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Feeding co-extruded flaxseed to pigs: Effects of duration and feeding level on growth performance and backfat fatty acid composition of grower–finisher pigs

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ABSTRACT

To examine the effect of co-extrusion on subsequent n-3 fatty acids in pig tissues, 8 pigs (barrows and gilts) were assigned to either a control treatment or one of nine treatments arranged in a 3 × 3 factorial design with 3 levels of co-extruded flaxseed (5%, 10% and 15%) and 3 durations of feeding (4, 8 and 12 weeks). Feed conversion improved slightly ($P = 0.01$) with increasing dietary flaxseed but feeding flax for more than 8 weeks reduced average daily gain ($P = 0.02$). In general, the duration and level of co-extruded flaxseed feeding affected ($P < 0.05$) most fatty acids except for 22:6n-3 ($P > 0.05$). Increasing the duration of flax feeding led to significant quadratic effects in backfat 18:3n-3 ($P < 0.001$) and total n-3 fatty acids ($P = 0.002$) when feeding 5% co-extruded flaxseed. Those increases were linear ($P < 0.001$) when feeding 10% and 15% co-extruded flaxseed. Consequently feeding higher levels of flax for shorter periods vs. lower levels for longer periods appears to be more efficient at increasing n-3 fatty acids in pig backfat, but increases appeared to be less consistent. Moreover the addition of a 50:50 mix of extruded flax/peas to pig diets provided a highly available source of 18:3n-3 yielding n-3 fatty acid enrichments in backfat comparable to studies where extracted flaxseed oil was fed. Feeding flax co-extruded with field peas can be used to optimize consistent enrichments of n-3 fatty acids in back fat and relatively small amounts of this fat could be used to manufacture pork products to meet Canadian standards for n-3 fatty acid enrichment.

1. Introduction

Historically humans consumed diets with an n-6 to n-3 fatty acid ratio of 1:1. In Western diets, this ratio has increased over the last 100 years to an average of 15:1 (Simopoulos, 2008). Consuming n-3 fatty acids may provide health benefits by reducing the risks for several diseases including heart disease (Ruxton, Reed, Simpson, & Millington, 2004), cancer (Larsson, Kumlin, Ingelman-Sundberg, & Wolk, 2004) and inflammatory diseases (Calder & Grimble, 2002). Thus the potential use of livestock products as vehicles to deliver n-3 fatty acids has been the subject of intensive research (Moghadasian, 2008). In this context, dietary strategies used to customize fatty acid composition of pig fat have been proven to be very effective because dietary fatty acids can be incorporated into pig fat with little modification (Bee, Jacot, Guex, & Biolley, 2008; D'Arrigo et al., 2002; Wood et al., 2008). However, differences in the relationships between intake and deposition exist, and these relate to whether the specific fatty acid can be synthesized *in vivo*. A strong relationship is seen for essential fatty acids [linoleic (18:2n-6) and α-linolenic (18:3n-3) acids] and their n-3 and n-6 elongation and desaturation products. On the other hand, saturated and monounsaturated fatty acids are synthesised *in vivo* and their deposition is less influenced by diet (Enser, Richardson, Wood, Gill, & Sheard, 2000).

Flax is the richest oilseed source of 18:3n-3 and feeding flax to pigs has been used to increase levels of n-3 fatty acids in pork, as reviewed by Nguyen, Nuijens, Everts, Salden, and Beynen (2003). Recently Thacker, Racz, and Soita (2004) reported feeding flax co-extruded with field pea could avoid problems with grinding and storage and Htoo, Meng, Patience, Dugan, and Zijlstra (2008) investigated extrusion techniques for a flax/pea mixture to optimize 18:3n-3 availability. Htoo et al. (2008) fed pigs a 50:50 mixture of flax and peas that was either ground through a knife mill with a 2 mm sieve or co-extruded using single or double screw techniques. Single screw extrusion with minimal water input increased 18:3n-3 availability in the flax/pea mix by 16% relative to ground flax/peas and by 10% relative to other extrusion techniques. In contrast, Thacker et al. (2004) found feeding 12% flax as a 50:50 mix of flax/peas reduced rates of body weight gain in finisher pigs whereas most other studies have found pigs can be fed up to 15% flax without affecting performance (e.g. Romans, Johnson, Wulf,

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Libal, & Costello, 1995a; Romans, Wulf, Johnson, Libal, & Costello, 1995b). However, regional variations in oil content and fatty acid distribution in flaxseed (Hall Iii, Tulbek, & Xu, 2006) makes comparison of results from different locations difficult. Moreover incorporation rates of 18:3n-3 when feeding flax have been quite variable and this can partly be attributed to processing challenges due to its high oil and 18:3n-3 content, but little attention has been paid to optimizing 18:3n-3 availability.

The objectives of the present research were, therefore, to feed single screw extruded flax/peas (50:50) to grow/finish pigs for different durations and levels and examine the effects on animal performance and n-3 fatty acid incorporation into backfat. The dietary inclusion and the duration of flax feeding were similar to those fed by Thacker et al. (2004) to determine if and when single screw extruded flax would negatively influence animal performance. In addition, a 3×3 factorial design was employed to determine if 18:3n-3 and n-3 fatty acids levels in backfat would plateau (Fontanillas, Barroeta, Baucells, & Guardiola, 1998; Matthews, Homer, Thies, & Calder, 2000) or remain linear (Cherian & Sim, 1995; Riley, Enser, Nute, & Wood, 2000; Romans et al., 1995a, 1995b) when feeding to a slaughter end point typical for Canada (i.e. 115 kg).

2. Materials and methods

2.1. Animal management

Animals involved in this study were cared for according to the guidelines set out by the Canadian Council of Animal Care (CCAC, 1993) under a protocol approved by the University of Saskatchewan Committee on Animal Care and Supply. The study included 80 pigs from the Prairie Swine Centre (Saskatoon, SK, Canada) with an initial body weight of 31 ± 3 kg (mean ± SD). All the animals were produced on-site and fed the same background diet prior to the experiment. Animals were stratified by weight, parentage and gender and included 4 gilts and 4 barrows per treatment. Pigs were housed individually in 0.91×1.83 m (1.67 m^2) pens with fully slatted floors and each pen was equipped with a single space feeder and nipple drinker.

Table 1
Ingredient composition and nutrient contents of experimental diets; weeks 1–4.

	Flax (%)			
Ingredients (% as fed)	0	5	10	15
Wheat	53.4	39.0	24.5	10.0
Barley	10.0	21.6	33.1	44.7
Soybean meal	17.0	15.3	13.5	11.8
Field pea	15.0	10.0	5.0	0.0
Co-extruded flaxseed and field pea[a]	0.0	10.0	20.0	30.0
Tallow	1.03	0.68	0.34	0.00
Di-calcium phosphate	0.85	0.83	0.82	0.80
Limestone	0.80	0.78	0.77	0.75
Vitamin premix[b]	0.50	0.50	0.50	0.50
Mineral premix[c]	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
l-Lysine-HCl	0.23	0.25	0.28	0.30
l-Threonine	0.10	0.10	0.10	0.11
DL-methionine	0.060	0.067	0.073	0.080
Calculated nutrient content (as fed)				
DE[d] (Mcal kg^{-1})	3.38	3.39	3.40	3.42
Digestible lysine: DE (g Mcal DE^{-1})[e]	2.80	2.80	2.80	2.80
Crude protein (%)	20.6	20.0	19.4	18.0
Calcium (%)	0.82	0.82	0.82	0.87
Phosphorus, total (%)	0.61	0.56	0.57	0.61

[a] Co-extruded 50% flaxseed and 50% field pea (Linpro; O&T Farms, Regina, Saskatchewan, Canada).

[b] Provided per kilogram of diet: vitamin A, 8250 IU; vitamin D_3, 825 IU; vitamin E, 40 IU; niacin, 35 mg; d-pantothenic acid, 15 mg; riboflavin, 5 mg; menadione, 4 mg; folic acid, 2 mg; thiamine, 1 mg; d-biotin, 0.2 mg; and vitamin B_{12}, 25 µg.

[c] Provided per kilogram of diet: Zn, 100 mg as zinc sulfate; Fe, 80 mg as ferrous sulfate; Cu, 50 mg as copper sulfate; Mn, 25 mg as manganous sulfate; I, 0.50 mg as calcium iodate; and Se, 0.10 mg as sodium selenite.

[d] Digestible energy.

[e] Other amino acids were formulated as a ratio to lysine (set at 100%): threonine, 62; methionine, 30; and tryptophan, 20 (NRC, 1998).

Table 2
Ingredient composition and nutrient contents of experimental diets; weeks 5–8.

	Flax (%)			
Ingredients (% as fed)	0	5	10	15
Wheat	44.2	31.1	18.1	5.0
Barley	20.2	30.9	41.6	52.3
Soybean meal	16.2	13.9	11.5	9.2
Field pea	15.0	10.0	5.0	0.0
Co-extruded flaxseed and field pea[a]	0.0	10.0	20.0	30.0
Tallow	1.00	0.67	0.33	0.00
Di-calcium phosphate	0.95	0.93	0.92	0.90
Limestone	0.80	0.78	0.77	0.75
Vitamin premix[b]	0.50	0.50	0.50	0.50
Mineral premix[c]	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
l-Lysine-HCl	0.12	0.16	0.19	0.23
l-Threonine	0.055	0.055	0.065	0.075
DL-methionine	0.015	0.025	0.035	0.045
Calculated nutrient content (as fed)				
DE[d] (Mcal kg^{-1})	3.34	3.35	3.37	3.38
Digestible lysine: DE (g Mcal DE^{-1})[e]	2.55	2.54	2.53	2.52
Crude protein (%)	19.2	19.2	16.8	16.2
Calcium (%)	0.86	0.88	0.79	0.72
Phosphorus, total (%)	0.64	0.66	0.60	0.58

[a] Co-extruded 50% flaxseed and 50% field pea (Linpro; O&T Farms, Regina, Saskatchewan, Canada).

[b] Provided per kilogram of diet: vitamin A, 8250 IU; vitamin D_3, 825 IU; vitamin E, 40 IU; niacin, 35 mg; d-pantothenic acid, 15 mg; riboflavin, 5 mg; menadione, 4 mg; folic acid, 2 mg; thiamine, 1 mg; d-biotin, 0.2 mg; and vitamin B_{12}, 25 µg.

[c] Provided per kilogram of diet: Zn, 100 mg as zinc sulfate; Fe, 80 mg as ferrous sulfate; Cu, 50 mg as copper sulfate; Mn, 25 mg as manganous sulfate; I, 0.50 mg as calcium iodate; and Se, 0.10 mg as sodium selenite.

[d] Digestible energy.

[e] Other amino acids were formulated as a ratio to lysine (set at 100%): threonine, 62; methionine, 30; and tryptophan, 20 (NRC, 1998).

Table 3
Ingredient composition and nutrient contents of experimental diets; weeks 9–12.

	Flax (%)			
Ingredients (% as fed)	0	5	10	15
Wheat	39.9	26.6	13.3	0.00
Barley	32.5	42.7	53.0	63.2
Soybean meal	8.70	7.13	5.57	4.00
Field pea	15.0	10.0	5.0	0.0
Co-extruded flaxseed and field pea[a]	0.0	10.0	20.0	30.0
Tallow	1.00	0.67	0.33	0.00
Di-calcium phosphate	0.50	0.48	0.47	0.45
Limestone	0.80	0.78	0.77	0.75
Vitamin premix[b]	0.50	0.50	0.50	0.50
Mineral premix[c]	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
l-Lysine-HCl	0.075	0.088	0.10	0.12
l-Threonine	0.005	0.005	0.005	0.005
DL-methionine	0.000	0.000	0.000	0.000
Calculated nutrient content (as fed)				
DE[d] (Mcal kg^{-1})	3.29	3.31	3.33	3.35
Digestible lysine: DE (g Mcal DE^{-1})[e]	1.98	1.97	1.95	1.94
Crude protein (%)	16.9	16.4	15.3	15.0
Calcium (%)	0.71	0.89	0.65	0.64
Phosphorus, total (%)	0.51	0.53	0.48	0.49

[a] Co-extruded 50% flaxseed and 50% field pea (Linpro; O&T Farms, Regina, Saskatchewan, Canada).

[b] Provided per kilogram of diet: vitamin A, 8250 IU; vitamin D_3, 825 IU; vitamin E, 40 IU; niacin, 35 mg; d-pantothenic acid, 15 mg; riboflavin, 5 mg; menadione, 4 mg; folic acid, 2 mg; thiamine, 1 mg; d-biotin, 0.2 mg; and vitamin B_{12}, 25 µg.

[c] Provided per kilogram of diet: Zn, 100 mg as zinc sulfate; Fe, 80 mg as ferrous sulfate; Cu, 50 mg as copper sulfate; Mn, 25 mg as manganous sulfate; I, 0.50 mg as calcium iodate; and Se, 0.10 mg as sodium selenite.

[d] Digestible energy.

[e] Other amino acids were formulated as a ratio to lysine (set at 100%): threonine, 62; methionine, 30; and tryptophan, 20 (NRC, 1998).

Table 4
Crude fat and fatty acid composition of diets.

	Weeks 1–4; flax (%)				Weeks 5–8; flax (%)				Weeks 9–12; flax (%)			
Item	0	5	10	15	0	5	10	15	0	5	10	15
Crude fat (g kg^{-1})[a]	26.8	41.8	64.0	71.0	26.9	34.4	64.7	72.5	21.8	38.0	60.2	79.3
Fatty acid (%)												
16:0	20.6	15.8	11.2	8.8	19.0	12.4	11.0	8.5	19.8	13.9	10.8	8.4
9*c*-16:1	1.3	0.9	0.5	0.2	1.2	0.2	0.6	0.2	0.9	0.6	0.3	0.1
18:0	8.6	6.5	4.5	3.1	6.8	3.0	4.8	3.4	5.2	4.6	3.8	3.0
9*c*-18:1	26.1	22.8	19.5	17.2	25.3	18.8	22.0	20.0	23.8	22.5	21.4	20.9
18:2n-6	37.6	30.4	24.7	23.2	40.6	36.3	25.8	24.9	43.8	33.4	27.7	24.9
18:3n-3	5.2	22.7	39.0	47.0	6.5	28.2	34.9	42.0	5.9	24.2	34.9	41.7
11*c*-20:1	0.6	0.9	0.5	0.5	0.7	1.0	1.1	1.0	0.7	0.9	1.1	0.9

[a] Measured.

The study included a control treatment (fed for 12 weeks) and nine treatments arranged in a 3 × 3 factorial design with 3 levels of extruded flaxseed (5%, 10% and 15%) and 3 durations of feeding (4, 8 and 12 weeks). Pigs on the 4 and 8 week treatments received control diets for 8 or 4 weeks, respectively prior to receiving their treatment diets.

Pigs had *ad libitum* access to feed and water. Pig weights and feed consumption were measured every two weeks and used to calculate average daily gain (ADG), average daily feed intake (ADFI) and feed conversion. Diets were formulated and adjusted every 4 weeks (0–4, 4–8 and 8–12 weeks; Tables 1–3) to meet the nutrient requirement of the pigs over the experiment (NRC, 1998). Diets were based on wheat, barley and soybean meal. Field peas were added to diets at a constant level within each feeding period taking into consideration the field pea included in co-extruded product, and tallow was used to balance energy levels among diets within phase. Diets within each feeding period had equal apparent ileal digestible lysine per Mcal of digestible energy (DE). Experimental diets included LinPro®, a commercial product (O&T Farms, Regina, Saskatchewan, Canada) containing a 50:50 mixture of co-extruded flaxseed and field peas using single screw extrusion at 400 psi without water and a barrel temperature of 135 °C (Htoo et al., 2008). Table 4 shows the crude fat content and fatty acid composition of the different diets.

2.2. Adipose tissue collection

One day prior to slaughter in a commercial abattoir, subcutaneous fat from each pig was collected using an 8 mm biopsy punch (Acuderm Inc., USA) at the 10th rib, approximately 5 cm from the backbone. The biopsy site was scrubbed and cleaned with isopropyl alcohol and iodine solution before injecting local anaesthesia [2 cc of 2% Lidocaine, (Gatlin, See, Hansen, Sutton, & Odle, 2002)]. An incision (~3–4 cm) was made at the biopsy site before applying the biopsy punch in order to minimize the quantity of intact skin with the fat samples. The subcutaneous fat samples were stored in a freezer at −80 °C until analyzed.

2.3. Chemical analyses

Feed fatty acid methyl esters (FAME) were prepared according to Sukhija and Palmquist (1988) and backfat (inner layer) FAME were prepared using the procedure described by Dugan et al. (2007). Feed and backfat FAME were analyzed according to Dugan et al. (2007).

2.4. Statistical analysis

The data were analyzed using the MIXED procedure within SAS (SAS, 2001). Individual pigs were the experimental unit. The statistical design used in the present experiment was an augmented factorial [i.e. 3 levels of flaxseed × 3 durations of flaxseed feeding × 2 genders and a single control group (i.e. 4 gilts and 4 barrows) fed the control diet for 12 weeks]. The statistical model included flaxseed level, length of feeding, gender and the interactions. Initial statistical analysis examined the main effects of flaxseed inclusion, length of feeding and the interactions. As such main effects could not be computed when the control was included in the model, therefore, initially control data were not used and the model included gender, diet, weeks of feeding and all their interactions. To further explore the effects of feeding flax at different levels for different durations on backfat fatty acid composition, linear and quadratic contrasts were conducted and this included data from control animals.

3. Results and discussion

3.1. Growth performance

There were no two or three-way interactions for ADG, ADFI or feed conversion ($P > 0.05$). As expected, barrows had higher ADG (7.8%), ADFI (14.3%) and lower feed conversion (7.9%) than gilts ($P < 0.01$; Table 5). With increasing dietary flaxseed, ADFI decreased from 2.62 to 2.45 kg/d ($P = 0.01$, Table 5) but ADG was unaffected ($P = 0.40$) and thus feed conversion improved slightly (from 0.38 to 0.39; $P = 0.01$). The lack of, or slight positive effects of adding flax to the diets on animal performance is in agreement with the majority of studies (Corino, Musella, & Mourot, 2008; Fontanillas et al., 1998; Kouba, Enser, Whittington, Nute, & Wood, 2003; Matthews et al., 2000; Riley et al., 2000; Romans et al., 1995a, 1995b; Wiseman, Redshaw, Jagger, Nute, & Wood, 2000). This is in contrast with Thacker et al. (2004) who found that pigs could tolerate up to 15% dietary flax in the grower period but 12% flax in the finishing diet reduced ADG. In the present experiment, increasing levels of co-extruded flaxseed, tended to reduce ADFI ($P = 0.06$; Table 5), but did not affect ADG. Prolonged feeding of co-extruded flaxseed, independent of its level of dietary inclusion, reduced ADG ($P = 0.01$). This observation together with the results of Thacker et al. (2004) would then suggest that long-term feeding of co-extruded flaxseed may marginally reduce ADG, but short-term feeding does not, even at high inclusion levels.

3.2. Backfat fatty acid composition

Diet by gender, feeding duration (weeks) by gender or diet by feeding duration by gender interactions were not found for backfat fatty acid composition ($P > 0.05$). Gender influenced backfat fatty acid composition with gilts having less ($P = 0.04$) saturated fatty acids (SFA) and more ($P = 0.03$) n-6 fatty acids (Table 6). Similar results were reported by Nuernberg et al. (2005) and likely relate to

Table 5
Performance of grower–finisher pigs fed different levels of extruded flaxseed for different durations.

Variable	Control	Diet (% flax)			Weeks			Gender		SEM[c]	P value		
		5	10	15	4	8	12	Gilt	Barrow		Diet	Weeks	Gender
Initial weight (kg)	31.1	30.8	30.9	31.4	30.9	31.2	31.0	30.7	31.3	1.48	0.31	0.74	0.08
Final weight (kg)	109.7	114.6	112.9	115.2	115.6a	115.7^{a}	111.4^{b}	110.0	118.5	2.09	0.36	0.02	<0.0001
ADG (kg d^{-1})	0.94	1.00	0.98	1.00	1.01^{a}	1.01^{a}	0.96^{b}	0.95	1.03	0.01	0.42	0.02	<0.0001
ADFI (kg d^{-1})	2.46	2.60^{a}	2.50^{ab}	2.47^{b}	2.58	2.55	2.45	2.33	2.72	0.03	0.01	0.06	<0.0001
Feed efficiency	0.38	0.39^{b}	0.39^{b}	0.41^{a}	0.39	0.40	0.39	0.41	0.38	0.01	0.01	0.80	<0.0001

No two or three-way interactions were observed ($P > 0.05$).
[ab]Means in rows with different letters are significantly different ($P < 0.05$).
[c] Standard error of the mean.

Table 6
Gender effects on the percent fatty acid composition of backfat.

Fatty acid (%)	Gender		SEM[a]	P value
	Gilt	Barrow		
18:2n-6	12.0	11.4	0.28	0.03
18:3n-3	9.3	9.3	0.38	0.99
20:2n-6	0.53	0.50	0.009	0.02
20:3n-3	1.27	1.25	0.023	0.58
20:4n-6	0.16	0.15	0.004	0.04
20:5n-3	0.13	0.12	0.007	0.06
22:5n-3	0.30	0.31	0.009	0.34
22:6n-3	0.07	0.07	0.002	0.98
n-3	11.1	11.0	0.39	0.95
n-6	12.7	12.1	0.290	0.03
n-6/n-3	1.39	1.36	0.025	0.15
SFA	31.6	32.8	0.39	0.04
MUFA	43.9	43.3	0.62	0.22
PUFA	23.8	23.2	0.67	0.26

SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids.
[a] Standard error of the mean.

barrows being fatter with larger adipocytes. This would result in a greater neutral lipid to phospholipid ratio in barrow backfat with neutral lipids being relatively rich in SFA and phospholipids relatively rich in n-6 fatty acids (Riley et al., 2000). All differences between genders were, however, less than 1% of total fatty acids and would be of limited practical significance.

Significant main effects ($P < 0.05$) for diet (% flax) and feeding duration (week) were found, in addition to diet by week interactions for most individual fatty acids except for 22:6n-3 ($P > 0.05$; Table 7). Several authors have reported a lack of effect of flax diets on tissue levels of 22:6n-3 (Ahn, Lutz, & Sim, 1996; Cherian & Sim, 1995; Fontanillas et al., 1998; Haak, De Smet, Fremaut, Van Walleghem, & Raes, 2008; Raes, De Smet, & Demeyer, 2004; Riley et al., 2000), and only a few studies have reported increased levels of 22:6n-3 after feeding a relatively low level (~2–2.5%) of dietary flax (Corino et al., 2008; Enser et al., 2000). Long chain fatty acid metabolism is controlled by a complex enzymatic system, consisting of desaturases and elongases (Raes et al., 2004). These enzymes act both on the n-6 and n-3 fatty acids but have a preference for the n-3 (Brenner, 1989). The lack of effect on 22:6n-3 fatty acid may be explained by competition for $\Delta 6$ desaturase activity between 18:3n-3 and the precursor for 22:6n-3 (i.e. 24:5n-3), when the dietary concentration of 18:3n-3 is high (Cameron et al., 2000).

Diet, feeding duration and diet by feeding duration effects ($P < 0.05$) were found for total polyunsaturated fatty acids (PUFA), monounsaturated fatty acids (MUFA), SFA, n-3 and n-6 fatty acids and the n-6/n-3 ratio (Table 7). In general, feeding flax containing diets for increasing durations prior to slaughter led to linear ($P < 0.05$) increases in 18:2n-6, 20:3n-3, 20:5n-3, 22:5n-3, total PUFA and n-6 fatty acids and reductions in 20:4n-6 and total MUFA (Table 8). The linear increase ($P < 0.001$) for 20:2n-6 and the linear decrease ($P < 0.001$) in SFA were only observed when 10% and 15% flax was included in the diet. The n-6/n-3 ratio decreased quadratically ($P < 0.001$), while the quadratic relationship for total n-3 content was stronger when feeding 5% ($P = 0.002$) than when feeding 10% ($P = 0.192$) or 15% ($P = 0.041$) co-extruded flaxseed containing diets. The quadratic relationship for n-3 fatty acids when feeding 5% flax in the diets was also observed for 18:3n-3 ($P < 0.001$), while increases in other 18:3n-3 elongation and desaturation products (20:3n-3, 20:5n-3 and 22:5n-3) were found to be linear.

Table 7
Diet, feeding duration and diet by feeding duration effects on percentages of fatty acids in backfat.

Flax (%)	Control	5			10			15			SEM[h]	P value		
Week		4	8	12	4	8	12	4	8	12		Diet	Weeks	Diet × week
18:2n-6	9.6	10.1e	10.9^{de}	10.6^{e}	10.1^{e}	11.9^{cd}	13.2^{ab}	11.7^{cd}	12.8^{bc}	14.0^{a}	0.3	<0.01	<0.01	0.01
18:3n-3	1.22	3.33^{f}	4.95^{e}	5.87^{e}	5.52^{e}	9.78^{c}	13.30^{b}	8.37^{d}	13.90^{b}	18.60^{a}	0.4	0.02	<0.01	<0.01
20:2n-6	0.46	0.47^{cd}	0.50^{bcd}	0.49^{cd}	0.46^{d}	0.54^{ab}	0.54^{ab}	0.51^{bcd}	0.51^{bc}	0.58^{a}	0.01	0.01	<0.01	0.1
20:3n-3	0.23	0.54^{f}	0.80^{e}	1.00^{d}	0.76^{e}	1.40^{c}	1.80^{b}	1.00^{d}	1.70^{b}	2.40^{a}	0.03	<0.01	<0.01	<0.01
20:4n-6	0.22	0.19^{ab}	0.18^{ab}	0.14^{d}	0.18^{bc}	0.15^{cd}	0.12^{d}	0.21^{a}	0.15^{d}	0.09^{e}	0.01	<0.01	<0.01	<0.01
20:5n-3	0.03	0.06^{e}	0.08^{d}	0.10^{cd}	0.07^{de}	0.12^{c}	0.19^{ab}	0.09^{d}	0.18^{b}	0.22^{a}	0.01	<0.01	<0.01	<0.01
22:5n-3	0.14	0.19^{g}	0.27^{ef}	0.29^{de}	0.22^{fg}	0.34^{cd}	0.42^{ab}	0.23^{fg}	0.36^{bc}	0.41^{ab}	0.01	<0.01	<0.01	0.07
22:6n-3	0.06	0.055	0.072	0.070	0.061	0.07	0.073	0.068	0.066	0.070	0.003	0.83	0.09	0.62
n-3	1.69	4.17^{f}	6.18^{e}	7.30^{e}	6.63^{e}	11.70^{c}	15.80^{b}	9.79^{d}	16.10^{b}	21.60^{a}	0.42	<0.01	<0.01	0.04
n-6	10.3	10.7^{e}	11.5^{de}	11.3^{e}	10.7^{e}	12.6^{cd}	13.9^{ab}	12.4^{cd}	13.4^{bc}	14.7^{a}	0.44	<0.01	<0.01	0.02
n-6/n-3	6.09	2.59^{a}	1.87^{b}	1.55^{c}	1.64^{c}	1.07^{e}	0.88^{f}	1.29^{d}	0.84^{f}	0.68^{g}	0.025	<0.01	<0.01	<0.01
SFA	36.2	35.6^{ab}	36.8^{ab}	34.9^{ab}	34.3^{b}	31.6^{cd}	29.2^{e}	31.8^{cd}	29.2^{de}	26.9^{e}	0.48	<0.01	<0.01	<0.01
MUFA	51.1	48.5^{ab}	44.8^{cd}	45.8^{bc}	47.6^{ab}	43.4^{d}	40.3^{e}	45.2^{cd}	40.5^{e}	36.3^{f}	0.67	<0.01	<0.01	0.02
PUFA	12.1	15.1^{e}	17.8^{d}	18.7^{d}	17.5^{d}	24.4^{c}	29.8^{b}	22.3^{c}	29.6^{b}	36.4^{a}	0.72	<0.01	<0.01	<0.01

SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids.
[a–g]Means in rows with different letters are significantly different ($P < 0.05$).
[h] Standard error of the mean.

Table 8
Linear and quadratic contrasts for fatty acids in backfat for length of feeding 5%, 10% or 15% flax to grower–finisher pigs prior to slaughter (0, 4, 8 and 12 weeks).

	P value					
Flax (%)	5		10		15	
Contrast[a]	L	Q	L	Q	L	Q
18:2n-6	0.011	0.257	<0.001	0.180	<0.001	0.295
18:3n-3	<0.001	<0.001	<0.001	0.238	<0.001	0.038
20:2n-6	0.186	0.520	<0.001	0.888	<0.001	0.777
20:3n-3	<0.001	0.076	<0.001	0.008	<0.001	0.267
20:4n-6	<0.001	0.540	<0.001	0.563	<0.001	0.127
20:5n-3	<0.001	0.145	<0.001	0.099	<0.001	0.277
22:5n-3	<0.001	0.200	<0.001	0.858	<0.001	0.332
22:6n-3	0.070	0.765	0.090	0.879	0.261	0.766
n-3	<0.001	0.002	<0.001	0.192	<0.001	0.041
n-6	0.019	0.260	<0.001	0.194	<0.001	0.297
n-6/n-3	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
SFA	0.508	0.463	<0.001	0.707	<0.001	0.217
MUFA	<0.001	0.048	<0.001	0.831	<0.001	0.323
PUFA	<0.001	0.046	<0.001	0.944	<0.001	0.090

SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids.
[a] Contrast: L = linear and or Q = quadratic relationship ($P < 0.05$).

The effect of feeding flaxseed on tissue deposition of n-3 fatty acids in pigs has been found to be quite variable (Nguyen et al., 2003). Feeding extracted flaxseed oil likely provides the most digestible form of 18:3n-3, but would be relatively expensive for inclusion in swine diets. D'Arrigo et al. (2002) fed 3% flaxseed oil (equivalent to 7.5% flaxseed containing 40% oil) to gilts for 8 weeks from 48 to 100 kg and found 9.29% n-3 fatty acids in backfat fatty acids. Results from the present experiment indicate co-extruded flax/pea mixture had a comparably high n-3 fatty acid availability yielding 6.2% and 11.7% (average 8.9%) n-3 fatty acids when feeding 5% and 10% flaxseed for 8 weeks. Fontanillas, Barroeta, Baucells, and Codony (1997) fed 4% flaxseed oil, equivalent to ~10% flaxseed to barrows for ~12 weeks and found 13.5% n-3 fatty acids in backfat and present results were equally as impressive yielding 15.8% n-3 fatty acids in backfat when feeding 10% flax for 12 weeks. Present findings of 9.79%, 16.1% and 21.6% n-3 fatty acids in backfat when feeding 15% co-extruded flaxseed for 4, 8 and 12 weeks, respectively appear higher than those reported by several authors. Romans et al. (1995b) found 6.92% n-3 fatty acids in inner backfat when feeding 15% flax to pigs for 4 weeks, while Thacker et al. (2004) found 13.5% n-3 fatty acids in backfat when feeding 15% flax for close to 14 weeks and Specht-Overholt et al. (1997) found 12% n-3 fatty acids in backfat when feeding 15% flax to finisher pigs for 7 weeks. Finally, Huang, Zhan, Luo, Liu, and Peng (2008) found 9.72% n-3 fatty acids in backfat fatty acids when feeding 10% flax to pigs for close to 13 weeks. Hence co-extruding flax with peas can, therefore, be used to optimize n-3 fatty acid availability from flax and optimization of flaxseed processing will no doubt have an important influence on the profitability and viability of producing n-3 enhanced pork.

In Canada, 300 mg n-3 fatty acids per serving (100 g) is required for an enrichment claim (CFIA & Health-Canada, 2009). Thus, when co-extruded flax/pea mixture was used as a source of dietary 18:3n-3: 3.5 g of backfat from pigs fed 10% or 15% flax for 8 or 4 weeks, respectively, 2.5 g of backfat from pigs fed 10% or 15% flax for 12 or 8 weeks, respectively, or 2.0 g of backfat from pigs fed 15% flax for 12 weeks would achieve the n-3 fatty acid levels required for an enrichment claim (assuming 85% fat in backfat). Additionally, besides the potential manufacturing of pork products enriched in n-3 fatty acids using backfat, commercial lean meat from pigs fed flax co-extruded with field peas could likely achieve the required n-3 levels by including the amount of fat (subcutaneous, intermuscular and intramuscular) usually present in retail cuts.

Adding flax to pig diets, does not however only affect backfat levels of n-3 fatty acids. Although there was a decrease in the relative content of 18:2n-6 in dietary fat when flax was added to the diet, the increased level of fat in flax containing diets actually led to increased absolute amounts (mg/g) of 18:2n-6 in the diet as the level of flax increased. Consequently this led to linear increases in backfat 18:2n-6 and total n-6 fatty acids when flax containing diets were fed for increasing durations. A linear increase in 20:2n-6 was, however, only found when the 10% and 15% flax diets were fed for increasing durations, and backfat levels of 20:4n-6 were linearly reduced when flax containing diets were fed for increasing durations (Table 8). The decrease in 20:4n-6 levels may be due to the competition between 18:2n-6 and 18:3n-3 for desaturation and elongation to form 20:4n-6 and 20:5n-3 (Cherian & Sim, 1995). The decrease in 20:4n-6 levels might be considered as beneficial, since increasing concentrations of this fatty acid in membrane phospholipids result in an overproduction of eicosanoids and induce platelet aggregation (Kristensen, Schmidt, & Dyerberg, 1989), which may contribute to hardening of the arteries and other chronic conditions (Simopoulos, 2002).

The overall changes in backfat levels of n-3 and n-6 when feeding the co-extruded flaxseed containing diets for increasing durations prior to slaughter were well reflected by significant linear ($P < 0.001$) increases in total PUFA. In general, increases in PUFA, however, meant either concomitant reductions in SFA and or MUFA. Remarkably, the level of SFA only showed a linear decline ($P < 0.001$) when feeding the 10% and 15% co-extruded flax diets for increasing durations. On the other hand, increases in PUFA when feeding flax containing diets for increasing duration were inversely mirrored by linear reductions in MUFA ($P < 0.001$), probably due to inhibition of Δ9 desaturase activity as the level of n-3 fatty acids increased in the diet (Waters, Kelly, O'Boyle, Moloney & Kenny, 2009).

3.3. n-3 Fatty acid enrichment paradox

Understanding the evolution of n-3 fatty acid enrichment in backfat is an important concept when trying to attain a certain level of enrichment for a particular weight of pig. When following the enrichment of n-3 fatty acids in backfat over time in pigs fed flax from 26 to 95 kg, Fontanillas et al. (1998) found a rapid enrichment leading to a plateau after about 4 weeks and similar observations were noted by D'Arrigo et al. (2002). Based on these data, the assumption might be made that to enrich n-3 fatty acids in market weight pigs, feeding pigs during the last 4 weeks of the finishing period would be enough to reach the maximum level of enrichment. The rate of enrichment and duration until plateau are however influenced by the level of flax supplementation and the starting weight of the pigs. The rate of fat deposition is higher in heavier pigs, and heavier pigs also have more body fat to dilute n-3 fatty acids originating from dietary flax. On the other hand, feeding higher levels of flax may provide 18:3n-3 in excess of its metabolism/catabolism processes, resulting in higher rates of n-3 fatty acid deposition. Thus, in the present study, when feeding 5% flax co-extruded with field peas a plateau in total n-3 fatty acids was observed for periods longer than 8 weeks, with no significant increase when the feeding duration was increased from 8 to 12 weeks (Table 7; Fig. 1). However, when feeding 10% and 15% flax the level of n-3 fatty acids did not plateau and continued to increase ($P < 0.01$) between 8 and 12 weeks. In addition, the quadratic component (Table 8) was stronger for the lowest level than for the higher levels of flax addition, which had clear linear relationships, indicating a lower dilution effect with increasing dietary flax. Similar results have been reported by Huang et al. (2008) when feeding 10% flaxseed for different periods prior to slaughter. n-3 Fatty acid levels can, therefore, be rapidly enriched in young

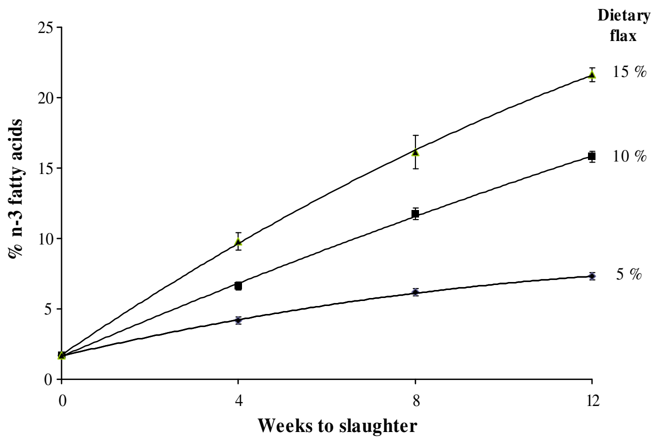

Fig. 1. Effect (mean ± standard error) of feeding 5%, 10% and 15% co-extruded flax different periods prior to slaughter on total n-3 fatty acid content.

pigs but paradoxically to achieve high levels of n-3 fatty acids in market weight pigs, relatively long feeding periods are still required. To yield the same level of n-3 fatty acid enrichment in backfat, however, shorter feeding periods with higher levels of dietary flax appear to be more efficient than longer feeding periods with lower levels of dietary flax. To produce n-3 enriched pork products, however, both the level and consistency of enrichment are important. Consistency from feeding lower flax levels for longer durations was observed to be higher than for high levels for shorter durations. As shown in Fig. 1, feeding 5% co-extruded flax for 12 weeks resulted in a lower standard error than feeding 10% for 8 weeks or 15% for 4 weeks (0.26 vs. 0.40 vs. 0.64, respectively). When feeding 5% flax for 8 weeks, the standard error value was lower than feeding 10% flax for 4 weeks (0.25 vs. 0.30), even if both diets resulted in a similar total n-3 fatty acid content (Table 7). A similar effect was observed when the variability from 10% to 12 weeks and 15% to 8 weeks diets were compared (0.38 vs. 1.18). Hence to more consistently yield the same level of n-3 fatty acid enrichment in backfat, shorter feeding periods with higher levels of dietary co-extruded flax appear to be more efficacious than longer feeding periods with lower levels of dietary flax. In practical terms, to be able to produce pork with consistently high levels of n-3 fatty acids, one will therefore likely need to strike a balance between feeding high levels of flax for short durations, which provides efficient n-3 fatty acid deposition, and feeding lower levels of flax for longer durations, which provides more consistent rates of deposition.

4. Conclusions

Feeding up to 15% flax for 8 weeks had no impact on live animal performance. Feeding any level of flax for 12 weeks reduced average daily gain but feeding higher levels of flax improved feed efficiency. The addition of a 50:50 mix of extruded flax/peas to pig diets provided a highly available source of 18:3n-3 yielding n-3 fatty acid enrichments in backfat comparable to reports when feeding supplemental flax seed oil. Enrichments of n-3 fatty acids, according to Canadian standards, can be obtained in pork products when relatively low levels of backfat from pigs fed flaxseed are included. Increasing the duration of flax supplementation resulted in a plateau in backfat total n-3 fatty acids when feeding 5% flax for 8 weeks but plateaus were not evident when feeding either 10% or 15% flax for 12 weeks. Hence feeding higher levels of co-extruded flaxseed for shorter periods vs. lower levels for longer periods appears to be more efficient at increasing n-3 fatty acids in pig backfat but more consistent enrichments were apparent when feeding lower levels of flaxseed for longer durations. Reaching desired n-3 fatty acid levels in pork will, however, also have to be done in concert with adequate levels of antioxidant protection to maintain pork quality. In the present experiment we added 40 IU of vitamin E per kilogram of feed, and although this appeared adequate for animal growth and performance, it was likely less than adequate to achieve optimal pork quality when feeding higher levels of flax for longer durations (Pettigrew & Esnaola, 2001). Designing flax feeding programs to increase n-3 fatty acids in pork will, therefore, also require optimization of vitamin E levels while taking into consideration economic returns to producers for added investments.

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