

## LIPID CHARACTERISTICS RELATED WITH FIBRE TYPE COMPOSITION IN MUSCLES OF MERTOLENGA YOUNG BULLS

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### Background

The relative proportions between the three types of fibre in muscle could be a major factor involved in the heterogeneity of meat quality. Some authors refer that meats are more tasty and juicy, and haem pigments and lipids oxidize faster in oxidative muscles than in glycolytic ones (Valin *et al*, 1982, Renner & Labas, 1987). On the other hand, lipid composition is considered an important meat quality attribute, and therefore, more knowledge is required to determine the relationships between lipid composition and metabolic type of the muscles.

In recent years the demand of meat from rustic cattle breeds increased among Portuguese consumers due to its high intrinsic quality but also because its production system has the advantage of being strictly related to the environment. Out of any significant genetic pressure so far, the biochemical characteristics of their muscles are expectable to be different from other domestic cattle breeds.

### Objective

The aim of this study was to evaluate the relationship between the fibre type composition and intramuscular lipid fraction characteristics in three muscles of Mertolenga young bulls.

### Material & Methods

Thirty Mertolenga males, ranging in age between 15 and 24 months were used in this work. Around 1 hour after slaughter, samples from *Longissimus dorsi* (Ld), *Supra spinatus* (Ss) and *Semi tendinosus* (St) were taken.

Total lipids were extracted by the procedure of Folch *et al* (1957) and then separated into neutral (NL) and polar (PL) fractions through a waters Sep-Pak silica cartridge (Millipore, Waters Chromatography Division, Milford, MA) as described by Juaneda & Rocquelin (1985). Fatty acid composition of triglycerides and phospholipids was determined by gas chromatography of methyl esters, prepared as described by Morrison & Smith (1964). The analysis was carried out using a Carlo Erba (Mega 5160) paired with a flame ionisation detector. The capillary column (DB 23, 60 m long, 0.32 mm i.d.), containing stationary phase cyanopropylmethylpolysiloxane, was used to separate fatty acids. The cholesterol was determined by HPLC, according to Roseiro *et al* (2002). Transverse serial sections (10µm) were cut in a cryostat at -24°C and stained for miofibrillar ATPase after pre-incubation at pH 4.45 as described by Brooke & Kaiser, 1970. The succinic dehydrogenase protocol described by Sheehan & Hrapchak (1987) was also used to define fibre metabolic traits. The percentage of each fibre type was calculated from a minimum of 400 units, by counting the total number of each type and dividing by the total number of fibres. Differentiation as type I, type IIA and type IIB was based on staining intensity.

### Results & Discussion

#### Fibre type composition and metabolic profile

The variation in the fibre type composition and metabolic profiles of the analysed muscles are indicated on Table 1. The St muscle showed mean percentages of fibres type I and IIB significantly lower and higher, respectively, than the other both muscles. This composition pattern is deeply reflected on its metabolic profile, which is more ( $p<0.05$ ) glycolytic (60.8%) than Ld (51.1%) and Ss (50.8%). Despite the similarity on fibre type distribution between Ld and Ss muscles, the last showed about 5% higher mean percentage of fibres type IIB and slightly lower mean counts of fibre types I and IIA. Contrarily to what it could be expectable, Ld muscle demonstrated an intermediate metabolic positioning. This apparent contradiction is possibly due to the fact that oxidative potential is not strictly determined by fibre type I content but also depends on prevailing metabolic characteristics of fibre type IIA (Zerouala & Stickland, 1991; Ruusunen & Puolanne, 1997). The data show that, accordingly to the oxidative/glycolytic relationship, the Ld (0.96) and Ss (0.97) can be considered as muscles of intermediate metabolism whereas the St should be classified as glycolytic.

#### Total fat, triglyceride and phospholipid contents intramuscular

Muscle type influenced significantly the intramuscular fat parameters (Table 1). The Ss muscle, the most oxidative among the analysed units, showed significantly higher total (1.94 mg/100g) and polar (0.69 mg/100g) lipid contents than Ld (1.55 and 0.61 mg/100g, respectively) and St (1.44 and 0.644 mg/100g, respectively). In relation to the neutral lipids, Ss muscle also presented higher mean values ( $p<0.05$ ) than St muscle but did not diverge from that obtained in Ld. These results indicate a certain positive relationship between the different intramuscular fat fractions and the oxidative profile of muscles. However it has to be emphasised that Ld muscle which is quite similar in their metabolic trait to Ss is, however much closer to St in terms of intramuscular fat composition. This unexpected trend is particularly evident in relation to the phospholipids. Oxidative cells are usually smaller in diameter and contain more mitochondria, giving rise to more membrane structure (Leseigneur-Meynier & Gandemer, 1991). According to Kauffman & Safanie (1967) the variation in intramuscular lipid content among muscles is determined by their different propensity to accumulate fat cells in the intercellular space. The cholesterol content was not significantly affected by the fibre type composition or metabolic profile of the muscle.

#### Fatty acid composition of neutral and polar lipid fractions

The intramuscular neutral and polar lipid fractions presented the fatty acid compositions described on Table 2. Regarding the former fraction, no significant variations have been verified in the proportions of total saturated fatty acids (SFA) and total monounsaturated fatty acids (MUFA) between muscles. Nevertheless, C14 and C18 compounds were higher ( $p<0.05$ ) in Ss (36.07) and Ld (35.19) muscles than in St muscle (21.87). Contrarily, the

total polyunsaturated fatty acid (PUFA) concentration differed significantly with the anatomical location of muscles, the higher and lower values being found in Ss (76.61) and in Ld, (52.47), respectively, with the St muscle in an intermediate position. This ordering trend is not totally in compliance with the metabolic profile of the muscles, since the St unit presented the lowest oxidative capacity among them. In relation to polar lipids, the variations verified in total SFA, MUFA and PUFA contents (Table 2) reflected better the oxidative activity of muscles, with the Ss muscle showing significantly higher mean values than the ST muscle. However and despite its metabolic "status", the Ld muscle presented, systematically, lower mean values in those composition parameters than the St muscle, the most glycolytic unit.

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References

Brooke, M.H. e Kaiser, K. 1970. Muscle fibre types: how many and what kind? *Arch. Neurology*, 23, 369-379.

Folch, J.; Lees, M.; Stanley, G., 1957. A simple method for the isolation and purification of total lipids from animal tissues. *Journal Biological Chemistry*, 226: 497.

Juaneda, P. & Rocquelin, G., 1985. Rapid and convenient separation of phospholipids and non phosphorus lipids from rat heart using silica cartridges. *Lipids*, 20 (1) 40:41.

Kauffman, R. & Safanie, A. 1967. Influence of porcine muscle structure on its lipid accumulation during growth. *J. Food Sci.*, 32:283.

Leseigneur-Meynier, A. & Gandemer, G. 1991. Lipid composition of pork muscle in relation to the metabolic type of the fibres. *Meat Science*, 29: 229-241.

Morrison, R. & Smith, L. 1964. Preparation of fatty acid metil esters and dimetil acetals from lipids with boron fluoride-methanol. *J. Lipid Res.*, 4: 600.

Renerre, M. & Labas, R. 1987. Biochemical factors influencing metmyoglobin formation in beef muscles. *Meat Science*, 19:151-165.

Roseiro L., Costa P. & Santos, C., 2002. Cholesterol and fat contents of barrosa meat as influenced by sex and muscle site. ICoMST, Italy.

Ruusunen, M. e Puolanne, E. (1997). Comparison histochemical properties of different pig breeds. *Meat Science*, 45, 119-125.

Valin, C., Touraile; C., Vigneron, P. & Ashmore, C. 1982. Prediction of lamb meat quality traits based on muscle biopsy fiber typing. *Meat Science*, 6, 257-263.

Zerouala, A.; Stickland, N. 1991. Cattle at risk for dark-cutting beef have a higher proportion of oxidative muscle fibres., *Meat Sci.*, 29:263.

Table 1 –Cholesterol and intramuscular fat mean contents in different muscles of Mertolenga young bulls.

	Muscle			F value	p
	St	Ld	Ss		
Total lipids (g/100g)	1.439 <sup>b</sup>	1.553 <sup>b</sup>	1.939 <sup>a</sup>	6.174	**
Neutral lipids (g/100g)	0.821 <sup>b</sup>	1.006 <sup>ab</sup>	1.236 <sup>a</sup>	3.625	*
Polar lipids (g/100g)	0.644 <sup>b</sup>	0.606 <sup>b</sup>	0.689 <sup>a</sup>	6.282	**
Cholesterol (mg/g)	0.440	0.485	0.483	1.542	ns
ATPase (pH 4.6)					
Fibre Type I (%)	17.84 <sup>b</sup>	31.98 <sup>a</sup>	30.28 <sup>a</sup>	17.825	***
Fibre Type IIa (%)	34.13	38.16	35.05	1.501	ns
Fibre Type IIb (%)	48.04 <sup>a</sup>	29.86 <sup>b</sup>	34.67 <sup>b</sup>	17.698	***
SDH					
Oxidatives (%)	39.23 <sup>b</sup>	48.86 <sup>a</sup>	49.17 <sup>a</sup>	8.293	**
Glycolitic (%)	60.77 <sup>a</sup>	51.14 <sup>b</sup>	50.84 <sup>b</sup>	8.293	**

In same row, means with different letters are significantly different. ns p>0.05; \* p<0.05; \*\* p<0.01; \*\*\* p<0.001.

Table 2 –Fatty acid composition (mg/100g muscle) of Neutral and Polar lipid fractions in different muscles of Mertolenga young bulls

Fatty acid	Neutral Lipids			Fatty acid	Polar Lipids		
	St	Ld	Ss		St	Ld	Ss
C14	21.87 <sup>b</sup>	35.19 <sup>a</sup>	36.07 <sup>a</sup>	C14	1.36	1.79	1.88
C16	206.73	297.10	291.58	C16	118.22	118.35	118.02
C17	9.52 <sup>b</sup>	12.92 <sup>a</sup>	14.55 <sup>a</sup>	C17	3.72 <sup>b</sup>	3.97 <sup>ab</sup>	4.49 <sup>a</sup>
C18	153.13	184.51	207.17	C18	80.77 <sup>b</sup>	73.42 <sup>b</sup>	95.35 <sup>a</sup>
Others	20.22	24.74	29.07	Others	7.04 <sup>b</sup>	7.72 <sup>b</sup>	8.81 <sup>a</sup>
Total SFA	401.95	515.70	563.87	Total SFA	211.11 <sup>ab</sup>	205.24 <sup>b</sup>	228.56 <sup>a</sup>
C16:1	30.57	36.43	38.13	C16:1	7.31	6.92	8.41
C18:1	370.97	417.83	496.44	C18:1	129.92 <sup>ab</sup>	127.91 <sup>b</sup>	146.32 <sup>a</sup>
Others	14.01	15.74	17.93	Others	12.11	9.73	17.00
Total MUFA	415.56	469.99	552.50	Total MUFA	149.34 <sup>b</sup>	144.55 <sup>b</sup>	171.73 <sup>a</sup>
C18:2	44.47	40.81	62.72	C18:2	180.78 <sup>b</sup>	171.82 <sup>b</sup>	221.19 <sup>a</sup>
C18:3	4.64 <sup>b</sup>	4.25 <sup>b</sup>	5.73 <sup>a</sup>	C18:3	7.94	9.93	10.58
Others	8.91	7.41	8.16	C20:3	15.13	12.74	15.10
Total PUFA	58.02 <sup>ab</sup>	52.47 <sup>b</sup>	76.61 <sup>a</sup>	C20:4	58.41	51.39	56.59
				C22:6	7.64	8.82	11.25
				Others	9.26	8.59	7.93
				Total PUFA	279.16 <sup>b</sup>	263.30 <sup>b</sup>	322.65 <sup>a</sup>

In same row, means with different letters are significantly different.