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Free-range rearing increases (n-3) polyunsaturated fatty acids of neutral and polar lipids in swine muscles

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Abstract

This study was conducted to determine the effect of rearing pigs on a free-range system, on fatty acid profile of neutral and polar lipids from *longissimus dorsi* (LD) and *masseter* (MS) muscles, with special reference to n-3 polyunsaturated fatty acids (PUFA). Food sources for free-reared pigs were basically acorn and pasture, which were high in oleic and linolenic acids, respectively whereas, indoors, animals were fed a concentrate high in oleic acid. The predominantly oxidative MS muscle had lower saturated fatty acid (SFA) and higher PUFA contents than LD in neutral lipids (NL), and higher total n-3 PUFA and arachidonic acid contents and lower linoleic acid content in polar lipids (PL). Rearing the animals outdoors significantly increased total n-3 and n-6 PUFA of NL and total n-3 of PL. All n-3 fatty acids detected in PL were significantly higher in free reared animals, including eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids.

Keywords: Free-range rearing; n-3 PUFA; Pasture; Muscle lipids; Phospholipids

1. Introduction

Livestock intensive farming has impressively enhanced performance of farm animals and reduced production costs. However, in the past few years, consumer demand for animal products produced under "organic" or "natural" systems has increased, and nowadays "organic" eggs, milk and meat are not uncommon in supermarkets (Sundrum, 2001). This new consumer demand is provoked by concerns about environment, animal welfare, food safety and food quality (Redman & Holden, 1994).

Consumers believe that organic pork is safer (Cowan, 1998) than intensively reared pork. However, there are few scientific studies about muscle composition of pigs reared under natural conditions to support this idea. In addition, there is a great variability of production systems included under the term "organic". Several traditional pig-rearing systems in Europe could be considered as organic production systems; examples are "Scharrel" pigs (van der Wall et al., 1993) Corsican pigs (Coutron-Gamboti, Gandemer, & Casabianca, 1998), Gascon and Limousin pigs (Simon et al., 1996) and Iberian pigs (Mayoral et al., 1999; Ruiz, Cava, Antequera, Martín, Ventanas, & López-Bote, 1998). All these animal managing systems show considerable differences from intensive farming. Basic features include free rearing in variable extension land, the utilization of autochthonous or assimilated pig breeds and the use of natural resources for the feeding of the animals, pasture being one of the major food sources in this type of management. Fatty acid composition of pasture is characterized by high levels of linolenic acid (C18:3 n-3; Andrés, Cava, Mayoral, Tejeda, Morcuende, & Ruiz, 2001; Ruiz et al., 1998). It is well known that fatty acid composition of the feed strongly influences the fatty acid composition of pig lipids, animals fed on diets high in C18:3 n-3 showing higher contents of most n-3 PUFA in different tissues (Ahn, Lutz & Sim, 1996; Matthews, Homer, Thies, & Clader, 2000). n-3 PUFA are believed to confer benefits to human health (British Nutrition Foundation, 1994). Therefore, pasture intake in pig organic production systems could lead to healthier pork by means of an increase of n-3 PUFA in muscle lipids.

Among the factors affecting composition of pork meat, several studies have shown the influence of muscle

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metabolism on fatty acid composition both triglycerides (Andrés et al., 2001; Morcuende, 2001) and phospholipids (Alasnier, Remignon, & Gandemer, 1996; Andrés et al., 2001). However, there is some controversy about such effects, since some studies indicate a positive influence of the oxidative character of the muscle on the n-3 and n-6 PUFA of phospholipids (Kriketos et al., 1995) whereas others show the opposite effect (Alasnier et al., 1996).

The present study was carried out to investigate the effect of rearing the pigs in natural conditions, pasture being one of the major originators of the fatty acid composition of muscle neutral and polar lipids, with special reference to n-3 PUFA, and to evaluate the differences in fatty acid composition of neutral and polar lipids in two muscles with different metabolic types of fibre profile.

2. Material and methods

2.1. Experiment and animals

This study was carried out with 70 Iberian pigs with an initial weight of 90 ± 5 kg. During the fattening period (60 days previous to slaughter), half the animals of each group were raised outdoors (OUT) in a 30 ha extension land, the exclusive feed source being acorn and pasture. The other half was raised indoors (IND) in a 6 ha extension land and fed on mixed diets. All the animals were slaughtered at about 150 ± 10 kg. The chemical composition of diets was determined according to AOAC (1984) methods (Table 1).

Table 1
Chemical composition and major fatty acids of acorn, pasture and formulated mixed diet

	Indoors	Outdoors	
	Mixed Diet	Acorn	Pasture
Dry matter (%)	88.8	60.8	27.4
Crude protein (% DM[a])	13.3	4.9	13.8
Fat (% DM)	6.1	9.6	2.6
Crude fibre (% DM)	7.1	4.4	22.8
Ash (% DM)	6.2	2.8	10.4
NFE[b] (% DM)	65.4	78.3	50.3
Fatty acid (%)			
C16:0	8.5	14.6	19.7
C16:1 (n-7)	0.3	0.1	2.5
C18:0	4.2	2.9	1.5
C18:1 (n-9)	69.2	68.0	9.2
C18:2 (n-6)	16.3	12.6	11.9
C18:3 (n-3)	0.7	0.4	54.6
C20:0	0.5	0.7	0.4
C20:1 (n-9)	0.3	0.7	0.2

[a] DM, dry matter.
[b] NFE, nitrogen-free extractives.

2.2. Sampling

Pigs were slaughtered by electrical stunning and exsanguination at a local slaughterhouse. Sampling was carried out within the hour following slaughter. A portion of the *longissimus dorsi* (LD) muscle, from the last lumbar to the first thoracic vertebra and the whole *masseter* (MS) muscles of all the 70 animals were taken and stored at -80 °C until analysis.

2.3. Intramuscular fat and fatty acid analysis

Samples were ground using a commercial grinder immediately before fat extraction. Intramuscular total lipids and lipids from diets were extracted and quantified according to the method described by Bligh and Dyer (1959). Polar and neutral lipids (PL and NL) from intramuscular fat were separated by using NH_2-aminopropyl minicolumns, following the method described by Kaluzny, Duncan, Merritt, and Epps (1985). Briefly, 0.1 g of intramuscular fat dissolved in 1ml of chloroform were added to the column, which was previously activated with 1 ml of chloroform. Neutral lipids were eluted with 3ml of chloroform:isopropanol (2:1). Polar lipids were subsequently eluted with 3ml of methanol. Fatty acid methyl esters (FAMEs) of muscle NL and PL and of diets were prepared by acidic-trans-esterification in the presence of sulphuric acid (5% sulphuric acid in methanol; Cava et al., 1997). FAMEs were analysed by gas chromatography using a Hewlett-Packard HP-5890A gas chromatograph, equipped with a flame ionisation detector (FID). Separation was carried out on a polyethylene glycol-TPA modified fused silica semicapillary column (30 m long, 0.53 mm id, 1 μm film thickness) maintained at 225 °C. Injector and detector temperatures were 230 °C. Carrier gas was nitrogen at a flow rate of 1.8 ml min^{-1}. Individual FAME peaks were identified by comparing their retention times with those of standards (Sigma, St Louis). All analyses were performed in duplicate.

2.4. Statistics

Data was subjected to a two-way analysis of variance (ANOVA) using the general linear model (GLM) procedure of the SPSS package (v10.0) to determine the overall effect of muscle (LD vs MS) and rearing system (OUT vs IND) and their interaction. Pairwise comparisons between means were carried out using the Tukey's test.

3. Results and discussion

3.1. Feeding composition

Table 1 shows chemical and fatty acid composition of feeding sources. Results from acorn and pasture, the

two basic food sources for free-reared Iberian pigs, are in agreement with previously published studies (Cava et al., 1997; Ruiz et al., 1998). Acorn showed high nitrogen-free extractives and low protein content, whereas pasture had elevated fibre and ash contents. High levels of oleic acid (C18:1 n-9) characterized the acorns (68%). As expected, pasture showed high levels of linoleic acid (18:3 n-3) (50.5%). A mixed commercial diet, typified the present formulations for Iberian pigs reared intensively. It had a high C18:1 n-9 content (69.2%), even higher than the acorn, whereas the 18:3 n-3 acid levels were rather low. Fatty acid compositions of mixed diets for Iberian pigs reared indoors have now changed markedly. Mixed diets for Iberian pigs used to have high linoleic acid (C18:2 n-6) and saturated fatty acid levels (Ruiz et al., 1998). Meat products from Iberian pigs reared outdoors and fed on acorns and pasture attain the highest quality and prices on the market. Partly due to several studies carried out in our department (Cava et al., 1997; Ruiz et al., 1998), in which animals reared outdoors and fed on acorns showed higher C18:1 n-9 and lower C18:2 n-6 contents, Iberian pigs are now categorized accordingly, animals showing higher levels of C18:1 n-9 and lower of C18:2 n-6 rating higher prices. This has led feed producers to formulate mixed feeds with higher levels of C18:1 n-9 and lower of C18:2 n-6.

3.2. Effect of rearing system on fatty acid composition of neutral and polar lipids

Fatty acid compositions of NL and PL from LD and MS muscles of swine reared outdoors and indoors are shown in Tables 2 and 3. Free-range pigs showed significantly higher PUFA content in NL of LD and MS muscles than pigs reared indoors and fed on concentrates. These differences were mainly produced by an almost twice higher n-3 PUFA content in NL of free-reared pigs ($P<0.001$), and also by slightly higher levels of n-6 PUFA ($P<0.05$). These led to significantly lower n-6:n-3 ratios in NL of pigs reared outdoors and fed on acorn and pasture ($P<0.001$). Therefore, although the n-6:n-3 ratio was higher than dietary recommendations in all cases (British Nutrition Foundation, 1994), free-rearing appears as an interesting way to reduce this ratio in pork. Levels of other fatty acids from the n-3 family in NL were either not detected or reached very low levels.

Rearing system did not significantly affect MUFA content of NL. This seems strange since, in previous studies, free-reared animals fed on acorns showed higher values than those fed on concentrates (Andrés et al., 2001; Ruiz et al., 1998;). Higher C18:1 n-9 levels in the concentrates used in the present study than in

Table 2
Effect of muscle (LD vs MS) and rearing system (outdoors vs indoors) on the fatty acids (% of total FAMEs) of neutral lipids from IMF of Iberian pigs[a]

	LD		MS		SEM	Significance level		
	OUT	IND	OUT	IND		Muscle	Rearing	Int.
C12:0	0.05ab	0.06a	0.04b	0.03c	0.001	***	NS[b]	**
C14:0	1.0a	1.1a	0.7b	0.8b	0.03	***	NS	NS
C15:0	0.05b	0.07b	0.04b	0.16a	0.01	*	***	**
C16:0	22.0a	22.2a	18.5b	19.5b	0.28	***	NS	NS
C16:1 (n-7)	4.6a	4.1ab	3.7b	3.9ab	0.11	*	NS	NS
C17:0	0.5a	0.2c	0.3b	0.3c	0.01	NS	***	***
C17:1 (n-9)	0.3a	0.2b	0.3ab	0.3a	0.01	NS	NS	***
C18:0	12.6a	12.3a	10.6b	11.1b	0.16	***	NS	NS
C18:1 (n-9)	48.9	51.2	50.6	50.1	0.41	NS	NS	NS
C18:2 (n-6)	7.2b	6.1b	10.5a	9.7a	0.22	***	**	NS
C18:3 (n-3)	0.7b	0.4c	0.9a	0.5c	0.03	**	***	NS
C20:0	0.4	0.4	0.3	0.3	0.01	NS	NS	NS
C20:1 (n-9)	1.3bc	1.0c	1.6a	1.4ab	0.03	***	**	NS
C20:4 (n-6)	0.6b	0.7b	1.8a	1.9a	0.08	***	NS	NS
Total SFA	36.4a	36.3a	30.6b	32.2b	0.40	***	NS	NS
Total MUFA	55.0	56.5	56.2	55.8	0.36	NS	NS	NS
Total PUFA	8.5b	7.1b	13.2a	12.0a	0.30	***	**	NS
Total (n-6)	7.8b	6.7b	12.3a	11.6a	0.30	***	*	NS
Total (n-3)	0.7b	0.4c	0.9a	0.5c	0.03	**	***	NS
Ratio (n-6)/(n-3)	10.7c	17.7ab	13.5bc	25.1a	0.9	***	***	*

[a] LD, *longissimus dorsi*; MS, *masseter*; OUT, outdoors, IND. indoors.
[b] NS, $P>0.05$.
* $P<0.05$.
** $P<0.01$.
*** $P<0.001$.
Means with different letters within the same row are significantly different ($P<0.05$).

those of previous ones [68% in the present study vs 24% in Ruiz et al. (1998) and 24.4% in Andrés et al. (2001)] are most likely why NL of indoors pigs were as high as animals fed on acorns and pasture and reared outdoors. Moreover, the latter rearing system is strongly determined by food availability, which varies from one year to another, depending on annual weather conditions. This leads to variations in fatty acid composition of muscle tissue; in this sense, muscular MUFA level in NL of animals reared outdoors and fed on acorns and pasture varied between 54 and 60% in studies carried out in our laboratory in different years (Andrés et al., 2001; Cava et al., 1997; Ruiz et al., 1998).

Nevertheless, C18:1 n-9 levels in muscle lipids of Iberian pigs reared outdoors are considerably higher than common levels of this fatty acid in meat from pigs reared under intensive conditions, which could also be considered a positive feature of this type of pork from a consumer health point of view.

PL (Table 3) showed higher levels of PUFA and lower of SFA and MUFA than NL, which agrees with previously reported results in different muscles from various pig breeds fed diverse diets (Andrés et al., 2001; Lauridsen, Nielsen, Henckel, & Sorensen, 1999).

Rearing system did not influence total SFA of PL. A similar trend has previously been reported in other muscles in pigs fed either outdoors on acorn and pasture or indoors on concentrates (Andrés et al., 2001). Ahn et al. (1996), studying the effect of increasing levels of dietary C18:3 n-3 on the fatty acid composition of phosphatidylcholine and phophatidylethanolamine of LD in pigs, did not detect any effect on SFA. Animals reared outdoors showed significantly ($P<0.05$) lower MUFA levels in MS muscle than those reared indoors and fed on concentrates, as a direct result of lower C18:1 n-9 and C16:1 n-7 levels. The same trend was detected in LD muscle although differences did not reach statistical significance. As discussed in NL, this is most likely a consequence of higher C18:1 n-9 levels in the mixed feed. Previous studies of Iberian pigs, fed either on concentrates or on acorns, showed the opposite tendency (Andrés et al., 2001). However, changes in the formulations of the diets of Iberian pigs, for the reasons explained earlier have led to similar or even higher C18:1 n-9 contents in the phospholipids of animals reared indoors.

Rearing system significantly affected total PUFA content of PL ($P<0.001$), free-reared animals showing higher levels than indoors ones. These differences were

Table 3
Effect of muscle (LD vs MS) and rearing system (outdoors vs indoors) on the fatty acids (% of total FAMEs) of polar lipids from IMF of Iberian pigs[a]

	LD		MS		SEM	Significance level		
	OUT	IND	OUT	IND		Muscle	Rearing	Int.
C12:0	0.1a	0.1a	0.1b	0.1b	0.001	***	NS[b]	NS
C14:0	0.3a	0.3a	0.2b	0.2b	0.01	***	NS	NS
C15:0	0.1a	0.1a	0.1b	0.1a	0.001	NS	*	**
C16:0	15.3a	14.4a	11.1c	13.2b	0.18	***	*	***
C16:1 (n-7)	1.1a	1.2a	0.8c	1.0b	0.02	***	***	**
C17:0	0.3c	0.4ab	0.4bc	0.5a	0.01	*	***	NS
C18:0	11.0[d]	12.9c	21.0a	18.8b	0.4	***	NS	***
C18:1 (n-9)	18.0a	18.5a	12.3c	14.1b	0.29	***	**	NS
C18:2 (n-6)	29.6a	30.6a	23.2c	27.8b	0.31	***	***	***
C18:3 (n-3)	1.1a	0.7b	1.1a	0.7b	0.04	NS	***	NS
C20:1 (n-9)	0.4	0.5	0.4	0.4	0.01	**	NS	NS
C20:3 (n-6)	1.4a	1.2b	0.9c	0.9c	0.02	***	*	*
C20:4 (n-6)	16.9b	15.3c	22.0a	18.4b	0.31	***	***	*
C20:5 (n-3)	1.2b	0.8c	1.6a	0.9c	0.04	***	***	**
C22:4 (n-6)	0.3	0.3	0.3	0.3	0.04	NS	NS	NS
C22:5 (n-3)	2.6b	2.2b	3.4a	2.2b	0.09	*	***	*
C22:6 (n-3)	0.7ab	0.3c	0.8a	0.4bc	0.05	NS	***	NS
Total SFA	27.1b	28.2b	32.8a	32.9a	0.28	***	NS	NS
Total MUFA	19.6a	20.1a	13.5c	15.5b	0.31	***	**	NS
Total PUFA	53.8a	51.4b	53.2a	51.5b	0.28	NS	***	NS
Total (n-6)	48.2a	47.4a	46.3b	47.4a	0.24	*	NS	NS
Total (n-3)	5.6b	4.0c	7.0a	4.1c	0.15	***	***	**
Ratio (n-6)/(n-3)	8.6b	11.8a	6.6c	11.4a	0.40	*	***	*

[a] LD, longissimus dovsi; MS, *masseter*; OUT, outdoors; IND, indoors.
[b] NS, $P>0.05$.
* $P<0.05$.
** $P<0.01$.
*** $P<0.001$.
Means with different letters within the same row are significantly different ($P<0.05$).

mainly produced by higher ($P<0.001$) n-3 PUFA levels in PL of animals reared outdoors, with slight or zero variation in n-6 PUFA. This led to lower n-6:n-3 ratios in PL of pigs reared outdoors in both muscles ($P<0.05$). However, individual PUFA followed different trends. C18:2 n-6 was the major fatty acid in PL of both muscles (23–30%). Animals reared outdoors showed lower levels of this fatty acid than those fed on concentrates, although these differences were significant only in the MS muscle ($P<0.05$). This was the opposite behaviour to that observed in NL, where animals fed on concentrates showed the lowest values in both muscles. However, the other major n-6 PUFA, C20:4 n-6, showed significantly ($P<0.05$) higher values in PL from animals reared outdoors in both muscles. Differences in n-6 PUFA content between food sources were scarce, and this explains the lack of variation in total n-6 PUFA in PL between rearing systems. However, the different trends followed by C18:2 n-6 and C20:4 n-6 are striking. Several factors could be responsible for this behaviour. Elongation and desaturation from C18:2 n-6 to C20:4 n-6 is controlled by an elongase enzyme and a series of fatty acid desaturases (Jeffcoat, 1979) which are influenced by a number of factors, including dietary factors such as level of MUFA in the diet or total fat content (Cho, Nakamura, & Clarke, 1999) and environmental factors (Gurr & Harwood, 1991). There may also be other factors influencing n-6 PUFA content in PL, such as metabolic competition with other long chain PUFA from the n-3 pathway (Pan & Storlien, 1993). Moreover, active metabolic processes aimed at incorporating specific long chain highly unsaturated lipids into membrane for particular metabolic purposes have also been proposed (Kriketos et al., 1995).

Total n-3 PUFA content was significantly affected by rearing system ($P<0.001$), animals reared outdoors showing higher contents than those reared indoors on concentrates in both muscles. This effect was more marked in the MS muscle, in which outdoor animals had around 70% higher n-3 PUFA levels than indoor ones. These differences are most likely occasioned by the higher C18:3 n-3 content of the pasture (around 50% of total fatty acids). Other authors have also found a higher total n-3 PUFA content in muscle phospholipids from animals fed diets high in C18:3 n-3 (Ahn et al., 1996; Specht-Overholt et al., 1997), the increasing levels of C18:3 n-3 being mainly responsible for the higher total n-3 PUFA in PL. In these studies, the higher C18:3 n-3 content of the diet led to increased amounts of some of the fatty acids of the n-3 pathway, especially EPA (C20:5 n-3) and C22:5 n-3, but not DHA (C22:6 n-3). In the present study, all individual n-3 PUFA, including EPA, DHA and C22:5 n-3, were significantly higher in animals reared outdoors and fed on acorns and pasture than in indoor animals fed on concentrates. Moreover, levels of DHA were twice higher in PL of animals fed acorn and pasture than in those fed concentrates. Previously published studies have shown a positive influence of rearing Iberian pigs outdoors on the C18:3 n-3 content of PL (Andrés et al., 2001), but this is the first study in which this positive effect has been observed in n-3 long chain PUFA. A role of EPA and DHA in the amelioration of a number of diseases, including coronary heart disease, is well recognized (British Nutrition Foundation, 1994). The increasing EPA and DHA contents and the decreasing n-6:n-3 ratio, together with the high MUFA levels of NL, indicate a possible beneficial effect of feeding the animals on pasture, and support the "healthy" image of "organic" pork. In fact, nutritional studies have already related the inclusion of meat products from Iberian pigs reared outdoors in the diet with improvement of plasmatic indicators of coronary and vascular disease (García et al., 1998).

Moreover, meat products from Iberian pigs reared outdoors attain the highest quality and prices in the market, which means that increasing n-3 PUFA content does not induce deleterious changes in the derived meat products. In fact, preliminary results of sensory and chemical analysis of dry-cured products prepared with meat from the animals of the present study, do not show any negative influence of rearing system on rancid flavour, flavour intensity or TBARS.

3.3. *Effect of muscle on fatty acid composition of neutral and polar lipids*

Muscle significantly affected total SFA ($P<0.001$) and PUFA ($P<0.001$) of NL, LD muscle showing higher levels of SFA and lower of PUFA than MS muscle. These differences are a direct consequence of higher amounts of palmitic (C16:0) ($P<0.001$) and stearic (C18:0) ($P<0.001$) acids in LD muscle, and higher amounts of C18:2 n-6 ($P<0.001$), C18:3 n-3 ($P<0.01$), and particularly arachidonic (C20:4 n-6) ($P<0.001$) acids in MS muscle. These results are totally in agreement with previous studies about fatty acid composition in MS and LD muscles, including the remarkably high arachidonic acid content in NL of MS muscle (Morcuende, 2001). The higher degree of unsaturation in MS muscle could be due to metabolic differences between muscles, although anatomical location should not be discarded as a factor influencing fatty acid composition of neutral lipids. In this sense, Malmforms, Lundström, and Hansson (1978) found higher unsaturation in outer layers of pig backfat, and proposed that the degree of unsaturation depends on the temperature gradient of the body. MS muscle is an outer muscle compared to LD muscle, and this could explain differences in SFA and PUFA contents of NL between these two muscles.

With regard to the possible muscle metabolic effect on fatty acid composition of NL, a negative influence of

the oxidative character of the muscle on SFA content has been previously reported (Andrés et al., 2001). *Masseter* is a predominantly oxidative muscle, showing 80–90% of type I + type IIa fibres (Tuxen & Rostrup, 1993) and, therefore, results from the present study support the negative relationship between SFA content of NL and the oxidative character of the muscle.

As far as muscle influence on fatty acid profile of PL is concerned, MS is a predominantly oxidative muscle, showing higher levels of SFA than LD, which has been described as a predominantly glycolytic muscle in the scientific literature. However, major individual SFA followed different trends. LD muscle showed significantly higher C16:0 ($P < 0.001$) and lower C18:0 ($P < 0.001$) levels than MS muscle.

LD had significantly higher levels of MUFA ($P < 0.001$) as a direct consequence of higher C18:1 n-9 content ($P < 0.001$). Individual PUFA followed different tendencies, C18:2 n-6 and C20:3 n-6 being higher in LD ($P < 0.001$), while MS showed higher levels of C20:4 n-6 ($P < 0.001$), C20:5 n-3 ($P < 0.001$), C22:5 n-3 ($P < 0.05$) and C22:6 n-3 ($P < 0.001$). These lead to a higher n-3 ($P < 0.001$) and lower n-6 ($P < 0.05$) PUFA content in MS muscle and, hence, a lower n-6:n-3 ratio ($P < 0.05$).

The cell membrane is a dynamic structure consisting basically of a phospholipid bilayer into which a range of proteins is inserted. The fatty acid profile of muscle membrane phospholipid will strongly influence cell function (Kriketos et al., 1995) and development of lipid oxidative changes during meat processing or storage (Pikul, Leszczynski, & Kummerow, 1984). There is some controversy about the influence of muscle metabolism on fatty acid composition of muscle membrane phospholipids. In several studies about the influence of muscle fibre type on fatty acid composition of muscle phospholipids, a positive relationship between oxidative fibres and SFA and a negative with PUFA content have been observed (Alasnier et al., 1996; Andrés et al., 2001). The present study supports the positive relationship between oxidative character of the muscle and SFA content, the predominantly oxidative MS muscle showing higher SFA levels than LD, despite the SFA of NL showing the inverse trend. However, there was no difference in PUFA content between LD and MS muscles.

Kriketos et al., (1995) found a significant influence of muscle on n-3 and n-6 PUFA contents of rat muscle membrane phospholipids, muscles with a more marked oxidative character, such as the soleus or the red part of the quadriceps, showing higher amounts of n-3 PUFA, especially C22:6 n-3. Furthermore, these authors found increased elongation and unsaturation of C18:2 n-6 in muscles with higher proportions of type IIb fibres. These observations were confirmed only in part in the present research, in which the predominantly oxidative MS muscle showed greater n-3 PUFA levels, but also higher levels of C20:4 n-6 and lower of C18:2 n-6, indicating an increased elongation and desaturation of the n-6 pathway. Alasnier et al. (1996) found a similar behaviour, to that in the present study, in rabbit muscle membrane phospholipids: higher C20:4 n-6 and total n-3 PUFA and lower total n-6 PUFA in oxidative muscles than in glycolytic ones. Information about a relationship between muscle metabolism and muscle membrane long chain PUFA is scarce in pigs. The present study partly confirms previous information obtained in rat and rabbit, although wider and more comprehensive studies of pig muscles with different metablolism are necessary.

4. Conclusion

Metabolic type of muscle influences fatty acid composition of NL and PL in swine, those muscles with a predominantly oxidative metabolism showing higher amounts of n-3 PUFA in both lipid classes. On the other hand, free-range rearing of pigs leads to increasing levels of total n-3 PUFA in neutral and polar lipids of pork, and of individual n-3 PUFA, including EPA and DHA. Therefore, feeding the animals outdoors on pasture appears as an interesting approach to improve the healthy image of "organic" pork. However, the effect of amount of pasture intake by pigs on n-3 PUFA content and n-6:n-3 ratio of pork should be studied further.

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