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Fatty acid composition of leg meat and perirenal fat of rabbits selected by growth rate

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Abstract

The effect of selection for growth rate on the fatty acid composition of edible rabbit fat and meat was studied. Two groups of contemporary animals, one selected for growth rate (S) during 14 generations and other unselected control group (C) were compared. Control group were the offspring of animals originated from embryos that were vitrified at the beginning of the experiment and thawed to produce a control group contemporary with the selected group. Forty four rabbits of both sexes of group C and forty of group S were used in the experiment. The composition of fatty acids of the meat of a hind leg and of the perirenal fat was determined by gas chromatography. Selection for growth rate modified the percentage of fatty acids both in meat and in perirenal fat, and increased the content of most fatty acids in meat, but the indices related to human health were only slightly modified by selection. The changes in percentage of fatty acids in meat affected myristic (2.24 and 2.48, for C and S, respectively) palmitic (25.38 and 26.50), *cis* n - 7 palmitoleic (2.08 and 2.79), oleic (22.52 and 23.51), linoleic (31.41 and 29.06) and arachidonic (2.10 and 1.77) acids. The highest change in indices related to human health was a 10% of reduction of the ratio polynsaturated:saturated fatty acids, which represents less than a 1% of change per generation. The ratio n - 6:n - 3 fatty acids slightly improved. Selection for growth rate would not damage the quality of meat and edible fat from a human health point of view.

Keywords: Fatty acids; Inter and intramuscular fat content; Rabbit meat; Genetics; Growth rate

1. Introduction

There is an increasing interest in the lipid composition of edible meat and fat of domestic animals because of its relationship with human health, particularly with cardiovascular illnesses (Hu & Willett, 2002). It is well known that the quantity and composition of the fatty acids found in monogastrics is directly influenced by the composition of the diet (see in rabbits, for example Bernardini, Dal Bosco, & Castellini, 1999; Oliver et al., 1997). However, there are no studies about the effect of selection for growth rate in the fatty acid composition of the meat. Rabbit genetic improvement programs are based in a three way cross in which the terminal sire is selected by growth rate (Blasco, 1996). Rabbit breeders and consumers are interested in knowing the consequences of this selection in the fatty acid composition of rabbit edible fat and meat. The only experiment carried out in monogastrics about the effect of selection on fatty acid composition is due to Cameron et al. (2000), who found differences in fatty acid composition of intramuscular fat of pig lines selected for different objectives (appetite, low food conversion rate and high lean growth tissue), but studies on rabbit meat have been focused in diet and there are no studies on the correlated response to selection for growth rate on the fatty acids composition.

Rabbit can be considered a model for other species like pigs or chicken. It has a short generation interval (6

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months) and some types of experiments in rabbits are not feasible in other monogastrics, for example those which require maintaining frozen control populations. The aim of this study was to assess the effect of selection for growth rate in fatty acid composition of rabbit meat and edible fat by means of comparing a population selected for growth rate with a contemporary control population that was frozen at the beginning of the experiment and thawed several generations later.

2. Materials and methods

2.1. Animals

Animals came from a synthetic line selected for high growth rate between the 4th and 9th weeks of life by individual selection (Estany, Camacho, Baselga, & Blasco, 1992). This line was formed by crossing a Californian line with a synthetic line created by mating commercial crossbred rabbits. Matings involved approximately 25 males and 90 females per generation.

Embryos belonging to generation seven of selection were vitrified and thawed to be contemporary of animals born in generation 21. The procedure is described by Vicente, Viudes-de-Castro, and García (1999). The control group (C) was formed from offspring of the thawed embryos, to avoid any effect of cryoconservation, and was maintained contemporary to rabbits from generation 21 of the selection line, fed with the same food and in the same farm. Forty four rabbits of group C and 40 rabbits of group S, chosen at random from litters of at least six young rabbits, one male and one female per litter, were used to asses the effects of selection for growth rate.

Animals were reared at the experimental farm of the Universidad Politécnica de Valencia. After weaning at 4 weeks of age, rabbits were placed in collective cages with 8 individuals each, and fed ad libitum with a commercial pelleted food (16.0% crude protein, 15.5% fibre, 3.4% fat) until 9 weeks of age, at which commercial slaughter weight in Spain (about 2 kg) was expected to be reached. Rabbits were slaughtered at a slaughter house in the farm, so there was no stress due to transport. No fastening was practised. Hot carcasses were put in a ventilated area for one hour, and were then stored at 3-5 °C during 24 h.

2.2. Fatty acid composition

Meat and perirenal fat samples were taken 24 h post mortem. A whole (deboned) hind leg was ground up using a domestic mincing machine and a 10 g sample was weighed. Another 20 g sample was then immediately taken from the remaining meat in order to predict its ethereal extract content using a NIR spectroscope (Model 5000, NIR Systems Inc., Silver Spring, MD, USA). Perirenal fat was weighed and a 10 g sample was taken. All samples were kept in vacuum-sealed aluminium bags and stored at -20 °C for later analysis at the Meat Technology Centre (IRTA) in Monells.

Fatty acids were extracted from meat using chloroform-methanol 2:1 (Folch, Lees, & Stanley, 1957). The solution was then treated in a separation funnel with a saturated solution of NaCl (40% in volume with respect to the organic phase). Once the two phases were well separated, the organic phase was added to the internal standard. Concentrated and methyl esters from the fatty acids were obtained through KOH-methanol (0.5 N) and boron trifluoride-methanol 14% (Sigma). The reaction was carried out at 100 °C for 1 h. The fatty acids methyl esters were extracted into hexane after addition of water saturated with NaCl (Díaz, 1994). The composition of the fatty acids was determined by gas chromatography (GC) using a Hewlett-Packard 5890 gas chromatograph equipped with a split flame ionisation detector and using a capillary column (DB 225 J&W, 30 m long, 0.25 mm internal diameter, 0.25 m fill thickness) and containing a polar stationary phase (cyano-propylphenyl-methylpolysiloxane). The carrier gas was helium at flow rate of 1 ml/min. Oven temperature remained at 110 °C for 1 min, increasing to 230 at a rate of 4 °C/min. The temperatures of the injector and detector were 250 °C. Meat fatty acids were quantified using pentadecanoic acid (C15:0) as an internal standard. The methyl esters were identified using a Sigma Chemical Co. Standard (Lipid Standard: Fatty Acid Methyl Ester Mixture # 189-19). Perirenal fat lipids were extracted and subsequently methyled following the same procedure as in the extraction of meat fatty acids. For the analysis of perirenal fat an internal standard was not used. The commercial diet used in the experiment was also analysed, and the analysis gave a high proportion of C18:2 n - 6 (45.5%), C18:1 n - 7 (22%) and C16:0 (17.7%). The percentage of C18:3 n - 3 was 6.4%.

2.3. Statistical analysis

Least square means were calculated to compare both groups. The GLM program of the SAS statistical package was used (SAS, 1988).

3. Results and discussion

Selection for growth rate was successful, improving growth rate about a 1% per generation (Piles & Blasco, 2003). This led to differences in carcass weight at the same age. Least square means and standard errors of carcass weight were 1230 ± 20 g and 1348 ± 20 g, for groups C and S, respectively (P < 0.01).

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3.1. Effect of selection on fatty acid composition of the hind leg meat

Selection for growth rate seems to have increased the percentage of fat content in the meat of the hind leg from 2.97 ± 0.10 (group C) to 3.21 ± 0.10 (group S), P < 0.10. This has lead to a general increase of the amount of fatty acids in the meat, although this increase has not been the same for all fatty acids. In a similar experiment in rabbits, made with the same line but in different generations, Piles, Blasco, and Pla (2000) did not find either clear differences in fat content of the hind leg between a control group and a group selected for growth rate.

Table 1 shows the fatty acid composition of the meat of the hind leg for both groups, whereas Table 2 shows the relative percentage of these fatty acids. Selection for growth rate did not decrease the content of any fatty acid, whereas clearly increased the content of myristic, palmitic, palmitoleic, C16:1 n - 9, margaric, C17:1, oleic and C20:2 n - 6 fatty acids. This increase was particularly high for palmitoleic (augmenting a 55%) and heptadecenoic (39%). The relative composition of fatty acids has been also modified; differences between C and S groups were found for myristic, palmitic, palmitoleic, oleic, linoleic and arachidonic acids, some of the changes being relevant.

Fatty acids C18:1 *trans*, C18:3 n - 6 (γ -linolenic) and C20:5 n - 3 (icosapentaenoic) were not detected in concentrations greater than 0.01%. When considering the percentages, the C18:1 trans, C18:3 n - 6 (γ -lino-

lenic) and long-chain fatty acids, C20:0, C20:3 n - 3 and C20:5 n - 3 (EPA), were below 0.01%.

There are no experiments of selection in the literature in which correlated response to selection for growth rate on fatty acid composition is assessed. Cameron et al. (2000) found in intramuscular fat of pigs some differences between lines selected for other criteria than ours: daily gain, food conversion rate and lean growth rate (i.e., growth rate corrected by fatness). Comparisons of the mean quantities with other experiments should be taken with caution, because fatty acid composition is affected by specie and diet (Oliver et al., 1997; Bernardini et al., 1999). When comparing rabbit meat to other meats, differences in fatty acid composition are large (see, for example Enser, Hallet, Hewett, Fursey, & Wood, 1996, for comparisons with beef, lamb and pork).

3.2. Fatty acid composition in perirenal fat

Table 3 shows the percentage of weight of total fatty acids of perirenal rabbit fat. Here selection also changed the proportions of several fatty acids; percentage of capric, lauric, pentadecanoic, palmitoleic, margaric and C20:0 was modified by selection for growth rate. The changes were not as pronounced as in the meat of the hind leg, with the exception of palmitoleic. As in the meat, the percentage of linoleic acid was very high as a consequence of the rabbits diet. Fatty acid C18:3 (α -linolenic) was detected in greater concentrations in perirenal fat compared with the concentration in the meat of

Table 1

Least square means and standard er	rors of fatty acids content in the	he hind leg meat (mg/100 g of meat	t)

Fatty acids	Group		Significance	
	Control $(n = 44)$	Selection $(n = 40)$		
C10:0 (capric)	3.75 ± 0.97	3.19 ± 1.01	ns	
C12:0 (lauric)	4.97 ± 0.65	6.27 ± 0.68	ns	
C14:0 (myristic)	54.21 ± 2.69	67.05 ± 2.82	**	
C16:0 (palmitic)	607.74 ± 23.42	712.28 ± 24.56	**	
C16:1 <i>cis</i> $n - 7$ (palmitoleic)	50.19 ± 4.92	78.00 ± 5.16	***	
C16:1 <i>n</i> – 9	7.88 ± 0.34	9.36 ± 0.36	**	
C17:0 (margaric)	14.69 ± 0.60	16.91 ± 0.63	*	
C17:1 (heptadecenoic)	4.84 ± 0.55	6.74 ± 0.58	*	
C18:0 (stearic)	169.12 ± 5.61	185.01 ± 5.88	†	
C18:1 $n - 9$ (oleic)	537.88 ± 23.16	635.27 ± 24.29	**	
C18:1 $n - 7$	31.79 ± 1.26	34.86 ± 1.32	t	
C18:2 $n - 6$ (linoleic)	757.89 ± 31.70	776.84 ± 33.24	ns	
C18:3 $n - 3$ (α -Linolenic)	77.92 ± 4.59	81.20 ± 4.81	ns	
C20:1 (icosaenoic)	8.24 ± 0.70	9.96 ± 0.73	t	
C20:2 $n - 6$	10.95 ± 0.56	12.78 ± 0.58	*	
C20:3 $n - 6$	6.67 ± 0.51	6.68 ± 0.54	ns	
C20:4 $n - 6$ (arachidonic)	47.98 ± 1.18	45.44 ± 1.24	ns	

ns, No significant.

 $p^* < 0.05.$

 $p^{**} < 0.01.$

p < 0.001.

 $^{^{\}dagger}p < 0.10.$

Table 2
Least square means and standard errors of relative percentage of fatty acids in rabbit hind leg meat

Fatty acids	Group		Significance	
	Control $(n = 44)$	Selection $(n = 40)$		
C10:0 (capric)	0.11 ± 0.02	0.11 ± 0.02	ns	
C12:0 (lauric)	0.20 ± 0.02	0.23 ± 0.02	ns	
C14:0 (myristic)	2.24 ± 0.05	2.48 ± 0.05	***	
C16:0 (palmitic)	25.38 ± 0.24	26.50 ± 0.25	**	
C16:1 $\overrightarrow{cis} n - 7$ (palmitoleic)	2.08 ± 0.12	2.79 ± 0.13	***	
C16:1 <i>n</i> – 9	0.33 ± 0.01	0.35 ± 0.01	ns	
C17:0 (margaric)	0.61 ± 0.02	0.63 ± 0.02	ns	
C17:1 (heptadecenoic)	0.21 ± 0.02	0.24 ± 0.02	ns	
C18:0 (stearic)	7.19 ± 0.13	6.91 ± 0.14	ns	
C18:1 $n - 9$ (oleic)	22.52 ± 0.38	23.51 ± 0.39	t	
C18:1 $n - 7$	1.35 ± 0.03	1.29 ± 0.03	ns	
C18:2 $n - 6$ (linoleic)	31.41 ± 0.62	29.06 ± 0.65	*	
C18:3 $n - 3$ (α -linolenic)	3.17 ± 0.12	3.02 ± 0.13	ns	
C20:1 (icosaenoic)	0.34 ± 0.02	0.36 ± 0.02	ns	
C20:2 $n - 6$	0.47 ± 0.02	0.48 ± 0.02	ns	
C20:3 $n - 6$	0.28 ± 0.02	0.26 ± 0.02	ns	
C20:4 $n - 6$ (arachidonic)	2.10 ± 0.08	1.77 ± 0.08	***	

ns, No significant. p < 0.05.

 $^{\dagger}_{**}p < 0.10.$

*** *p* < 0.01.

p < 0.001.

Table 3

Least square means and standard errors of relative percentage of total fatty acids in rabbit perirenal fat

Fatty acids	Group ¹		Significant level
	Control $(n = 44)$	Selection $(n = 40)$	
C10:0 (capric)	0.37 ± 0.04	0.50 ± 0.05	*
C12:0 (lauric)	0.38 ± 0.04	0.51 ± 0.04	*
C14:0 (myristic)	2.63 ± 0.06	2.74 ± 0.07	ns
C14:1	0.12 ± 0.05	0.24 ± 0.05	ns
C15:0 (pentadecanoic)	0.58 ± 0.01	0.64 ± 0.01	**
C16:0 (palmitic)	25.08 ± 0.27	25.66 ± 0.29	ns
C16:1 <i>cis</i> $n - 7$ (palmitoleic)	2.18 ± 0.12	2.61 ± 0.13	*
C16:1 <i>n</i> – 9	0.35 ± 0.01	0.33 ± 0.01	ns
C17:0 (margaric)	0.60 ± 0.01	0.67 ± 0.01	***
C17:1 (heptadecenoic)	0.24 ± 0.02	0.29 ± 0.02	ns
C18:0 (stearic)	5.31 ± 0.11	5.33 ± 0.13	ns
C18:1 <i>n</i> – 9 (oleic)	23.16 ± 0.41	23.00 ± 0.46	ns
C18:1 $n - 7$	1.20 ± 0.03	1.13 ± 0.03	t
C18:2 $n - 6$ (linoleic)	32.82 ± 0.70	31.47 ± 0.77	ns
C18:3 $n - 6$ (γ -linolenic)	0.09 ± 0.01	0.06 ± 0.01	t
C18:3 $n - 3$ (α -linolenic)	3.92 ± 0.13	3.87 ± 0.14	ns
C20:0	0.10 ± 0.01	0.05 ± 0.01	**
C20:1 (icosaenoic)	0.32 ± 0.02	0.32 ± 0.02	ns
C20:2 $n - 6$	0.27 ± 0.02	0.28 ± 0.02	ns
C20:3 $n - 6$	0.03 ± 0.01	0.03 ± 0.01	ns
C20:3 <i>n</i> – 3	0.03 ± 0.00	0.01 ± 0.00	Ť
C20:4 $n - 6$ (arachidonic)	0.21 ± 0.01	0.23 ± 0.01	ns

ns, No significant.

 $p^{\dagger} p < 0.10.$

* *p* < 0.05.

p < 0.03.

 $p^* < 0.001.$

the hind leg. Some Fatty acids not found in the meat of the hind leg (C14:1, C15:0, γ-linolenic, C20:0 and C20:3 n-3) were detected in perirenal fat, though in very low

percentages. C18:1 *trans* and C20:5 n-3 (EPA) fatty acids were detected in concentrations lower than 0.01%.

3.3. Indices related to human health

Tables 4 and 5 show the indices related to human health for the meat of the hind leg and for perirenal fat, respectively. The nutritional quality of fat has been evaluated in terms of the ratio polyunsaturated:saturated fatty acids (P:S), and the ratio (n - 6):(n - 3) fatty acids. In a balanced diet, the recommended ratio for P:S is 0.45 or higher (Department of Health & Social Security UK, 1994), and that for the ratio n - 6:n - 3 a maximum of 4.0 is recommended.

Selection for growth rate modified some of the indices, although these modifications were small. The highest effect of selection was for the P:S ratio of the meat of the hind leg, which only decreased a 10% in 14 generations of selection, which represents less than a 1% per generation. Selection seems to have improved (P < 0.10) the ratio n - 6:n - 3 fatty acids in the meat, but no differences were found for this ratio in perirenal fat. This ratio is very high in rabbit meat, due to the high content in linoleic acid. (for example, our values were similar to the results published by Dalle Zote (2002), who reported a n - 6:n - 3 index of 11.6 for the meat of rabbit hind leg). As we said before, there are no other selection experiments for which our results could be compared. In the experiment with pigs quoted before, Cameron et al. (2000) found very small differences between the selected lines and the control line for the ratio

Table 4

Least Square means and standard errors of indices related to human health. in rabbit hind leg	meat
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	Group		Significance	
	Control	Selection		
S	35.73 ± 0.31	36.86 ± 0.33	*	
M + P	64.27 ± 0.31	63.14 ± 0.33	*	
P:S	1.06 ± 0.03	0.95 ± 0.03	**	
S:M	1.35 ± 0.0211	1.30 ± 0.02	ns	
S:(M+P)	0.56 ± 0.01	0.59 ± 0.01	*	
Total $n-6$	34.26 ± 0.62	31.58 ± 0.65	**	
n - 6:n - 3	11.47 ± 0.30	10.67 ± 0.32	†	
(M + P):S	1.81 ± 0.02	1.72 ± 0.03	*	

S (saturated fatty acids) = C10:0 (capric) + C12:0 (lauric) + C14:0 (myristic) + C16:0 (palmitic) + C17:0 (margaric) + C18:0 (stearic). M (monounsaturated fatty acids) = C16:1 n - 7 (palmitoleic) + C16:1 n - 9 + C17:1 (heptadecenoic) + C18:1 n - 9 (oleic) + 18:1 n - 7 + C20:1 (icosaenoic).

P (polyunsaturated fatty acids) = C18:2 n - 6 (γ -linoleic) + C18:3 n - 3 (α -linolenic) + C20:2 n - 6 + C20:3 n - 6 + 20:4 n - 6 (arachidonic). n - 6 = C18:2 n - 6 (linoleic) + C20:2 n - 6 + C20:3 n - 6 + C20:4 n - 6 (arachidonic).

n - 3 = C18:3 n - 3 (\$\alpha\$-linolenic).

- ns, No significant.
- $^{\dagger} p < 0.10.$
- $p^* < 0.05.$
- $p^{**} p < 0.01.$

Table 5

Least Square means and standard errors of fatty acids indices of perirenal rabbit fat related with human health

	Group		Significance	
	Control	Selection		
S	35.04 ± 0.34	36.11 ± 0.37	*	
M + P	64.96 ± 0.34	63.89 ± 0.37	*	
P:S	1.08 ± 0.03	1.01 ± 0.04	ns	
S:M	1.29 ± 0.02	1.30 ± 0.02	ns	
S:(M+P)	0.54 ± 0.01	0.57 ± 0.01	*	
Total $n - 6$	33.44 ± 0.71	32.08 ± 0.77	ns	
n - 6:n - 3	8.73 ± 0.15	8.38 ± 0.16	ns	
(M + P):S	1.87 ± 0.03	1.78 ± 0.03	*	

S (saturated fatty acids) = C10:0 (capric) + C12:0 (lauric) + C14:0 (myristic) + C16:0 (palmitic) + C17:0 (margaric) + C18:0 (stearic). M (monounsaturated fatty acids) = C16:1 n - 7 (palmitoleic) + C16:1 n - 9 + C17:1 (heptadecenoic) + C18:1 n - 9 (oleic) + 18:1 n - 7 + C20:1 (icosaenoic).

P (polyunsaturated fatty acids) = C18:2 n - 6 (γ -linoleic) + C18:3 n - 3 (α -linolenic) + C20:2 n - 6 + C20:3 n - 6 + 20:4 n - 6 (arachidonic).

n-6 = C18:2 n - 6 (linoleic) + C20:2 n - 6 + C20:3 n - 6 + C20:4 n - 6 (arachidonic).

n - 3 = C18:3 n - 3 (α -linolenic).

ns, No significant.

 $p^* < 0.05.$

n - 6:n - 3 after seven generations of selection. In the same experiment, the ratio P:S only changed for the line selected for low food conversion rate. Lines selected for growth rate corrected for fatness cannot be properly compared with our line, but they did not change their P:S ratio.

In conclusion, selection for growth rate changes the fatty acid composition of both meat and edible fat. This change can be high for some fatty acids (for example palmitoleic), but changes of indices related to human health are very small. Some of the changes in these indices are favourable and some of them are not favourable, but as a whole, the effects of selection for growth rate would not lower the meat and fat quality from a human health point of view.

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