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Use of re-esterified palm oils, differing in their acylglycerol structure, in fattening pig diets

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Re-esterified oils are new fat sources obtained from the chemical esterification of acid oils with glycerol (both economically interesting by-products from oil refining and biodiesel industries, respectively). The different fatty acid (FA) positional distribution and acylglycerol composition of re-esterified oils may enhance the apparent absorption of saturated fatty acids (SFA) and, therefore, their overall nutritive value, which might lead to an increased deposition of SFA. The aim of the present study was to investigate the potential use of re-esterified palm oils, in comparison with their corresponding acid and native oils in fattening pig diets, studying their effects on fatty acid apparent absorption, acylglycerol and free fatty acid (FFA) composition of feces, growth performance, carcass-fat depots and fatty acid composition of backfat. Seventy-two crossbred boars and gilts (average weight of 24.7 ± 2.55 kg) were blocked by initial BW (nine blocks of BW for each gender), housed in adjacent individual boxes, and fed one of the four dietary treatments, which were the result of a basal diet supplemented with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL), or re-esterified palm oil high in mono- and diacylglycerols (PEH). Regarding results from the digestibility balance, PA and PN showed similar apparent absorption coefficients (P > 0.05), despite the high, FFA content of the former. However, re-esterified palm oils (both PEL and PEH) showed a higher apparent absorption of total FA than did their corresponding native and acid oils (P < 0.001), mainly due to the increased apparent absorption of SFA (P < 0.001). This resulted in a greater feed efficiency and an increased deposition of SFA in backfat of pigs fed PEH, when compared with those fed PA (P < 0.05), although no differences were found for carcass-fat depots (P > 0.05). We conclude that re-esterified oils are interesting fat sources to be considered in fattening pigs.

Keywords: absorption, diacylglycerols, fatty acids, monoacylglycerols, pigs

Implications

The chemical esterification of acid oils with glycerol (both economically interesting by-products from oil refining and biodiesel industries, respectively), generates a new type of fats with a different acylglycerol structure, that can provide valuable characteristics in animal nutrition, apart from the environmental benefits associated with their use. The results of this study indicate that re-esterified palm oils are interesting fat sources to be considered in fattening pigs.

Introduction

Palm oil is one of the few vegetable oils rich in saturated fatty acids (SFA). Thus, its use in fattening pig diets is attractive because it may be associated with a positive influence on fat firmness (Wiseman and Agunbiade, 1998). However, its high SFA content impairs its nutritive value (Powles *et al*., 1993). Besides the importance of the fat degree of saturation, the fat acylglycerol structure also plays an important role in the nutritive value of fats. Given the specificity of pancreatic lipase for the hydrolysis of fatty acids (FA) located at the *sn*-1,3 positions (Mattson and Beck, 1956), SFA are better absorbed if they are located at the *sn*-2 position, because they are absorbed as 2-monoacylglycerols, instead of as free SFA. Free SFA are highly hydrophobic and have a great ability to form insoluble soaps with divalent cations in the gut (Small, 1991). This fact might become more relevant with the use of acid palm oils (by-products from oil-refining industry), which are rich in free SFA.

The chemical esterification of acid palm oil with glycerol (another economically interesting by-product from biodiesel

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industry) results in the formation of re-esterified palm oils. These alternative fat sources show an increased proportion of SFA located at the acylglycerol *sn*-2 position, and a higher amount of mono- (MAG) and diacylglycerol (DAG) molecules than their corresponding native oil (Vilarrasa *et al*., 2015). Therefore, it was hypothesized that the different acylglycerol structure of re-esterified palm oils may enhance SFA apparent absorption and, thus, their overall nutritive value, which might lead to an increased deposition of SFA. The aim of the present study was to investigate the potential use of re-esterified palm oils, differing in their acylglycerol structure, in comparison with their corresponding acid and native oils in fattening pig diets, studying their effects on FA apparent absorption, acylglycerol and free fatty acid (FFA) composition of feces, growth performance, carcass-fat depots and FA composition of backfat.

Material and methods

Experimental fats

Experimental fats were supplied by SILO S.p.a. (Florence, Italy). Re-esterified palm oils were produced using, as raw materials, acid palm oil (a by-product obtained from the refining process of crude palm oil, with a high FFA content) and glycerol (a by-product obtained from the methylation process applied in biodiesel production), which were processed under high vacuum conditions (1 to 3 mmHg), for 4 to 6 h, at 190°C to 250°C, and without chemical catalysts. According to the stoichiometric proportion of FFA and glycerol, fats with the same FA profile (Table 1), but with different FA positional distribution (Table 2), and triacylglycerol (TAG), DAG and MAG proportions (Table 1) were obtained.

Oil samples were analyzed in triplicate for moisture (Method 926.12 of the Association of Official Analytical Chemists (AOAC), 2005), impurities (ISO 663:2007), unsaponifiable matter (Method 933.08 of the AOAC, 2005), acylglycerol and FFA composition (ISO 18395:2005), MAG and DAG positional isomers (Sacchi *et al*., 1997), total FA composition (Guardiola *et al*., 1994), *sn*-2 FA composition (Commission Regulation (EEC) No. 2568/91 – Annex VII), and gross energy content (IKA-Kalorimeter system C2000 basic; Staufen, Germany), as described in more detail in our previous report (Vilarrasa *et al*., 2014).

Animals and diets

The study was performed at the experimental farm of *IRTA Mas de Bover* (Constantí, Tarragona, Spain). The experimental procedure received the prior approval from the Ethical Committee of the same institution. All animal housing and husbandry conformed to the European Union Guidelines (2010/63/EU).

A total of 72 crossbred ((Landrace × Duroc) × Pietrain) boars and gilts, with an initial BW of 24.7 ± 2.55 kg, were

Table 1 *Chemical analyses of the experimental fats*[1]

Item	PN oil	PA oil	PEL oil	PEH oil
Moisture (%)	0.10	0.17	0.36	1.13
Impurities (%)	<0.50	<0.50	0.50	<0.50
Unsaponifiable matter (%)	0.58	1.14	1.92	1.78
Fatty acid composition (%)				
C16:0	43.4	45.0	45.1	46.4
C18:0	4.34	4.50	4.54	4.57
C18:1 n-9	39.3	37.7	37.9	37.2
C18:2 n-6	10.1	9.66	9.19	8.59
Minor fatty acids	2.89	3.23	3.30	3.26
SFA	49.1	50.8	51.1	52.5
MUFA	40.5	39.1	39.4	38.7
PUFA	10.4	10.1	9.49	8.79
Acylglycerol and FFA composition (%)				
TAG	87.5	16.3	41.9	11.8
DAG	7.72	22.3	44.3	45.8
1(3),2-DAG %[2]	26.7	31.8	27.1	31.3
MAG	0.70	8.26	12.6	42.4
2-MAG %[3]	40.0	8.85	7.25	7.74
FFA	4.04	53.2	1.22	0.00
Glycerol-to-fatty acid ratio[4] (mol/mol)	0.34	0.23	0.47	0.67
Gross energy (MJ/kg)	39.2	38.4	38.2	36.2

SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; TAG = triacylglycerols; DAG = diacylglycerols; MAG = monoacylglycerols; FFA = free fatty acids.
[1]Native palm oil (PN oil), acid palm oil (PA oil), re-esterified palm oil low in mono- and diacylglycerols (PEL oil), and re-esterified palm oil high in mono- and diacylglycerols (PEH oil).
[2]The proportion of 1(3),2-DAG *v.*1,3-DAG.
[3]The proportion of 2-MAG *v.*1(3)-MAG.
[4]Estimated calculation based on the values of the acylglycerol and FFA composition.

obtained from the swine herd of the same experimental station, blocked by initial BW (nine blocks of BW for each gender), and randomly assigned to one of the four dietary treatments. Animals were housed in adjacent individual boxes, had free access to feed and water, and were raised under controlled conditions of light and temperature.

The experiment was planned to cover the BW range from ~25 to 100 kg of BW. For this purpose, the feeding program consisted of three diets (in pelleted form): a starter diet (from ~25 to 50 kg of BW; from 0 to 35 days of the experiment), a grower diet (from ~50 to 75 kg of BW; from 35 to 70 days of the experiment) and a finisher diet (from ~75 to 100 kg of BW; from 70 to 100 days of the experiment). Basal diets were formulated to meet or exceed (National Research Council, 2012) requirements and to minimize basal fat levels. One-half of the starter diet was supplemented with 1% of Celatom (Jesús Riesgo; Madrid, Spain) to increase the amount of HCl-insoluble ash as an inert digestibility marker and, thus, perform the digestibility balance. The four dietary treatments were the results of including 4% (as-fed basis) of one of the following experimental fats in the basal diets: native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in MAG and DAG (PEL) or re-esterified palm oil high in MAG and DAG (PEH). The composition of experimental diets is presented in Table 3.

Analytical determinations of feeds were performed according to the methods of AOAC (2005): dry matter (Method 934.01), ash (Method 942.05), CP (Method 968.06), crude fat (Method 2003.05) and crude fiber (Method 962.09). Gross energy was determined as described previously for experimental fats and HCl-insoluble ash of pre-starter feeds was determined following the method of McCarthy *et al.* (1974). Lipids from feeds were extracted with chloroform/methanol (2 : 1, by vol) according to the Folch *et al.* (1974) procedure. Nonadecanoic acid (C19:0, Sigma-Aldrich Chemical Co.; St. Louis, MO, USA) was added as an internal standard before processing. The FA content was analyzed following the method of Morrison and Smith (1964). The macronutrient and the FA composition of the experimental diets are presented in Table 4.

Table 2 *Fatty acid positional distribution[1] within acylglycerol molecules of the experimental fats[2] (%)*

Item	PN oil	PA oil	PEL oil	PEH oil
C16:0	8.53	6.81	17.5	11.1
C18:0	5.33	8.74	19.5	15.1
C18:1 n-9	52.2	11.3	24.8	19.1
C18:2 n-6	59.3	11.2	24.8	19.0
SFA	8.33	6.92	17.6	11.6
MUFA	51.4	11.4	24.8	19.1
PUFA	58.4	11.2	24.8	19.0

SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.
[1]Expressed as the proportion of each fatty acid located at the *sn*-2 position of the acylglycerol molecules, calculated as follows: *sn*-2% = (*sn*-2/Total) × *a* × 100, where *sn*-2 is the FA composition at the *sn*-2 position, Total is the total FA composition in the original fat, and *a* is the ratio between the moles of FA located at the *sn*-2 position and the moles of total FA. *a* was 0.30, 0.09, 0.21 and 0.15 for PN, PA, PEL and PEH oils, respectively.
[2]Native palm oil (PN oil), acid palm oil (PA oil), re-esterified palm oil low in mono- and diacylglycerols (PEL oil) and re-esterified palm oil high in mono- and diacylglycerols (PEH oil).

Table 3 *Ingredient composition of the experimental diets (as-fed basis)[1]*

Ingredients (%)	Starter diet (from 0 to 35 days)		Grower diet (from 35 to 70 days)	Finisher diet (from 70 to 100 days)
	With marker	Without marker		
Wheat	38.44	29.68	22.00	17.61
Barley	30.00	39.97	52.22	59.51
Soybean meal 44%	15.40	14.85	10.43	7.62
Experimental fats[1]	4.00	4.00	4.00	4.00
Biscuit meal	4.00	4.00	4.00	4.00
Sunflower meal	3.69	4.00	4.00	4.00
Dicalcium phosphate	1.12	1.10	0.93	0.82
Celatom[2]	1.00	–	–	–
Calcium carbonate	0.71	0.77	0.71	0.70
Sodium bicarbonate	0.55	0.54	0.67	0.76
Vitamin and mineral premix[3]	0.40	0.40	0.40	0.40
L-Lysine	0.44	0.43	0.41	0.39
L-Threonine	0.16	0.15	0.16	0.14
DL-Methionine	0.11	0.10	0.09	0.06
Ethoxyquin 66%	0.02	0.02	0.02	0.02

[1]Native palm oil (PN oil), acid palm oil (PA oil), re-esterified palm oil low in mono- and diacylglycerols (PEL oil) or re-esterified palm oil high in mono- and diacylglycerols (PEH oil).
[2]Jesús Riesgo; Madrid, Spain.
[3]Provides per kg of feed: vitamin A (from retinol), 5500 IU; vitamin D_3 (from cholecalciferol), 1100 IU; vitamin E (from alfa-tocopherol), 7 mg; thiamine, 0.5 mg; riboflavin, 1.4 mg; pyridoxine, 1.0 mg; cobalamine, 0.008 mg; calcium pantothenate, 5.6 mg; nicotinic acid, 8 mg; menadione, 0.5 mg; Ca, 0.72 g; Mg, 57 mg; Fe (from $FeSO_4 \cdot 7H_2O$), 80 mg; Cu (from $CuSO_4 \cdot 5H_2O$), 10 mg; Co (from $2CoCO_3 \cdot 3Co(OH)_2 \cdot H_2O$), 0.4 mg; Zn (from ZnO), 100 mg; Mn (from MnO), 40 mg; I (from $Ca(I_2O_3)_2$), 0.5 mg; Se (from Na_2SeO_3), 0.25 mg; ethoxyquin, 2.67 mg.

Table 4 *Analyzed[1] macronutrient content and fatty acid composition of the experimental diets (as-fed basis)[2]*

Item	Starter diet (from 0 to 35 days)								Grower diet (from 35 to 70 days)				Finisher diet (from 70 to 100 days)			
	With marker				Without marker											
	PN	PA	PEL	PEH	PN	PA	PEL	PEH	PN	PA	PEL	PEH	PN	PA	PEL	PEH
Macronutrient content (%)																
Dry matter	88.8	88.4	88.7	88.5	88.4	88.3	88.3	88.2	88.9	88.2	89.0	88.7	89.8	89.7	89.6	89.7
CP	15.8	16.3	16.0	16.1	16.3	16.5	16.3	16.3	15.3	15.3	15.0	15.4	14.7	14.9	14.3	14.4
Crude fat	6.13	6.08	5.89	6.04	5.88	5.90	5.95	6.04	5.95	6.20	6.01	5.70	6.03	6.35	6.04	6.18
Crude fiber	3.88	3.84	3.97	3.79	4.12	3.79	3.78	3.76	4.36	4.38	4.51	4.07	4.49	4.03	4.13	4.20
Ash	5.63	5.59	5.52	5.60	4.77	4.70	4.82	4.63	4.39	4.32	4.48	4.39	4.32	4.40	4.24	4.42
Gross energy (MJ/kg)	16.9	16.9	16.9	16.8	17.1	17.1	17.1	17.0	17.1	17.1	17.1	17.0	17.3	17.3	17.3	17.2
Fatty acid composition (%)																
C16:0	32.9	33.5	34.3	34.2	33.5	34.7	33.7	33.9	33.5	33.4	34.0	33.7	32.7	32.6	33.0	31.4
C18:0	3.37	3.45	3.41	3.48	3.56	3.50	3.38	3.38	3.23	3.35	3.37	3.32	3.24	3.32	3.26	3.15
C18:1 n-9	36.0	34.1	33.5	33.6	35.3	34.4	35.2	34.0	34.5	34.4	34.3	33.4	36.4	36.3	36.1	35.9
C18:2 n-6	23.3	24.5	24.3	24.2	23.3	23.0	23.2	24.2	24.3	24.4	23.9	25.0	23.3	23.3	23.2	25.0
C18:3 n-3	1.62	1.82	1.73	1.70	1.56	1.71	1.66	1.73	1.70	1.82	1.73	1.81	1.65	1.78	1.70	1.87
Minor fatty acids	2.79	2.68	2.77	2.78	2.79	2.67	2.82	2.80	2.72	2.64	2.65	2.74	2.73	2.67	2.73	2.69
SFA	37.5	38.2	39.1	39.0	38.3	39.4	38.4	38.6	38.1	38.0	38.7	38.3	37.2	37.1	37.6	35.8
MUFA	37.4	35.5	34.9	35.0	36.7	35.8	36.7	35.4	35.9	35.8	35.6	34.8	37.8	37.7	37.5	37.3
PUFA	25.0	26.3	26.1	26.0	24.9	24.7	24.9	26.0	26.0	26.2	25.7	26.9	25.0	25.2	24.9	26.9

SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.
[1]All samples were analyzed, at least, in duplicate.
[2]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL), or re-esterified palm oil high in mono- and diacylglycerols (PEH).

Controls and sampling

The amount of feed and BW of the animals were recorded at the beginning and at the end of each phase (0, 35, 70 and 100 days of the experiment), which enabled the calculation of average daily feed intake, average daily gain and gain-to-feed ratio (G : F) for each period and for the overall study.

From day 6 to 9, a digestibility balance was undertaken. Feces were collected once a day by rectal stimulation and were immediately frozen at −20°C. On the last day of the balance, feces from each animal were pooled, homogenized, and a representative sample was freeze-dried, ground and kept at 5°C until further analyses. Fecal samples were analyzed by the same methods as those described for feeds (HCl-insoluble ash, gross energy and FA content) to determine the digestible energy of the diets and the apparent absorption of FA. The apparent absorption coefficient of a particular nutrient (*X*) was calculated as follows:

$$\% \text{ apparent absorption of } X = \{1 - [(F_X/F_M)/(D_X/D_M)]\} \times 100$$

where F_X is the concentration of a particular nutrient in feces, F_M is the concentration of the inert marker in feces, D_X is the concentration of a particular nutrient in the diet and D_M is the concentration of the inert marker in the diet. The digestible energy was calculated from the product of energy apparent absorption and its corresponding feed gross energy.

The acylglycerol and FFA composition of feces was analyzed according to ISO 18395:2005, in which TAG, DAG, MAG and FFA are separated according to their molecular size by size-exclusion HPLC. Before analysis, fat was extracted from feces with diethyl ether after acidification with HCl 1N. The acylglycerol molecules were quantified by internal normalization. Finally, these values were expressed as grams per 100 g of fat intake, based on values of fat apparent absorption.

At the end of the experimental period, three boars and three gilts of each treatment (those with BW close to the treatment average weight) were transported to the *IRTA* experimental abattoir (Monells, Girona, Spain). Animals were fasted (deprived of feed but not water) for ∼16 h. The following morning, they were reweighed to obtain the fasted live weight, stunned using 85% CO_2 for 120 s, and killed by exsanguination. The carcasses were then scalded, mechanically dehaired before evisceration, split along the mid line and weighed. Fat and muscle thickness were measured using the Fat-O-Meat'er probe (Carometec A/S; Soeborg, Denmark) between the third- and the fourth-last ribs at 6 cm to the midline. Values obtained were used to calculate the lean meat percentage according to the Spanish official equation (Font-i-Furnols and Gispert, 2009). The liver and perirenal fat were weighed, and were expressed in absolute values and as a percentage of the live animal weight. Backfat samples (from the dorsal midline overlaying the *m. longissimus dorsi*) were taken, vacuum packed, and stored at −20°C until chemical analyses. The FA composition of backfat samples was analyzed following the same method as that described for feed and feces.

Statistical analysis

Normality of the data and homogeneity of the variance were verified. The experiment was designed as a randomized complete block design with nine blocks of initial BW within sexes and four dietary treatments. All data were subjected to ANOVA using the GLM procedure of SAS (version 9.2, SAS Institue Inc.; Cary, NC, USA). For carcass-fat depots and FA composition of backfat, the model only included dietary treatment as fixed effect and sex as a blocking factor. No interaction between sex and treatments was found to be significant for any of the variables studied. Differences between treatment means were tested using Tukey's correction for multiple comparisons. Individual pig data served as the experimental unit, so there were 18 replicates per dietary treatment. For carcass-fat depots and FA composition of backfat, there were only six observations per dietary treatment. For perirenal fat and liver weights, the BW before slaughter was included as a covariate in the model. Results in tables are reported as least square means and differences were considered significant at $P < 0.05$.

Results and discussion

Characterization of experimental fats

The chemical analyses of the experimental fats are presented in Table 1. Experimental fats showed a similar FA composition, indicating that the esterification of acid palm oil with glycerol did not substantially modify the FA composition of fat. More than 80% of the total FA was composed of palmitic (45.0 ± 1.23%) and oleic (38.0 ± 0.65%) acids.

Regarding the fat acylglycerol and FFA composition, PN oil, considered the positive control, was mainly composed of TAG. Acid palm oil, as a negative control, showed a high FFA content. Re-esterified oils contained 45.1 ± 1.06% of DAG, almost no FFA, and variable amounts of TAG and MAG. Re-esterified palm oil high in MAG and DAG showed a higher amount of MAG (42.4% *v.* 12.6%) and a lower amount of TAG (11.8% *v.* 42.4%) than did PEL oil. In both re-esterified oils, 1(3)-MAG and 1,3-DAG were the major isomers.

The FA positional distribution within acylglycerol molecules (Table 2) found in PN oil was in agreement with that reported by Mattson and Volpenhein (1963), SFA being preferentially esterified at the *sn*-1,3 positions and unsaturated FA at the *sn*-2 position, as FA distribution in vegetable oils is genetically determined. However, the chemical esterification process does not allow a selective esterification of FA in the different glycerol positions. Therefore, compared with PN oil, re-esterified palm oils showed a certain redistribution of FA within acylglycerol molecules. Although this process did not result in a complete random distribution of FA (i.e. 33% of each FA should appear in the *sn*-2 position), re-esterified palm oils showed a higher percentage of SFA and, therefore, a lower percentage of MUFA and PUFA located at the *sn*-2 position, when compared with

their corresponding PN oil. The reason for the lower proportion of *sn*-2 FA in PEH oil than in PEL was the higher 1(3)-MAG content of the former. In this sense, PA oil, due to its high FFA content, showed the lowest proportions of *sn*-2 FA.

Finally, indices of lipid quality (Table 1) showed low levels of moisture, impurities, and unsaponifiable matter for PN and PA oils, but yielded values above 2.5% for re-esterified palm oils.

Digestibility balance

The effects of dietary fat sources on the digestible energy of the diets and the apparent absorption of individual FA are presented in Table 5. We only performed one digestibility balance at the beginning of the study. In a previous study, using the same experimental fats with weaning piglets (Vilarrasa *et al*., 2015) only small differences were observed, and it was considered that this and the measurement in the current study would be representative of the digestibility of the fats under study. As expected, unsaturated FA were better absorbed than SFA, and stearic acid was less well absorbed than was palmitic acid. According to Doreau and Chilliard (1997), as unsaturation increases, digestibility is increased, and as FA chain-length increases, digestibility is reduced. However, it must be taken into account that the apparent fecal absorption of individual FA is affected by microflora biohydrogenation of unsaturated FA in the hindgut, resulting in an underestimation of the apparent absorption of stearic acid and an overestimation of the apparent absorption of unsaturated FA of the C18-family (Duran-Montgé *et al*., 2007). In any case, the total FA apparent absorption is not affected (Jørgensen *et al*., 1992).

Contrary to what was expected, no differences in FA apparent absorption were found between PN and PA treatments ($P > 0.05$). The same results were found by DeRouchey *et al.* (2004), who examined the effects of different levels of FFA in choice white grease in weaning piglets. In contrast, Powles *et al.* (1993) reported that the FFA content of fats appeared to be one of the major determinants of the digestible energy values of fats when given to growing/finishing pigs. One possible explanation for the lack of differences between PN and PA oils could be related to the particular FA positional distribution of these fat sources. Both PN and PA oils showed a very similar proportion of SFA located at the *sn*-2 position of acylglycerol molecules (8.33% and 6.92% for PN and PA oils, respectively). This means that the amount of FFA as SFA after hydrolysis of PN oil by pancreatic lipase gives approximately the same amount of SFA as FFA as the PA oil. The difference of FFA was mainly relevant in the case of UFA (the proportion of UFA located at the *sn*-2 position of acylglycerol molecules was 52.8% and 11.3% for PN and PA oils, respectively). However, the effect of FFA on UFA absorption is lower in comparison with that of SFA (Powles *et al*., 1993). Thus, acid palm oils may be an economically interesting alternative to native palm oils, provided they have a moderate amount of FFA and a low percentage of moisture, impurities and unsaponifiable matter, as we have also observed in weaning piglets (Vilarrasa *et al*., 2015).

Table 5 *Digestible energy of the diets (MJ/kg) and apparent total-tract digestibility coefficients (%) of fattening pigs according to different fat sources in diet*

	Dietary treatments[1]					
Item	PN	PA	PEL	PEH	r.m.s.e.[2]	*P*-values
DE (MJ/kg)	14.4	14.4	14.5	14.5	0.3	ns
Total fatty acids	84.3^{b}	85.1^{b}	88.7^{a}	88.9^{a}	2.8	***
SFA	69.1^{b}	70.9^{b}	81.2^{a}	80.8^{a}	6.0	***
MUFA	92.9^{b}	93.4^{ab}	94.2^{a}	94.4^{a}	1.3	**
PUFA	94.3	94.6	94.4	94.1	1.5	ns
C16:0	78.0^{b}	78.7^{b}	87.7^{a}	89.2^{a}	4.6	***
C18:0	−2.69	4.77	18.2	9.32	26.63	ns
C18:1 n-9	96.0^{b}	96.3^{ab}	96.8^{a}	97.0^{a}	0.9	**
C18:2 n-6	94.9	95.0	94.8	94.6	1.5	ns
C18:3 n-3	94.2	94.8	94.1	93.9	1.4	ns

DE = digestible energy; SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; ns = not significant.
[a,b]Values within a row with different superscripts differ significantly at $P < 0.05$.
[1]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL) or re-esterified palm oil high in mono- and diacylglycerols (PEH).
[2]$n = 18$.
$^{**}P < 0.01$; $^{***}P < 0.001$.

Concerning the use of re-esterified palm oils, animals fed both PEL and PEH diets showed a higher total FA apparent absorption, compared with those fed their corresponding native (PN) and acid (PA) oil-enriched diets ($P < 0.001$). These differences were mainly given by the enhanced SFA apparent absorption observed in both PEL and PEH treatments ($P < 0.001$), although small differences were also found for the apparent absorption of monounsaturated FA ($P = 0.003$). Nevertheless, these differences were not enough to be reflected in the feed digestible energy ($P > 0.05$), probably because fat only accounted for 14% of feed gross energy.

The improved SFA apparent absorption observed in re-esterified palm oils may be related to their higher content of SFA located at the *sn*-2 position (Table 2), and also to their higher amount of MAG and DAG molecules (Table 1). The importance of the FA positional distribution within acylglycerol molecules on fat absorption has already been demonstrated by several authors (Carnielli *et al*., 1995; Renaud *et al*., 1995; Lien *et al*., 1997). For example, although lard and palm oil have similar levels of SFA, Renaud *et al.* (1995) reported that rats fed with lard showed a higher total FA apparent absorption than did those fed with palm oil, because in lard the majority of palmitic acid is esterified at the *sn*-2 position, whereas in palm oil this SFA is mainly located at the *sn*-1,3 positions (Mattson and Lutton, 1958) and, as a consequence, palmitic acid is easily absorbed as 2-MAG in lard or poorly absorbed as FFA in palm oil. In any case, it is important to note that our levels of *sn*-2 SFA were not as high as were those used in the studies cited above (from 33% to 84% of palmitic acid located at the *sn*-2 position). On the other hand, the importance of the presence of MAG and DAG molecules in dietary fats has been less studied. However, due to their amphiphilic

properties, they may act as emulsifiers and might thus contribute towards improved digestion of fat. In this sense, Jones *et al.* (1992) observed that the addition of distilled MAG to tallow increased the digestibility of total FA and long-chain SFA in weaning pigs.

Acylglycerol and FFA composition of feces

The acylglycerol and FFA composition of feces (in grams per 100 g of fat intake) is shown in Table 6. Feces contained low levels of TAG, DAG and MAG. This means that TAG and DAG were almost completely hydrolyzed along the gastrointestinal tract, and MAG were absorbed due to their hydrophilic character (2-MAG and some 1(3)-MAG) or completely hydrolyzed to glycerol and FFA (mainly 1(3)-MAG). FFA constituted the major lipid fraction in feces for all treatments, suggesting that the formation of mixed micelles may be the rate-limiting step of fat absorption in fattening pigs, as has also been observed in weaning piglets (Jones *et al.*, 1992; Vilarrasa *et al.*, 2015), but less severely.

Regarding dietary treatments, differences were only observed for FFA content. Animals fed PEL and PEH excreted a lower amount of FFA than did those fed PN and PA ($P > 0.05$). The amount of FFA excreted per 100 g of fat intake paralleled the results of SFA apparent absorption, suggesting that the emulsification of SFA is the most critical step in fat utilization, due to their strong tendency to form insoluble soaps with calcium and magnesium, at the alkaline pH of the small intestine (Renaud *et al.*, 1995; Lien *et al.*, 1997).

Table 6 *Acylglycerol and FFA composition of feces (g/100 g of fat intake) according to different fat sources in diet*

	Dietary treatments[1]					
Item	PN	PA	PEL	PEH	r.m.s.e.[2]	*P*-values
TAG	0.73	0.76	0.81	0.81	0.29	ns
DAG	4.45	4.17	4.38	4.41	1.18	ns
MAG	2.58	2.76	2.18	2.14	0.75	*
FFA	14.8[a]	14.8[a]	12.1[b]	12.0[b]	3.1	**

TAG = triacylglycerols; DAG = diacylglycerols; MAG = monoacylglycerols; FFA = free fatty acids; ns = not significant.
[a,b]Values within a row with different superscripts differ significantly at $P < 0.05$.
[1]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL) or re-esterified palm oil high in mono- and diacylglycerols (PEH).
[2]$n = 18$.
*$P < 0.05$; **$P < 0.01$.

Growth performance and carcass-fat depots

The effects of dietary fat sources on growth performance are presented in Table 7. Differences were only found for the G : F ratio in the starter period and in the overall experiment. Animals fed PEH showed a higher G : F than did those fed PA ($P = 0.001$), and PN and PEL showed intermediate values. Thus, for the overall period, to gain 1 kg of BW, animals from the PA treatment needed to consume 10.7% more feed than did those from PEH ($P < 0.001$), probably related to the

Table 7 *Growth performance of fattening pigs according to different fat sources in diet*

	Dietary treatments[1]					
Item	PN	PA	PEL	PEH	r.m.s.e.[2]	*P*-values
From 0 to 35 days						
ADFI (g)	1449	1504	1438	1438	149	ns
ADG (g)	707	691	690	727	105	ns
G : F (g/g)	0.48[ab]	0.46[b]	0.49[ab]	0.51[a]	0.04	*
BW at 35 days (kg)	49.5	48.9	49.0	50.1	3.7	ns
From 35 to 70 days						
ADFI (g)	2024	2023	1963	1987	204	ns
ADG (g)	820	795	809	823	93	ns
G : F (g/g)	0.40	0.39	0.41	0.42	0.03	ns
BW at 70 days (kg)	78.1	76.8	77.3	79.0	5.9	ns
From 70 to 100 days						
ADFI (g)	2702	2591	2592	2577	381	ns
ADG (g)	1031	935	936	996	146	ns
G : F (g/g)	0.39	0.37	0.36	0.39	0.06	ns
BW at 100 days (kg)	109	105	105	109	9	ns
From 0 to 100 days						
ADFI (g)	2022	2045	1966	1972	168	ns
ADG (g)	844	793	806	842	85	ns
G : F (g/g)	0.41[ab]	0.39[b]	0.41[ab]	0.43[a]	0.03	**

ADFI = average daily feed intake; ADG = average daily gain; G : F = gain-to-feed ratio; ns = not significant.
[a,b]Values within a row with different superscripts differ significantly at $P < 0.05$.
[1]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL) or re-esterified palm oil high in mono- and diacylglycerols (PEH).
[2]$n = 18$.
*$P < 0.05$; **$P < 0.01$.

Table 8 *Carcass yield and carcass-fat depots of fattening pigs according to different fat sources in diet*

	Dietary treatments[1]					
Item	PN	PA	PEL	PEH	r.m.s.e.[2]	*P*-values
Carcass yield (%)	80.1	80.2	81.1	80.0	1.4	ns
Backfat thickness[3] (mm)	15.8	14.8	15.8	16.0	2.4	ns
Muscle depth[3] (mm)	60.0	58.0	58.3	58.3	5.1	ns
Lean percentage[4] (%)	61.4	62.0	61.1	61.0	2.3	ns
Perirenal fat						
(kg)	0.86	0.80	0.97	1.00	0.28	ns
(g/kg BW)	7.87	7.23	9.12	9.08	2.60	ns
Liver						
(kg)	2.06	2.17	2.01	2.28	0.34	ns
(g/kg BW)	18.8	19.4	18.3	21.0	3.1	ns

ns = not significant.
[1]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL) or re-esterified palm oil high in mono- and diacylglycerols (PEH).
[2]$n = 6$.
[3]Measurement made between the third and fourth last ribs at 6 cm to the carcass midline with Fat-O-Meat'er (Carometec A/S; Soeborg, Denmark).
[4]Calculated from backfat thickness and muscle depth between third and fourth last ribs using the Spanish official equation (lean percentage (%) = 66.91–0.895 × backfat thickness + 0.144 × muscle depth; Font-i-Furnols and Gispert, 2009).

lower coefficient of total FA apparent absorption reported in animals fed PA.

In comparing our results with those of the literature, Scheeder *et al.* (2003) reported that the FA positional distribution of dietary fats did not affect growth performance in growing–finishing pigs. In relation to the fat acylglycerol composition, Murata *et al.* (1997), Taguchi *et al.* (2001) and Murase *et al.* (2005) also did not find differences in the performance of rats fed DAG or TAG oil-enriched diets. In contrast, Meng *et al.* (2004) reported that the weight gain was lower in rats fed a DAG oil-rich diet, while Kamphuis *et al*. (2003) observed a lesser feeling of hunger and appetite in women fed a DAG oil-enriched diet.

The effects of dietary fat sources on carcass yield and carcass-fat depots are presented in Table 8. Carcass yield, backfat thickness, muscle depth, carcass lean percentage and liver and perirenal fat proportions were not affected by dietary fat sources ($P > 0.05$). Regarding the fat FA positional distribution effect, Ponnampalam *et al.* (2011) did not find differences in body fatness (evaluated by backfat thickness) in weaning piglets fed palm oil-enriched diets with a different FA positional distribution (native palm oil, chemically modified palm oil or enzymatically modified palm oil). However, animals fed lard-enriched diets showed a greater external adipose tissue thickness than did those fed native palm oil, which has a lower *sn*-2 SFA content. In relation to the fat acylglycerol composition, Murase *et al.* (2002) and Meng *et al.* (2004) have reported a reduced accumulation of fat in visceral adipose and subcutaneous tissue, and a reduced fat content in the liver of animals fed 1,3-DAG-enriched diets, as compared with those fed TAG of the same FA composition. The presence of dietary 1,3-DAG has been shown to increase β-oxidation of FA in the liver (Murata *et al*., 1997) and in the small intestine (Murase *et al*., 2002). We might not have encountered significant differences in fat depots of animals fed re-esterified palm oils because the effect of MAG and DAG might have counteracted the effect of *sn*-2 SFA.

FA composition of backfat

The effect of dietary fat sources on FA composition of backfat is presented in Table 9. The FA composition of backfat was a clear reflection of the FA composition of diets, as has been extensively reviewed by Madsen *et al.* (1992). However, although finisher diets showed a very similar FA composition, PEH animals tended to deposit more SFA than did PN and PA ones, which in turn resulted in a lower unsaturated-to-saturated FA ratio ($P < 0.05$). This difference may be related to the higher SFA apparent absorption observed for PEH treatment, although a higher SFA content should have also been expected in animals from PEL treatment. In agreement with our results, Smink *et al.* (2008) demonstrated that the feeding of randomized palm oil instead of native palm oil significantly raised the palmitic acid content of breast meat and abdominal fat in broiler chickens. Ponnampalam *et al.* (2011) also observed that weaning piglets fed diets with chemically or enzymatically modified palm oil deposited more SFA in the subcutaneous adipose tissue than did those fed a diet with native palm oil. In contrast, Innis *et al.* (1996) and Scheeder *et al.* (2003) did not find differences in the FA composition of adipose tissue of pigs fed inter-esterified and native fats.

Taken together, results of the present study indicate that feeding fattening pigs with diets supplemented with re-esterified palm oils resulted in a higher total FA apparent absorption, mainly due to the increased absorption of SFA, than did their corresponding native and acid oils. This resulted in a greater feed efficiency and an increased SFA deposition in backfat of pigs fed PEH when compared with those fed PA. Thus, we conclude that re-esterified oils

Table 9 *Fatty acid composition (%) of backfat according to different fat sources in diet*

	Dietary treatments[1]					
Item	PN	PA	PEL	PEH	r.m.s.e.[2]	*P*-values
C12:0	0.06	0.06	0.05	0.06	0.01	ns
C14:0	1.13	1.11	1.04	1.11	0.12	ns
C15:0	0.05	0.06	0.05	0.06	0.02	ns
C16:0	23.6^{ab}	23.2^{b}	23.7^{ab}	24.7^{a}	0.8	*
C16:1	1.53	1.50	1.49	1.58	0.19	ns
C17:0	0.37	0.41	0.34	0.39	0.10	ns
C17:1	0.23	0.27	0.22	0.26	0.07	ns
C18:0	11.3	11.7	12.4	12.8	1.0	ns
C18:1t + C18:1 n-11	0.14	0.45	0.31	0.19	0.29	ns
C18:1 n-9	42.8	43.1	43.3	42.2	1.4	ns
C18:1 n-7	1.71	1.77	1.73	1.76	0.15	ns
C18:2 n-6	14.8	13.9	13.1	12.5	1.5	ns
C18:3 n-3	0.77	0.79	0.71	0.69	0.10	ns
C20:0	0.15	0.15	0.18	0.20	0.04	ns
C20:1 n-9	0.63	0.70	0.75	0.76	0.11	ns
C20:2 n-6	0.50	0.53	0.51	0.47	0.05	ns
C20:4 n-6	0.22	0.28	0.21	0.23	0.07	ns
SFA	36.7^{b}	36.8^{b}	37.7^{ab}	39.4^{a}	1.6	*
MUFA	47.0	47.8	47.8	46.8	1.6	ns
PUFA	16.3	15.5	14.5	13.9	1.7	ns
UFA : SFA	1.73^{a}	1.73^{a}	1.65^{ab}	1.54^{b}	0.11	*

SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; UFA : SFA = unsaturated-to-saturated fatty acid ratio; ns = not significant.
[a,b]Values within a row with different superscripts differ significantly at $P < 0.05$.
[1]Diets with 4% (as-fed basis) of native palm oil (PN), acid palm oil (PA), re-esterified palm oil low in mono- and diacylglycerols (PEL) or re-esterified palm oil high in mono- and diacylglycerols (PEH).
[2]$n = 6$.
*$P < 0.05$.

are interesting fat sources to be considered in fattening. However, further studies are needed to determine whether the use of re-esterified palm oils can affect the FA positional distribution of meat and fat depots, because a higher content of SFA at the *sn*-2 position of TAG in meat could lead to increased SFA availability for the consumer.

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