FATTY ACID COMPOSITION OF RABBIT MEAT WHEN FED A LINSEED BASED DIET DURING DIFFERENT PERIODS AFTER WEANING

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ABSTRACT

The aim of the study was to investigate the dietary use of extruded linseed on the fatty acid (FA) profile of rabbit meat when fed during different weeks of the fattening period. Two diets with comparable energy and protein content but largely different in ω-6/ω-3 ratio were fed to weaned rabbits (29 d old). The ω-3 enriched diet (ω-6/ω-3=1.03) was obtained by incorporation (12.8%) of a concentrate, based on extruded linseed (L) at the expense of raw materials rich in ω-6 PUFA, in a control (C) fattening diet (ω-6/ω-3=4.22). Both diets were fed continuously during 6 weeks (L and C groups) or rabbits shuttled after 4 weeks from one diet to the other (LC and CL groups). At the age of 71 days, out of each treatment group, 12 rabbits with a weight near to the average group weight were slaughtered and the FA profile in the hind leg was determined. Differences in slaughter yield, pH_{24} or colour characteristics were not significant. However, significant differences in FA profile according to the diet as well as the duration of feeding were observed. The α-linolenic acid content amounted to 6.33, 20.91, 12.39, and 15.51%, for C, L, CL and LC treatments, respectively. Thus, even a short distribution (2 weeks) of an ω-3 rich diet allows to produce rabbit meat with a twice as high content of ω-3 FAs. The ω-6/ω-3 ratio dropped from 5.39 to 1.26, 2.61 and 1.84% in C, L, CL and LC hind legs, respectively. The same pattern was found for DPA and DHA, while EPA content was not significantly different between treatments. These results indicate that the decrease of ω-3 FAs is slower compared to the incorporation and that rabbits are able to elongate FAs.

Key words: Fatty acids, Rabbit meat, Feeding, Linseed.

INTRODUCTION

Today's consumers diet is characterised by a high ω-6/ω-3 ratio of polyunsaturated fatty acids (PUFA's) leading to increasing risks of cardiovascular related diseases and even some cancers (Brasseur *et al*., 2004). Scientific evidences on the beneficial effects of the very long-chain ω-3 fatty acids (FA) have been demonstrated for the last half century with e.g. cardio-protective action of eicosapentaenoic acid (EPA, C20:5ω-3) and docosahexaenoic acid (DHA, C22:6ω-3) (Williams, 2000; Ruxton *et al*., 2005). Furthermore it has been shown that linolenic acid (LNA) has immunomodulation properties and serves as a precursor for the synthesis of very long chain (≥20C) PUFA's (Bazinet *et al*., 2004).

In many experiments it has been shown that the PUFA content of meat (especially non-ruminants) can largely be managed by the diet and by consequence can contribute to a reduction of the above mentioned diseases (Raes *et al.*, 2004; Givens, 2005). Because of its high C18:3ω-3 content, linseed is frequently used as a vegetable source of ω-3 PUFA while fish oil is commonly used to enrich the meat with very long chain fatty acids. Rabbit meat is known for its quite low ω-6/ω-3 ratio due to its herbivorous diet (Dalle Zotte, 2002; Combes, 2004). However, less is known about the deposition rate of the dietary PUFA's (Hernández and Gondret, 2006).

The aim of our study was to assess the effect of an ω-3 enriched diet, using extruded linseed at the expense of raw materials rich in ω-6 PUFA, on females' performance and milk composition and the

meat composition of the fattening rabbits. The present paper is restricted to the results related to the effect on the carcass composition and FA profile of the hind leg. Doe performances, milk composition and viability of the progeny have been previously described (Maertens *et al.*, 2005).

MATERIALS AND METHODS

Animals and experimental design

Two isoenergetic and isonitrogenous diets but largely different in ω-6/ω-3 ratio were fed to weaned rabbits (29 d old). The ω-3 enriched diet (ω-6/ω-3 = 1.03) was obtained by incorporation (12.8%) of a concentrate, based on extruded linseed (Nutex®) in a standard fattening diet (Table 1). The extruded linseed replaced mainly soybeans (rich in ω-6 PUFA), wheat, animal fat and beet pulp.

Table 1: Ingredient , chemical and fatty acid composition of experimental diets and extruded linseed

Ingredients	Control diet	ω-3 diet	Nutex®
Nutex® (Extruded linseed)	-	12.8	
Wheat	5.6	1.6	
Soybeans full-fat	6,8	-	
Sunflower meal 27	9.0	11.0	
Beet pulp	1.5	-	
Flax chaff	7.6	6.1	
Animal fat	1.0	-	
Common part[1]	68.5	68.5	
Crude protein (%)	17.2	17.2	18.7
Crude fat (%)	5.5	5.9	22.4
Crude fiber (%)	17.1	16.2	8.2
ADF (%)	17.9	17.6	9.0
ADL (%)	5.2	5.3	3.4
DE (MJ/kg)[2]	10.06	10.04	18.03
C16:0 (%)	14.11	9.30	5.53
C18:0 (%)	4.51	2.82	3.48
C18:2 (%)	48.42	35.05	18.36
C18:3 (%)	11.58	34.41	51.15
SFA (%)	20.34	13.24	9.26
MUFA (%)	19.21	16.89	21.03
PUFA (%)	60.45	69.87	69.71
ω-6/ω-3	4.22	1.03	0.36

[1]Alfalfa meal, wheat shorts, cane molasses, amino acids, minerals and vitamin and trace mineral premix
[2]Determined in a digestibility trial with growing rabbits

Both experimental diets (control diet and linseed, C and L groups respectively) were fed continuously during 6 weeks or rabbits shuttled after 4 weeks from one diet to the other (CL and LC groups, respectively). In total 16 litters of 8 weanlings were used for the study and each litter was randomly allocated to one of the 4 dietary treatments. Feed and water (nipple drinkers) were always available *ad libitum*. Rabbits were caged per four and housed together in an experimental room equipped with central heating and an underpressure ventilation system allowing to maintain the temperature between 16 and 22°C during the entire experiment. At the age of 71 days, out of each treatment group, 12 rabbits with a weight near to the average group weight were slaughtered. Carcass determinations were performed in accordance with the recommendations of Blasco and Ouhayoun (1996).

Determinations and analyses

Colour determinations L* (lightness), a* (redness) and b* (yellowness) were determined by means of reflectance colorimetry thereby using a Labscan II (3) 24 h *post mortem* in the loin (*Longissimus lumborum*). The loin was obtained after cutting the carcass between the 7th and 8th thoracic vertebrae and between the 6th and 7th lumbar vertebrae. pH was measured at 24 hours post-mortem (pH_{24}) by using a glass electrode with a conic tip (SP24, Consort P600), by averaging two independent measures in the hind leg.

A chloroform/methanol (2:1) solution was used to extract the fat of the hind leg muscles. The FA composition was determined by gas-liquid chromatography (VARIAN 3400). The fatty acid methyl esters were prepared by HCl/CH_3-OH transesterification and analysed on a DB-23 capillary column (l=60 m; ID=0.25 mm; FD=0.25 µm) (Chrompack, UK).

Data were subjected to an analysis of variance (Statistica 7 program; Snedecor and Cochran, 1989). An LSD test was used to detect significant ($P<0.05$) differences between treatments.

RESULTS AND DISCUSSION

In Table 2, results related to the carcass and meat characteristics are presented. Differences between dietary treatments in slaughter yield and fat percentage were small and not significantly different ($P>0.05$). pH measured after 24h in the *Longissimus lumborum* (LL) was between 5.76 and 5.80 in all treatments and also no significant ($P>0.10$) differences were measured for the different colour parameters.

The FA profile of the hind leg muscle is presented in Table 3. The dietary inclusion of extruded linseed during the whole fattening period had a very strong impact on the overall FA composition reflected in both the total SFA (30.65 vs. 27.92% for C and L rabbits, respectively) and PUFA (44.67 vs. 49.80% for C and L rabbits, respectively). This clear effect has previously been reported in several other species and also in rabbits by Dal Bosco *et al*. (2004) and Bianchi *et al*. (2006).

Rabbits fed continuously the *ω*-3 diet had a 330% higher α-linolenic acid content compared with the rabbits fed the C diet while their linoleic content was 26.5% lower. This corresponds with the change of both fatty acids in control and linseed diet (297 and 27.6%, respectively) and confirms that the dietary fatty acid content is very well reflected in the meat of especially monogastrics as was recently demonstrated in rabbits by Gigaud and Combes (2007). As a result the *ω*-6/*ω*-3 ratio decreased significantly from 5.39 towards 1.26 when fed the L diet.

Table 2: Carcass and meat characteristics according to the dietary treatment

Feeding scheme	C[1]	L[2]	CL (4 + 2 w)[3]	LC (4 + 2 w)[4]	SEM	P
Live weight (kg)	2.64	2.67	2.63	2.63	0.02	>0.10
Yield (%)	58.3	58.0	59.0	58.2	0.20	>0.10
Carcass fat (%)	4.28	4.32	4.25	4.47	0.13	>0.10
pH_{24h} in the hind leg	5.79	5.77	5.80	5.76	0.	>0.10
Colour of LL						
L*	57.3	56.6	56.3	57.5	0.34	>0.10
a*	-1.93	-2.10	-1.81	-1.89	0.08	>0.10
b*	4.90	4.86	5.14	5.36	0.17	>0.10

[1]C= Control diet during 6 weeks; [2]L= Linseed diet during 6 weeks; [3]CL= C diet (4 weeks) followed by L diet during 2 weeks; [4]LC= L diet (4 weeks) followed by C diet

Also the very long fatty acid profile was significantly influenced by the dietary inclusion of 12.8% of the extruded linseed. Levels of arachidonic acid (C20:4*ω*-6) in the meat dropped significantly when fed the linseed diet during 6 weeks while DPA (C22:5*ω*-3) and DHA (C22:6*ω*-3) significantly increased and EPA (C20:5*ω*-3) remained at the same level. These findings corroborate with the trend found by Bianchi *et al*. (2006). However, Dal Bosco *et al*. (2004) found also a significant increase in EPA while arachidonic acid did not drop in rabbits fed a diet with 8% of linseed. Anyway, the present results confirm that rabbits have the ability to elongate and desaturate LNA due to the activity of caecal microflora and caecotrophe reingestion (Dal Bosco *et al.*, 2004).

If the rabbits were fed the *ω*-3 rich diet only during the 2 last weeks before slaughtering (CL), C18:3*ω*-3 was already twice as high as ($P<0.01$) in the C-rabbits (12.39 vs. 6.33, respectively). Thus even a short distribution of an *ω*-3 rich diet allows to produce meat with a very low *ω*-6/*ω*-3 ratio, in line with the results of Gigaud and Combes (2007). The opposite dietary treatment (LC) resulted in a significant higher C18:3*ω*-3 content (15.51%) compared to the CL treatment. This indicates that the

decrease of LNA when fed an *ω*-6 rich diet is slower than the incorporation of LNA when fed the linseed diet during the same 2 weeks period. The same effect was found for DHA but not for arachidonic acid. It is known that the long chain metabolites of LNA are highly incorporated in muscle phospholipids, of which turnover is slow and more susceptible to change in early growth period (Raes *et al*., 2004).

Table 3: Fatty acid profile (% of total methyl esters) of the hind leg muscles

Feeding scheme[1]	C	L	CL	LC	SEM	P
C14:0	1.82	1.73	1.69	1.77	0.03	NS
C16:0	19.84 a	17.84 b	18.69 ab	18.72 ab	0.21	<0.05
C16:1	2.67	2.31	2.15	2.36	0.09	NS
C18:0	6.31	5.94	6.36	6.27	0.08	NS
C18:1	21.32 a	18.77 b	19.83 bc	20.42 ac	0.22	<0.01
C18:2*ω*-6	33.93 a	24.95 b	30.72c	27.32 b	0.44	<0.001
C18:3*ω*-3 (LNA)	6.33 a	20.91 b	12.39 c	15.51 d	0.20	<0.001
C20:3*ω*-6	0.22	0.20	0.22	0.21	0.01	NS
C20:4*ω*-6 (ARA)	3.46 a	2.70 b	3.64 a	2.75 b	0.11	<0.001
C20:5*ω*-3 (EPA)	0.34	0.28	0.31	0.31	0.04	NS
C22:5*ω*-3 (DPA)	0.14 a	0.41 b	0.25 c	0.26 c	0.02	<0.001
C22:6*ω*-3 (DHA)	0.17 a	0.41 b	0.29 c	0.35 d	0.02	<0.001
∑SFA	30.65 a	27.92 b	29.33 ab	29.25 ab	0.29	<0.05
∑MUFA	24.36 a	21.37 b	22.30 bc	23.11 ac	0.32	<0.01
∑PUFA	44.49 a	49.88 b	47.84 bc	46.71 ab	0.51	<0.01
∑PUFA*ω*-6	37.61 a	27.85 b	34.59 c	30.28 b	0.72	<0.001
∑PUFA*ω*-3	6.98 a	22.03 b	13.25 c	16.43 d	0.81	<0.001
ω-6/*ω*-3	5.39 a	1.26 b	2.61 c	1.84 d	0.24	<0.001

[1]See Table 2; NS: Not significant (P>0.10); a, b: means in the same row differ significantly (P<0.05)

When we compare the FA profile determined in the this study with average literature data of rabbit meat, SFA's represent only about 30% of total FA instead of 39% and PUFA's about 45% instead of 34% (Review of Combes, 2004). This different profile can be explained by the high (unsaturated) fat level in our experimental diets. The *ω*-6/*ω*-3 ratio of rabbits fed the control diet (5.39) was quite in line with the average value found for rabbit meat (5.77) (Combes, 2004). This can be explained by the dietary inclusion of alfalfa meal (25.5%) which is known to be rich in C18:3*ω*-3 besides the soybeans (rich in *ω*-6 FA's) (Sauvant *et al*., 2002).

CONCLUSIONS

From this experiment, it can be concluded that rabbits' FA profile can be easily managed by the dietary FA profile and the use of extruded linseed (12.8%) results in meat with nearly the same amount of *ω*-3 as *ω*-6 FA's. If such diet is fed only 2 weeks before slaughtering, the *ω*-3 level is already twice as high as that of the control group. When the decrease was compared with the incorporation, a slower reduction of *ω*-3 FA's was observed in the same 2-weeks period. Finally rabbits have the ability to elongate FA's and with the *ω*-3 rich diet, significant higher levels of DPA and DHA were determined.

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REFERENCES

Bazinet R.P., Douglas H., Mc Millan E.G., Wilkie B.N., Cunnane S.C. 2004. Dietary c18:3ω3 influences immune function and the tissue fatty acid response to antigens and adjuvant. *Immunology Letters, 95, 85-90.*

Blasco A., Ouhayoun J. 1996. Harmonization of criteria and terminology in rabbit meat research. Revised proposal. *World Rabbit Sci., 4, 93-98.*

Bianchi M., Petracci M., Cavani C. 2006. Effects of dietary inclusion of dehydrated lucerne and whole linseed on rabbit meat quality. *World Rabbit Sci., 14, 247-257.*

Brasseur D., Delzenne N., Henderickx H., Huyghebaert A., Kornitzer M. 2004. Recommandations et allégations concernant les acides gras omega-3. *Conseils Supérieur D'hygiène, Belgique, 108p. (www.health.fgov.be/csh_hgr).*

Combes S. 2004. Valeur nutritionelle de la viande de lapin. *Inra Prod. Anim., 17, 373-383.*

Dal Bosco A., Castellini C., Bianchi L., Mugnai C. 2004. Effect of dietary α-linolenic acid and vitamin e on the fatty acid composition, storage stability and sensory traits of rabbit meat. *Meat Sci., 66, 407-413.*

Dalle Zotte A. 2002. Perception of rabbit meat quality and major factors influencing the rabbit carcass and meat quality. Review. *Livest. Prod. Sci., 75, 11-32.*

Gigaud V., Combes S. 2007. Effect of a decreasing omega 6/omega 3 ratio on fatty acid content of rabbit meat. *In: Proc. 12èmes Journ. Recherche Cunicole, 2007 November, Le Mans, France, 191-194.*

Givens D.I. 2005. The role of animal nutrition in improving the nutritive value of animal-derived foods in relation to chronic disease. *Proc. Nutrition Society, 64, 395-402.*

Hernández P., Gondret F. 2006. Rabbit meat quality. *In: Maertens and Coudert (Eds.) Recent Advances in Rabbit Sciences. COST and ILVO Publication, Melle, Belgium, 269-290.*

Maertens L., Aerts J.M., De Brabander D.L. 2005. Effet d'un aliment riche en acides gras omega-3 sur les performances et la composition du lait des lapines et la viabilite de leur descendance. *In: Proc. 11èmesJourn. Recherche Cunicole, 2005 November, Paris, France, 205-208.*

Ruxton C.H.S., Calder P.C., Reed S.C., Simpson M.J.A. 2005. The impact of long-chain n-3 polyunsaturated fatty acids on human health. *Nutrition Research Reviews, 18, 113-129.*

Sauvant D., Perez J.M., Tran G. 2002. Tables de composition et de valeur nutritive des matières premières destinées aux animaux d'élevage. *INRA Editions, Rue Claude Bernard, Paris, 301 p.*

Snedecor G.W., Cochran W.G. 1989. Statistical Methods (8th Edn.). *Iowa State University Press. Ames, IA, USA.*

Williams C.M. 2000. Dietary fatty acids and human health. *Ann. Zootech., 49, 165-180.*