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# Influence of finishing diet on fatty acid profile in *Psoas major* muscle of Nero Siciliano pig

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**Abstract.** Skeletal muscle is a heterogeneous tissue composed of different fibre types, and differences in meat quality traits between "white" and "red" muscles and between certain breeds are well established. Therefore, the study examined the fatty acid composition of the red (*Psoas major* - PM) muscle in twenty-four Nero Siciliano pigs fed with two different diets. The trial was carried out in the Nebrodi mountain area of Sicily (Italy). During the fattening period, the animals were divided into two groups, kept in two distinct wooded areas of 6 hectares each, and fed exclusively on acorn (A) and germinated barley (B). Results showed that the concentrations of the long chain n-6 and n-3 fatty acids (which were recently found to play an important role in human nutrition) differed significantly between the two groups ( $P=0.015$  for long chain n-6 and  $P=0.048$  for long chain n-3). The linoleic acid (A 5.96 vs B 5.10;  $P=0.006$ ) and  $\alpha$ -linolenic acid (A 0.30 vs B 0.25;  $P=0.028$ ) were higher in group A. Both groups had high concentrations of C18:1 n-9, but in group A (43.27) concentrations were significantly ( $P=0.006$ ) higher than in group B (40.10).

**Keywords.** Nero Siciliano pig – Acorn – Fatty acids – *Psoas*.

## ***Influence de l'alimentation de finition sur le profil en acides gras du muscle Psoas major du porc Nero Siciliano***

**Résumé.** Le muscle squelettique est un tissu hétérogène comprenant divers types de fibres. Les différences existantes entre les muscles « blancs » et « rouges » et entre les races qui influencent les caractéristiques qualitatives de la viande sont bien connues. Le profil en acides gras du muscle rouge (*Psoas major* - PM) de 24 porcs de race Nero Siciliano alimentés avec deux rations différentes de finition a été examiné. L'épreuve a été réalisée en Sicile (Italie), dans le parc des «Nebrodi» ; les animaux ont été subdivisés en deux groupes homogènes et tenus dans deux parcelles boisées différentes de 6 ha chacune, dans lesquelles ils recevaient respectivement glands (A) et orge germée (B). Les résultats obtenus ont mis en évidence que les concentrations en acides gras à chaîne longue n-6 et n-3 (dont le rôle important en nutrition humaine a été vérifié récemment) sont significativement différentes entre les deux groupes ( $P=0,015$  pour le n-6 et  $P=0,048$  pour le n-3). Les concentrations en acide linoléique (A 5,96 vs B 5,10 ;  $P=0,006$ ) et en acide  $\alpha$ -linoléique (A 0,30 vs B 0,25 ;  $P=0,028$ ) ont été plus élevées dans le groupe A. Les deux groupes (glands et orge germée) ont montré des teneurs élevées en C18:1n-9, mais pour A (43,27) elle a été significativement plus élevée ( $P=0,006$ ) que pour B (40,10).

**Mots-clés.** Porc Nero Siciliano – Glands – Acides gras – *Psoas*.

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## **I – Introduction**

The Nero Siciliano pig is a rustic pig breed reared in Sicily (Southern Italy). Compared with industrial pig production systems, the Nero Siciliano pig production process lasts longer. As to the diet, the final months before slaughter represent the crucial phase of the production process, because the feed characteristics (e.g. of acorn, grass or local concentrate feeds) during the critical finishing period significantly affect meat quality. In recent years, the consumers' interest in the so-called 'natural', 'bio' or 'organic' meats has been increasing. Therefore, meat from pig production systems in which pigs are free-range reared and fed on natural feeds with no growth promoters and antibiotics, has become an important field of interest. But the high quality of meat and meat products is the result of various factors, including

genetics, rearing system and muscle type. Muscles are composed of different type of fibres (aW, aR and bR) with different contractile and metabolic properties (Cava *et al.*, 2003). Depending on the types of fibre that constitute a muscle, it differ in fat deposition, heme pigment concentration and phospholipid and fatty acid composition. Meats are more tasty and juicy, and total heme pigments and lipids oxidise faster in oxidative muscles than in glycolytic ones; this contributes to different textural properties (Wood, Wiseman and Cole,1994). The aim of this study was to examine the characteristics and the acidic profile of *Psoas major* muscle, a fast oxidative-glycolytic  $\alpha$ -red muscle, in pigs fed with two different fattening diets.

## II – Materials and methods

Twenty-four “Nero Siciliano” pigs were reared in outdoors in the Nebrodi mountain region of Sicily. Animals were assigned to two groups called Acorn (A) and Barley (B), consisting of 12 animals each, homogenous for sex (castrated males) and body weight (BW,  $79.48 \pm 0.15$  kg). Animals of group A were kept in a wooded area of 6 hectares, appropriately enclosed, and fed with acorn during the fattening period (90 days). Animals of group B were reared within an open-air system in the same rural region and fed with germinated barley on a basis of 2.5 kg/pig/d during the fattening period. After 90 days and a fasting period of 18 hours (ASPA, 1991), animals were slaughtered. A sample of *Psoas major* (PM) muscle tissue was taken from each of the 24 carcasses. Each sample was examined for its crude fat (AOAC, 2005), followed by an analysis of its acidic composition. The fatty acid compositions were determined on lipids extracted by an automatic extractor Foss Model Soxtec. Fatty acids methyl esters of the intramuscular fat were prepared by direct transesterification with sulphuric acid : methanol 1:2 (Christie, 1993) and analysed using an Agilent Technologies 6890N (U.S.A) gaschromatograph operated with a split/splitless injector, a Gerstel autosampler MPS2 (Germany), a flame ionization detector and fused silica capillary column OMEGAWAX 250 (Supelco, U.S.A.), 30 m x 0.25 mm I.D., 0.25  $\mu$ m film thickness. Column temperature was programmed: initial isotherm of 160 °C (6 min.), increment of 3°C/min and final isotherm of 250°C (30 min.). Temperature of the injector and detector: 250°C. Injection volume: 1.0  $\mu$  L. Carrier gas: helium (1 m L/min). Split ratio: 1:50. Identification of fatty acids was made by comparing the relative retention times of FAME peaks from samples with standards from Supelco (U.S.A.). Peak areas were acquired and calculated by Chemstation software (Agilent, U.S.A.) and expressed in percentage of the total fatty acid identified. On the basis of the fatty acid identified, the quality indices were calculated using the equations proposed by Hlbricht and Southgate (1991). Data were analysed by GLM procedure of SAS (2001).

## III – Results and conclusions

Relative percentages of individual fatty acids in intramuscular neutral lipids fractions of *Psoas major* muscle and of two diets are shown in Tables 1 and 2.

**Table 1. Fatty acid classes in two diets**

Fatty acids	Acorn		Barley	
	Mean	$\pm$ SD	Mean	$\pm$ SD
$\Sigma$ SFA	20.76	0.06	30.31	0.53
$\Sigma$ MUFA	56.43	0.31	17.89	0.07
$\Sigma$ PUFA	22.81	0.37	51.80	0.61
$\Sigma$ PUFA n -3	2.87	0.05	7.18	0.28
$\Sigma$ PUFA n -6	19.93	0.32	44.62	0.33

Results are expressed as percentage of total fatty acid methyl esters identified; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids).

**Table 2. Fatty acid profiles (% methyl esters listed) of intramuscular fat of *Psoas major* muscle from Nero Siciliano pigs fed with different fattening diets**

Fatty Acids	Feeding system				P-value
	Acorn		Barley		
	Mean	±SD	Mean	±SD	
C12	0.07	0.01	0.07	0.01	ns
C14	1.23	0.11	1.17	0.16	ns
C15	0.07	0.02	0.06	0.01	ns
C16	22.87	1.23	23.74	0.90	0.0001
C16:1	3.44	0.34	3.50	0.31	ns
C17	0.30	0.04	0.28	0.04	0.045
C17:1	0.29	0.17	0.29	0.11	ns
C18	13.23	3.08	13.66	1.01	ns
C18:1 n-9	39.58	3.38	43.06	1.48	0.011
C18:1 n-7	7.82	3.33	4.61	0.44	0.016
C18:2 n-6	7.11	1.03	6.19	0.84	0.001
C18:3 n-3	0.36	0.09	0.26	0.03	0.0002
C20	0.26	0.07	0.27	0.06	ns
C20:1 n-9	1.00	0.10	0.93	0.22	ns
C20:2 n-6	0.38	0.05	0.31	0.12	0.006
C20:3 n-6	0.09	0.04	0.11	0.09	ns
C20:4 n-6	1.30	0.65	0.98	0.20	0.005
C22	0.14	0.07	0.09	0.03	0.003
C22:1 n-9	0.08	0.07	0.06	0.01	ns
C22:5 n-3	0.15	0.08	0.18	0.06	ns
C24	0.14	0.03	0.09	0.06	0.0001
C22:6 n-3	0.09	0.04	0.09	0.04	ns
∑SFA	38.42	3.86	39.51	1.73	ns
∑MUFA	52.21	2.95	52.39	1.32	ns
∑PUFA	9.50	1.80	8.16	1.12	0.001
∑PUFA n-3	0.61	0.15	0.53	0.10	0.038
∑PUFA n-6	8.89	1.68	7.63	1.05	0.001
AI	0.45	0.05	0.47	0.03	ns
TI	1.16	0.21	1.22	0.10	ns

Results are expressed as percentage of total fatty acid methyl esters identified. AI, atherogenic index; TI, thrombogenic index; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids).

The polyunsaturated fatty acids (PUFA) showed an increase (P=0.001) in group A compared with group B, accompanied by a decrease in the total saturated fatty acids (SFA). Particularly, the PUFA n-3 group was highest in muscle of pigs fed with acorn (0.61% vs 0.53%; P=0.038) as well as PUFA n-6 group (8.89% vs 7.63%; P=0.001). This difference is due mainly to the essential fatty acids (EFA) (Givens *et al.*, 2006), present in greater proportion in the acorn group. The  $\alpha$ -linolenic acid (C18:3 n-3) and the linoleic acid (C18:2 n-6), in fact, were 0.36% and 7.11% respectively in acorn group and 0.26% and 6.19% in barley group. Also the arachidonic acid (important product from EFA by the action of  $\Delta 5$  and  $\Delta 6$  desaturase and elongase

enzymes), which has various metabolic roles including eicosanoid production (Wood *et al.*, 2008) is increased in pigs fed with acorn (1.30% vs 0.98%;  $P=0.005$ ). The PUFA content in muscle from pig fed with barley (8.16%) is significantly ( $P=0.001$ ) lower than in acorn group (9.50%); this results suggest that in A group, the fast-oxidative metabolism of *Psoas* muscle, was slowed by polyphenols, and/or some derived antioxidant metabolites, present in acorn.

In recent years there has been much interest in the beneficial effects of the very long chain (VLC, carbon chain length  $\geq 20$ ) PUFA, in particular eicosapentaenoic acid (EPA, C20:5) and docosahexaenoic acid (DHA, C22:6). The beneficial effects have been well documented and include anti-atherogenic, anti-thrombotic and anti-inflammatory effects. Crucially, it is now evident that in vivo synthesis of EPA and DHA from dietary  $\alpha$ -linolenic acid (ALNA; C18:3 n-3) is very limited in adult humans, especially in men (Burdge *et al.*, 2003), so it is interesting to examine how increasing intake of ALNA in human diet through animal nutrition. A number of intervention studies do suggest that high intakes of ALNA can beneficially affect a number of CVD risk factors including LDL cholesterol (e.g. Zhao *et al.*, 2004). The result for  $\alpha$ -linolenic acid content in acorns analysed for this study (2.08%) was higher than previously reported (1.8%) by Petrović *et al.* (2004) and higher than amounts of C18:3 n-3 (0.7-1.0%) usually found in acorns of Mediterranean forest oaks (*Q. ilex*, *Q. Rotundifolia* and *Q. suber*) (Cava *et al.*, 1997; López-Bote, 1998). Feeding acorn raised the n-3 content in the meat, being of interest from the consumer's health point of view. Additional researches however are needed.

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