# ASSOCIATION BETWEEN MEAT QUALITY, RESIDUAL GLYCOGEN AND PROTEIN STATUS

V. Santé-Lhoutellier<sup>1\*</sup>, P. Gatellier<sup>1</sup>, S. Traoré<sup>1</sup>, K. Kajak-Siemaszko<sup>2</sup>, D. Jaworska<sup>2</sup>, W. Przybylski<sup>2</sup>, D. Kołożyn-Krajewska<sup>2</sup>

<sup>1</sup> INRA, UR370 Qualité des Produits Animaux, F-63122 Saint Genès-Champanelle, France

<sup>2</sup>Warsaw University of Life Sciences – SGGW, Faculty of Human Nutrition and Consumer Sciences, Nowoursynowska 159c Street, 02-776 Warsaw, Poland

\* Corresponding author (phone: +33-473-062-4708; fax: +33-473-624-268; e-mail: veronique.sante@clermont.inra.fr)

*Abstract* – We aimed to analyse the relationship between technological and sensory quality of meat and the effect of residual glycogen on protein status (hydrophobicity, protein aggregates and myofibrillar protein oxidation) after cooking. The research was carried out on 24 P76 synthetic line of pigs (12 males and 12 females). Slaughter value of carcass was evaluated by CGM apparatus. Meat quality was evaluated on the basis of pH value (measured in 1, 3 and 24 h post mortem), meat colour (in Lab system), natural drip loss and cooking yield. Intramuscular fat and glycolytic potential (in them residual glycogen) was also determined. Sensory quality of meat 26 h *post mortem* after heat treatment was estimated on the basis of: odour, intensity and homogenity of meat colour, tenderness and juiciness, fat perceptibility and flavour. In cooked meat protein oxidation and aggregates and also hydrophobicity were evaluated. The results showed that sensory quality of meat depends mainly on the level of intramuscular fat, ultimate pH and natural drip loss. Significant relationships between glycogen, ultimate pH, drip loss and marbling and protein status after cooking were also observed. More particularly, we found that protein oxidation process impacted sensorial attributes such as odour and colour of meat after cooking.

Index Terms - pig quality, glycogen, oxidation of protein, aggregation of protein

## I. INTRODUCTION

Pork meat quality depends on genetic and environmental factors, which influenced technological and sensory pork quality, mainly by glycogen content, intramuscular fat level, extend and rate of *post mortem* glycogenolysis and also ante mortem and slaughter condition [1], [2], [3]. As shown in many studies glycogen level plays a key role *in post mortem* glycogenolysis that determined many meat quality traits. Glycogen content influences ultimate pH which determined many meat quality traits as colour, drip loss, water holding capacity, cooking loss and sensory attributes.

Myofibrillar proteins are the major constituents of muscle proteins and play a key in tenderness determinism and water holding capacity of meat. Therefore, information related to their denaturation or modification pattern is very important in meat technology. Endogenous proteolytic enzymes, including hydrolysis of structural proteins, influence also meat tenderisation. Proteolysis in meat is mainly affected by cathepsins, calpains, and also by proteasome activity [4], [5]. Postmortem changes in muscle impact the antioxidant defense system [6], [7] and therefore the degree of lipid and protein oxidation [8]. As mentioned by [9] heat treatment of food rich of protein could induce reactions between peptides and aminoacids with carbohydrates and oxidized lipids and protein. Oxidative processes are known to be the major cause of meat quality deterioration, affecting flavour, colour and nutritional composition.

The negative effect of residual glycogen on technological yield of cooked ham processing was demonstrated by [10], [3], [11]. [12] showed the negative effects of high residual glycogen on beef qualities. This residual glycogen is also a source of glucose in meat, which could potentially promote molecular interactions.

The aim of the study was to analyse the relationship between technological and sensory quality of meat and the effect of residual glycogen on protein status (hydrophobicity, protein aggregates and myofibrillar protein oxidation) after cooking.

# II. MATERIALS AND METHODS

The research was executed on 24 pigs (12 males and 12 females) of P76-PenArLan hybrids line. The animals were produced and reared under the same conditions and were fed a standard diet. They were slaughtered at about 105 kg live

weight in commercial slaughterplant in accordance to the legally binding procedure (the distance from the farm to the slaughterhouse 200km, a rest of about 2h, automatic electric stunning of high voltage and exanguination in a vertical position). Backfat and loin thickness were measured with CGM apparatus on the 7<sup>th</sup> rib. On the basis of this measurement meatiness was evaluated according to [13]

Colour parameters (CIE L\*a\*b\*) were assessed using CR310 Minolta 48h after slaughter. The decline of pH was measured at 1, 3 and 24h *post mortem*. Drip loss was determined at 48h *post mortem* according to [14]. Glycogen, glucose and glucose-6-phosphate after glycogen hydrolysis with amyloglucosidase [15] and lactate [16] in the muscle were also determined. On the basis of them the glycolytic potential (GP) was calculated according to [17]. Glucose content was measured 48 h post mortem using glucometer Glucard 2, GT12 (KDK, Corporation, Japan). Muscle fat was determined according to Soxhlet's method (PN-73/A-85111 Polish Norm). Cooking yield of meat was determined by subjecting 500g of meat sample to heat treatment until the core of the sample reached 72°C.

The sensory quality of meat (96 h after slaughter) was determined after heat treatment – cooking in a salt solution (0,8% NaCl) according to [18] method. The heat treatment process was conducted to achieve a temperature of  $72^{\circ}$ C inside the meat, and then the samples were covered and kept until reaching a temperature of  $75^{\circ}$ C. After cooling the meat samples were evaluated in terms of odour of cooked meat, intensity and homogeneity of colour, marbling, sensory fat perceptibility, tenderness and juiciness. On the basis of the above mentioned quality characteristics the overall sensory quality was evaluated. For sensory assessment the sensory scaling method was used, an unstructured graphical scale with precisely determined edge definitions was applied.

After tenderization of meat (in 96 h after slaughter) meat was treatment to thermal processing by contact grill in controlled temperature during 3-4 minutes (for 72°C inside sample). In these meat protein carbonyl groups were evaluated by the method of [19] with slight modifications for measurement in meat samples [20]. Hydrophobicity of myofibrillar proteins was determined using the hydrophobic chromophore bromophenol blue (BPB) according to [21] with slight modifications. Protein aggregates were detected by a front face fluorescence technique. Before measurement, myofibrillar protein concentration was adjusted to 0,04 mg/mL of 20 mM phosphate buffer at pH6. The fluorescence was measured with a standard spectrofluorometer Perkin-Elmer LS 50B fitted with a front surface accessory (Perkin-Elmer Plate Reader).

Specific protein oxidation was evaluated by labeling protein carbonyls with DNPH followed by immunobloting (Oxyblot kit from Chemicon International) of proteins separated by 12.5 % SDS-PAGE [22]. Image analysis was performed using Quantity One software (Biorad)

The results were analysed using Statistica 6.0 PL system and the correlations between analysed traits were calculated.

## **III. RESULTS AND DISCUSSION**

Carcass quality, technological and sensory qualities of meat are presented in Table 1. We observed a high meatiness, normal ultimate pH value, high cooking yield (CY), low drip loss, appropriate sensory qualities. Protein oxidation evaluated by carbonyl group content are similar to values reported by [23] for lamb meat after 7 day storage and lower than reported by [24] after 10 days of meat ageing. Numerous significant correlations were obtained between analysed traits. In order to clarify we mentioned in this paper few of them. It was observed that the level of IMF and ultimate pH and also drip loss was correlated with sensory quality of pork. High drip loss resulted statistically significant (r=-0.61; p<0.05) in lower juiciness. Similar observation was reported by [25]. The authors observed that the level of IMF in meat was related with higher juiciness and overall sensory quality in pork meat. Glycolytic potential represents the amount of glycogen present in the muscle at time of slaughter. In our study a relatively high drip loss in meat was related to higher level of glycolytic potential. Such a correlation was also reported by [26]. The results showed that meat with high level of glycogen at slaughter lead to higher residual glycogen. We observed also a positive correlation between level of glucose in meat juice and residual glycogen (r=0.82; p<0.01). Interestingly a significant relationship between glucose level and protein aggregates was also found (r=0.54, p <0.05).

Protein oxidation is positively correlated to drip loss (r=0.43; p<0.05), marbling (r=0.57; p<0.05) and glycogen level (r=0.45; p<0.05). A negative relationship between protein aggregates and marbling of raw meat was noticed (r=-0.47; p<0.05). This result could be partially explained the fact that generally IMF and glycogen are negatively correlated in meat [27]. Protein oxidation and more particularly oxidized actin was promoted in our case by higher pH [5] mentioned that high pH favors  $O_2^-$  production in meat. Protein oxidized products were positively correlated to the level of glucose (r=0.42; p<0.05) and drip loss (r=0.72; p<0.01). The results showed also a relationship between sensory attributes such as odour of cooked meat and carbonyls content (r=0.65; p<0.05) and colour and protein oxidized products (r=0.67; p<0.05).

Traits	Average	SD
Hot carcass weight- HCW (kg)	86,31	8,92
Loin thickness - LD (mm)	65,17	4,79
Backfat thickness – BF (mm)	12,30	2,38
Meatiness (%)	60,02	1,67
pH 1	6,50	0,21
pH <sub>3</sub>	6,33	0,21
pH <sub>24</sub>	5,60	0,07
Colour L	55,24	1,72
a	16,08	0,89
b	9,28	0,77
Cooking yield - CY	72,12	1,48
Drip loss <sub>48</sub> (%)	3,42	1,85
IMF (%)	1,50	1,15
Glycolytic potential (µmol/g)	128,24	16,98
Residual glycogen (µmol/g)	16,41	6,47
Lactate (µmol/g)	95,41	9,70
Glucose in meat juice (mg/dl)	148,77	41,21
Eating quality of cooked meat 96h	post mortem [0	-10c.u.]
Odour	7,76	0,21
Homogenity of colour	8,17	0,31
Ton of colour	7,97	0,36
Marbling	2,59	0,89
Tenderness	6,30	1,18
Juiciness	4,51	0,84
Fat perceptibility	2,30	0,29
Flavour	6,40	0,57
Overall quality	6,15	0,78
Protein status of cook	ked meat	
Hydrophobicity (µg bound BBP)	75,78	1,63
Carbonyls (nmol carbonyls/mg protein)	3,12	1,47
Protein aggregates (FI)	452,65	98,53
Myosin oxidized (AU)	5,55	0,73
Actin oxidized (AU)	3,99	1,46
Oxydation products (AU)	7,44	4,30

Table 1. Characteristics of carcass slaughter parameters, technological value, sensory attributes and protein status of meat.

## **IV. CONCLUSION**

We showed that sensory quality of meat depends mainly of intramuscular fat, ultimate pH and natural drip loss. Significant relationships between glycogen, ultimate pH, drip loss and marbling and protein status after cooking were also observed. We also found that protein oxidation process impacted sensorial attributes such as odour and colour of meat after cooking. Glycogen is known to affect technological meat quality, but in our experiment, we showed it may impact also the nutritional value of meat. Indeed glycogen seemed to favor oxidative processes, either by forming glucose when meat is heated or by enhancing protein interactions.

#### ACKNOWLEDGEMENT

The authors thanks Egide project "POLONIUM" for their support in this joined French and Polish experiment.

### REFERENCES

[1] De Vries, A. G., Faucitano, L., Sosnicki, A. & Plastow, G. S. (2000). The use of gene technology for optimal development of pork meat quality. Food Chemistry 69, 397-405.

[2] Rosenvold, K. & Andersen, H. J. (2003) Factors of significance for pork quality - review. Meat Science 64, 219-237.

[3] Przybylski, W., Sieczko, L., Jaworska, D., Czarniecka-Skubina, E. & Niemyjski, S. (2007). Estimation of conditionality of pork sensory quality by using multivariate analysis. Archiv Tierzucht, 50,125-135.

[4] Santé-Lhoutellier, V., Aubry, L., & Gatellier, P. (2007). Effect of oxidation on in-vitro digestibility of skeletal muscle myofibrillar proteins. *Journal of Agricultural and Food Chemistry*, 55, 5343–5348.

[5] Chelh, I., Gatellier, P. & Santé-Lhoutellier, V. (2007). Characterisation of fluorescent Schiff bases formed during oxidation of pig myofibrils. *Meat Science*, 76, 210-215.

[6] Renerre, M., Dumont, F., & Gatellier, P. (1996). Antioxidant enzyme activities in beef in relation to oxidation of lipid and myoglobin. *Meat Science*, 43, 111-121.

[7] Renerre, M., Poncet, K., Mercier, Y., Gatellier, P. & Metro, B. (1999). Influence of dietary fat and vitamin F on antioxidant status of muscles of turkey. *Journal of Agricultural and Food Chemistry*, 47, 237-244.

[8] Morzel, M., Gatellier, P., Sayd, T., Renerre, M. & Laville, E. (2006). Chemical oxidation decreases proteolytic susceptibility of skeletal muscle myofbrillar proteins. *Meat Science*. 73, 536-543.

[9] Sikorski, Z. E (2007). Chemia żywności, t. 2 Sacharydy, lipidy, białka, Wyd. WNT, Warszawa (in Polish)

[10] Fernandez X., Monin G., Talmant A., Mourot J. & Lebret B. (1999). Influence of intramuscular fat content on the quality of pig meat – 2. Consumer acceptability of m. *longissimus lumborum. Meat Science* 53, 67-72

[11] Przybylski, W., Jaworska, D., Czarniecka-Skubina, E., Kajak-Siemaszko, K. & Wachowicz, I. (2007). Using of multidimensional analysis in the evaluation of technological value and sensory quality of pork meat. *Polish Journal of Food and Nutrition Science*, Vol. 57, No. 4(B), 449-455.

[12] Immonen, K., Ruusunen, M. & Puolanne, E. (2000). Some effects of residual glycogen concentration on the physical and sensory quality of normal beef. Meat Science, 55, 33-38.

[13] Borzuta, K. (1998) - The study of variable methods usefulness to meatiness estimation in Europ system. Roczn. Inst. Przem. Mięs 35, 2, 1.

[14] Prange, H., Juggrt, L. & Scharner, E. (1977). Untersuchungen zur Muskel fleischqualitaet beim Schwein. Archives Experim. Vet. Med. 30(2) 235-248.

[15] Dalrymple, R. H. & Hamm, R. (1973). A method for the extraction of glycogen and metabolites from a single muscle sample. J. Food Technol., 8, 439-444.

[16] Bergmeyer, H. U. (1974). Methods of Enzymatic Analysis. Academic Press, New York, pp. 1127, 1196, 1238, 1464.

[17] Monin, G. & Sellier, P. (1985). Pork of low technological quality with a normal rate of muscle pH fall in the immediate post-mortem period: The case of the hampshire breed. *Meat Science*, 13, 49-63.

[18] Baryłko-Pikielna, N., Kossakowska, T. & Baldwin, Z. (1964). The choice of optimal method of samples beef and pork meat preparation for sensory analysis. *Roczn. Inst. Przem. Mięs.* 1, 111-131 (in Polish).

[19] Oliver, C. N., Alin, B. W., Moerman, E. J., Goldstein, S., & Stadtman, E. R. (1987). Age-related changes in oxidized proteins. Journal of Biological Chemistry, 262, 5488-5491.

[20] Mercier, Y., Gatellier, P., Viau, M., Remignon, H., & Renerre, M. (1998). Effect of dietary fat and vitamin E on lipid and protein oxidation in turkey meat during storage. *Meat Science*, 48, 301-317.

[21] Chelh, I., Gatellier, P. & Santé-Lhoutellier, V. (2006). Technical note: A simplified procedure for myofibril hydrophobicity determination, *Meat Science*, 74, 681-684.

[22] Santé-Lhoutellier, V., Aubry, L. & Gatellier, P. (2007). Effect of oxidation on in vitro digestibility of skeletal muscle myofibrillar proteins. Journal of Agricultural Food Chemistry 55(13), 5343-5348.

[23] Santé-Lhoutellier, V., Engel, E. Aubry, L., & Gatellier, P. (2008). Effect of animal (lamb) diet and meat storage on myofibrillar protein oxidation and in vitro digestibility *Meat Science* 79, 777-783.

[24] Martinaud, A., Mercie, Y., Marinova, P., Tassy, C., Gatellier, P., & Renerre, M. (1997). Comparison of oxidative process on myofibrillar proteins from beef during maturation and by different model oxidation system. *Journal of Agricultural and Food Chemistry*. 45, 2481-2487

[25] Jaworska, D., Przybylski, W., Kołożyn-Krajewska, D., Czarniecka-Skubina, E., Wachowicz, I., Trząskowska, M., Kajak, K., Lech, A. & Niemyjski, S. (2006). Relationships between traits determining technological and sensory quality of pork. *Animal Science Papers and Reports*, 24 Sup.2, 121-135,

[26] Krzecio, E., Miszczuk, B., Podsiadla-Kocwin., Sieczkowska, H., Zybert, A., Antosik, K. & Lyczynski, A. (2006). Slaughter value of carcasses and technological quality of meat from porkers differing in drop loss from the *longissimus lonborum* muscle. *Animal Science papers and Reports*. 24(2) 159-169

[27] Przybylski, W., Gromadzka-Ostrowska, J., Olczak, E., Jaworska, D., Niemyjski, S. & Sante-Lhoutellier, V. (2009). Analysis of variability of plasma lepton and lipids concentration In relations to glycolytic potential, intramuscular FAT and meat quality In P76 pigs. J. Anim. Feed Sci. 18, 296-403.