meat Science, 25*, 241-249.

Schivazappa, C., Degni, M., Costa, L. N., Russo, V., Buttazoni, L., & Virgili, R. (2002). Analysis of raw meat to predict proteolysis in Parma ham. *Meat Science, 60*, 77-83.

Schivazappa, C., Virgili, R., Simoncini, N., Tiso, S., Álvarez, J., & Rodríguez, J. M. (2017). Application of the magnetic induction technique for the non-destructive assessment of salt gain after the salting process of Parma ham. *Food Control, 80*, 92-98. doi:https://doi.org/10.1016/j.foodcont.2017.04.017

Serra, X., Ruiz-Ramírez, J., Arnau, J., & Gou, P. (2005). Texture parameters of dry-cured ham m. biceps femoris samples dried at different levels as a function of water activity and water content. *Meat Science, 69*, 249-254.

Tomažin, U., Škrlep, M., Prevolnik Povše, M., Batorek Lukač, N., Karolyi, D., Červek, M., & Čandek-Potokar, M. (2020). The effect of salting time and sex on chemical and textural properties of dry cured ham. *Meat Science, 161*, 107990. doi:https://doi.org/10.1016/j.meatsci.2019.107990

FIGURES

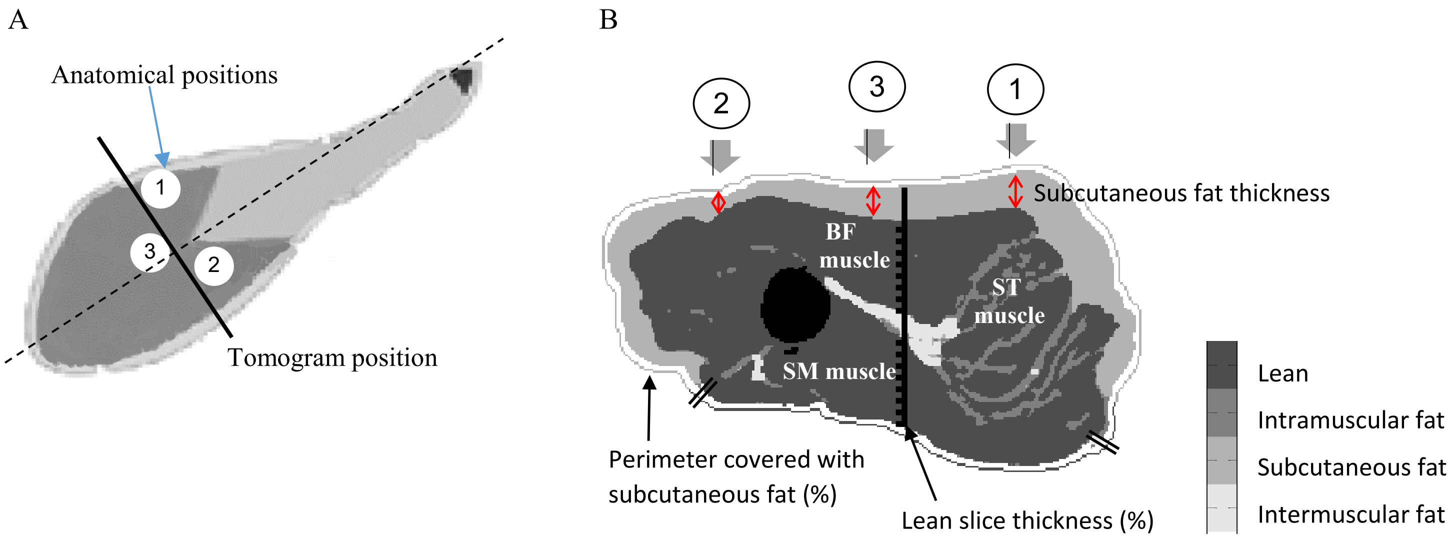

Figure 1. Anatomical positions (1, 2 and 3) in the whole dry-cured ham (A) and its correspondence to a tomogram (B).

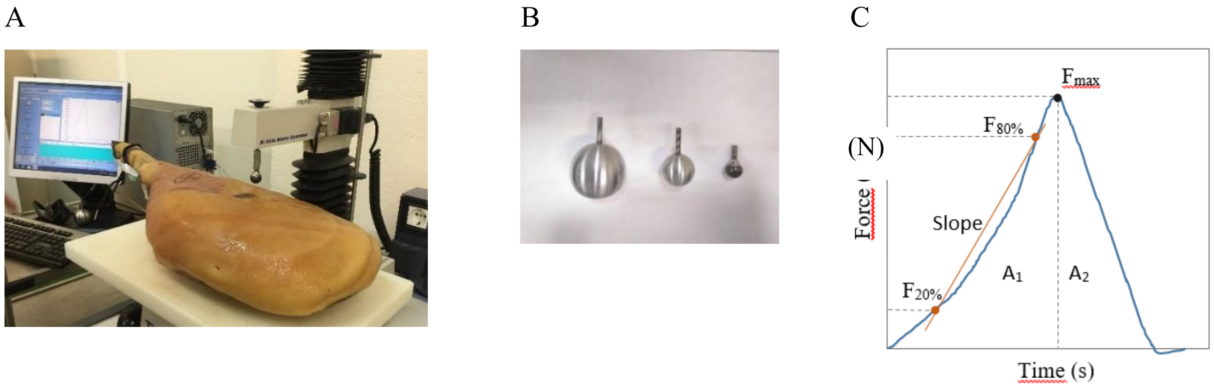

Figure 2. Instrumental texture analysis on the ham surface (ITAS) (A). Large, medium and small probes used for instrumental texture evaluation on the surface of the ham (B). Example of acquired force/time curve and textural parameters determined (C).

TABLES

Table 1. Distribution of hams according to texture group and probe size.

<table>
<tr><td colspan="2"></td><td colspan="3">Probe size</td></tr>
<tr><td>Texture group
by expert evaluation
(Reference values)</td><td>Anatomical positions for instrumental test</td><td>Large
(ø = 5.5 mm)</td><td>Medium
(ø = 3.5 mm)</td><td>Small
(ø = 1.9 mm)</td></tr>
<tr><td rowspan="3">Hard texture</td><td>1</td><td rowspan="3">n = 20</td><td rowspan="3">n = 20</td><td rowspan="3">n = 20</td></tr>
<tr><td>2</td></tr>
<tr><td>3</td></tr>
<tr><td rowspan="3">Soft texture</td><td>1</td><td rowspan="3">n = 20</td><td rowspan="3">n = 20</td><td rowspan="3">n = 20</td></tr>
<tr><td>2</td></tr>
<tr><td>3</td></tr>
</table>

Table 2. Mean ± standard error of weight loss, ham surface measurements (F_{max}, A_1, A_2 and slope using the small probe in position 2) and internal texture in BF muscle (F_0, Y_2 and Y_{90} from SR test and Hardness, cohesiveness and springiness from TPA test) for dry-cured ham experts' texture groups.

Texture group	**Weight loss (%)**	**Measurements at ham surface (small probe - position 2)**					**Texture in BF muscle (SR test)**				**Texture in BF muscle (TPA test)**		
		n	**F_{max} (N)**	**A_1 (N.mm)**	**A_2 (N.mm)**	**Slope (N/mm)**	**n**	**F_0 (N)**	**Y_2**	**Y_{90}**	**Hardness (N)**	**Cohesiveness**	**Springiness**
Soft	34.1 ±1.78	20	139.25[b] ±4.98	787.42[b] ±29.49	469.71[b] ±17.12	11.28[b] ±0.40	5	19.88[b] ± 0.97	0.326 ± 0.005	0.624[a] ± 0.007	90.22[b] ±5.24	0.52[b] ±0.013	0.61 ±0.023
Hard	37.5 ±1.46	20	195.53[a] ±7.10	1137.50[a] ±38.92	635.43[a] ±26.16	15.49[a] ±0.66	4	32.75[a] ± 0.75	0.299 ± 0.001	0.584[b] ± 0.001	142.19[a] ±4.78	0.60[a] ±0.005	0.71 ±0.007

[ab] Different letters between texture groups indicate significant differences ($p<0.05$).

Table 3. Characterisation of hams used in experiments performed with different probes.

Probe size batch	Texture group	N	Weight green ham (kg)		Salt content (%)		Weight loss (%)		Subcutaneous fat thickness (mm)*		Lean slice thickness (%)		Intermuscular fat area (%)		Intramuscular fat area (%)		Perimeter covered with subcutaneous fat (%)	
			Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Large	Soft	20	13.0[a]	0.20	5.3[b]	0.12	33.6[a]	0.48	11.5[a]	1.19	78.0	1.89	4.0	0.30	4.9	0.21	69.6[a]	0.96
	Hard	20	13.1[a]	0.18	6.4[a]	0.17	38.2[b]	0.42	8.7[b]	0.94	79.7	1.89	3.4	0.29	5.4	0.24	63.3[c]	0.88
Medium	Soft	20	13.4[a]	0.23	5.4[b]	0.10	32.6[a]	0.44	13.8[a]	1.37	83.5	2.10	2.9	0.35	5.4	0.14	69.6[a]	1.04
	Hard	20	12.9[b]	0.19	6.3[a]	0.11	38.4[b]	0.38	8.7[b]	0.95	80.9	2.04	3.1	0.25	5.2	0.22	65.3[bc]	1.03
Small	Soft	20	12.9[a]	0.15	5.5[b]	0.09	33.4[a]	0.35	13.1[a]	1.15	82.0	1.83	3.0	0.23	5.1	0.20	68.7[ab]	0.68
	Hard	20	13.2[a]	0.15	6.4[a]	0.13	38.1[b]	0.31	11.2[a]	1.04	84.7	2.24	2.3	0.38	5.3	0.18	62.6[c]	0.77

[ab] Different letters between texture groups within each probe size batch indicate significant differences ($p<0.05$).
*Mean value for the three anatomical positions. The lean slice thickness is the percentage of pixels containing lean in a line perpendicular to the tray and at 3 cm from the edge of the bone over the number of pixels containing lean and fat. The percentage of the perimeter of the slice covered with subcutaneous fat is the number of pixels of the perimeter of the slice segmented as subcutaneous fat into the total number of pixels of the perimeter.

Table 4. F values and significance (p-values) for differences between texture groups in each textural parameter on the surface of the ham when using different probe sizes in the different anatomical positions (n=20).

Probe size	Anatomical position		F_{max} (N)	A_1 (N.mm)	A_2 (N.mm)	Slope (N/mm)
Small	1	F-value	152.6	178.7	106.6	120.0
		p-value	0.000	0.000	0.002	0.001
	2	F-value	**407.3**	**504.9**	277.0	290.8
		p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001
	3	F-value	240.6	253.5	202.7	190.2
		p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001
Medium	1	F-value	248.2	293.7	261.1	166.0
		p-value	< 0.0001	< 0.0001	< 0.0001	0.000
	2	F-value	302.4	333.7	330.6	290.7
		p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001
	3	F-value	227.5	238.0	250.7	184.8
		p-value	< 0.0001	< 0.0001	< 0.0001	0.000
Large	1	F-value	328.6	309.1	**328.9**	**310.5**
		p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001
	2	F-value	157.2	143.8	134.4	210.0
		p-value	0.000	0.000	0.001	< 0.0001
	3	F-value	267.1	286.8	281.2	232.6
		p-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001

Table 5. Classification scores (% correctly classified in cross-validation) of dry-cured ham production in soft and hard experts' texture groups using weight loss, textural parameters from the surface of the ham and subcutaneous fat thickness.

Used parameters for classification	Used statistical analysis	n	Whole weight loss range of the production (28 to 42%)	n	Medium weight loss range of the production (34.5 to 37.5%)
Weight loss	DA	136	80.4 %	56	66.7 %
Textural parameters (F_{max}, A_1, A_2, slope)	PLS-DA	56	80.4 %	27	70.4 %
Force/time curve	PLS-DA		78.6%		66.7%
Textural parameters (F_{max}, A_1, A_2, slope) + Weight loss	PLS-DA		82.1 %		70.4 %
Textural parameters (F_{max}, A_1, A_2, slope) + subcutaneous fat thickness	PLS-DA		82.1 %		74.1 %
Textural parameters (F_{max}, A_1, A_2, slope) + Weight loss + subcutaneous fat thickness	PLS-DA		82.1 %		74.1 %
Force/time curve + Weight loss	PLS-DA		78.6%		66.7%

Textural parameters were obtained using the small probe in anatomical position 2.