

## FATTY ACID COMPOSITION OF TWO DIFFERENT MUSCLES IN RABBITS : ALTERATIONS IN RESPONSE TO SATURATED OR UNSATURATED DIETARY FATTY ACID COMPLEMENTATION

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**ABSTRACT** : Two experiments were carried out to determine differences in the fatty acid profile of two rabbit muscles, *longissimus dorsi* (l.d.) and *vastus lateralis* (v.l.). Fatty acid analysis was carried out by gas chromatography on the samples of ten 8-week-old rabbits, in the first experiment after a 4-week feeding period with a commercial pelleted diet, to obtain information about possible differences. In the second experiment 4-week treatments were performed with two high-fat experimental feeds, from which one was complemented with a saturated animal fat source (FAT) and the other with an unsaturated fat source, full-fat soya (SOYA). Samples

were obtained from eighteen 8-week-old rabbits per diet. With the commercial diet, significant differences were evidenced between the 2 muscles for C14:0, C18:1 (20.9 and 17.5 % for l.d. and v.l.), C18:2 n-6 (24.5 and 27.9 % for l.d. and v.l.) and C20:4 (n-6). On average C16:0 proportion was 26.8 %, that of C18:0 was 8.5% and that of C18:3 (n-3) was 2.32 %. After FAT feeding C14:0, C16:1, C18:0, C18:1, C18:2(n-6) and C20:4 were found to differ. SOYA feeding caused alterations in C14:0, C16:0, C18:0, C18:1 and C18:3. With SOYA feeding proportion of C18:2 increased up to 35 % in both muscles.

**RÉSUMÉ** : Composition en acides gras de deux muscles chez le lapin: modifications en réponse à l'addition de lipides saturés ou insaturés dans l'alimentation.

Deux expériences ont été conduites pour déterminer les différences de profils en acides gras de 2 muscles chez le lapin le *longissimus dorsi* (l.d.) et le *vastus lateralis* (v.l.). Les analyses ont été conduites par chromatographie en phase gazeuse sur des échantillons prélevés sur des lapins âgés de 8 semaines. Avant le sacrifice, les lapins avaient reçu pendant 4 semaines un aliment standard commercial dans l'expérience 1. Dans la seconde expérience, les lapins ont reçu dans les mêmes conditions un aliment riche en lipides supplémenté soit avec des graisses animales saturées (FAT),

soit avec des lipides insaturés fournis par de la graine de soja entière (SOYA). Avec l'aliment commercial, des différences de profil ont été observées entre les 2 muscles pour le C:14, le C18:1 (20,9 et 17,5 % pour l.d. et v.l.), le C18:2 n-6 (24,5 et 27,9 % pour l.d. et v.l.), ainsi que pour le C20:4 n-6. En moyenne la proportion de C16:0 était de 26,8%, celle du C18:0 de 8,5% et celle du C18:3 n-3 de 2,68%. L'addition de FAT a modifié les proportions de C14:0, C16:1, C18:0, C18:1, C18:2 n-6 et de C20:4. L'addition SOYA a modifié les proportions de C14:0, C16:0, C18:0, C18:1 et de C18:3 n-3. Elle a aussi accru la proportion de C18:2 n-6 jusqu'à lui faire représenter environ 35% des acides gras dans chacun des 2 muscles étudiés.

### INTRODUCTION

The relative amounts of fatty acids in animal tissues are of determining importance from the viewpoint of functional properties and tissue quality. Regarding fatty acid analysis in rabbit fat tissue (LIN *et al.*, 1993), muscles (OUHAYOUN *et al.*, 1985) and blood plasma (FEKETE *et al.*, 1989) have been the most widely investigated.

There are plenty of data available from comparative studies on the fatty acid profile of different rabbit tissues. It is an interesting fact that there are great differences between diverse tissue types, because the origin of fatty acids is mainly the blood plasma pool which derives in first line from the feed (DI MARINO *et al.*, 2000). However, fatty acid profile is dependent on numerous factors. COBOS *et al.* (1995) published differences of the fatty acid composition between species (Spanish wild rabbits and hares), and BOCCIGNONE *et al.* (1986) between breeds (Burgundy Fawn, Californian and New Zealand White). In addition, differences can also be measured depending on slaughter weight and age (PARIGI-BINI *et al.*, 1992).

Differences in the fatty acid profile can be detected between tissues with divergent functional properties; WAKATA *et al.* (1990) published results between red and white muscle types. Differences in muscle design and functional properties hypothesize various fatty acid profiles. Not only muscle phospholipids (ALASNIER *et al.*, 1998), but also triglycerides are proven to alter in fatty acid composition in different muscles.

Based on the fact that dietary fatty acids are effectively detectable in numerous organs, our investigation was designed to determine possible differences between two muscles. In the experiments *muscle longissimus dorsi* (l.d.) and *muscle vastus lateralis* (v.l.) of Pannon white rabbits were analysed, which both are fast twitch white muscles. The first aim of our trials was to establish the differences existing in the fatty acid profile of the two muscles after feeding a commercial diet and the second to measure the variations induced by two experimental diets, containing either saturated or unsaturated fat sources.

### MATERIALS AND METHODS

First, the fatty acid composition of l.d. and v.l. from 10 Pannon white rabbits fed a commercial diet

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**Table 1 : Composition of the diets applied in the examinations**

Ingredients %	Diet		
	Commercial	FAT	SOYA
Wheat	un.	10	10.5
Barley	un.	10	10
Soybean	un.	11	-
Full fat soybean	un.	-	24.5
Alfalfa	un.	40	40
Bran	un.	17	13
Fat powder (40%)	un.	10	-
Vitamins, mineral premix, etc.	un.	2	2
<i>Chemical Composition (% DM)</i>			
Dry matter (DM)	88.0	89.33	89.4
Crude protein	16.0	16.6	18.9
Crude fat	2.3	6.04	6.07
Crude fibre	15.5	13.2	13.5
DE rabbit (calc.) MJ/kg	10.3	10.8	12.5
<i>Fatty acid composition % total</i>			
C14:0	0.91	1.22	0.23
C16:0	23.03	21.90	12.30
C16:1	0.24	1.96	0.42
C17:0	0.28	0.14	trace
C18:0	4.10	9.60	3.66
C18:1	20.20	31.95	22.34
C18:2 (n-6)	44.60	24.38	50.73
C18:3 (n-3)	5.83	7.40	9.37
Σ sat. / Σ unsat.	0.40	0.50	0.19

was determined. In the second trial 36 rabbits were fed two experimental diets: the former was complemented with 10% of animal fat powder (FAT) and the other with 24.5% of full-fat soybean (SOYA). Ingredients, chemical composition and fatty acid composition of the feeds are shown in Table 1.

The experiment started at the age of 4 weeks and lasted until the age of 8 weeks. Male rabbits were housed in wire net cages and were fed *ad libitum*. After 4 weeks rabbits were slaughtered and samples of 5g from the left l.d. (always from the same location, i.e. from the 2<sup>nd</sup> to the 3<sup>rd</sup> lumbar vertebrae) and v.l. were taken and were stored at -20°C until fatty acid analysis. Prior to gas chromatography samples were homogenized, fat content was extracted (chloroform:methanol (2:1)) and extracted lipids were methyl-esterified. Individual fatty acids were identified

using qualitative standards.

Results were given in percentage of total fatty acids. Statistical analysis was carried out with SPSS 8 for Windows (1996), applying independent samples t-test, Chi<sup>2</sup>-test and two-way ANOVA, where the effects of muscle type and feed were involved into the model.

## RESULTS AND DISCUSSION

### Diets

In order to determine the possible differences between the fatty acid compositions of the diets Chi<sup>2</sup>- test was performed. Results showed clear differences between the fatty acid composition of the commercial and the FAT diet ( $P < 0.05$ ) and between the two experimental diets, i.e. FAT and SOYA ( $P < 0.01$ ).

### Comparison of l.d. and v.l. using commercial diet

Results of the fatty acid analysis are shown in Table 2. Pentadecanoic (C15:0) and margaric acid (C17:0), representing only a small proportion of the saturated fatty acids did not show significant differences between muscles. Also palmitic acid (C16:0) and stearic acid (C18:0), detectable in relatively high proportions did not reach the significant level between muscles. In contrast C14:0 (miristic acid), C18:1 (oleic acid), C18:2(n-6) (linoleic acid) and C20:4(n-6) (arachidonic acid) showed statistically provable differences.

### Comparison of l.d. and v.l. using saturated fatty acid complementation

The experimental diet FAT had considerable effect onto the fatty acid profile of l.d. and v.l. Statistically provable differences between the muscles were found in miristic acid, palmitoleic acid (C16:1), stearic acid, oleic acid, linoleic acid and arachidonic acid. Significance levels are shown in Table 3.

The great influence of dietary stearic acid might be of interest as its proportion in the two muscles was relatively high (9.5 and 10.1% in l.d. and v.l.) and was significantly different. Dietary stearic acid (9.6%) seemed to have a strong effect on muscles' stearic acid content, in agreement with the results of KESSLER *et al.* (1994). Oleic acid showed significantly higher

**Table 2 : Results of the muscle fatty acid analysis obtained with commercial diet.**

		n	C14:0	C15:0	C16:0	C16:1	C17:0	C18:0	C18:1	C18:2	C18:3	C20:4
l.d. Commercial	mean	10	1.73	0.43	27.14	2.35	0.51	8.12	20.90	24.52	2.48	5.61
	SD		0.31	0.09	0.78	0.62	0.12	0.82	1.55	1.27	0.56	1.00
v.l. Commercial	mean	10	1.38	0.47	26.40	2.17	0.53	8.95	17.50	27.90	2.16	6.43
	SD		0.24	0.04	1.41	0.48	0.07	1.36	1.85	1.45	0.29	1.08
Significance level			*	NS	NS	NS	NS	NS	**	**	NS	*

**Table 3 : Incidence of dietary fat on muscles fatty acid composition.**

		n	C14:0	C15:0	C16:0	C16:1	C17:0	C18:0	C18:1	C18:2	C18:3	C20:4
l.d. FAT	mean	18	1.70	0.39	25.59	2.67	0.45	9.48	27.91	20.74	2.68	4.95
	SD		0.29	0.02	0.93	0.59	0.06	0.62	1.02	0.98	0.32	0.77
v.l. FAT	mean	18	1.26	0.37	25.36	2.14	0.45	10.11	23.92	24.42	2.16	5.70
	SD		0.26	0.03	1.23	0.38	0.06	0.97	1.26	1.03	0.27	0.94
<b>Significance level</b>			***	NS	NS	*	NS	*	***	***	NS	*
l.d. SOYA	mean	18	1.31	0.37	19.35	1.91	0.29	7.28	22.66	34.83	4.10	4.50
	SD		0.29	0.03	4.71	0.68	0.06	1.13	1.13	1.24	0.66	1.71
v.l. SOYA	mean	18	0.98	0.38	21.65	1.56	0.36	8.77	18.98	35.21	2.98	5.90
	SD		0.31	0.04	0.75	0.62	0.09	1.05	1.34	1.21	0.61	1.34
<b>Significance level</b>			*	NS	***	NS	NS	***	***	NS	***	NS
Feed		36	***	NS	***	***	***	***	***	***	***	***
Muscle		36	***	NS	NS	**	*	***	***	***	***	***
Feed x Muscle		36	NS	*	*	NS	NS	NS	NS	***	*	NS

\*\*\* P< 0.001 ; \*\* P< 0.01 ; \* P< 0.05 ; NS: P> 0.05 ; un.: unknown

proportion in l.d. and reached firmly elevated amounts in both muscles according to the relatively high oleic acid content of the FAT diet. The levels of linoleic and arachidonic acids were significantly higher in v.l. than in l.d., showing the same tendency after feeding the commercial diet.

#### Comparison of l.d. and v.l. using unsaturated fatty acid complementation

SOYA feed had also distinctly detectable effects on the fatty acid composition of both muscles investigated. Contents of the two muscles in miristic acid, palmitic acid, stearic acid, oleic acid and linolenic acid (C18:3 n-3), were statistically provable as different (see Table 3).

The relative amount of miristic acid was markedly lower compared to the level after FAT diet, however, differences found in the other two feeding groups persisted.

Stearic acid showed a higher proportion in v.l. than in l.d.

Concerning oleic acid, the ca. 4% difference (experienced after the commercial and the FAT diets) was also seen when fed SOYA. Results obtained in the present study are accordant with those of RAIMONDI *et al.* (1975) who observed great increase in the proportion of oleic acid after dietary unsaturated fatty acid complementation. Oleic acid in our experiments also sensitively reflected the proportion of supplement in the feed. However, oleic acid was present in a significant higher proportion in l.d. after feeding all three diets. CAVANI *et al.* (1996) found an elevation of oleic acid followed by a decrease of linoleic acid after full-fat soybean addition to the feed. The same tendency was observed in our trials.

The SOYA diet, containing 50.7% linolenic acid, which was twice more than in FAT, strongly increased the linolenic acid proportion in muscles, mainly in l.d., which showed a significantly higher proportion.

Arachidonic acid did not reach the significance level of 5% between muscles after SOYA feeding, possibly caused by relatively high standard deviation.

#### Differences between muscles of different feeding groups

From the results above it can be seen that the diets affected the proportion of all the fatty acids except of pentadecanoic and margaric acid. Muscles showed very similar proportions of C15:0, however, its amount was slightly higher in v.l. than l.d. by feeding commercial and SOYA diets and minimally lower after feeding the FAT diet.

Furthermore, the two muscles differed in palmitic acid, this difference was only affected by the SOYA diet. It is presumable that m.v.l. is richer in palmitic acid than l.d. and the difference might be masked when the dietary amount is relatively high.

This tendency was also observed in linoleic acid which showed significantly higher percentages in v.l. following commercial and FAT diets, while significance was not reached after SOYA diet, containing 50.7% of this fatty acid.

Linolenic acid always showed higher concentrations in l.d. than v.l., but the difference was significant only after feeding the SOYA diet, containing the highest amount of it. In this case it is hypothesized that to attain the optimal function of l.d. a higher level of linolenic acid is needed that was only reached after high dietary assumption, in the SOYA diet.

Significant feed-muscle interactions were observed for C15:0, C16:0, C18:2 and C18:3, clearly indicating the cases where the effect of muscle type and feed can not be handled independently.

### CONCLUSION

In the authors' opinion l.d. and v.l. can be characterized taking into account the differences found after the feeding of three diets with seriously different fatty acid compositions. The high-fat experimental diets with saturated or unsaturated fatty acid profiles made it possible to obtain information regarding the response of the investigated muscles to dietary fatty acid complementation. From the results of our trials it can be clearly seen, that after four-week trial periods dietary fatty acids are clearly detectable in the muscle tissue of rabbits. Results obtained could serve as comparison basis in our further investigations, where analysis of the same muscles is planned.

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