INFLUENCE OF GENDER, GENETIC BACKGROUND AND REARING METHOD ON THE RELATIONSHIP BETWEEN PROTEIN EXPRESSION AND MEAT QUALITY

A. Kwasiborski*[1], T. Sayd[1], C. Chambon[1], V. Santé-Lhoutellier[1], D. Rocha[2] and C. Terlouw[1]

[1] *INRA, Unité Qualité des Produits Animaux, Centre de Theix, 63122 St Genès Champanelle, France*
[2] *Genus plc, Genus Cambridge Research Laboratory, Department of Pathology, University of Cambridge CB2 1QP Cambridge, United kingdom. Email: anthony@clermont.inra.fr*

Keywords: pigs, proteome, meat quality, *longissimus dorsi*

Introduction

With a mean consumption of 43.5kg per year and per capita in 2001, pork is the most consumed meat in Europe (Devine, 2003), but its technological and sensory qualities are still insufficiently controlled. Rearing method, breed and gender influence raw material and end-product qualities. Meat quality depends on post-mortem metabolism, involving many proteins: enzymes, transport, stress or chaperone proteins. This study evaluated effects of rearing method, genetic background and gender on protein expression and its relationship with meat quality in pigs.

Materials and Methods

Twelve castrated male and 12 female pigs, sired by Duroc or Large White PIC purebred, were reared either outdoors or indoors in a 2 x 2 x 2 factorial design. Dams were Large White x Landrace. Immediately after slaughter, *Longissimus lumborum* (LL) samples were taken to determine various meat quality parameters including ultimate pH, colour (L*, a*, b*), and drip loss at 3 days and cooking loss. LL proteomic maps were established with 2-dimensional electrophoresis, using an immobilised pH 5-8 gradient for the first dimension and a SDS-PAGE gel for the second dimension. After staining with Coomassie blue, each gel was scanned and intensity of spots was evaluated using PDQuest software. The identifications were made by MALDI-ToF mass spectrometry.

Results and Discussion

ANOVA found that for 122 of a total of 248 spots intensity varied significantly: 18 were influenced by rearing method, 10 by breed, 90 by gender and 32 by various interactions between these factors (Table 1). Eighty spots were identified and could be classified in different categories including glycolysis enzymes, stress and chaperone proteins.

Table 1: Number of spots of which intensity varied due to rearing method, sire breed or gender(left) and due to their interactions (right).

	Factors		
	rearing	sire	gender
Total	18	10	90
Spots affected by one factor	12	6	74

	Interactions			
	rearing x sire	rearing x gender	sire x gender	rearing x sire x gender
Total	8	10	10	4
Spots affected by interactions of factors	4	4	7	2

ANOVA (Table 2) found that ultimate pH was influenced by gender and by the interaction between rearing method and breed. Drip loss was influenced by breed. The other meat qualities were not influenced by the treatment factors.

Table 2: Means and significant treatment effects on meat quality parameters.

	Mean by factors						influencing factors	
	rearing		sire		gender			
Variable	Outdoor	Indoor	Duroc	Large White	Male	Female	Gender p=0.035	Rearing x sire p= 0.050
Ultimate pH	5,56	5,53	5,56	5,53	5,63	5,47		
L*	53,01	51,74	52,07	52,68	51,48	53,27		
a*	8,10	8,50	8,35	8,24	7,96	8,64		
b*	4,84	4,02	4,37	4,49	4,10	4,76		
Drip day 3	4,52	4,17	3,38	5,21	3,59	5,15	sire p=0.020	
Cooking loss	18,48	19,85	18,11	20,23	18,31	20,03		

Multiple forward stepwise regression analysis was used to identify proteins most strongly correlated with meat quality parameters. When means of explanatory or explained variables varied according to treatment group, or when their regression coefficients differed (Figure 1), multiple regressions were carried out separately for each group. Explanatory variables were retained when $p<0.05$. In castrated males, 72% of ultimate pH variability was explained by 2 unidentified proteins. In females, a fragment of the creatine kinase and one other unidentified protein explained 76% of ultimate pH variability (Table 3). Significant multiple regression models were also found for the other meat quality parameters (Table 3).

As these parameters are also influenced by ultimate pH (Renerre, 1990; Offer and Cousins, 1992), the multiple regression models were tested again adding ultimate pH as a possible explanatory variable. Ultimate pH was retained in 3 regressions in addition to the proteins (Table 3). For example, in outdoor pigs, intensity of a myoglobin chemical form explains 57% of L* variability, addition of ultimate pH improves the model with 14% (Table 3). The relationship between L* and myoglobin is well known and related to the proportions of oxymyoglobin MbO_2 and metmyoglobin MetMb, which depend on the activity of oxygen-consuming enzymes and on MetMb reducing activity (Lindhal et al, 2001). L* is low when myofilament spacing is low due to high ultimate pH (Bendall and Swatland, 1988). For indoor pigs, L* is uncorrelated with this myoglobin chemical form, suggesting that the latter is not correlated with relative amounts of MbO_2 and MetMb.

Table 3: Proteins explaining the meat quality variations. Arrows indicate model improvement after inclusion of ultimate pH.

variables	experimental group	retained proteins	Correlation coefficient R	% variability explained
ultimate pH	Male	5021= unidentified	0.73	49
		4305= unidentified	0.73	72
		7220=Creatine kinase	-0.39	81
		8026=unidentified	0.46	90
	Female	7220=Creatine kinase	-0.81	62
		3101= unnidentified	0.74	78
L*	Indoor	None		0 ⟶ 56
	Outdoor	6004= Myoglobin	-0.78	57 ⟶ 71
a*	Male	7107= Creatine kinase	0.72	46
		5021=unidentified	-0.67	81
	Female	38= Ca2+ binding protein	0.69	42
		3303= Guanosine diphosphate dissociation inhibitor	-0.01	62
		5021= unidentified	-0.49	79
		7107= Creatine kinase	-0.63	87
		4408= Leucine aminopeptidase	-0.58	93
b*	Male	6211= unidentified	0.83	67 ⟶ 86
	Female	6117= unidentified	-0.75	51
Drip day 3	Large White sire	6106= Creatine kinase	0.83	62
		1425= unidentified	0.66	94
		8026= unidentified	0.48	99
	Duroc sire	None		0
Cooking loss	Male	2004= Dj1 protein	-0.74	51
		2615= Heat shock protein 70	-0.43	73
	Female	5613= Transferrin	-0.68	41
		7019= Glyceraldehyde-3-phosphate dehydrogenase	0.18	60

Figure 1: Scatter plot and regression lines for cooking loss and transferrin expression.

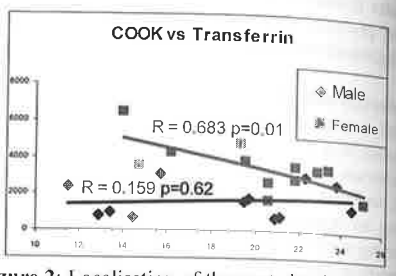
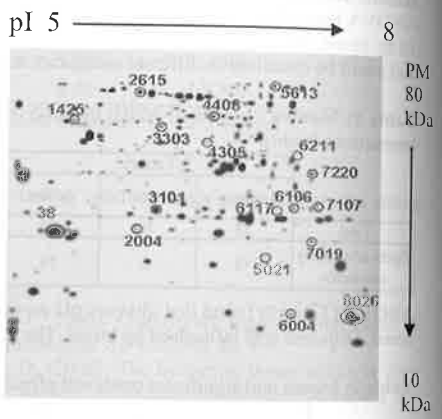


Figure 2: Localisation of the spots implicated in the meat quality variability



Conclusion

Protein expression may explain up to 99% of the variability in certain meat quality parameters. Relationships between protein expression and meat quality depend on gender, breed and rearing method. This study is a first step towards a better understanding of the role of certain proteins in meat quality.

References

Bendall JR. and Swatland HJ (1988). A review of the relationships of pH with physical aspects of pork quality. *Meat Science*, 24: 85-126.

Devine R. (2003). La consommation des produits carnés. INRA production Animal, 16 (5) : 325-327.

Ledward DA. (1992). Colour of raw and cooked meat. In Johnston DE, Knight MK & Ledward DA. The chemistry of muscle-based foods (pp 128-144). Cambridge: Royal Society of chemistry.

Lindhal G., Ludstrom K. and Tornberg E. (2001). Contribution of pigment content, myoglobin forms and internal reflectance to the colour of pork loin and ham from pure breed pigs. *Meat Science*, 59: 141-151.