# TOWARDS BUILDING OF A "NUMERICAL HAM" FOR SIMULATING PROTEOLYSIS AND SALT AND WATER TRANSFERS DURING THE DRY-CURED HAM ELABORATION PROCESS

R. Harkouss<sup>1\*</sup>, C. Chevarin<sup>1</sup>, P. Gatellier<sup>1</sup>, A. Lebert<sup>2</sup>, J-D. Daudin<sup>1</sup> and P-S. Mirade<sup>1</sup>

<sup>1</sup> INRA, UR370 Qualité des Produits Animaux, Saint-Genès-Champanelle, France

<sup>2</sup> Institut Pascal, UMR6602 UBP/CNRS/IFMA, Aubière, France

Abstract – Salting is essential during the dry-cured ham elaboration process. The reduction of salt content affects the final product quality. This study aimed to build a 3D numerical model which predicts the biochemical evolution and the salt and water distributions during different stages of dry-curing hams. This model will be then used to test scenarios aiming of reducing the salt content without affecting the final product quality. The biochemical measurements and the statistical analyses showed that temperature accelerates the activity of enzymes responsible for the protein degradation, while salting and drying slowed it down; furthermore, the geometrical position of the muscles inside the ham plays an important role on the proteolysis evolution during the process. The first approach of the simulation showed a good prediction of salt penetration towards the inner zones as well as of water migration to the outside of the ham. The calculated proteolysis index (PI) values were very close to PI experimental measurements made on Bayonne dry-cured ham samples.

Key Words – Dry-cured ham, Simulation, Salt reduction, Transfers, Proteolysis

## I. INTRODUCTION

Generally talking, although salt is important for regulation of blood pressure, an excessive sodium intake may lead to hypertension and increases the risk of stroke and cardiovascular diseases. For that, the major issue in food industry is to decrease the salt content without altering the final product quality. In this perspective that comes the french "Na moins" project, involving studies on sodium reduction in dry-cured and cooked hams to obtain healthier products. In dry-cured ham elaboration, salt affects both quality and safety of the final product: sensory properties and microbiological stabilization (Benedini et al. [1], Taormina [2]). Furthermore, the final product quality is affected by proteolysis evolution (Toldra et al. [3]) which depends on several factors, like temperature and water content (Arnau et al. [4], Serra et al. [5]). This study aims first to describe the proteolysis evolution as a function of temperature and water and salt contents; and second, to combine these proteolysis models with salt/water diffusion and heat transfer models into COMSOL Multiphysiscs 4.3a software, to obtain finally a "numerical ham". The global objective is to build a 3D numerical model, describing the proteolysis evolution, combined to salt penetration, water migration and heat transfer phenomena occurring during all stages of dry-curing ham elaboration. It would help research and technical institutes to try different scenarios in order to reduce the final salt content in dry-cured ham, without altering the final quality.

## II. MATERIALS AND METHODS

The preparation of small samples mimicked closely the different steps used in dry-cured ham processing. Five different muscles were studied: Biceps femoris (BF), Semimembranosus (SM), Semitendinosus (ST), Gluteus medius (GM) and Rectus femoris (RF). Figure 1 resumes the experimental protocol of this preparation.

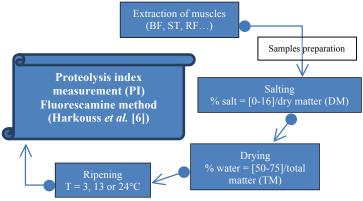


Figure 1. Samples preparation protocol

The based studv was on Doehlert-type experimental design, built with three factors: temperature (3 levels), water content (5 levels) and NaCl content (7 levels). Using this design reduced the final number of kinetics from 105 to 15 per muscle (13, plus 2 repetitions of the center of Doehlert design). The proteolysis index (PI) of samples was determined using the fluorescamine method (Harkouss et al. [7]). The rapidity and the specificity of this method make it a good choice for testing ham quality. For more details about the experimental protocol of samples preparation and the fluorescamine method, please refer to [6]. Using the version "2.12.11" of the software R, an ANOVA and a multiple linear regression (MLR) were performed on all slopes of all the kinetics obtained for the 5 muscles to get models relating the proteolysis evolution to temperature and water and salt contents for each type of muscle.

The obtained statistical models were coupled with other physical models (mass and heat transfers) into Comsol Multiphysics 4.3a software. For that, a segmentation of 181 X-ray tomography images of green ham was made using the software Mimics, to obtain the geometry of a complete ham. This 3D ham geometry was then smoothed many times and meshed with approximately 111000 tetrahedral meshes, before being imported to Comsol Multiphysics software (Figure 2).

Figure 2. Pork ham meshing

Both Fourier and Fick laws were introduced in the numerical simulation using Comsol to model heat and mass transfers, respectively, as well as the function of proteolysis evolution statistically obtained.

The Fourier equation is:

 $\rho^*Cp^*\Delta T/\Delta t - k^*\Delta^2 T = 0$  (1) where  $\rho$  is density, Cp is the specific heat capacity and k is

the thermal conductivity of ham. The initial temperature was the temperature of green ham at the beginning of the process/salting stage, i.e.  $3^{\circ}$ C. The mass transfer models (for salt or water) followed the Fick equation:

 $\Delta C/\Delta t - D^*\Delta^2 C = 0$  (2) where C is the salt or water content and D is the diffusion coefficient of salt or water. The initial salt content was 0% MT (beginning of salting stage) and the initial water content was 75% MT (fresh meat).

The phenomenological models of proteolysis velocity followed the obtained function as:

Vp = f (temperature, salt and water contents).

The first approach of calculation aimed to simulate only the salting and the post-salting stages at low temperature (3°C) without taking into account any ham volume variation resulting from either the swelling of the meat structure due to salt penetration or the reduction due to water evaporation (air drying). Respect to the limited input salting method used widely in industry, salting was for 15 days with a brine solution of C<sub>0</sub> = 280 g salt/l. It was followed by 55 days of postsalting, where salt diffused inside the ham without any salt adding at the surface. For that purpose, several boundary conditions were imposed on the different physical models as reported in Figure 3.

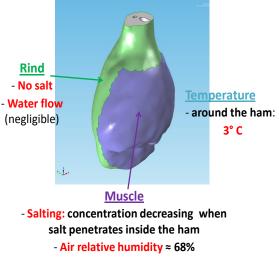


Figure 3. Boundary conditions used for modelling the first 70 days in the dry-curing ham elaboration process.

# III. RESULTS AND DISCUSSION

The biochemical measurements showed the effect of temperature, drying and salting on the proteolytic evolutions in pork meat. As an example, Figure 4 shows both effect of salting and drying, at 13°C, in the case of ST muscle.

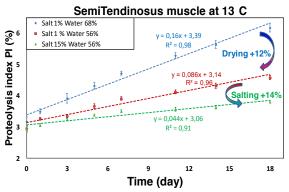


Figure 4. Proteolysis kinetics of ST muscle, at 13°C.

In Figure 4, the blue and red curves show the effect of the drying (water content at 68% and 56% TM, respectively, and salted at 1% DM) and the red and green curves show the effect of salting (salt content at 1% and 15% MS, respectively, and dried at 56% TM) on the enzymes proteolytic activity in ST muscle. These results indicate that drying acts as an inhibitor for the protein degradation. The graph also shows that salting is an important stage in the dry-curing ham, since it slows down the proteolysis. In general, biochemical results proved that proteolysis is (1) strongly accelerated by increasing temperature, (2) slowed down by the reduction of water content due to the drying and (3) effectively inhibited by the addition of a high quantity of NaCl.

It is clear from Figure 4 that kinetics were well fitted by straight lines, where slopes represented the proteolysis velocity. Statistical analyses treated all slopes of all kinetics and revealed that the studied factors as temperature, salt and water contents had significant effect on the evolution of the proteolysis; on the other hand, the muscle type had not shown any significant effect on proteolysis for BF, SM and ST muscles (RF and GM muscles having their own behavior). MLR allowed to obtain the coefficients needed to build the proteolysis velocity models for the five types of muscles. As an example, the proteolysis evolution model of ST muscle is described as: Slope (IP) =  $4.84.10^{-3} - 9.18.10^{-3} \text{ T} - 7.28.10^{-4} \text{ S} +$  $1.69.10^{-4}$  W -  $3.92.10^{-4}$  T.S +  $3.25^{-4}$  T.W +  $5.21.10^{-5}$  S.W (3) where T is the temperature, S is the salt content and W is the water content.

Figure 5 is an illustration of the response surface of ST muscle at 2% salt (DM), as function of temperature and water content. It shows clearly that by raising temperature, proteolysis greatly progressed and by reducing water, proteolysis decreased.

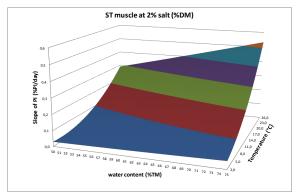


Figure 5. Response surface of proteolysis velocity of ST muscle at 2% salt (DM)

Figure 6 shows the results of the first simulation approach of salting and post-salting.

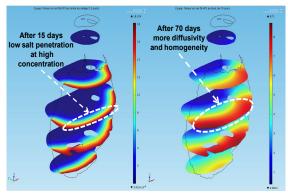


Figure 6. Salt distribution after 15 and 70 days of process.

After 15 days, the salt penetration was just observed in the first few cm from the surface, but the concentration was relatively high (16-18% of TM); whereas the salt concentration was very low in the middle and negligible near to the fatty region inside the ham. At the end of postsalting (70 days), salt showed more ability to penetrate deeply towards the middle region and inside the ham. The salt content obviously decreased at the surface (7-8% TM), gently increased in the middle (4-5% TM) and slightly inside the ham. At the end of salting, the mean average of salt content was approximately 4% of TM equivalent to a salt intake of 408 g for a green ham weighing 10 kg, which is in agreement with the industrial practice.

Figure 7 highlights the heterogeneity of salt and water distribution, as well as the proteolysis evolution inside the ham after 70 days.

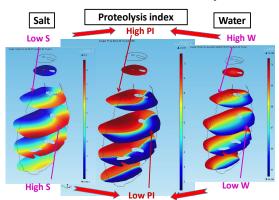


Figure 7. Proteolysis evolution, salt and water diffusion models, after 70 days of process

Results showed that low salt content (1-2% of TM) and high water content (70% of TM) inside the ham favored proteolysis, with PI values reaching 10%. On the other hand, a higher salt content (6-8% of TM) and a lower water content (65% of TM), as near the surface of the ham - the muscle exposed to the air dries faster than that surrounded by fat near to the rind inside - slowed down the proteolytic evolution thus PI did not exceed 3-4%, which is considered as a low value by comparing with the inner PI values. These calculated PI values were in agreement with those measured on industrial Bayonne drycured ham samples taken after post-salting stage.

## IV. CONCLUSION

Experimental results and statistical analyses confirmed that high temperature and water content favored proteolysis, whereas high salt content slowed it down. Furthermore, although the difference of physico-chemical properties between different types of muscles influences the protein degradation, the geometrical position of the muscle in the ham (SM near to surface and BF deeply inside) also affects the proteolytic evolution, due to the various salt and water transfer behaviors in each of them during the drying and curing of hams. The first results of the "3D numerical ham" showed a good prediction of the distribution of salt and water contents. The calculated PI values were in agreement with the experimental results already obtained and in line with the PI measurements made on industrial drycured hams samples at the same processing time.

The development of this model combining the salt and water transfers with the statisticallydetermined models of proteolysis evolution will be improved by introducing a function to predict the evolution of water activity and by taking into account the geometry of the different muscle types forming the ham as well as the volume reduction (up to 30%) due to drying which is the main phenomenon affecting the ham volume. The complete 3D numerical model would be very useful to attempt the global objective mentioned at the end of the introduction.

## ACKNOWLEDGEMENTS

This work was funded by the Na<sup>-</sup> integrated programme (ANR-09-ALIA-013-01) financed by the French National Research Agency.

## REFERENCES

- Benedini, R., Parolari, G., Toscani, T. & Virgili, R. (2012). Sensory and texture properties of Italian typical dry-cured hams as related to maturation time and salt content. Meat Science 90: 431–437.
- Taormina, P. J. (2010). Implications of salt and sodium reduction on microbial food safety. Critical Reviews in Food Science and Nutrition, 50: 209–227.
- 3. Toldrá, F. & Flores, M. (2000). The use of muscle enzymes as predictor of pork meat quality. Food Chemistry 69: 387-395.
- 4. Arnau, J., Guerrero, L. & Gou, P. (1997). Effects of temperature during the last month of ageing and of salting time on dry-cured ham aged for six months. Journal of the Science of Food and Agriculture 74: 193–198.
- Serra, X., Ruiz-Ramirez, 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.
- Harkouss, R., Mirade, P-S. & Gatellier, P. (2012). Development of a rapid, specific and efficient procedure for the determination of proteolytic activity in dry-cured ham: Definition of a new proteolysis index. Meat Science 92: 84–88.