Omega-3 polyunsaturated fatty acid from extruded linseed influences the fatty acid composition and sensory characteristics of dry-cured ham from heavy pigs

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Omega-3 polyunsaturated fatty acid from extruded linseed influences the fatty acid composition and sensory characteristics of dry-cured ham from heavy pigs[1]

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ABSTRACT: Twenty-four barrows (78.1 ± 1.7 kg of initial BW) were fed a control diet (2.5% sunflower oil) or an experimental diet containing 5% whole extruded linseed. Both diets were supplemented with 170 mg of vitamin E/kg of feed. At slaughter (160 ± 9.2 kg of BW), 6 hams were collected per group and delivered to Stazione Sperimentale per l'Industria delle Conserve Alimentari of Parma for seasoning. There was no effect ($P > 0.05$) of diet on the physicochemical characteristics of dry-cured ham. The linseed diet increased ($P < 0.05$) the content of n-3 PUFA in trimmed fat (green ham), semimembranosus muscle, subcutaneous adipose tissue, and the whole slice (dry-cured ham). Moreover, there was a decrease in the n-6:n-3 PUFA ratio from 12 to 3 in all of those tissues. In contrast, the greater α-linolenic acid content in linseed caused an increase in the iodine value of green hams to a value that is not accepted by the Parma Ham Consortium. A sensory consumer test indicated that linseed-enriched dry-cured ham had the least acceptance score for odor, taste, and overall acceptability descriptors. These data suggest that the use of extruded linseed for pig feed is an acceptable way to improve the nutritional quality of long-cured pork products but could be limited by negative effects on the sensory characteristics of dry-cured ham.

Key words: dry-cured ham, linseed, n-3 polyunsaturated fatty acid, pig, pork quality, sensory evaluation

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INTRODUCTION

Long-chain n-3 PUFA consumption is considered to have beneficial effects on human health (Simopoulos, 2001). However, people in the Western world currently consume an inadequate quantity of some PUFA; they consume too much linoleic acid (**LA**, C18:2n-6) and too little α-linolenic acid (**ALA**, C18:3n-3) and long-chain n-3 PUFA (C20:5n-3 + C22:5n-3 + C22:6n-3; Albert et al., 1998). One way to increase the intake of n-3 PUFA without changing the nutritional behavior of consumers would be to fortify traditional food items such as meat and meat products with n-3 PUFA (e.g., feeding pigs diets containing linseed). Linseed is rich in ALA, a precursor of the longer chain n-3 PUFA. However, the conversion from ALA to n-3 PUFA is not efficient in adult humans (Burdge et al., 2002) and the incorporation of n-3 PUFA into foods, including muscle food products, poses some difficulties. Polyunsaturated fatty acids are known to be more susceptible to oxidation than MUFA or SFA. Moreover, the increased degree of unsaturation of these fatty acids makes them susceptible to oxidative damage (Higgins et al., 1999), which negatively affects the processing characteristics and sensory quality and acceptability of processed meat products (Warnants et al., 1998). Incorporation of n-3 PUFA into meat may lead to oxidation (one of the main causes of functional, sensory, and nutritional quality deterioration in meat and meat products) because of the development of off-flavors, the insolubilization of proteins, and the formation of free radicals and other oxidized compounds, such as cholesterol oxidation products (O'Keefe et al., 1995; Morrissey et al., 1998). Oxidative phenomena in mus-

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cle-based foods take place immediately after slaughter (or even preslaughter), when cellular mechanisms controlling lipid oxidation no longer function (Morrissey et al., 1998). However, fatty acids and AA contribute to the development of aromas characteristic of processed meat products such as dry-cured ham. The objective of the present study was to study lipid composition, nutritional properties, and sensory characteristics of green and dry-cured ham produced from pigs fed either a diet containing 5% extruded linseed or a control diet; the pigs were slaughtered when they reached 160 kg of BW.

MATERIALS AND METHODS

All experimental procedures were carried out in accordance with the guidelines of the French Institut National de la Recherche Agronomique (**INRA**) for the care and use of laboratory animals.

Animals and Diets

A total of 24 castrated crossbred pigs [Large White × (Landrace × Pietrain)] with a BW of 78.1 kg (±1.7 kg) were chosen on the basis of their BW and were assigned to 1 of 2 dietary treatments. All pigs (12 per treatment) were slaughtered at 160.0 kg (±9.2 kg) of BW. The diets, formulated to be isoenergetic and isolipidic, differed in lipid source and in fatty acid composition: 2.5% sunflower oil was present in the control diet, and 5% extruded linseed was included in the linseed diet. The fatty acid composition of each diet is shown in Table 1. Both diets contained 170 mg of vitamin E and 250 μg of selenium (170 μg as selenite, and 80 μg as selenium-methionine) per kilogram of feed. The animals were kept in individual cages with a slatted floor, in total confinement, in an environmentally controlled building at the experimental station of the INRA center of Saint Gilles (France), with ad libitum access to feed and water until slaughter. Details of the diet composition, experimental procedure, and data on animal growth and meat quality were reported in a previous paper (Corino et al., 2008).

Ham Measurements and Tissue Sampling

At 24 h postmortem, samples of trimmed fat were cut from the hams of the right side of each carcass of all pigs (total of 24) and were then vacuum-packed, frozen, and stored at −20°C for analysis of total lipid and fatty acid composition. Six randomly selected barrows per treatment (total of 12) were slaughtered, and 24 h after slaughter and cold-trimming, the left hams were weighed and sent to the Stazione Sperimentale per l'Industria delle Conserve Alimentari (Parma, Italy), where they were processed and ripened using the conventional technology for transformations such as Parma ham (Italian DPO), as specified by the regulation [Council Regulation (EEC) No. 2081/92] stated by the Parma Ham Consortium (1992). The production process for Parma ham is the same as reported by Corino et al. (2003). After 16 mo of seasoning, the thighs were weighed to assess the percentage drop-weight. The percentage drop-weight is expressed as the percentage of weight loss between the green and seasoned product. Dry-cured hams were sectioned perpendicularly to the bone at the knee level. The samples containing essentially 3 muscles [semimembranosus (**SM**), biceps femoris, and semitendinosus] were vacuum-packaged and stored at 4°C for sensory and chemical analyses. Samples of the SM and subcutaneous adipose tissue from all hams were taken, vacuum-packed, frozen, and stored at −20°C pending analysis of chemical composition, total lipids, and fatty acid composition. At sampling, the color of the dry-cured ham was measured on the SM muscle and on the subcutaneous adipose tissue. Tristimulus color coordinates (L*, a*, b*) were record-

Table 1. Fatty acid composition (g/100 g of fatty acids) of diets

	Diet	
Fatty acid	Control	Linseed
C14:0	0.17	0.16
C14:1	0.01	0.01
C16:0	14.51	14.22
C16:1n-7	0.39	0.36
C18:0	3.33	3.15
C18:1n-9	25.64	20.91
C18:2n-6	51.34	39.61
C20:0	0.02	0.02
C18:3n-3	2.58	19.96
C20:1n-9	0.43	0.44
C20:2n-6	0.28	0.27
C20:3n-3	0.65	0.34
C20:4n-6	0.03	0.04
C22:1	0.11	0.15
C20:5n-3	0.12	0.07
C24:0	0.22	0.10
C24:1	0.07	0.06
C22:5n-3	0.01	0.02
C22:6n-3	0.09	0.11
Total	100	100
SFA[1]	18.25	17.65
MUFA[2]	26.65	21.93
PUFA[3]	55.10	60.42
n-6 PUFA[4]	51.65	39.92
n-3 PUFA[5]	3.45	20.50
n-6:n-3 PUFA	14.97	1.95

[1]The percentage of SFA is the sum of C14:0, C16:0, C18:0, C20:0, and C24:0.

[2]The percentage of MUFA is the sum of C14:1, C16:1, C18:1, C20:1, C22:1, and C24:1.

[3]The percentage of PUFA is the sum of C18:2, C18:3, C20:2, C20:3, C20:4, C20:5, C22:5, and C22:6.

[4]The percentage of n-6 PUFA is the sum of C18:2, C20:2, and C20:4.

[5]The percentage of n-3 PUFA is the sum of C18:3, C20:3, C20:5, C22:5, and C22:6.

ed using a CR-300 Chroma Meter (Minolta Cameras, Osaka, Japan).

Chemical Analysis

All chemicals used in this study were of reagent grade. Unless otherwise indicated, reagents were purchased from Sigma Chemical Co. (St. Louis, MO). The chemical composition (DM, CP, ether extract, ash; AOAC, 2000) was determined using the SM muscle of the dry-cured ham as the sample. The proteolysis index was calculated as the percentage ratio between nitrogen soluble in 5% trichloroacetic acid and total nitrogen (AOAC, 2000). The sodium chloride concentration in the processed product was detected by titration with silver nitrate according to the method of Mohr (AOAC, 2000).

Fatty Acid Analysis

Lipid extraction was conducted using chloroform: methanol (2:1) according to the method of Folch et al. (1957). Lipids were extracted from 10 g of each sample of the SM, from the whole slice, and from 1.5 g of each sample of trimmed and subcutaneous adipose tissue. The extracts were dried under vacuum on a rotary evaporator (Laborota 4000, Heidolph Instruments, Milan, Italy).

Fatty acid composition was measured after methylation of the samples. Fatty acid methyl esters were prepared with boron trifluoride methanol according to procedures developed by Morrison and Smith (1964). Trimmed fat samples of green hams were analyzed on a gas chromatograph equipped with a fused-silica capillary column (25 m × 0.25 mm internal diameter) with a base-deactivated silica stationary phase (a 0.25-μm film thickness), filled with a stationary phase (80% biscyanopropyl and 20% cyanopropylphenyl), and using margaric acid (C17:0) as the internal standard. The furnace temperature was 180°C, and injector and detector temperatures were 240°C. For all samples, retention times and peak areas were determined by chromatography (Nelson Analytical, Manchester, NH). The identities of the peaks were verified by comparison with the retention times of standard fatty acid methyl esters. The results were expressed as the percentage of the total fatty acid composition of trimmed and subcutaneous adipose tissue, SM muscle, and the whole slice. When the whole slice was analyzed, results were also expressed as milligrams per 100 g of sample.

The iodine value (**IV**) was calculated from the fatty acid composition (g/100 g of total FA) according to the AOAC (2000) equation:

$$\begin{aligned}\text{IV} = {} & (\text{C16:1n-7} \times 0.950) + (\text{C18:1n-9} \times 0.860) \\ & + (\text{C18:2n-6} \times 1.732) + (\text{C18:3n-3} \times 2.616) \\ & + (\text{C20:1n-9} \times 0.785) + (\text{C22:1n-9} \times 0.723).\end{aligned}$$

Sensory Analysis

An affective method of preference (Association Française de Normalisation, 2000) has been used to evaluate consumer acceptability. The panel consisted of 100 consumers distributed evenly by age and sex. The consumers were asked to evaluate the product visually (appearance, fat color, and muscle color) and then organoleptically (odor, taste, saltiness, and consistency) to evaluate the 2 classes of product, finally expressing a judgment on overall acceptability. The samples were cut into 0.5-mm slices and served in random order at room temperature in Petri dishes coded with 3-digit numbers. The judgments were expressed individually on a 9-point hedonic scale ranging from disliked extremely (score = 1) to liked extremely (score = 9; Peryam and Pilgrim, 1957); consumers were isolated in individual booths to reduce collaboration, and distilled water and unsalted crackers were provided to the panelists between successive ham samples. All assessments were carried out in a sensory laboratory equipped according to UNI-ISO 8598 recommendations (International Organization for Standardization, 1989).

Statistical Analysis

The statistical model for physicochemical characteristics of the subcutaneous adipose tissue and SM of dry-cured ham and for sensory analysis of the whole slice used the variable diet as the only difference between samples. The model included diet for analysis of total lipid content, fatty acid composition, and IV of the trimmed fat and whole slice. Concerning total lipid content and fatty acid composition of subcutaneous adipose tissue and SM of dry-cured ham, the statistical model included the main effect of the diet (control and linseed), the tissue (muscle and fat), and their interaction. Statistical analysis of the data was performed by ANOVA and the least separation distance procedure was used for separation of means (SPSS Inc., Chicago, IL). The individual carcass was considered the experimental unit. Data are presented as the means of each group and SEM. Differences were considered significant if $P \leq 0.05$.

RESULTS

Ham Physicochemical Characteristics

Green ham weight and weight loss were not affected by dietary treatment ($P > 0.05$). Dietary treatment did not affect the coordinates L*, a*, and b* of the SM muscle and subcutaneous adipose tissue color of dry-cured hams (Table 2). The linseed-containing diet did not affect ($P > 0.05$) the DM, CP, ether extract, and ash content of dry-cured hams (Table 2). The proteolytic index and the salt content of dry-cured hams were not affected ($P > 0.05$) by dietary linseed supplementation (Table 2).

Table 2. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on ham weight loss, color, chemical composition (as percentage of fresh weight basis), salt concentration, and proteolysis index of dry-cured ham

Item	Control diet	Linseed diet	SEM[1]	*P*-value
Green ham weight, kg	14.08	14.56	0.302	0.437
Ham weight loss, %	31.72	32.47	0.399	0.371
Subcutaneous adipose tissue color[2]				
L*	69.63	65.38	5.567	0.434
a*	6.00	7.29	0.847	0.472
b*	6.00	5.43	0.240	0.247
Semimembranosus muscle color				
L*	33.86	35.60	0.576	0.137
a*	11.59	11.47	0.232	0.807
b*	4.77	4.95	0.223	0.709
Chemical composition				
DM, %	46.95	47.08	0.590	0.917
CP, %	29.41	31.37	0.756	0.210
Ether extract, %	5.69	4.83	0.388	0.289
Ash, %	7.72	8.11	0.201	0.369
Salt concentration, %	8.92	9.45	0.180	0.152
Proteolysis index, %	33.19	32.48	0.990	0.737

[1]n = 6.
[2]L* = lightness (0 = black, 100 = white), a* = redness (+50 = red; −50 = green), b* = yellowness (+50 = yellow; −50 = blue).

The total lipid content in trimmed fat of green ham (73.10 vs. 76.05%; SEM = 1.099) and in SM (7.68 vs. 6.88%; SEM = 4.022), subcutaneous adipose tissue (75.44 vs. 80.27%; SEM = 4.022), and the whole slice (26.96 vs. 25.34%; SEM = 1.048) of dry-cured hams was not affected ($P > 0.05$) by dietary treatment.

Green Ham Fatty Acid Composition

No differences were observed in total SFA, MUFA, and PUFA content in the trimmed fat of green ham (Table 3; $P > 0.05$). The increase in total n-3 and the decrease in total n-6 PUFA content reduced the n-6:n-3 PUFA ratio ($P < 0.001$) in the trimmed fat of linseed-fed pigs (−78% vs. the control values). However, differences ($P < 0.05$) between the dietary treatments were detected in the content of individual SFA (C20:0 and C24:0), MUFA (C22:1n-9 and C24:1n-9), n-6 PUFA (C18:2n-6, C20:2n-6, and C20:4n-6), and n-3 PUFA (C18:3n-3, C20:3n-3, C20:5n-3, and C22:5n-3). In particular, the percentage of ALA ($P < 0.001$) increased 4-fold, eicosatrienoic acid (C20:3n-3; $P < 0.001$) doubled, eicosapentaenoic acid (**EPA**, C20:5n-3; $P < 0.001$) increased 5-fold, and docosapentaenoic acid (**DPA**, C22:5n-3; $P < 0.001$) increased 3-fold in green ham from linseed-fed pigs compared with control pigs. The fatty acid composition also affected the IV of trimmed fat. Specifically, the greater content in ALA caused a greater IV ($P < 0.01$) in green ham from linseed-fed pigs than in the control hams.

Dry-Cured Ham Fatty Acid Composition

The dietary treatment caused some changes in the fatty acid composition of SM of dry-cured ham (Table 4). The SM of linseed dry-cured ham showed a greater ($P < 0.05$) content of C16:0, total n-3 PUFA, and particularly ALA, EPA, and DPA than the SM of control ham. The percentage of ALA increased 4-fold, and EPA and DPA were nearly doubled in dry-cured ham from linseed-fed pigs compared with ham from control pigs. The SM composition of control dry-cured ham showed a greater content of n-6 PUFA than the SM of linseed dry-cured ham; in particular, LA ($P < 0.05$) was greater in control than linseed dry-cured ham. As a consequence of these changes in the fatty acid composition of the SM, the n-6:n-3 PUFA ratio was markedly reduced (−68% vs. the control values) in the muscle of dry-cured ham from pigs fed linseed diets ($P < 0.001$).

The dietary treatment produced changes in the fatty acid composition of subcutaneous adipose tissue from dry-cured ham (Table 4). The SFA (C14:0, C16:0) and MUFA (C16:1n-7) were greater ($P < 0.05$) in linseed dry-cured ham, whereas C20:0 was decreased ($P < 0.05$) compared with the control ham. The subcutaneous adipose tissue of linseed dry-cured ham showed an increased content of total n-3 PUFA ($P < 0.001$), ALA ($P < 0.001$), and DPA ($P < 0.001$). The percentage of ALA in the linseed dry-cured ham increased 3-fold and DPA doubled compared with the control dry-cured ham. The subcutaneous adipose tissue composition of the control dry-cured ham showed a greater content of n-6 PUFA ($P < 0.001$), particularly of LA ($P < 0.001$), compared with the subcutaneous adipose tissue of linseed dry-cured ham. As a consequence of these changes in the fatty acid composition of subcutaneous adipose tissue, the n-6:n-3 PUFA ratio was markedly reduced, by 76% relative to control values, in the subcutaneous adipose tissue of linseed dry-cured ham ($P < 0.001$).

Table 3. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on fatty acid composition (g/100 g of fatty acids) and iodine value (IV) of trimmed fat

Fatty acid	Control diet	Linseed diet	SEM[1]	*P*-value
C14:0	1.22	1.19	0.018	0.669
C14:1	0.06	0.06	0.003	0.087
C16:0	23.48	23.46	0.152	0.937
C16:1n-7	2.00	2.00	0.058	0.991
C18:0	12.58	12.26	0.229	0.499
C18:1n-9	40.73	40.60	0.267	0.818
C18:2n-6	16.50	13.31	0.405	0.001
C20:0	0.20	0.16	0.012	0.047
C18:3n-3	0.93	4.21	0.344	0.001
C20:1n-9	0.85	0.85	0.019	0.881
C20:2n-6	0.77	0.62	0.020	0.001
C20:3n-3	0.41	0.83	0.045	0.001
C20:4n-6	0.03	0.06	0.003	0.001
C22:1n-9	0.02	0.04	0.004	0.045
C20:5n-3	0.01	0.05	0.004	0.001
C24:0	0.10	0.06	0.006	0.001
C24:1n-9	0.02	0.03	0.002	0.001
C22:5n-3	0.06	0.18	0.014	0.001
C22:6n-3	0.03	0.03	0.002	0.082
SFA[2]	37.58	37.13	0.278	0.435
MUFA[3]	43.68	43.58	0.298	0.868
PUFA[4]	18.74	19.29	0.288	0.353
n-6 PUFA[5]	17.30	13.99	0.419	0.001
n-3 PUFA[6]	1.44	5.30	0.405	0.001
n-6:n-3	12.01	2.64	0.981	0.001
IV[7]	68.61	71.57	0.530	0.003

[1]n = 12.
[2]The percentage of SFA is the sum of C14:0, C16:0, C18:0, C20:0, and C24:0.
[3]The percentage of MUFA is the sum of C14:1, C16:1, C18:1, C20:1, C22:1, and C24:1.
[4]The percentage of PUFA is the sum of C18:2, C18:3, C20:2, C20:3, C20:4, C20:5, C22:5, and C22:6.
[5]The percentage of n-6 PUFA is the sum of C18:2, C20:2, and C20:4.
[6]The percentage of n-3 PUFA is the sum of C18:3, C20:3, C20:5, C22:5, and C22:6.
[7]IV = (C16:1n-7 × 0.950) + (C18:1n-9 × 0.860) + (C18:2n-6 × 1.732) + (C18:3n-3 × 2.616) + (C20:1n9 × 0.785) + (C22:1n-9 × 0.723).

Differences ($P < 0.05$) were observed between the SM and subcutaneous adipose tissue. The SM had a greater content of C16:1n-7 ($P < 0.001$), C20:1n-9 ($P < 0.01$), total MUFA ($P < 0.01$), and some n-6 PUFA and n-3 PUFA: arachidonic acid (C20:4n-6; $P < 0.001$), EPA ($P < 0.001$), and DPA ($P < 0.001$). However, the ALA ($P < 0.001$) and LA ($P < 0.01$) content was greater in subcutaneous adipose tissue, as well as the content of total PUFA ($P < 0.001$), n-6 PUFA ($P < 0.001$), and n-3 PUFA ($P < 0.001$). The interaction of tissue × diet was significant ($P < 0.001$) for ALA, DPA, total n-6 PUFA, and total n-3 PUFA.

Dietary treatment caused changes in fatty acid composition of the whole slice (Table 5). The whole slice of linseed dry-cured ham showed an increased content of total n-3 PUFA ($P < 0.001$), ALA ($P < 0.001$), EPA ($P < 0.05$), and DPA ($P < 0.05$). The percentages of ALA and EPA in the whole slice of linseed dry-cured ham tripled and the percentage of DPA doubled compared with the percentages in the whole slice of control dry-cured ham. The whole-slice composition of the control dry-cured ham had a greater content of n-6 PUFA ($P < 0.001$), particularly LA ($P < 0.001$), compared with the whole slice of linseed dry-cured ham. Because of these changes in the fatty acid composition of the whole slice, the n-6:n-3 PUFA ratio was considerably reduced (−72% vs. the control values) in the whole slice of linseed dry-cured ham ($P < 0.001$). In milligrams or grams per 100 g of product (Figure 1), it was observed that the hams in the control group had greater values for LA ($P < 0.01$), total n-6 PUFA ($P < 0.01$), and the n-6:n-3 PUFA ratio ($P < 0.001$) than hams in the linseed group, and had smaller values for ALA ($P < 0.001$), EPA ($P < 0.05$), DPA ($P < 0.05$), and total n-3 PUFA ($P < 0.001$).

The decosahexaenoic acid content (**DHA**, C22:6 n-3; $P > 0.05$) and the arachidonic acid ($P > 0.05$) content were different in SM, in subcutaneous adipose tissue, and in the whole slice of control vs. linseed dry-cured ham. No differences in the content of total SFA, MUFA, and PUFA ($P > 0.05$) were observed in SM, in subcutaneous adipose tissue, or in the whole slice because of the different diets.

Dry-Cured Ham Sensory Analysis

Consumer tests revealed a consumer preference ($P < 0.05$) for descriptors such as odor and taste of the

Table 4. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on fatty acid composition (g/100 g of fatty acids) of semimembranosus muscle and of subcutaneous adipose tissue of dry-cured ham

	Semimembranosus muscle		Subcutaneous adipose tissue			*P*-value[2]		
Fatty acid	Control diet	Linseed diet	Control diet	Linseed diet	SEM[1]	T	D	T × D
C14:0	1.27[a]	1.32[ab]	1.29[a]	1.41[b]	0.060	0.113	0.010	0.234
C14:1	0.03	0.04	0.02	0.04	0.019	0.881	0.183	0.537
C16:0	23.59[a]	24.11[b]	23.60[a]	24.13[b]	0.496	0.991	0.049	0.971
C16:1n-7	3.26[c]	3.28[c]	2.53[a]	2.80[b]	0.128	0.001	0.033	0.069
C18:0	11.13	11.68	11.21	11.23	0.527	0.511	0.279	0.333
C18:1n-9	47.55	46.28	46.35	45.88	0.874	0.086	0.064	0.372
C18:2n-6	11.19[b]	9.67[a]	12.89[c]	10.28[ab]	0.663	0.003	0.001	0.120
C20:0	0.09[ab]	0.03[a]	0.15[b]	0.03[a]	0.078	0.440	0.028	0.416
C18:3n-3	0.55[a]	2.02[c]	0.81[b]	3.07[d]	0.160	0.001	0.001	0.001
C20:1n-9	0.09[b]	0.08[b]	0.07[a]	0.08[ab]	0.022	0.008	0.250	0.187
C20:2n-6	0.49	0.45	0.64	0.41	0.332	0.734	0.445	0.563
C20:3n-3	0.07	0.07	0.06	0.04	0.058	0.613	0.653	0.717
C20:4n-6	0.04[b]	0.04[b]	0.03[a]	0.03[a]	0.007	0.001	0.814	0.725
C22:1n-9	0.04	0.03	0.01	0.02	0.026	0.191	0.896	0.284
C20:5n-3	0.12[b]	0.22[c]	0.03[a]	0.07[ab]	0.044	0.001	0.009	0.168
C24:0	0.02	0.01	0.01	0.01	0.014	0.386	0.202	0.445
C24:1n-9	0.19	0.19	0.14	0.20	0.042	0.456	0.212	0.205
C22:5n-3	0.14[b]	0.33[c]	0.06[a]	0.15[b]	0.026	0.001	0.001	0.001
C22:6n-3	0.14	0.15	0.10	0.12	0.045	0.129	0.436	0.750
SFA[3]	36.10	37.15	36.26	36.81	0.951	0.838	0.108	0.616
MUFA[4]	51.16[b]	49.90[ab]	49.12[a]	49.02[a]	0.880	0.005	0.143	0.207
PUFA[5]	12.74[a]	12.95[a]	14.62[b]	14.17[b]	0.642	0.001	0.692	0.311
n-6 PUFA[6]	11.72[b]	10.16[a]	13.56[c]	10.72[a]	0.555	0.001	0.001	0.034
n-3 PUFA[7]	1.02[a]	2.79[b]	1.06[a]	3.45[c]	0.170	0.001	0.001	0.002
n-6:n-3 PUFA	11.49[b]	3.64[a]	12.79[b]	3.11[a]	1.146	0.366	0.001	0.075

[a–d]Means in the same row without common superscripts differ ($P < 0.05$).
[1]n = 6.
[2]T = tissue; D = diet; T × D = tissue × diet. Differences were considered significant if $P \leq 0.05$.
[3]The percentage of SFA is the sum of C14:0, C16:0, C18:0, C20:0, and C24:0.
[4]The percentage of MUFA is the sum of C14:1, C16:1, C18:1, C20:1, C22:1, and C24:1.
[5]The percentage of PUFA is the sum of C18:2, C18:3, C20:2, C20:3, C20:4, C20:5, C22:5, and C22:6.
[6]The percentage of n-6 PUFA is the sum of C18:2, C20:2, and C20:4.
[7]The percentage of n-3 PUFA is the sum of C18:3, C20:3, C20:5, C22:5, and C22:6.

samples coming from animals fed the control diet. Other descriptors did not exhibit differences ($P > 0.05$), although overall acceptability was consistently oriented toward samples originating from animals fed the control diet ($P < 0.05$; Table 6).

DISCUSSION

Effects of Diet on Ham Physicochemical Characteristics

In agreement with the results of Wilfart et al. (2004), dietary treatment did not affect the weight of green ham. Additionally, no differences in fat and muscle color of dry-cured ham were observed after cutting (time of air exposure = 0), confirming the results observed by Santos et al. (2008) in dry-cured ham. These results are in general agreement with those reported for other meat products, such as dry-cured loin (Hoz et al., 2007) and dry fermented sausages (Hoz et al., 2004). The same applied to the chemical composition of dry-cured ham; our study showed no effects of dietary treatment on DM, CP, ether extract, ash, or salt concentration, or on the proteolysis index of the SM in the 2 different dry-cured ham batches. These data are consistent with the results of Santos et al. (2008) in dry-cured ham and Hoz et al. (2007) in dry-cured loin.

Effects of Diet on Green Ham Fatty Acid Composition

Diet is known to affect the fatty acid composition of pig adipose tissue, particularly back and perirenal adipose tissue (Fontanillas et al., 1997; Apple et al., 2008). In the current study, the fatty acid composition of the trimmed fat was dependent on the diet. As noted by Vorin et al. (2003), there is a relationship between the amount of n-3 PUFA consumed and the amount stored. The amount of n-3 PUFA deposited in the trimmed fat was greater in linseed dry-cured ham. Consequently, in the trimmed fat of pigs fed the control diet, the n-6:n-3 PUFA ratio value was 12 and the n-6:n-3 PUFA ratio of the trimmed fat of pigs fed the linseed diet was approximately 2.5. In several studies in light pigs (110.0 ± 10 kg of BW; Cunnane et al., 1990; Ahn et al., 1996), the DHA content in adipose tissue was not affected by

Table 5. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on fatty acid composition (g/100 g of fatty acids) of a whole slice of dry-cured ham

Fatty acid	Control diet	Linseed diet	SEM[1]	*P*-value
C14:0	1.31	1.36	0.018	0.134
C14:1	0.02	0.03	0.001	0.120
C16:0	23.63	24.20	0.171	0.091
C16:1n-7	2.74	2.79	0.042	0.531
C18:0	11.24	11.77	0.148	0.073
C18:1n-9	46.29	45.39	0.290	0.125
C18:2n-6	12.52	10.29	0.433	0.001
C20:0	0.15	0.02	0.032	0.036
C18:3n-3	0.80	2.87	0.347	0.001
C20:1n-9	0.07	0.07	0.003	0.839
C20:2n-6	0.75	0.64	0.023	0.012
C20:3n-3	0.10	0.04	0.016	0.049
C20:4n-6	0.03	0.04	0.003	0.629
C22:1n-9	0.01	0.02	0.003	0.181
C20:5n-3	0.03	0.09	0.014	0.021
C24:0	0.07	0.04	0.015	0.389
C24:1n-9	0.06	0.04	0.023	0.706
C22:5n-3	0.09	0.18	0.018	0.004
C22:6n-3	0.09	0.12	0.014	0.293
SFA[2]	36.40	37.39	0.305	0.104
MUFA[3]	49.19	48.34	0.284	0.142
PUFA[4]	14.41	14.27	0.252	0.793
n-6 PUFA[5]	13.30	10.97	0.451	0.001
n-3 PUFA[6]	1.11	3.30	0.368	0.001
n-6:n-3 PUFA	11.98	3.32	1.455	0.001

[1]n = 6.
[2]The percentage of SFA is the sum of C14:0, C16:0, C18:0, C20:0, and C24:0.
[3]The percentage of MUFA is the sum of C14:1, C16:1, C18:1, C20:1, C22:1, and C24:1.
[4]The percentage of PUFA is the sum of C18:2, C18:3, C20:2, C20:3, C20:4, C20:5, C22:5, and C22:6.
[5]The percentage of n-6 PUFA is the sum of C18:2, C20:2, and C20:4.
[6]The percentage of n-3 PUFA is the sum of C18:3, C20:3, C20:5, C22:5, and C22:6.

a greater amount of ALA in the diet, which agrees with our results. Cunnane et al. (1990; 5% of dietary linseed supplementation) and Ahn et al. (1996; 1.5 to 3.5% of ALA dietary supplementation) reported increased concentrations of ALA and EPA, but not of DHA, in subcutaneous adipose tissue. In contrast with our results, Romans et al. (1995) observed an increase in the quantities of DHA (approximately +200%) in the inner and outer layers of subcutaneous adipose tissue in light pigs fed diets with linseed at 15% (3.5% ALA) for different lengths of time (7, 14, 21, or 28 d). In addition, Enser et al. (2000) observed increased DHA in subcutaneous adipose tissue (+50%) and in muscle (+35%) in light pigs with long-term linseed feeding (0.45% ALA from 25 to 95 kg LW). The reason for these differences is unclear. In the present and other long-term feeding experiments (Cunnane et al. 1990; Ahn et al., 1996), dietary amounts of ALA probably resulted in greater ALA and EPA tissue concentrations, indicating a competitive exclusion of DHA from tissue lipids, particularly from phospholipids, which normally contain only small amounts of ALA (Enser et al., 2000). In addition, genotype affects the conversion rate of ALA to EPA and DPA and DHA, which is greater for Large White pigs relative to the crossbreds, as was recently reported by Kloareg et al. (2007).

The IV is an important qualitative characteristic of green hams undergoing processing and ripening using the conventional technology specified by regulations [Council Regulation (EEC) No. 2081/92] set out by the Parma Ham Consortium (1992). In fact, the Parma Ham Consortium stipulates that the trimmed fat of green hams must have an IV score equal to or less than 70 to avoid fat quality problems. In our experiment, only the control trimmed fat had an IV of less than 70 (IV = 68.6), whereas the linseed trimmed fat had an IV of approximately 71. This value would lead the Parma Ham Consortium to discard the linseed green hams for processing as Parma hams.

Effects of Diet on Dry-Cured Ham Fatty Acid Composition

During processing, lipids undergo intense lipolysis controlled by both lipase and phospholipase. Because of the long processing time (often at relatively high temperatures), a large amount of the volatile compounds, generated in dry-cured hams, is formed through fatty acid oxidation both through autooxidation processes and by incomplete bacterial β-oxidation (Gandemer, 2002). Moreover, the variation in fatty acid composition of subcutaneous adipose tissue found in green ham

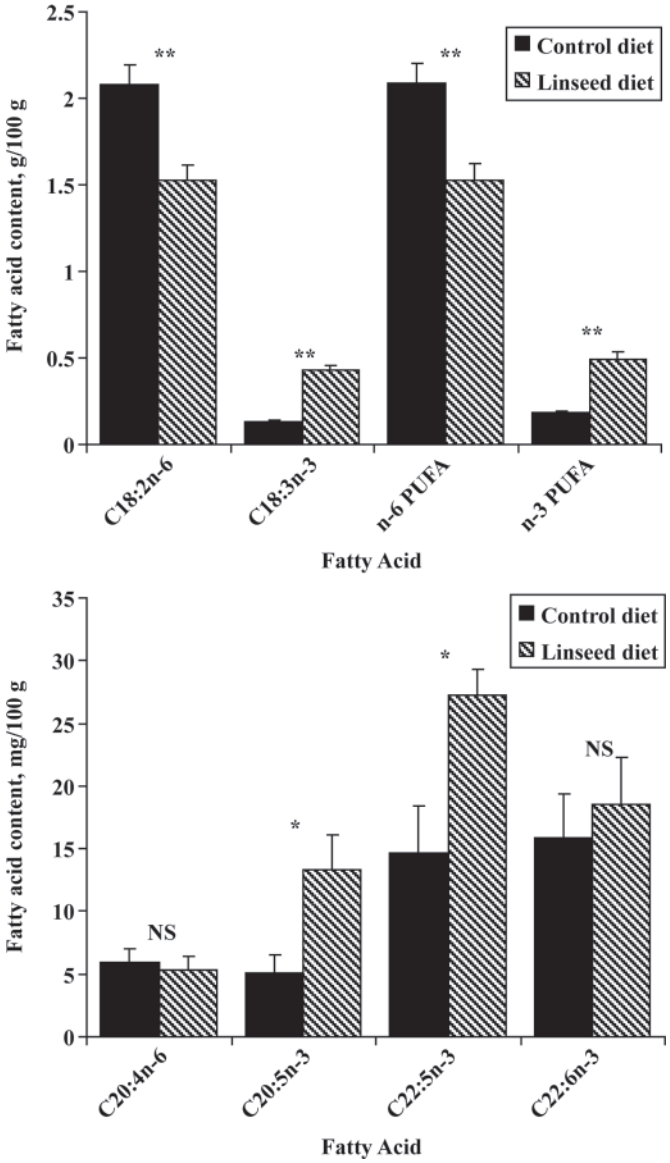

Figure 1. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on fatty acid content of a whole slice of dry-cured ham (g or mg/100 g). Values are means ± SE, 6 dry-cured hams per group (*$P < 0.05$; **$P < 0.01$; NS = not significant).

was observed in dry-cured ham. In fact, linseed dry-cured hams showed a numerically greater content of total n-3 PUFA, ALA, EPA, and DPA, and showed a reduced LA content and n-6:n-3 PUFA ratio compared with control dry-cured hams. This suggests that a dietary integration of antioxidants can preserve the long-chain fatty acids that remain in products with a long shelf life, such as dry-cured ham.

As previously reported in fresh pork (Leszczynski et al., 1992; Warnants et al., 1996; 1998; Kouba and Mourot, 1999; Corino et al., 2002), dietary fat has a smaller effect on the fatty acid composition of the intramuscular adipose tissue of dry-cured ham than on the subcutaneous adipose tissue. This may be due to the weak deposition of absorbed fat in muscle tissue (Vernon, 1992) or to the greater amount of membrane lipids in intramuscular adipose tissue and in myofibers containing elevated quantities of PUFA, which are less sensitive to dietary variations. However, in the present study, some variations were observed in the fatty acid composition of SM of dry-cured ham (particularly LA and ALA). As reported by Pastorelli et al. (2003) in heavy pigs (160 ± 10 kg of BW) and by Santos et al. (2008) in light pigs, the fatty acid composition of dry-cured ham depends on the dietary fatty acid composition. Both studies observed an increase in ALA content in dry-cured ham obtained from pigs fed diets enriched in n-3 PUFA. Santos et al. (2008) also observed an increase in the EPA and DPA in SM of light pigs fed diets supplemented with 3% linseed, which is in agreement with results of the present study. Pastorelli et al. (2003), in a study of heavy pigs fed diets supplemented with 3% rapeseed oil until 110 kg of BW and with 2.5% rapeseed oil until 160 kg of BW, observed an increase in only ALA content because rapeseed oil, which has less n-3 PUFA compared with products derived from linseed, was used. The increase in n-3 PUFA content is especially desirable for the ideal n-6:n-3 PUFA ratio (between 5:1 and 10:1) suggested for human health (International Society for the Study of Fatty Acids and Lipids, 2004).

It should be noted that the PUFA have different modes of incorporation into tissues. The present results for LA are in agreement with those reported by Nguyen et al. (2003) for light pigs fed diets supplemented with 1 to 3% linseed. In fact, LA is stored preferentially in adipose tissue rather than in muscle. It is possible that the intake of LA is greater than the amount required

Table 6. Effects of diet (control diet: 2.5% sunflower oil; linseed diet: 5% extruded linseed) on the sensory acceptability of dry-cured ham[1]

Descriptor	Control diet	Linseed diet	SEM	*P*-value
Overall acceptability	5.8	5.1	0.15	0.015
Appearance	6.5	6.3	0.15	0.635
Fat color	5.8	5.8	0.16	0.852
Muscle color	6.1	6.3	0.14	0.554
Odor	6.6	5.6	0.15	0.001
Salty	6.1	6.4	0.14	0.261
Taste	6.3	5.2	0.17	0.001
Consistency	6.1	5.8	0.15	0.231

[1]The judgments were expressed individually on a 9-point hedonic scale ranging from disliked extremely (score = 1) to liked extremely (score = 9).

for membrane synthesis, so a greater proportion of the intake is channeled into storage sites (adipose tissue). In contrast, EPA and DPA are preferentially stored in the organs or muscle rather than in the adipose tissue. However, in contrast with Nguyen et al. (2003), in our experiment, much more ALA was stored in adipose tissue than in muscle, whereas slightly more EPA, DPA, and DHA were deposited in the muscle than in subcutaneous adipose tissue. These differences may be due to the different amounts of n-3 PUFA in the diets, different sources of n-3 PUFA, and length of feeding. Rustan et al. (1988), Øverland et al. (1996), and Wood et al. (2008) have argued that the greater concentrations of EPA, DPA, and DHA in the lipid of muscle tissue compared with subcutaneous adipose tissue are probably due to a greater content of phospholipids in intramuscular fat than in subcutaneous fat and that PUFA are preferentially incorporated into phospholipids rather than triacylglycerol.

The whole slice fatty acid composition (which is of interest from a dietetic point of view) was shown to be more similar to that of subcutaneous adipose tissue than to that of SM, with values slightly less than in subcutaneous adipose tissue. There was also an increase in the content of ALA, EPA, and DPA, as observed by Hoz et al. (2004) in dry-fermented sausages from light pigs fed diets supplemented with 3% linseed. Moreover, an important modification was observed in the n-6:n-3 PUFA ratio, with values decreasing from approximately 12 (control dry-cured ham) to approximately 3 (linseed dry-cured ham) because of a substantial increase in the total content of n-3 PUFA and a decrease in n-6 PUFA. In fact, the dry-cured ham enrichment in n-3 PUFA was concomitant with a decrease in the n-6 PUFA content (mainly in the LA), in which a reduction of approximately 20% was shown and in agreement with observations of Hoz et al. (2004) in dry-fermented sausages.

Concerning the whole slice fatty acid content, 100 g of a whole slice of linseed dry-cured ham contained from 30 to 50% of the daily requirements of n-3 PUFA. Indeed, the recommended daily intake of n-3 PUFA is 1 g for women and 1.5 g for men, as reported by the Istituto Nazionale della Nutrizione/Società Italiana di Nutrizione (1996), the World Health Organization/Food and Agriculture Organization of the United Nations (2002), and the Food and Nutrition Board, Institute of Medicine, National Academy of Sciences (2005).

Effects of Diet on Dry-Cured Ham Sensory Analysis

The sensory quality of pork meat products is affected by the fatty acid composition of the adipose tissue and the muscle from which it comes. To investigate the degree of acceptance of the 2 different dry-cured hams, a preference test was carried out: overall acceptability, appearance, fat and muscle color, odor, saltiness, taste, and consistency descriptors were assessed. Muscle and fat color, as observed by CIELAB color analysis, was not different between the 2 dietary treatments. The same results were observed by Pastorelli et al. (2003) in heavy pigs and by Santos et al. (2008) in light pigs. Concerning the saltiness, Pastorelli et al. (2003) reported values not consistent with those measured in our experiment. However, the descriptor "salty" depends greatly on the technology used for processing raw meat. Contrary to the reports by Pastorelli et al. (2003) and Santos et al. (2008), the n-3 PUFA supplementation of the diet caused differences in odor and taste descriptors, which were judged as more intense by the consumer panel in linseed dry-cured ham than in control ham. Concerning the results of Pastorelli et al. (2003), this different result could be explained by the fact that rapeseed oil has a very small n-3 PUFA content compared with extruded linseed, and the supplementation of vitamin E was also different. Regarding the results of Santos et al. (2008), differences could be explained by the different slaughter weights of animals (100 vs. 160 kg) and the different lengths of the ripening period of hams (12 vs. 16 mo). Linseed dry-cured hams, with the greatest IV and greatest n-3 PUFA content, showed the least acceptance for odor, taste, and overall acceptability. These results are in agreement with previous findings in other pork products, such as dry-fermented sausages (Hoz et al., 2004) and dry-cured loin (Hoz et al., 2007). Moreover, some authors reported several differences in sensory evaluation negatively related to different unsaturation grades of the fatty acids (C18:3n-3 vs. C18:2n-6 and C18:1n-9; Campo et al., 2003) and to a low oxidative stability (Santos et al., 2008). The consumer judgment of overall acceptability was oriented toward the product obtained from animals fed the control diet. In particular, the taste and odor might have been impaired by the occurrence of off-flavors originating from the oxidative breakdown of unsaturated fatty acids, which is especially true for long-cured meat products such as dry-cured ham.

This study confirmed that it is possible to improve the nutritional quality of long-cured pork products through supplementation of the feed of live pigs. In particular, dietary enrichment with raw materials rich in linolenic acid increased not only linolenic acid content, but also concentrations of its derivatives EPA and DPA. Furthermore, dietary extruded linseed reduced the n-6:n-3 PUFA ratio and a value less than 5 was obtained (as recommended by nutritionists). Quality problems that might have been expected because of tissue enrichment in n-3 PUFA, which increases the risk of oxidative phenomena of long-cured pork products, appear to have been partially controlled by dietary supplementation with vitamin E and selenium. It may be possible to improve the nutritional quality of long-cured meat products (e.g., Parma ham) by increasing dietary extruded linseed and antioxidants related to the supply of n-3 PUFA in pig feed. In fact, the con-

trol of lipid oxidation and the balance between volatile compounds arising from lipid oxidation, carbohydrate fermentation, AA degradation, and additives such as spices are the critical points to control in producing high-quality dry-cured meat products. Further studies are required for a better understanding of the contribution of individual molecules, and the processes involved in their generation, to the typical aroma of dry-cured meat products (aged and dry-cured flavors).

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