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Original Research Article

Fatty acids, retinol and cholesterol composition in various fatty tissues of Celta pig breed: Effect of the use of chestnuts in the finishing diet

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ABSTRACT

This investigation was designed to evaluate the effects of chestnuts use in the finishing diet of Celta pigs. Thirty-six pigs were separated in three different groups (three different diets: compound feed, mixed and chestnut); fatty acid profile, and cholesterol and retinol content of intramuscular, adipose, perirenal and hepatic fat were evaluated. The inclusion of chestnuts in the diet fundamentally affected the content of oleic (C18:1*n*–9), linoleic (C18:2*n*–6) and α-linolenic (C18:3*n*–3) acids and other *n*–6 PUFA with different intensity depending on the anatomical location. The cholesterol and retinol contents were lower in pigs fed on chestnuts. These pigs showed higher contents of C18:1*n*–9 in hepatic fat, perirenal fat and intramuscular fat of *Psoas major* (PM) muscle. The greatest differences of C18:3*n*–3 were found in the liver. The amount of intramuscular fat showed higher values in the pigs fed with chestnuts. Using discriminant statistical techniques it was possible to classify, in PM muscle and liver, 100% of the pigs fed with chestnut.

1. Introduction

The Celta pig is an autochthonous porcine breed raised traditionally in Galicia (northwest of Spain). Because of their poor growth rate, carcass conformation and crossbreeding between local pigs and improved breeds, the Celta breed became extinct during the second half of the 20th century. Presently, their population is part of a project of recuperation, conservation, and promotion. The aim of the recovery of the Celta pig breed is not only to obtain high-quality meat but also contribute to the environmental preservation and recovery of traditional practices. The use of local breed and extensive or semi-extensive feeding systems based on natural feed resources could also help to maintain the development in rural populations of northwest Spain.

Several authors (Alasnier et al., 1996; Franco et al., 2006; Monziols et al., 2007) have reported variation in fatty acid composition of different anatomical locations. The influence of diet on the fatty acid composition of animal tissues has been the subject of many investigations (Olivares et al., 2009a,b; Rey et al., 2006; Wood et al., 2008). Some fatty acids respond faster to dietary change than others. Saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) are synthesised *in vivo* and are less influenced by diet than the linoleic (C18:2*n*–6) and α-linolenic (C18:3*n*–3) acids, which cannot be synthesised and, therefore, reflect dietary change. The longer chain (C20–22) fatty acids of the *n*–6 and *n*–3 series can also be synthesised from C18:2*n*–6 and C18:3*n*–3, respectively (Enser et al., 2000).

In order to increase the amount of intramuscular fat content (IMF) numerous studies have been carried (D'Souza et al., 2003; Ruusunen et al., 2007). Several reports indicated a negative association between dietary vitamin A and IMF in pigs (D'Souza et al., 2003). On the other hand, the vitamin A concentration can alter tissue fatty acid saturation. Olivares et al. (2009a,b) observed that vitamin A supplementation increased the proportion of saturated fatty acids in subcutaneous back fat and liver lipids in pigs, but produced no effect in intramuscular fatty acid composition. Also, they showed that the magnitude of the induced change and the regulation of fat saturation by vitamin A are dependent on pig genotype.

Other studies have shown that the amount of intramuscular lipids can be increased, with less effect on subcutaneous fat deposition, by feeding on low protein diets (Wood et al., 2004). One of the possible explanations could be that lower dietary protein levels stimulate the activity of muscle lipogenic enzymes, and hence increase *de novo* fatty acid synthesis (Doran et al., 2006).

The cholesterol content of pig meat reported in the literature varies greatly between different studies. The discrepancy can be attributed to natural variation brought about by factors such as

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age, breed of the animals, diet and rearing system (Bragagnolo and Rodríguez-Amaya, 2002). Modification of the fatty acid composition of livestock by dietary means is important, not only because of the probable relationship between fatty acid intake, cholesterol and heart disease, but also because of the effect of the fatty acid profile on the oxidative stability of animal tissues (Monahan et al., 1992; Rey et al., 2001).

Tissue fatty acid composition is not modified to the same extent in different locations and in different depots. Different studies have focused on the effect of diet composition on different fat depots, but an extensive study has not been performed. The presence of other components in fat depots has been linked to their presence in the diet. Cholesterol and retinol are common components in commercial feed whereas they are not present in chestnuts. The NW region (Galicia) is the main area of chestnut production in Spain. The use of chestnuts in the feeding of the Celta pig breed, in an extensive management system, would reduce production costs and bring a high quality product to the market, well differentiated, with high added value and with healthier fat content.

Therefore, the aim of this research was the study of the inclusion of chestnut finishing diet on the fatty acid composition, cholesterol, and retinol contents in adipose tissues, perirenal, intramuscular and hepatic fat.

2. Material and methods

2.1. Pigs, samples and diets

A total of 36 castrated Celta pigs were used in the study. Piglets, which were vaccinated and deparasitised following the usual protocols, were suckled until an age of 40 days. All pigs were reared and fattened until the age of 16 months in an extensive regime, with a livestock density of 12 animals per hectare. After weaning, the pigs were fed with a commercial compound feed. At the age of 12 months, the pigs were randomly divided into three different groups each comprising 12 animals: Group A was fed with a commercial compound feed (3 kg per animal per day) during the 4 months prior to slaughter; Group B was fed with a mixed diet (commercial compound feed/chestnuts; 1.5 kg commercial compound feed + 2.5 kg chestnuts per animal per day) for the remaining 4 months, and Group C was fed with a mixed diet (commercial compound feed/chestnuts; 1.5 kg commercial compound feed + 2.5 kg chestnuts per animal per day) until the age of 13 months and then a diet based only on chestnuts (5 kg chestnut per animal per day) 3 months prior to slaughter. The average weights (mean ± SD) of carcasses were 145.2 ± 19.8 kg for pigs fed on chestnut (170.1 ± 16.7 kg, live weight), 151.7 ± 15.3 kg for pigs fed on mixed diet (173.6 ± 13.0 kg, live weight) and 144.3 ± 18.2 kg for pigs fed on commercial compound feed (166.3 ± 14.6 kg, live weight). No significant differences in the carcass and live weights of the three groups were found.

After slaughtering and 24 h of refrigeration, samples from intramuscular fat (*Longissimus dorsi* (LD) and *Psoas major* (PM) muscles), adipose tissues (rump, dorsal, ventral and covering the *Biceps femoris* muscle) and perirenal fat from each carcass were obtained. Also, a liver sample (100 g) from the lateral hepatic lobe was taken. All samples were vacuum-packed and deep frozen at −20 °C for no longer than 4 months until their analysis.

Chestnuts and commercial compound feed were sampled and their chemical composition was determined according to the Association of Official Analytical Chemist (1990). The fatty acids profile of total lipids was determined using the procedure described by Franco et al. (2006). The commercial compound feed, mixed diet and the chestnut composition are shown in Table 1.

Table 1
Chemical composition and fatty acids of chestnut, mixed diet and commercial compound feed.

	Chestnut[a]	Mixed diet[b]	Compound feed[c]
Energy (kcal/100 g)	275.5	343.1	456.6
Composition (g/100 g)			
Dry matter	51.9	66.0	89.5
Crude protein	4.2	8.4	15.3
Ether extract	3.3	3.5	3.9
Crude fibre	2.0	3.0	4.6
Starch	32.0	34.9	39.7
Ash	1.3	3.2	6.3
Cholesterol (mg/100 g)	0.0	12.1	32.3
Retinol (μg/100 g)	0.0	67.5	180
Fatty acids (mg/100 g)			
	Mean ± SD	Mean ± SD	Mean ± SD
C12:0	1.23 ± 0.04	2.85 ± 0.05	5.56 ± 0.06
C14:0	3.63 ± 0.11	17.9 ± 0.14	41.7 ± 0.19
C14:1	0.04 ± 0.01	0.09 ± 0.01	0.17 ± 0.02
C15:0	2.35 ± 0.02	3.30 ± 0.02	4.87 ± 0.00
C15:1	1.00 ± 0.01	0.99 ± 0.01	0.97 ± 0.03
C16:0	501 ± 1.25	641 ± 0.92	874 ± 0.37
C16:1*n*–7	12.0 ± 0.03	28.9 ± 0.12	57.1 ± 0.26
C17:0	3.20 ± 0.01	6.87 ± 0.09	12.9 ± 0.24
C17:1	1.86 ± 0.02	3.86 ± 0.05	7.19 ± 0.08
C18:0	32.5 ± 0.31	155.8 ± 0.22	361 ± 0.08
C18:1*n*–9	912 ± 3.37	963 ± 2.32	1049 ± 0.57
C18:2*n*–6	1510 ± 0.61	1324 ± 1.65	1013 ± 3.37
C18:3*n*–6	2.50 ± 0.04	2.35 ± 0.06	2.12 ± 0.08
C18:3*n*–3	190 ± 5.58	156 ± 3.58	97.5 ± 0.24
C20:0	7.54 ± 0.09	6.58 ± 0.07	4.98 ± 0.04
C20:1*n*–9	12.9 ± 0.24	13.7 ± 0.19	15.1 ± 0.09
C20:2*n*–6	1.59 ± 0.15	2.91 ± 0.22	5.10 ± 0.34
C20:3*n*–6	1.28 ± 0.01	1.36 ± 0.03	1.50 ± 0.06
C20:4*n*–6	0.37 ± 0.10	1.63 ± 0.13	3.73 ± 0.20
C20:3*n*–3	0.53 ± 0.51	0.97 ± 0.44	1.71 ± 0.31
C20:5*n*–3	0.43 ± 0.10	0.89 ± 0.08	1.66 ± 0.05
C22:0	7.43 ± 0.08	6.85 ± 0.07	5.89 ± 0.07
C22:1*n*–9	2.40 ± 0.25	2.94 ± 0.57	3.84 ± 1.09
C22:2*n*–6	85.6 ± 3.31	169 ± 3.81	307 ± 4.63
C23:0	1.61 ± 0.07	8.75 ± 0.14	20.6 ± 0.27
C24:0	4.90 ± 0.22	3.75 ± 0.73	1.82 ± 1.57
C24:1*n*–9	0.32 ± 0.09	0.58 ± 0.19	1.01 ± 0.36
SFA	566 ± 1.23	854 ± 0.89	1333 ± 0.33
UFA	2734 ± 1.24	2671 ± 0.89	2567 ± 0.32
MUFA	941 ± 3.31	1014 ± 2.67	1134 ± 1.60
PUFA	1793 ± 2.08	1658 ± 2.03	1433 ± 1.93
∑*n*–6	1602 ± 0.70	1501 ± 1.01	1332 ± 1.45
∑*n*–3	191 ± 2.06	157 ± 1.37	101 ± 0.20

SFA: sum of saturated fatty acids; UFA: sum of unsaturated fatty acids; PUFA: sum of polyunsaturated fatty acids; MUFA: sum of monounsaturated fatty acids; SD: standard deviation.
[a] 5 kg of chestnuts/animal and day.
[b] (1.5 kg of commercial compound feed + 2.5 kg of chestnuts)/animal and day.
[c] 3 kg of commercial compound feed/animal and day.

2.2. Fatty acids analysis

The intramuscular and hepatic fat was extracted using chloroform and methanol (2:1) according to the method of Folch et al. (1957), whereas the adipose and perirenal fat was extracted using a microwave oven of 700 W power and 2450 MHz microwave frequency, for 5 min, following the procedure described by De Pedro et al. (1997). In order to determine if there are significant differences between the two methods of fat extraction, fat was extracted in triplicate from two different locations with both methods, and the lipid profile was analysed. There were no significant differences ($p > 0.05$) in the fatty acid profile between the two extraction methods. The method of De Pedro et al. (1997) does not affect the determination of adipose tissue fatty acids and