Dietary fat type, meat quality and fatty acid metabolism in swine

Jamlong Mitchaothai

Utrecht, 2007

Dietary fat type, meat quality and fatty acid metabolism in swine

Jamlong Mitchaothai

Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands

Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong Chok, Bangkok, Thailand

Ph.D. Thesis, Utrecht University. With summary in English, Dutch and Thai

ISBN 978-90-393-4681-5

Key words: pigs, diet, polyunsaturated fatty acids, meat quality, body composition, energy balance, fatty acid synthesis

Cover: Jitsupang Tree-oat and Jamlong Mitchaothai
Lay-out: Jamlong Mitchaothai

Dietary fat type, meat quality and fatty acid metabolism in swine

(with a summary in English)

Type voedingsvet, vleeskwaliteit en vetzuurstofwisseling bij varkens

(met een samenvatting in het Nederlands)

ชนิดของไขมันในอาหาร, คุณภาพเนื้อ และเมทาบอลิซึมของกรดไขมันในสุกร

(พร้อมบทสรุปภาษาไทย)

Proefschrift

ter verkrijging van de graad van doctor aan de Universiteit Utrecht
op gezag van de rector magnificus, prof. dr. J.C. Stoof,
ingevolge het besluit van het college voor promoties
in het openbaar te verdedigen op
donderdag 8 november 2007
des middags te 3.45 uur

door

Jamlong Mitchaothai
geboren op 29 juli 1973 te Angthong, Thailand

Promotor: Prof. dr. ir. A. C. Beynen

Co-promotoren: Dr. H. Everts
Dr. C. Yuangklang

This thesis was accomplished with financial support from

Mahanakorn University of Technology, Thailand

"Imagination is more important than knowledge"

"จินตนาการสำคัญกว่าความรู้ที่มี"

--Albert Einstein--

To

my Mother, Father and Sisters for their eternal love

List of abbreviations

ADFI	Average daily feed intake
ADG	Average daily gain
ALA	α-Linolenic acid
BF	Back fat
BT	Beef tallow
BW	Body weight
CF	Crude fibre
CP	Crude protein
DM	Dry matter
EE	Ether extract
EFA	Essential fatty acid
FA	Fatty acids
FCR	Feed conversion ratio
GE	Gross energy
DE	Digestible energy
IMF	Intramuscular fat
LA	Linoleic acid
LO	Linseed oil
LSQ	Lenden-Speck-Quotient
ME	Metabolisable energy
MUFA	Monounsaturated fatty acids
ND	Not detectable
NEFA	Non-essential fatty acid
NS	Not significant
PUFA	Polyunsaturated fatty acids
SD	Standard deviation
SE	Standard error
SFA	Saturated fatty acids
SO	Sunflower oil

Contents

CHAPTER 1

General introduction

Dietary fat sources for swine

Diets for swine are generally low in fat, the level being less than 50 g/kg DM (Doreau and Chilliard, 1997). The use of diets low in fats and high in carbohydrates is more economic than the use of diets high in fats and low in carbohydrates. Fats provide energy and thus contribute to meeting the energy requirements of pigs. When compared with the other energy sources, carbohydrates and proteins, fats provide about twice as much on a weight basis. More importantly, dietary fats provide essential fatty acids and are required for the formation of micelles in the duodenum. The essential fatty acids, linoleic acid and α-linolenic acid, cannot be synthesized in the body and must be ingested to prevent disease symptoms of deficiency. Micelles not only facilitate the absorption of fats, but also are required to enable the absorption of the fat-soluble vitamin A, D, E, and K.

Dietary fat sources can be of either animal or plant origin. Different fat sources contain different fatty acids as illustrated in Table 1. In general, vegetable oils are rich in polyunsaturated fatty acids (PUFA), whereas animal fats are rich in saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA). However, the plant fats, coconut oil and palm kernel oil, also contain high amounts of SFA. Olive oil has a high content of MUFA. Fish oils are typified by high contents of polyunsaturated fatty acids with long carbon chains (LC-PUFA).

In order to reduce the risk of cardiovascular disease in humans, it is recommended to lower the intake of saturated fats and cholesterol (Jakobsen, 1999). Replacement of SFA in the diet by MUFA or PUFA will decrease the blood concentrations of low density lipoprotein (LDL), which is the major carrier of cholesterol in humans. High levels of LDL are associated with an increased risk of coronary heart disease. Individual fatty acids may have differential effects on blood cholesterol concentrations. The SFA's lauric acid (C12:0), myristic acid (C14:0) and palmitic (C16:0) raise cholesterol, whereas the SFA stearic acid (C18:0) does not affect blood cholesterol levels (Jakobsen, 1999). The MUFA oleic acid (C18:1) has cholesterol-lowering activity (Yu et al., 1995) and increases the oleic acid content of LDL (Seiquer et al., 1995), resulting in LDL's being less susceptible to oxidation (Reaven et al., 1993; Reaven et al., 1991) and thereby less atherosclerosis (Nicolosi et al., 2002). The PUFA linolenic acid (LA; C18:2 n-6) is more susceptible to oxidation than is oleic acid. The cholesterol-lowering effect of α-linolenic acid (ALA; C18:3 n-3) may be somewhat greater than that of linoleic acid (Arjmandi et al., 1998; Garg and Blake, 1997; Goyens and Mensink, 2006). In addition, high intakes of the omega-3 PUFA's such as ALA, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), are considered to have beneficial effects on blood pressure and platelet aggregation through their role as precursor of eicosanoids (Jakobsen, 1999). The ratio of n-6:n-3 PUFA's is considered to be an index of an healthy diet and the recommended ratio is less than 4 (Wood et al., 2004).

In the light of the above-mentioned, vegetable oils rich in PUFA, especially LA and ALA, have received extra attention with regard to the human diet. The same holds for the diet of swine. As shown below, the intake of PUFA's by pigs is reflected in their edible meat. Thus, there is increased interest in the use of plant oils, at the expense of animal fats, for the production of swine diets. There is an additional reason for plant oils becoming preferred dietary fats. The contamination with toxic, fat-soluble agents is more likely to occur with fats of animal origin than with oils of plant origin. Animal fats may become contaminated due to the deposition of pesticides in adipose tissue (Table 2). The pesticides ingested with contaminated feedstuffs of plant

Table 1. Fatty acid profile of common fats and oils (%) (modified from Reese (2003))

Name	**Lauric**	**Myristic**	**Palmitic**	**Palmioleic**	**Stearic**	**Oleic**	**Linoleic**	**α-Linolenic**	**Arachidic**	**Gadoleic**	**Arachidic**	**Eicosapentaenoic (EPA)**	**Docosahexaenoic (DHA)**
Structure[a]	12:0	14:0	16:0	16:1	18:0	18:1	18:2	18:3	20:0	20:1	20:4	20:5	22:6
Type				n-7		n-9	n-6	n-3		n-9	n-6	n-3	n-3
Vegetable oil													
Canola oil	-	0.1	4.1	0.3	1.8	60.9	21.0	8.8	0.7	1.0	-	-	-
Coconut oil	47.1	18.5	9.1	-	2.8	6.8	1.9	0.1	0.1	-	-	-	-
Corn oil	-	0.1	10.9	0.2	2.0	25.4	59.6	1.2	0.4	-	-	-	-
Cottonseed oil	0.1	0.7	21.6	0.6	2.6	18.6	54.4	0.7	0.3	-	-	-	-
Linseed (flax) oil	-	-	5.3	-	4.1	20.2	12.7	53.3	-	-	-	-	-
Olive oil	-	-	9.0	0.6	2.7	80.3	6.3	0.7	0.4	-	-	-	-
Palm oil	0.3	1.1	42.9	0.2	4.6	39.3	10.7	0.4	0.3	-	-	-	-
Palm kernel oil	48.2	16.2	8.4	-	2.5	15.3	2.3	-	0.1	0.1	-	-	-
Peanut oil	-	0.1	11.1	0.2	2.4	46.7	32.0	-	1.3	1.6	-	-	-
Safflower oil	-	0.1	6.8	0.1	2.3	12.0	77.7	0.4	0.3	0.1	-	-	-
Soybean oil	-	0.1	10.6	0.1	4.0	23.2	53.7	7.6	0.3		-	-	-
Sunflower oil	-	0.1	7.0	0.1	4.5	18.7	67.5	0.8	0.4	0.1	-	-	-
Sunflower oil(high oleic)	-	-	3.7	0.1	5.4	81.3	9.0	-	-	-	-	-	-
Animal fat													
Beef tallow	0.1	3.2	24.3	3.7	18.6	42.6	2.6	0.7	0.2	0.3	-	-	-
Chicken fat	0.1	0.8	25.3	7.2	6.5	37.7	20.6	0.8	0.2	0.3	-	-	-
Lard (pork fat)	0.1	1.5	26.0	3.3	13.5	43.9	9.5	0.4	0.2	0.7	-	-	-
Milk fat	3.1	10.8	28.8	2.5	13.3	27.6	2.5	1.6	0.1	0.1	-	-	-
Fish oil													
Anchovy oil	-	10.6	16.1	11.4	2.8	10.2	1.0	0.4	0.4	0.5	1.7	24.6	9.8
Cod liver oil	-	6.2	10.5	7.4	1.6	14.3	0.9	0.5	-	18.6	0.4	12.8	8.0
Menhaden oil	-	8.6	21.2	10.6	3.3	15.0	7.0	1.3	0.4	1.2	1.9	13.4	1.4
Salmon oil (wild)	-	5.3	15.8	9.3	3.3	15.5	3.4	1.0	2.5	1.0	0.3	16.6	13.4
Sardine oil	-	6.7	18.9	8.8	3.4	17.1	1.1	0.1	0.1	2.5	1.6	19.1	11

[a]number of carbons : number of double bounds; - = no data available

origin will be deposited in adipose tissue. This not only holds for body fat of pigs, but for body fat of broiler chickens and yolk fat of eggs. Fish oils are not only becoming scarce due to overfishing, but are becoming richer in toxic compounds due to the pollution of oceans; these compounds being heavy metals, dioxins, and plasticizers (Napier, 2007). The idea of producing healthier swine meat when using fats of plant origin rather than of animal origin was a major reason for carrying out the studies described in this thesis.

Table 2. Accumulation of pesticides in body fat of pigs after intake during 4 to 13 weeks (modified from Kan and Meijer (2007))

Pesticide	Concentration ratio (body fat/feed)
PCB	5-6
Polychlorinated dioxins/furans	1.4-2.0
Toxaphene	6-16

Essential fatty acids

In mammals, fatty acids can be synthesised *de novo* from acetyl-Co A, the end product being palmitic acid (C16:0), which can be elongated to stearic acid (C18:0). The enzyme Δ9-desaturase can add a double bound between carbon atoms 9 and 10, resulting in the conversion of stearic acid into oleic acid (C18:1 n-9). Both plants and animals have the enzyme Δ9-desaturase, but animal cells lack the enzyme Δ-12 desaturase for the conversion of oleic acid into linoleic acid (C18:2 n-6). Animal cells also lack the enzyme Δ-15-desaturase and thus cannot convert linoleic acid into α-linolenic acid (C18:3 n-3). Some plants do possess a Δ-12 desaturase and Δ-15 desaturase. Therefore, animals can neither synthesize linoleic acid (LA) nor α-linolenic acid (ALA) acid and thus have to ingest these fatty acids with their diet. Consequently, LA and ALA are essential fatty acids for pigs.

LA and ALA are the parent fatty acids of the families of n-6 and n-3 PUFA, respectively (Figure 1). The two groups of PUFA's are metabolically not interconvertable (Zollner and Tato, 1992). However, the desaturases and elongases that convert LA and ALA are identical (Figure 1). Thus, among the two families of fatty acids there is competition for the same desaturating and elongating enzymes (Zollner and Tato, 1992), but the enzymes have a preference for the n-3 PUFA's (Raes et al., 2004). Additionally, there also is competition between n-3 and n-6 PUFA's for the incorporation into phospholipids. The most important products of LC-PUFA biosynthesis are arachidonic acid (AA, C20:4 n-6), which is derived from LA, and eicosapentaenoic acid (EPA, C20:5 n-3) and docosahexaenoic acid (DHA, C22:6 n-3), which are derived from ALA. Both AA and EPA are substrates for the enzymes cyclo-oxygenase and lipoxygenase that convert them into the eicosanoids, comprising the prostaglandins, thromboxanes and leukotrienes. The eicosanoids play a crucial role in the regulation of inflammation and the immune responses. DHA is of particular importance for the structure and function of the retina and the central nervous system (Zollner and Tato, 1992). DHA is required for maximum cognitive development and for minimizing its loss with aging (Innis, 2007). There is evidence that pigs fed diets rich in n-3 PUFAs are healthier than those fed diets poor in n-3 PUFA's (Nguyen, 2002). In addition, the meat of pigs fed diets rich in PUFA's may promote health of the consumers.

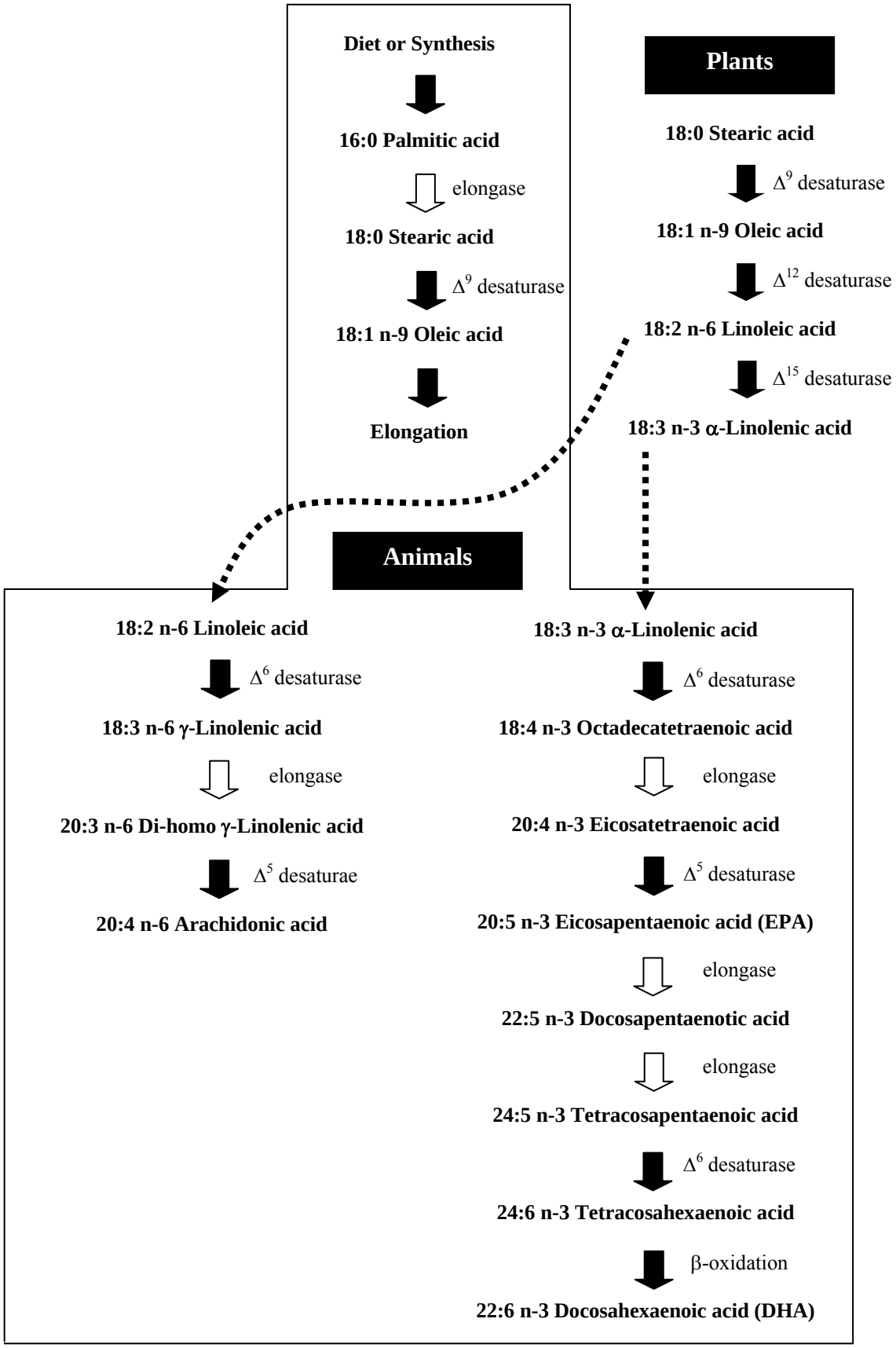

Figure 1. Generalised scheme for LC-PUFA biosynthesis (modified from Napier, 2007)

Fatty acid composition of meat

Lipids in mammalian cells can be classified into two major components, triacylglycerols (neutral fats) and phospholipids. Triacylglycerols (triglycerides) contain a molecule of glycerol esterified with molecules of fatty acids. Glycerophospholipids belong to the phospholipids and are crucial components of cell membranes. In glycerophospholipids, there are two fatty acids attached to the glycerol molecule while the remaining hydroxyl group is bound to a phosphate group that can be esterified with on of several alcohols. Most of the triacylglycerols in the body of animals are stored in the adipose tissues. Intramuscular fat refers to the lipids present in the intramuscular adipose tissues and in the muscle fibres (Raes et al., 2004).

The phospholipids in muscle are characterised by a high PUFA content, the PUFA's comprising 20 to 50% of the total fatty acids in the phospholipids. The phospholipid content is relatively constant and only little influenced by species, breed, age and nutrition, but the phospholipid content depends on the metabolic fibre type of the muscle (Raes et al., 2004). Red muscles have a greater proportion of phospholipids than white muscles, resulting in higher percentage of PUFA in the red muscles (Wood et al., 2003). When compared to phospholipids, triacylglycerols in the body are rich in SFA and MUFA. In triacylglycerols, the content of PUFA, which are mainly represented by LA and ALA, may vary between 2 and 30 % of total fatty acids (Raes et al., 2004). However, there is a marked species difference for content of PUFA in body triacylglycerols. Beef triacylglycerols contain 2 to 3 % PUFA, whereas pork triacylglycerols contain 7 to 15 % PUFA (Gandemer, 1999). Enser et al. (1996) reported PUFA/SFA ratios of 0.58 for pork, but only 0.11 and 0.15 for beef and lamb, respectively (Wood et al., 2003). As described below, the composition of the diet is a major determinant of the n-6/n-3 PUFA ratio in pork.

Dietary fat and meat quality

Pork meat is mainly comprised of myofibrils which are rich in protein. As explained above, it is considered advantageous to increase the amount of polyunsaturated fatty acids in pork. However, a change in the fatty acid composition may alter certain aspects of meat quality. The firmness of fat and cohesiveness are dependent on a high fraction of stearic acid, whereas these characteristics of meat quality are negatively influenced by a high fraction of linoleic acid (Wood et al., 1985). Firm fat is desirable for meat processing, but it is disadvantageous for consumers' health. The basic pigment of fresh meat is myoglobin while tenderness is mainly dependent on the amount and characteristics of connective tissues. As a consequence, changing the amount of fat in pork may not have a noticeable impact on colour and tenderness (Scheeder et al., 2000). However, an increased concentration of PUFA in pork would imply an increased risk of lipid oxidation, leading to off-odours and changes of flavour and colour (Wood et al., 2003), but supplementation swine diets with extra vitamin E may reduce oxidation of PUFAs in pork fat (Asghar et al., 1991). Nevertheless, the fatty acid composition of pork can influence meat quality.

Digestibility of dietary fat and fatty acids

Digestibility is a measure for the availability of nutrients from food. In fact, digestibility is an index of the net amount of fat taken up between mouth and anus. This index is called faecal fat digestibility. The digestibility of dietary fat by pigs

depends on the fatty acid composition of the fat source (Table 3). Fats of animal origin have a lower digestibility than vegetable oils rich in PUFA. The digestibility of individual fatty acids depends on the fat source and on the type of other fats in the diet (Jørgensen et al., 2000; Li et al., 1990). The former is explained by differences in the position of an individual fatty acid on the glycerol molecule. In triacylglycerols, the fatty acids are bound in the glycerol molecule to either C1, C2 and C3, that is the so-called *sn*1-, *sn*2- and *sn*3-position, respectively. After consumption of triacylglycerols, the lingual and gastric lipase will releases the fatty acids at the *sn*3-position. Liberated fatty acids with less than 12 carbon atoms are protonated at the low pH in the stomach and are only partially ionized at higher pH in the small intestine. Longer-chain fatty acids with more than 12 carbon atoms and are totally ionized at high pH and are less soluble due to the length of the molecule. The lingual and gastric lipase action results in 1,2-diacylglycerols and fatty acids entering the duodenum (Bracco, 1994; Mu and Porsgaard, 2005) (Figure 2). In the duodenum, pancreatic lipase and colipase liberate *sn*1- and *sn*3-fatty acids so that fat droplets are formed in combination with bile acids (Bracco, 1994). The fat droplets and the *sn*2-monoacylglycerols that have been produced lead to the formation of micelles which can diffuse through the intestinal contents to be taken up by the enterocytes. Within the enterocytes, the free fatty acids and monoacylglycerols are esterified and as such incorporated into chylomicrons. The chylomicrons are secreted into the lymph and will reach the blood for transport to the tissues that take up the fatty acids. Mu and Porsgaard (2005) have reviewed the literature and concluded that the intestinal absorption of dietary fats is influenced by the structure of triacylglycerols. The absorption of fatty acids is enhanced when they are located at the *sn*2-position. This is especially the case under conditions that fat digestion and absorption are compromised. The higher the proportion of an individual fatty acid bound at the *sn*2-position, the higher will be the digestibility of that fatty acid.

Table 3. Influence of fat source on fat digestibility in pigs[a]

Diet composition			Estimated fat digestibility
Basal diet	Supplemental fat source	Ratio of U/S fatty acids[b]	
Barley-soy	Beef tallow	1.0	70-85
Corn-soy	Beef tallow	1.5	85-92
Barley-soy	Soy oil	4.0	90-95
Corn-soy	Soy oil	4.8	90-95

[a]Modified from Stahly (1984)
[b]Total dietary ratios of unsaturated/saturated fatty acids

Apparent ileal and faecal digestibility

Apparent ileal digestibility is a measure of the availability of nutrients before the terminal ileum, whereas apparent faecal digestibility relates to availability in the whole intestinal tract. In the large intestine, carbohydrates, protein and fats that escaped absorption in the small intestine are further degraded or transformed by microbes. Thus, apparent ileal digestibility reflects the absorbed nutrients, whereas apparent faecal digestibility is also influenced by the activity of microbes in the large bowel. Figure 3 gives a representative diagram for the absorption, deposition, and transformation of dietary LA and ALA (Jakobsen, 1999). Consumed and absorbed LA and ALA are deposited in tissues in unchanged form or after desaturation and

elongation as mentioned earlier. However, LA and ALA that have escaped from absorption in the small intestine will be hydrogenated by microbes in the large bowel, resulting in additional saturated fatty acids in the faeces. It should be noted that the faecal fats include secreted bile lipids, microbial lipids, lipids associated with sloughed intestinal cells and short chain fatty acids produced by microbes (Mu and Porsgaard, 2005).

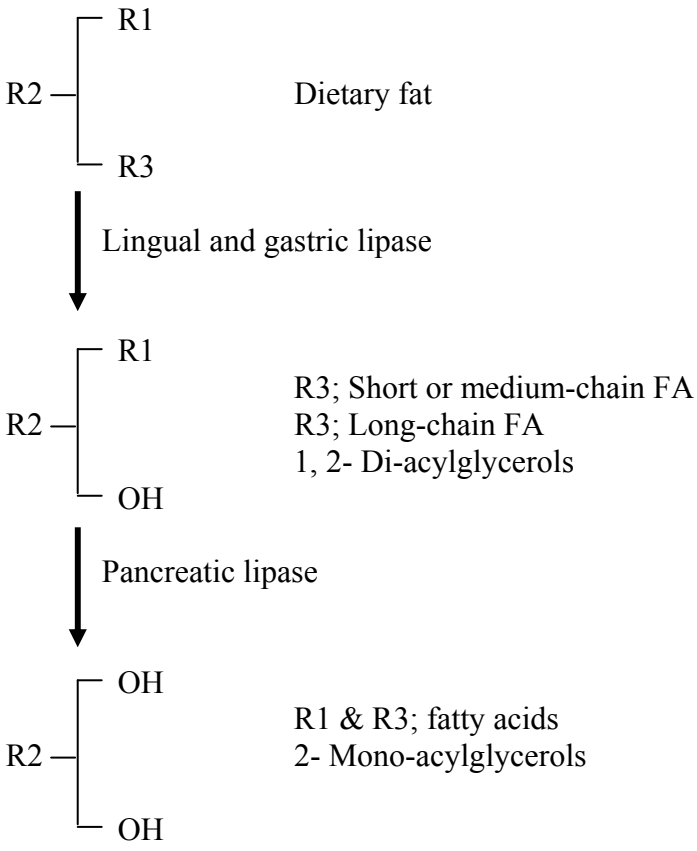

Figure 2. Scheme for enzymatic digestion of dietary triacylglycerols (modified from Bracco (1994))

Apparent ileal digestibility can be measured by the use of pigs with T-cannula at the terminal ileum and using an indigestible marker. An ideal marker for digestibility studies should have the following properties (Maynard, 1979): (1) totally indigestible and unabsorbable, (2) pharmacologically inactive within the digestive tract, (3) pass through the digestive tract at a uniform rate, (4) readily determined chemically and (5) preferably a substance naturally present in the feed. In nutritional studies, indigestible markers can be classified as internal and external markers. Internal markers are integral components of a feedstuff, meeting the properties mentioned above. Lignin and acid insoluble ash (AIA) are commonly used as an internal marker. External markers are indigestible substances added to a diet or feedstuff. Chromic oxide (Cr_2O_3) is a well-known external marker, but other external markers are titanium dioxide (TiO_2) and yttrium oxide (Y_2O_3). The study of Jagger et al. (1992) has compared the efficiency of Cr_2O_3, TiO_2 and lignin as indigestible marker for pigs and they concluded that TiO_2 is most suitable. Cr_2O_3 gave a low recovery and high variation and lignin moved out of phase with amino acids. It was suggested that the most appropriate concentration of TiO_2 is 1 g/kg feed. In addition, TiO_2 , unlike Cr_2O_3, can be legally added to the diet of animals (Titgemeyer et al.,

2001) and it lacks the carcinogenic property of Cr_2O_3 (Peddie et al., 1982). To quantify the amount of TiO_2 in samples, there is a new procedure that is practical, accurate and expedient (Myer et al., 2004). Previous studies have focused on the apparent digestibility of protein, amino acids, carbohydrates and minerals ((Donkoh et al., 1994; Houdijk et al., 1999; Jagger et al., 1992; Jørgensen et al., 1997; Lindberg and Cortova, 1995; Pettersson and Lindberg, 1997; Yin et al., 1991), but only few studies have looked at the digestibility of individual fatty acids (Jørgensen et al., 2000; Li et al., 1990). Thus, further studies are required to determine the apparent digestibility of individual fatty acids.

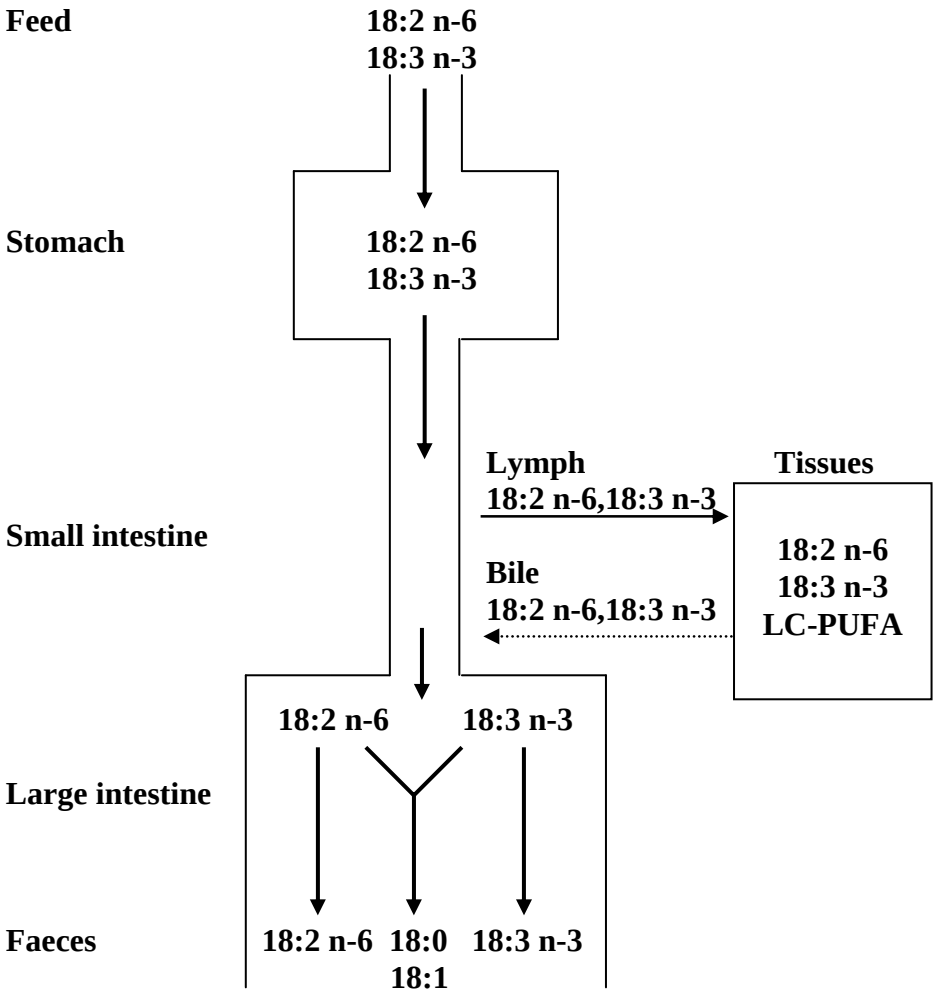

Figure 3. Fate of C18:2 n-6 and C18:3 n-3 in monogastric animals and preruminants (modified from Jakobsen, 1999)

Metabolism and biosynthesis of fatty acids

Dietary and endogenously synthesized fatty acids can serve for the generation of energy or can be stored in the body for oxidation at a later stage. The metabolic pathways are well known, but the quantitative aspects have not been studied extensively. Especially, the quantitative differences in metabolic pathways for individual saturated, monounsaturated and polyunsaturated n-6 and n-3 fatty acids have not been described for pigs fed diets either rich or poor in PUFA. The proportion of body fat and the efficiency of energy storage can be reduced by high intakes of

PUFA at the expense of SFA (Okuno et al., 1997; Sanz et al., 2000a; Sanz et al., 2000b). This may be explained by the observation that LA and ALA are preferentially oxidised (Cunnane and Anderson, 1997; Leyton et al., 1987). Previous studies (Cunnane and Anderson, 1997; Kabir and Ide, 1996) have also shown that the feeding of ALA may increase fatty acid oxidation. Thus, a higher oxidation rate of ALA could explain the observed lower rate of ALA deposition in body of pigs, when compared with LA (Nguyen et al., 2003).

Fatty acids that are consumed in the form of the triacylglycerols will be digested, absorbed and reformed into triacylglyerols. Subsequently, the dietary fatty acids taken up by tissues can be oxidized, elongated to longer chain fatty acids or deposited as such (Figure 4). *De novo* synthesis of fatty acids mainly results in the deposition of palmitic, stearic and oleic acid. The losses of fatty acids with faeces can be quantified by measuring fatty acids in faeces, but these fatty acids are not necessarily derived from the diet as explained above. Thus, calculating the amount fatty acids digested by using the apparent ileal digestibility of fatty acids (Kloareg et al., 2005) would give more realistic values. Fatty acids can be synthesized *de novo* from precursors such as glucose and amino acids and can be itself precursors for fatty acid elongation (Figure 5) (Kloareg et al., 2005). However, only the non-essential fatty acids can be synthesized and not the essential fatty acids (EFA), LA and ALA. This means that the disappearance (oxidation) of EFA can be estimated by subtracting the amount of EFA retained from the amount of EFA intake (Bazinet et al., 2003; Javadi et al., 2007b). Likewise, *de novo* synthesis of NEFA can be estimated by subtracting the amount of NEFA intake from the amount of NEFA retained (Javadi et al., 2007b; Kloareg et al., 2005). However, the actual amount of fatty acid oxidation, transformation, and synthesis cannot be quantified using whole carcass analysis. Therefore, the indicated calculations of oxidation and *de novo* synthesis should be defined as maximum disappearance and minimum *de novo* synthesis, respectively (Javadi et al., 2007b).

Energy metabolism

Carbohydrates, fats and proteins can be used to generate energy in the form of ATP for the functioning of animal cells. Animals are not capable to capture all of the energy available in the feed consumed, because the efficiency of digestion and energy transfer is less than 100% (Figure 6). Energy loss occurs at all metabolic conversions steps. Maynard (1979) estimated that the losses of gross energy in a typical pig diet were 20 % in the form of faecal losses, 2 – 3 % as urinary losses, negligible gaseous losses and 15 – 40 % losses as heat. Total heat production of an animal is composed of heat increment and heat used for maintenance (Bondi and Drori, 1987). Thus, energy expenditure equals total heat production (Figure 6). Because the amounts of gaseous losses in pigs are very small, the energy expenditure can be calculated as follows: energy intake – faecal energy – urine energy – energy stored in body. The efficiency of the conversion of glucose or protein into body fat is considerably lower than that of feed fat into body fat (Bondi and Drori, 1987; Newsholme and Leech, 1986). Energetic efficiency is also influenced by the type of fat and is related to (1) chain length; (2) degree of saturation; (3) degree of esterification; and (4) the combinations of (1), (2) and (3) (Stubbs and Tolkamp, 2006). Beynen and Katan (1985) have suggested that PUFA are more preferentially oxidised than SFA, which has an impact on the oxidation of carbohydrates in that they may shift from the oxidative into the lipogenic pathway. As a consequence, the feeding of pigs with

PUFA instead of SFA would lead to more heat expenditure because of the lower energetic efficiency of the conversion from carbohydrates into body fat.

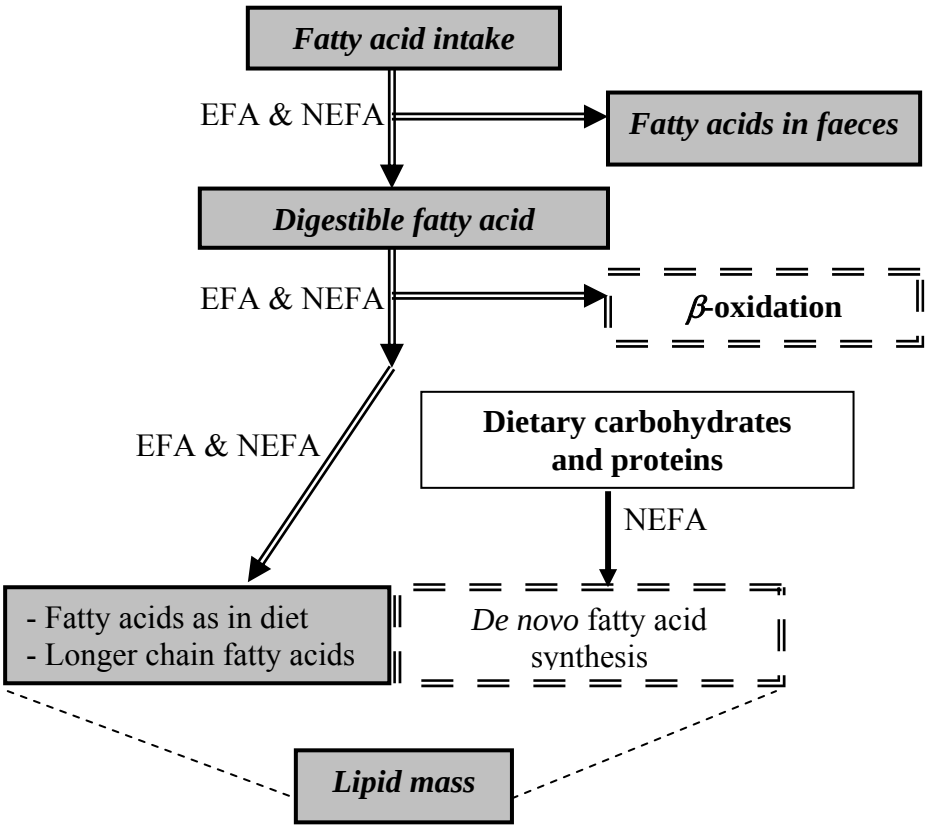

Figure 4. The flow diagram for lipid deposition (modified from Lizardo (2002)). EFA = Essential fatty acids; NEFA = Non-essential fatty acids. Using a digestibility trial and whole carcass analysis, the gray box can be quantified by fat extraction and gas chromatography, and the box with dashed and double lines can be calculated as minimum synthesis or maximum disappearance for NEFA and EFA, respectively .

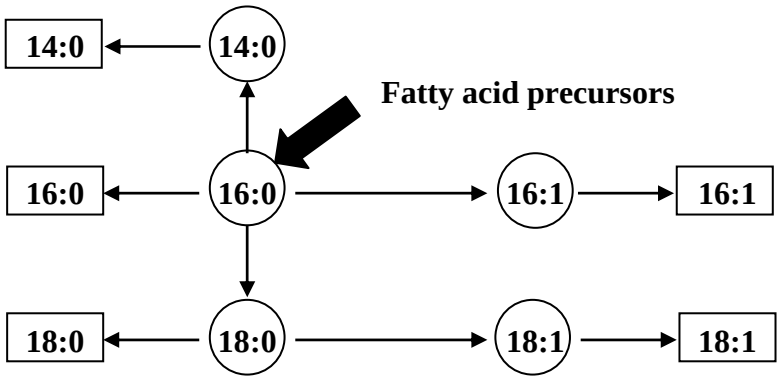

Figure 5. Schematic illustration of *de novo* synthesis, deposition and oxidation of fatty acids in growing pigs. Rectangles = deposited fatty acids; Circles = intermediate fatty acids undergoing deposition or further metabolism (modified from Kloareg et al. (2005))

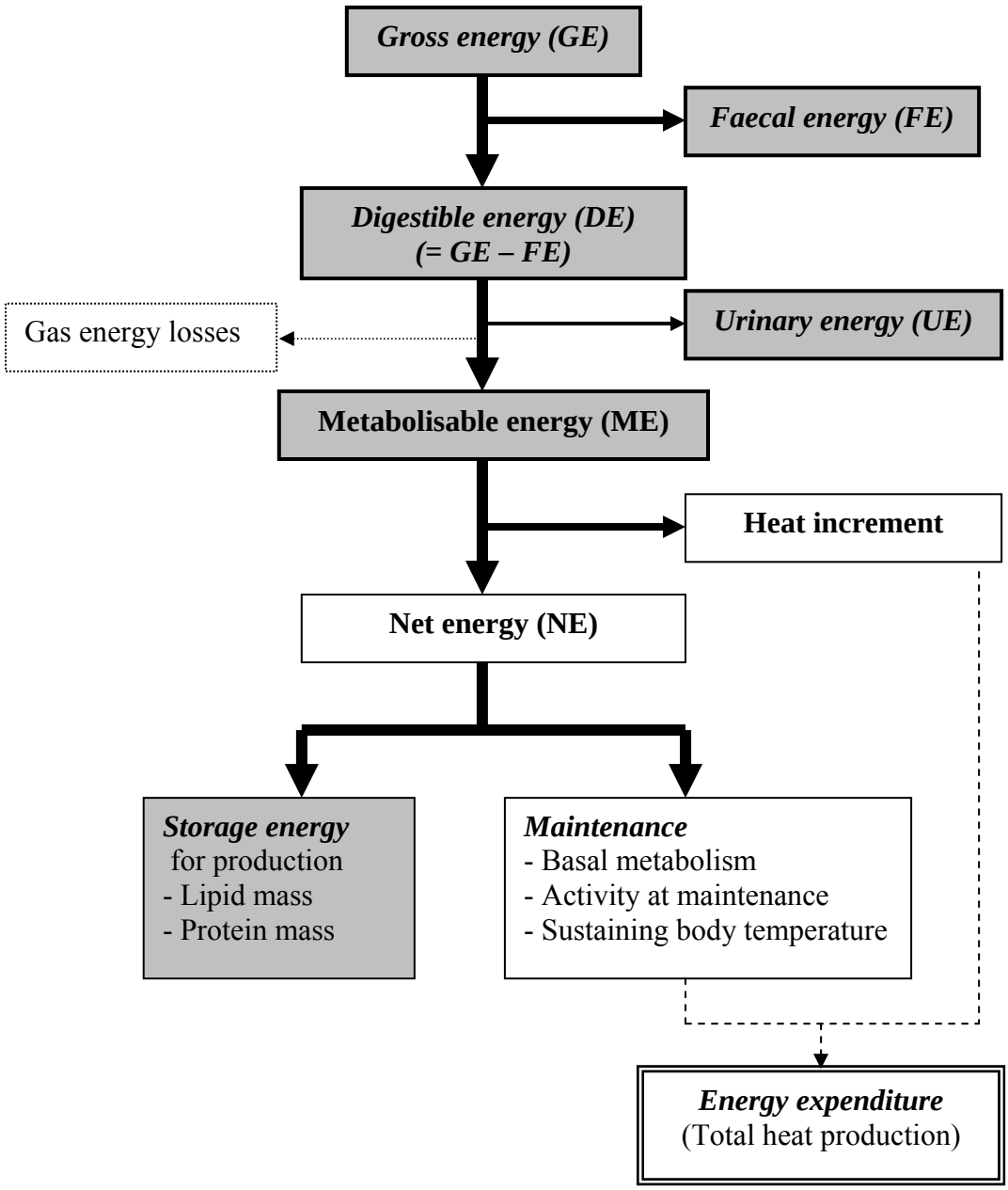

Figure 6. The partitioning of food energy within the growing-finishing pig (modified from Bondi and Drori (1987) and Maynard (1979)). Using a digestibility trial and whole carcass analysis and bomb calorimetry the gray boxes can be quantified; the box with dotted lines can be ignored because of the small amount, and the box with double lines can be estimated based on the known values for the gray boxes.

OUTLINE OF THE THESIS

The general scope of the research described in this thesis is the impact of replacement of animal fat by vegetable oil in the diet of growing-finishing pigs with emphasis on meat composition and meat quality characteristics, incorporation of dietary fatty acids into the body, *de novo* synthesis of fatty acids in whole body and energy expenditure. It was anticipated that the data collected would provide new information on fatty acid and energy metabolism and the possibility to improve pork meat with regard to its effect on the health of consumers. For each chapter, the topic or objective may be summarized as follows.

Chapter 1 provides a summary of the current knowledge on dietary fat type in relation to pork meat quality, fatty acid digestibility, fatty acid and energy metabolism.

Chapter 2 shows the effects of feeding growing-finishing pigs on diets with either sunflower oil or beef tallow on meat quality and fatty acid composition of various tissues.

Chapter 3 describes the interaction between metabolism and intake of individual fatty acids from sunflower oil or beef tallow.

Chapter 4 illustrates how iso-energetic intakes of diets containing either sunflower oil or beef tallow influence fatty acid composition of pork meat and the digestibility, deposition and *de novo* synthesis of individual fatty acids.

Chapter 5 is an account of an investigation of how a diet high in α-linolenic acid or linoleic acid would affect energy metabolism, fatty acid deposition and fatty acid metabolism in the whole body of native Thai pigs.

Chapter 6 describes a study similar to that in Chapter 5, but the diets used were high in α-linolenic acid and contained either sunflower oil or beef tallow as fat background.

Chapter 7 documents the quantification of the apparent ileal and faecal digestibility in pigs fed diets containing either sunflower oil or beef tallow. In addition, the whole body oxidation of fatty acids and energy expenditure were determined by taking into account ileal digestibility of individual fatty acids.

Chapter 8 lists the general conclusions of the various studies performed.

REFERENCES

Arjmandi, B.H., D.A. Kahn, Shanil-Juma, M.L. Drum, Sreevidya-Venkatesh, E. Sohn, L. Wei, and R. Derman. 1998. Whole flaxseed consumption lowers serum LDL-cholesterol and lipoprotein(a) concentrations in postmenopausal women. Nutrition Research 18:1203-1214.

Asghar, A., J.I. Gray, A.M. Booren, E.A. Gomma, M.M. Abouzied, and E.R. Miller. 1991. Effects of supranutritional dietary vitamin E levels on subcellular disposition of a-tocopherol in the muscle and on pork quality. Journal of the Science of Food and Agriculture 57:31-41.

Bazinet, R.P., E.G. McMillan, R. Seebaransingh, A.M. Hayes, and S.C. Cunnane. 2003. Whole-body beta-oxidation of 18:2omega6 and 18:3omega3 in the pig varies markedly with weaning strategy and dietary 18:3omega3. J Lipid Res 44:314-9.

Beynen, A.C., and M.B. Katan. 1985. Why do polyunsaturated fatty acids lower serum cholesterol? Am J Clin Nutr 42:560-3.

Bondi, A.A., and D. Drori. 1987. Animal nutrition. English ed. Wiley, Chichester, New York,.

Bracco, U. 1994. Effect of triglyceride structure on fat absorption. Am J Clin Nutr 60:1002S-1009S.

Cunnane, S.C., and M.J. Anderson. 1997. The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. Journal of Nutrition 127:146-152.

Donkoh, A., P.J. Moughan, and W.C. Smith. 1994. Comparison of the slaughter method and simple T-piece cannulation of the terminal ileum for determining ileal amino acid digestibility in meat and bone meal for the growing pig. Animal Feed Science and Technology 49:43-56.

Doreau, M., and Y. Chilliard. 1997. Digestion and metabolism of dietary fat in farm animals. Br J Nutr 78 Suppl 1:S15-35.

Enser, M., K. Hallett, B. Hewitt, G.A.J. Fursey, and J.D. Wood. 1996. Fatty acid content and composition of english beef, lamb and pork at retail. Meat Science 44:443-458.

Gandemer, G. 1999. Lipids and meat quality: lipolysis, oxidation, maillard reaction and flavour. Science des Aliments 19:439-458.

Garg, M.L., and R. Blake. 1997. Cholesterol dynamics in dynamics in rats fed diets containing either canola oil or sunflower oil. Nutrition Research 17:485-492.

Goyens, P.L., and R.P. Mensink. 2006. Effects of alpha-linolenic acid versus those of EPA/DHA on cardiovascular risk markers in healthy elderly subjects. Eur J Clin Nutr 60:978-84.

Houdijk, J.G., M.W. Bosch, S. Tamminga, M.W. Verstegen, E.B. Berenpas, and H. Knoop. 1999. Apparent ileal and total-tract nutrient digestion by pigs as affected by dietary nondigestible oligosaccharides. J Anim Sci 77:148-58.

Innis, S.M. 2007. Dietary (n-3) fatty acids and brain development. J Nutr 137:855-9.

Jagger, S., J. Wiseman, D.J. Cole, and J. Craigon. 1992. Evaluation of inert markers for the determination of ileal and faecal apparent digestibility values in the pig. Br J Nutr 68:729-39.

Jakobsen, K. 1999. Dietary modifications of animal fats: status and future perspectives. Lipid-Fett 101:475-483.

Javadi, M., M.J.H. Geelen, H. Everts, R. Hovenier, S. Javadi, H. Kappert, and A.C. Beynen. 2007. Effect of dietary conjugated linoleic acid on body composition and energy balance in broiler chickens. British Journal of Nutrition in press.

Jørgensen, H., J.E. Lindberg, and C. Andersson. 1997. Diurnal variation in the composition of ileal digesta and the ileal digestibilities of nutrients in growing pigs. J. Sci. Food Agric. 74:244-250.

Jørgensen, H., V.M. Gabert, M.S. Hedemann, and S.K. Jensen. 2000. Digestion of fat does not differ in growing pigs fed diets containing fish oil, rapeseed oil or coconut oil. J Nutr 130:852-7.

Kabir, Y., and T. Ide. 1996. Activity of hepatic fatty acid oxidation enzymes in rats fed alpha-linolenic acid. Biochim Biophys Acta 1304:105-19.

Kan, C.A., and G.A.L. Meijer. 2007. The risk of contamination of food with toxic substances present in animal feed. Animal Feed Science and Technology 133:84-108.

Kloareg, M., L. Le Bellego, J. Mourot, J. Noblet, and J. van Milgen. 2005. Deposition of dietary fatty acids and of de novo synthesised fatty acids in growing pigs: effects of high ambient temperature and feeding restriction. Br J Nutr 93:803-11.

Leyton, J., P.J. Drury, and M.A. Crawford. 1987. Differential oxidation of saturated and unsaturated fatty acids in vivo in the rat. Br J Nutr 57:383-93.

Li, D.F., R.C. Thaler, J.L. Nelssen, D.L. Harmon, G.L. Allee, and T.L. Weeden. 1990. Effect of fat sources and combinations on starter pig performance, nutrient digestibility and intestinal morphology. J Anim Sci 68:3694-704.

Lindberg, J.E., and Z. Cortova. 1995. The effect of increasing inclusion of lucerne leafmeal in a barley-based diet on the partition of digestion and on nutrient utilization in pigs. Animal Feed Science and Technology 56:11-20.

Lizardo, R., J. Van Milgen, J. Mourot, J. Noblet, and M. Bonneau. 2002. A nutritional model of fatty acid composition in the growing-finishing pig. Livestock Production Science 75:167-182.

Maynard, L.A. 1979. Animal nutrition. 7th ed. McGraw-Hill, New York ; Sydney.

Mu, H., and T. Porsgaard. 2005. The metabolism of structured triacylglycerols. Prog Lipid Res 44:430-48.

Myer, R.O., J.H. Brendemuhl, F.W. Leak, and J.B. Hess. 2004. Evaluation of a rendered poultry mortality-soybean meal product as a supplemental protein source for pig diets. J Anim Sci 82:1071-8.

Napier, J.A. 2007. The production of unusual Fatty acids in transgenic plants. Annu Rev Plant Biol 58:295-319.

Newsholme, E.A., and A. Leech. 1986. Biochemistry for the Medical Sciences. Reprint and correction ed. John Wiley & Sons, New York.

Nguyen, LQ. 2002. Fatty acid supply of growing pigs in Central Vietnam. PhD thesis, Utrecht University.

Nguyen, L.Q., M.C.G.A. Nuijens, H. Everts, N. Salden, and A.C. Beynen. 2003. Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. Meat Science 65:1399-1406.

Nicolosi, R.J., T.A. Wilson, G. Handelman, T. Foxall, J.F. Keaney, and J.A. Vita. 2002. Decreased aortic early atherosclerosis in hypercholesterolemic hamsters fed oleic acid-rich TriSun oil compared to linoleic acid-rich sunflower oil. J Nutr Biochem 13:392-402.

Okuno, M., K. Kajiwara, S. Imai, T. Kobayashi, N. Honma, T. Maki, K. Suruga, T. Goda, S. Takase, Y. Muto, and H. Moriwaki. 1997. Perilla oil prevents the excessive growth of visceral adipose tissue in rats by down-regulating adipocyte differentiation. J Nutr 127:1752-7.

Peddie, J., W.A. Dewar, A.B. Gilbert, and D. Waddington. 1982. The use of titanium dioxide for determining apparent digestibility in mature domestic fowls (Gallus domesticus). The Journal of Agricultural Science 99:233-236.

Pettersson, A., and J.E. Lindberg. 1997. Ileal and total tract digestibility in pigs of naked and hulled barley with different starch composition. Animal Feed Science and Technology 66:97-109.

Raes, K., S. De Smet, and D. Demeyer. 2004. Effect of dietary fatty acids on incorporation of long chain polyunsaturated fatty acids and conjugated linolenic acid in lamb, beef and pork meat: a review. Animal Feed Science and Technology 113:199-221.

Reaven, P., S. Parthasarathy, B.J. Grasse, E. Miller, D. Steinberg, and J.L. Witztum. 1993. Effects of oleate-rich and linoleate-rich diets on the susceptibility of low density lipoprotein to oxidative modification in mildly hypercholesterolemic subjects. J Clin Invest 91:668-76.

Reaven, P., S. Parthasarathy, B.J. Grasse, E. Miller, F. Almazan, F.H. Mattson, J.C. Khoo, D. Steinberg, and J.L. Witztum. 1991. Feasibility of using an oleate-rich diet to reduce the susceptibility of low-density lipoprotein to oxidative modification in humans. Am J Clin Nutr 54:701-6.

Reese, D.E. 2003. Omega-3 fatty acids and swine reproduction: a review. University of Nebraska-Lincoln, Nebraska.

Sanz, M., A. Flores, and C.J. Lopez-Bote. 2000a. The metabolic use of energy from dietary fat in broilers is affected by fatty acid saturation. Br Poult Sci 41:61-8.

Sanz, M., C.J. Lopez-Bote, D. Menoyo, and J.M. Bautista. 2000b. Abdominal fat deposition and fatty acid synthesis are lower and beta-oxidation is higher in broiler chickens fed diets containing unsaturated rather than saturated fat. J Nutr 130:3034-7.

Scheeder, M., K.R. Gläser, B. Eichenberger, and C. Wenk. 2000. Influence of different fats in pig feed on fatty acids composition of phospholipids and physical meat quality characteristics. European Journal of Lipid Science and Technology 102:391-401.

Seiquer, I., M. Manas, E. Martinez-Victoria, M.C. Ballesta, and J. Mataix. 1995. The influence of dietary fat source (sunflower oil or olive oil) on LDL composition and serum lipid levels in miniature swine (Sus scrofa). Comp Biochem Physiol B Biochem Mol Biol 111:163-9.

Stahly, T.S. 1984. Use of fats in diets for growing pigs. 1st ed. Anchor Bredon Ltd., Essex.

Stubbs, R.J., and B.J. Tolkamp. 2006. Control of energy balance in relation to energy intake and energy expenditure in animals and man: an ecological perspective. Br J Nutr 95:657-76.

Titgemeyer, E.C., C.K. Armendariz, D.J. Bindel, R.H. Greenwood, and C.A. Loest. 2001. Evaluation of titanium dioxide as a digestibility marker for cattle. J Anim Sci 79:1059-63.

Wood, J.D., R.C.D. Jones, J.A. Bayntun, and E. Dransfield. 1985. Backfat quality in boars and borrow at 90 kg live weight. Animal Production 40:481-487.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2003. Effects of fatty acids on meat quality: a review. Meat Science 66:21-32.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: a review. Meat Science 66:21-32.

Yin, Y.-L., R. Hunan, H. Zhong, C.-M. Chen, and H. Dai. 1991. Influence of different cannulation techniques on the pre-caecal digestibility of protein, amino acids and cell wall constituents from diets, containing different protein meals, in pigs. Animal Feed Science and Technology 35:271-281.

Yu, S., J. Derr, T.D. Etherton, and P.M. Kris-Etherton. 1995. Plasma cholesterol-predictive equations demonstrate that stearic acid is neutral and monounsaturated fatty acids are hypocholesterolemic. Am J Clin Nutr 61:1129-39.

Zollner, N., and F. Tato. 1992. Fatty acid composition of the diet: impact on serum lipids and atherosclerosis. Clin Investig 70:968-1009.

CHAPTER 2

Effect of dietary fat type on meat quality and fatty acid composition of various tissues in growing-finishing swine

J. Mitchaothai[a, c, *], C. Yuangklang[b], S. Wittayakun[b], K. Vasupen[b], S. Wongsutthavas[b], P. Srenanul[b], R. Hovenier[c], H. Everts[c], A. C. Beynen[c]

[a]Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand
[b]Sakon Nakhon Agricultural Research and Training Center, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand
[c]Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, Utrecht The Netherlands

Meat Science 76 (2007) 95-101

Abstract

Thirty-six castrated male growing pigs were used to study the effect of dietary beef tallow (BT) versus sunflower oil (SO) on meat quality and fatty acid composition of various tissues. The diets used contained either 5% (w/w) of the variable fat source. The fat type had no significant effect on carcass traits (carcass weight, back-fat thickness, fat-lean ratio) and meat quality (colour, pH_1, pH_U, drip losses, cooking losses, shear force, sacromere length, loin moisture, loin marbling). The diet with SO instead of BT significantly increased the incorporation of polyunsaturated fatty acids in adipose tissues, loin and liver at the expense of the sum of saturated and monounsaturated fatty acids. In erythrocytes, the diet containing SO raised the contents of saturated and polyunsaturated fatty acids and lowered that of monounsaturated fatty acids. In particular, the SO diet produced an increase in the content of linoleic acid (C18:2 n-6) in the various tissues. It is concluded that feeding a diet with SO instead of BT altered the fatty acid composition of tissues without simultaneously affecting various characteristics of meat quality.

Keywords: Swine; Sunflower oil; Beef tallow; Meat quality; Fatty acid composition

1. Introduction

In non-ruminants, the fatty acid pattern of dietary lipids is reflected in the fatty acid composition of tissues (Jorgensen, Jensen, & Eggum, 1996). Therefore, it would be anticipated that the dietary fat type affects the meat quality of pigs. For adipose tissue and intramuscular fat of swine, mathematical relationships between the dietary concentration of polyunsaturated fatty acids and the fatty acid composition of these tissues have been established (Nguyen, Nuijens, Everts, Salden, & Beynen, 2003). The consumer increasingly prefers products with a higher unsaturated fatty acid composition because of their beneficial effects in preventing cardiovascular diseases (Van Oeckel, Casteels, Warnants, Damme, & Boucque, 1996). Pig diets supplemented with vegetable oils such as soybean oil, sunflower oil, and corn oil, contain a high percentage of unsaturated fatty acids and should lead to healthy products for consumers. However, the impact of these oils on meat quality is not well known. In two studies the feeding of diets rich in sunflower oil (SO) to pigs had no clear influence on various characteristics of meat quality (Miller, Shackelford, Hayden & Reagan, 1990; Rhee, Davidson, Knabe, Cross, Ziprin & Rhee, 1988). It was the aim of the present study to repeat the two earlier studies, but extending the number of meat quality variables and the types of tissues for determination of their fatty acid composition. In the present study, the effect of SO was compared with that of beef tallow (BT). In samples taken from each pig both meat quality characteristics and tissue fatty acid composition were measured.

2. Materials and Methods

2.1. Animals, diets and feeding

Thirty-six barrows, Landrace × Large White × Duroc crossbred, with 25.9 ± 2.5 kg of body weight fed a commercial diet for 10 days before the commencement of the present study. Then, the pigs, weighing 31.0 ± 3.1 kg, were allotted to one of two treatments on the basis of body weight in a completely randomised design by dividing them into 2 groups that were penned in groups of 3 animals. The pigs were allowed *ad libitum* access to feed and water throughout the experiment. Feed consumption was recorded twice a day. There were two experimental diets with either beef tallow (BT) or sunflower oil (SO) as shown in Table 1. Ingredients and nutrient composition of the experimental diets are shown in Table 1. In Table 2 the analysed fatty acid composition of the diets is shown. The pigs were weighed weekly until the end of the experiment which lasted 91 days. All pigs were slaughtered at body weight of 96 ± 7.2 kg in a commercial slaughterhouse. The pigs were put on transport by truck in the evening and killed the next morning after a fasting and resting period of 8 hours upon arrival. Immediately after killing the pigs were exsanguinated.

2.2. Carcass traits, meat quality and chemical analyses

Hot carcass weight was measured after removing all internal organs including kidney. The thickness of back fat was determined according to standard methods described previously (Sripromma, 1984). Fat-lean ratio was expressed according to the LSQ (Lenden-Speck-Quotient) system of Pfeiffer & Falkenberg (1972). Briefly, the LSQ is calculated as (B1 + B2)/(2 x B3) where B1 = back fat thickness at the front base of gluteus muscle, B2 = back fat thickness on top of gluteus muscle (at the

thinnest part of back fat) and B3 = shortest distance from the front base of gluteus muscle to the dorsal border of the spinal cord. The right *M. longissimus* was used for meat quality assessing. The colour score of *M. longissimus thoracic* was measured by using the 6-point Japanese pork colour scale (Nakai, Saito, Ikeda, Ando, & Komatsu, 1975). The scoring was done at 30 min after slaughtering. The pH-value of muscle was determined using the Meat pH meter (Model HI99163, Hanna Instruments, Portugal) at 45 min (pH_1) and 24 hr (pH_U) after slaughter. Drip loss was assessed as the proportionate weight loss of a slice of muscle (175 – 185 g with thickness of 2.54 cm) that had been suspended in a plastic bag for 24 h at 2°C (Honikel, 1987). Right *M. longissimus lumborum* (loin) was collected and stored at –20°C until analysing the remaining meat quality parameters. Pork chops were weighed before and after cooking to determine percentage of cooking loss. The pork chops were put in a plastic bag and then cooked for 40 min in a water bath with constant temperature of 70 C. After the chops had cooled to room temperature (25°C), five 1.27-cm-diameter cores from each chop were removed with a cylindrical core parallel to the muscle fiber orientation. Cores were sheared perpendicular to the muscle fiber orientation using a Warner-Bratzler shear V-blade attached to an Texture Analyser (Stable Micro System Ltd., Surrey, England) fitted with a 10-g compression load cell with a crosshead speed of 900 mm/min. Peak force values of cores sheared through the centre were used to determine the mechanical tenderness of the sample. Sarcomere length was assessed (in four replicates) by taking the slice from the central part of small *M. longissimus thoracic* (3 to 4 g) according to a method described previously (Monin, Larzul, Le Roy, Culioli, Mourot, Rousset-Akrim, Talmant, Touraille, & Sellier, 1999). The diet and meat samples were dried at 60°C for 72 hr in a forced-hot air oven and then were analysed for crude protein, crude fiber, and ash (AOAC, 1990). The dried meat samples were quantified for the percentage of moisture and fat contents according to standard chemical analyses (AOAC, 1990) and then the percentage of fat was calculated and expressed as loin intramuscular fat.

2.3. Fatty acid composition

Total lipids in the fresh and dried samples (subcutaneous fat from the inguinal region and the back, loin, liver, and diet) were extracted according to a procedure described previously (Horwitz, 1975). In the total lipid fraction the fatty composition was determined. Each sample was added to a flask and 2 ml of ethanol was added to moisturise the sample. Subsequently, 10 ml of HCl (8 mol/l) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80°C for 30-40 min. The tubes were cooled down, 10 ml of ethanol (96%) and 25 ml of petroleum ether (boiling point between 40 and 60°C) were added and the tube was again vigorously shaken for another 1 min. The fat-containing upper layer was decanted into a 150 ml round-bottom flask. The extraction procedure was repeated twice with 15 ml of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath at 40°C. The round-bottom flasks with the lipids were dried overnight at 60°C and the total lipids were measured gravimetrically. Total lipids were saponified and methylated according to the procedure of Metcalfe, Schmitz and Pelka (1966) followed by gas liquid chromatography using a flame ionisation detector, a Chrompack column (Fused silica, no.7485, CP.FFAPCB 25m * 0.32 mm, Chrompack, Middelburg, The Netherlands) and H_2 as carrier gas. The individual fatty acids are expressed as weight percentage of total methyl esters. Blood samples were collected by vena cava puncture. For the analysis of the fatty acid

composition of erythrocyte membranes, blood was collected in heparinised tubes. Erythrocytes were separated by centrifugation for 5 min at 3000 rpm. The erythrocytes were haemolysed and stored at –20°C until fatty acid analyses. From the erythrocyte membranes fatty acids were extracted, methylated (Metcalfe, Schmitz, & Pelka, 1966), and determined by gas chromatography (Nelson, 1975).

2.4. Statistical analyses

The effect of dietary fat type was evaluated for statistical significance by the Student's *t* test (SPSS, 1999). All results are expressed as means ± SD for 18 animals per group except for ADFI and feed:gain because the pigs were housed three in a pen.

Table 1
Composition of the diets

	BT	SO
Raw materials (%)		
Cassava chip	45.53	45.53
Soybean meal (44% CP)	34.00	34.00
Extruded soy beans	7.00	7.00
Beef tallow	5.00	-
Sunflower oil	-	5.00
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Di-calcium phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Premix	0.25	0.25
Analysed nutrients (%)		
Dry matter	86.5	86.3
Crude protein	19.3	19.2
Crude fat	6.8	6.7
Crude fiber	4.7	4.9
Ash	7.7	8.2
Calculated ME (MJ/kg)	13.8	14.0

BT = beef tallow; SO = sunflower oil

3. Results and Discussion

3.1. Animal performance

Final body weight, average daily feed intake (ADFI), average daily gain (ADG), and feed to gain ratio were not significantly different ($P > 0.05$) between pigs fed either the BT diet or the SO diet (Table 3). These results agree with previous research (Bryhni, Kjos, Ofstad, & Hunt, 2002; Cera, Mahan, & Reinhart, 1990; Smith, Knabe, & Smith, 1996; Wiseman & Agunbiade, 1998) in which the experimental pigs were fed either restricted amounts of feed or *ad libitum*. Hence, in the present study, the fat type in the diet for growing-finishing pigs did not affect growth performance.

3.2. Meat quality

Table 2
Fatty acid composition of experiment diets

	BT	SO
Fatty acids, g methylester/100 g methylesters		
C 8:0	0.02	0.02
C 10:0	0.05	0.05
C 14:0	1.48	0.05
C 14:1	0.06	0.06
C 15:0	0.51	0.06
C 15:1	0.37	0.37
C 16:0	19.23	9.26
C 16:1	0.68	0.04
C 17:0	1.59	0.05
C 17:1	0.28	0.00
C 18:0	29.48	4.34
C 18:1 n-9	21.91	31.52
C 18:1 n-7	4.71	1.11
C 18:2 n-6	11.65	49.07
C 18:3 n-6	0.32	0.00
C 18:3 n-3	1.63	1.42
C 18:4n-3	0.00	0.00
C 20:0	0.44	0.34
C 20:1 n-9	0.48	0.39
C 20:5n-3	0.05	0.05
C 22:0	0.02	0.77
C 22:1 n-9	0.00	0.11
C 24:0	0.10	0.33
Unidentified	4.75	0.57
Σ SFA	52.92	15.27
Σ MUFA	28.49	33.60
Σ PUFA	13.65	50.54

BT = beef tallow; SO = sunflower oil

Carcass traits did not differ ($P > 0.05$) for the two fat supplements (Table 3). These results agree with reports published earlier (Bee, Gebert, & Messikommer, 2002; Miller, Shackelford, Hayden, & Reagan, 1990; Nuernberg, Fischer, Nuernberg, Kuechenmeister, Klosowska, Eliminowska-Wenda, Fiedler, & Ender, 2005). Meat quality traits were not statistically different between the BT and SO treatments. These results agree with previous results of Scheeder, Gläser, Eichenberger, and Wenk (2000) who found that feeding 7% pork fat, 4.95% olive oil or 3.17% soybean oil to growing-finishing pigs did not affect pH, cooking losses, texture, or colour of pork. Miller, Shackelford, Hayden, and Reagan (1990) also found no difference in drip losses, cooking losses, shear force, and marbling between pigs fed animal fat and sunflower oil. The sacromere length (1.97-1.98 μm) was close to the values of pigs carrying the nn type of halothane gene as reported by Monin, Larzul, Le Roy, Culioli, Mourot, Rousset-Akrim, Talmant, Touraille, and Sellier (1999). The intramuscular fat or marbling of loin was not statistically different between both treatments which is in accordance with the results of Nuernberg, Fischer, Nuernberg, Kuechenmeister, Klosowska, Eliminowska-Wenda, Fiedler, and Ender (2005) when the effect of olive

and linseed oil were compared. From overall carcass and meat quality results, it may be concluded that adding SO instead of BT to the diet did not alter the quality of pig meat.

Table 3
Animal performance, carcass traits, and meat quality

	BT	SO	P-value
Growth performance			
Initial BW, kg	30.9 ± 3.4	31.1 ± 3.1	0.858
Final BW, kg	95.6 ± 6.1	96.5 ± 8.4	0.728
ADFI, kg/d (n=6)	2.30 ± 0.3	2.33 ± 0.1	0.803
ADG, kg/d	0.790 ± 0.06	0.809 ± 0.07	0.476
Feed : gain (n=6)	3.1 ± 0.4	2.9 ± 0.2	0.420
Carcass quality			
Hot carcass, kg	72.42 ± 12.0	76.62 ± 9.0	0.294
Back fat thickness, cm	2.52 ± 0.4	2.60 ± 0.3	0.509
Fat-lean ratio (LSQ)	0.25 ± 0.1	0.27 ± 0.1	0.709
Meat quality (loin)			
Colour[1]	2.78 ± 0.7	3.00 ± 0.7	0.340
pH_1	6.04 ± 0.3	6.13 ± 0.3	0.467
pH_U	5.74 ± 0.4	5.72 ± 0.5	0.880
Drip loss, %	3.16 ± 2.4	2.20 ± 1.9	0.204
Cooking loss, %	18.38 ± 2.9	17.17 ± 2.6	0.222
Shear force, N	56.16 ± 7.1	56.49 ± 9.1	0.917
Sacromere length, μm	1.97 ± 0.2	1.98 ± 0.1	0.875
Moisture, %	69.12 ± 3.2	69.65 ± 3.8	0.660
IMF, % wet weight	3.03 ± 0.5	2.97 ± 0.6	0.731

BT = beef tallow; SO = sunflower oil
[1]Japanese colour score: 1 = pale, pinkish grey, 6 = dark, purplish red (Nakai, Saito, Ikeda, Ando & Komatsu, 1975).
IMF = intramuscular fat

3.3. Fatty acid composition

Fatty acid profiles of fat in subcutaneous fat, back fat, retroperitoneal fat, loin, erythrocytes, and liver are shown in Tables 4-6. The fatty acid composition of erythrocytes and liver was determined for comparative reasons because in metabolic studies these measurements are often considered relevant. The relative concentration of C16:0 (palmitic acid) after feeding the BT diet was higher for adipose tissue ($P < 0.001$) and liver ($P < 0.05$), but there was no statistical difference for loin and erythrocytes. Similarly, the relative concentrations of C18:0 and C18:1 n-9 (oleic acid) after feeding BT diet were higher for adipose tissues ($P < 0.01$) and loin ($P < 0.05$) (except for lower concentration of C18:0 in erythrocytes) while no difference was found for liver. Conversely, the relative concentration of C18:2 n-6 (linoleic acid) upon SO diet treatment was higher ($P < 0.001$) for adipose tissues, loin, erythrocytes,

and liver. Similar observations have been reported by Miller, Shackelford, Hayden & Reagan (1990). As would be expected on the basis of the concentrations of C18:3 n-3 (α-linolenic acid) in the experimental diets, the relative concentration of C18:3 n-3 in adipose tissues of pigs fed the SO diet was lower ($P < 0.05$) than that in pigs fed the BT diet. However, the concentrations of α-linolenic acid in the diet and adipose tissue may not be correlated strongly especially when the level in the diet is quite low as in this study. The essential α-linolenic acid may be desaturated and elongated in the body. Probably more importantly, α-linolenic acid may be preferentially oxidized for energy generation, leading to a diminished incorporation into adipose tissue as has been suggested previously (Leyton, Drury, & Crawford, 1987). The relative percentage of C20:4 n-6 (arachidonic acid) in the adipose tissues of pigs fed the SO diet was higher, but there was no significant difference for loin, erythrocytes, and liver. In contrast, the relative percentage of C22:6 n-3 (docosahexaenoic acid) was higher ($P < 0.001$) in liver of pigs fed BT whereas there was no difference in the loin and it was not detectable in adipose tissues and erythrocytes.

In the current study, the content of saturated fatty acids (SFA) in adipose tissues ($P < 0.01$) and loin ($P < 0.05$) of the animals fed BT were higher, but it was lower ($P < 0.01$) in the erythrocytes and there was no change for liver. In all tissues of the pigs fed BT, the levels of monounsaturated fatty acids (MUFA) were higher ($P < 0.001$) whereas the levels of polyunsaturated fatty acids (PUFA) were higher ($P < 0.001$) for the pigs fed the diet containing SO. These results are in agreement with a previous study (Hartman, Costello, Libal, & Walhlstrom, 1985). The ratio of MUFA/SFA was not different between both treatments whilst the PUFA/SFA ratio was markedly increased by feeding sunflower oil. In all tissues except for erythrocytes which contained no detectable n-3 polyunsaturated fatty acids, the ratios of n-6/n-3 and C18:2 n-6/C18:3 n-3 were about two to three times higher for pigs fed the SO diet than for those fed the BT diet. In the current study the ratios of n-6/n-3 and C18:2 n-6/C18:3 n-3 were markedly higher than the recommended value of 4, which is based on the presumed effects of dietary polyunsaturated fatty acids on human health (Wood, Richardson, Nute,, Fisher, Campo, Kasapidou, ., Sheard, & Enser, 2003). However, the higher incorporation of C18:2 n-6 had no adverse effect on meat quality which is in accordance with previous results of Hartman, Costello, Libal, & Walhlstrom (1985) and those of West and Myer (1987). It is known that a high n6/n3 PUFA ratio is a risk factor of cancer and coronary heart disease (Assman, Cullen, Jossa, Lewis & Mancini, 1999). Feeding crushed whole linseed had no adverse effect on meat quality and could reduce the n-6/n-3 ratio closed to the target level (less than 4) (Enser, Richardson, Wood, Gill, & Sheard, 2000; Sheard, Enser, Wood, Nute, Gill, & Richardson, 2000). Therefore, in further experiments attention may be directed to supplements rich in α-linolenic acid in order to produce healthier pork.

Nguyen, Nuijens, Everts, Salden, and Beynen (2003) have published mathematical relationships for the intake of n-6 and n-3 PUFA and their contents in adipose tissue and muscle by using the data of different feeding trials. We used the relationships to predict the incorporation of linoleic and α-linolenic acids in adipose tissue of the swine in the current study. The predicted levels of linoleic acid and α-linolenic acid in adipose tissue were close to those actually measured in this study.

4. Conclusion

In this study, the supplementation of SO at an inclusion level of 5% as fed in the diet for growing-finishing pigs had no clear effect on the selected carcass traits

Table 4
Fatty acid composition of subcutaneous and back fat

	Subcutaneous fat		Signi-ficance	Back fat		Signi-ficance
	BT	SO		BT	SO	
Analysed fatty acids (g/100g methylester)						
C 14:0	1.56 ± 0.14	1.24 ± 0.21	***	1.47 ± 0.11	1.02 ± 0.14	***
C 16:0	22.97 ± 1.73	20.89 ± 2.56	***	22.04 ± 1.53	18.67 ± 2.00	***
C 17:0	0.66 ± 0.16	0.32 ± 0.09	***	0.82 ± 0.19	0.36 ± 0.08	***
C 18:0	12.80 ± 1.75	11.04 ± 1.75	**	13.41 ± 1.86	10.64 ± 1.69	***
C 20:0	0.16 ± 0.09	0.21 ± 0.03	*	0.15 ± 0.09	0.19 ± 0.05	**NS**
C 22:0	ND	ND	-	ND	ND	-
C 24:0	ND	ND	-	ND	ND	-
∑ SFA	**38.19 ± 3.35**	**33.70 ± 4.43**	**	**38.01 ± 3.21**	**30.88 ± 3.67**	***
C 16:1	2.13 ± 0.36	1.59 ± 0.35	***	1.78 ± 0.32	1.10 ± 0.21	***
C 17:1	0.50 ± 0.11	0.20 ± 0.08	***	0.55 ± 0.15	0.19 ± 0.07	***
C 18:1n-7	3.44 ± 0.40	2.40 ± 0.42	***	3.50 ± 0.50	2.06 ± 0.22	***
C 18:1n-9	39.05 ± 2.28	35.96 ± 1.99	***	38.06 ± 1.84	34.47 ± 1.76	***
C 20:1n-9	0.88 ± 0.10	0.81 ± 0.11	*	0.92 ± 0.09	0.81 ± 0.07	***
C 24:1	ND	ND	-	ND	ND	-
∑ MUFA	**46.00 ± 2.99**	**40.95 ± 2.55**	***	**44.80 ± 2.56**	**38.66 ± 2.14**	***
C 18:2n-6	11.48 ± 3.22	22.29 ± 5.26	***	12.24 ± 3.13	26.99 ± 3.52	***
C 18:3n-3	1.19 ± 0.23	1.01 ± 0.18	*	1.25 ± 0.16	1.08 ± 0.14	**
C 18:3n-6	ND	ND	-	0.00 ± 0.00	0.01 ± 0.03	**NS**
C 20:2n-6	0.46 ± 0.11	0.89 ± 0.18	***	0.47 ± 0.13	1.06 ± 0.09	***
C 20:3n-6	0.00 ± 0.00	0.04 ± 0.06	*	0.00 ± 0.00	0.05 ± 0.07	**
C 20:3n-3	0.12 ± 0.08	0.05 ± 0.07	**	0.13 ± 0.08	0.07 ± 0.07	*
C 20:4n-6	0.15 ± 0.10	0.27 ± 0.06	***	0.12 ± 0.08	0.25 ± 0.06	***
C 20:5n-3	ND	ND	-	ND	ND	-
C 22:3	0.00 ± 0.00	0.03 ± 0.06	*	0.00 ± 0.00	0.03 ± 0.06	**NS**
C 22:4n-6	0.00 ± 0.00	0.01 ± 0.03	**NS**	0.00 ± 0.00	0.02 ± 0.05	**NS**
C 22:5n-3	0.04 ± 0.07	0.01 ± 0.03	**NS**	0.02 ± 0.05	0.00 ± 0.00	**NS**
C 22:6n-3	ND	ND	-	ND	ND	-
∑ PUFA	**13.43 ± 3.50**	**24.59 ± 5.64**	***	**14.23 ± 3.23**	**29.56 ± 3.79**	***
unknown	2.38 ± 0.63	0.75 ± 0.33	***	2.96 ± 0.78	0.91 ± 0.26	***
MUFA/SFA	1.22 ± 0.16	1.23 ± 0.15	NS	1.19 ± 0.15	1.27 ± 0.20	NS
PUFA/SFA	0.36 ± 0.11	0.76 ± 0.26	***	0.38 ± 0.11	0.98 ± 0.26	***
∑ n6/∑ n3	9.31 ± 2.88	18.35 ± 4.13	***	9.46 ± 4.10	24.97 ± 2.76	***
18:2n6/18:3n3	9.81 ± 2.99	22.17 ± 3.48	***	10.00 ± 3.65	25.16 ± 1.57	***

BT = beef tallow; SO = sunflower oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
NS: $P > .05$; *$P < .05$; **$P < .01$; ***$P < .001$.

Table 5
Fatty acid composition of retroperitoneal fat and loin intramuscular fat

	Retroperitoneal fat		Signi-ficance	Loin		Signi-ficance
	BT	SO		BT	SO	
Analysed fatty acids (g/100g methylester)						
C 14:0	1.73 ± 0.14	1.22 ± 0.17	***	1.29 ± 0.23	1.27 ± 0.17	NS
C 16:0	25.31 ± 2.11	21.74 ± 2.82	***	23.40 ± 1.90	22.40 ± 1.89	NS
C 17:0	0.85 ± 0.17	0.33 ± 0.09	***	0.34 ± 0.07	0.23 ± 0.05	***
C 18:0	18.67 ± 2.61	14.73 ± 2.52	***	12.32 ± 1.15	11.59 ± 1.15	*
C 20:0	0.22 ± 0.07	0.23 ± 0.04	NS	0.11 ± 0.10	0.17 ± 0.07	NS
C 22:0	ND	ND	-	ND	ND	-
C 24:0	0.01 ± 0.02	0.00 ± 0.00	NS	ND	ND	-
∑ SFA	**46.89 ± 4.26**	**38.27 ± 5.37**	***	**37.93 ± 2.79**	**35.32 ± 2.83**	**
C 16:1	1.56 ± 0.36	0.91 ± 0.22	***	2.99 ± 0.38	2.23 ± 0.35	***
C 17:1	0.43 ± 0.08	0.13 ± 0.07	***	0.35 ± 0.08	0.14 ± 0.08	***
C 18:1 n-7	2.98 ± 0.34	1.59 ± 0.19	***	3.73 ± 0.30	2.73 ± 0.30	***
C 18:1 n-9	32.71 ± 2.24	30.39 ± 1.44	**	38.45 ± 3.10	35.80 ± 2.12	**
C 20:1 n-9	0.63 ± 0.09	0.62 ± 0.08	NS	0.76 ± 0.12	0.73 ± 0.11	NS
C 24:1	ND	ND	-	ND	ND	-
∑ MUFA	**38.32 ± 2.72**	**33.63 ± 1.80**	***	**46.55 ± 3.52**	**41.63 ± 2.63**	***
C 18:2 n-6	10.40 ± 2.18	25.26 ± 5.27	***	9.83 ± 2.66	17.54 ± 3.97	***
C 18:3 n-3	1.17 ± 0.25	1.00 ± 0.23	*	0.65 ± 0.11	0.58 ± 0.17	NS
C 18:3 n-6	0.01 ± 0.04	0.00 ± 0.00	NS	0.01 ± 0.06	0.00 ± 0.00	NS
C 20:2 n-6	0.31 ± 0.05	0.78 ± 0.09	***	0.28 ± 0.04	0.65 ± 0.13	***
C 20:3 n-6	0.00 ± 0.00	0.05 ± 0.06	**	0.29 ± 0.16	0.23 ± 0.06	NS
C 20:3n-3	0.02 ± 0.05	0.02 ± 0.04	NS	0.00 ± 0.00	0.01 ± 0.03	NS
C 20:4n-6	0.13 ± 0.08	0.26 ± 0.05	***	1.60 ± 0.91	1.58 ± 0.60	NS
C 20:5n-3	ND	ND	-	0.08 ± 0.14	0.00 ± 0.00	***
C 22:3	0.00 ± 0.00	0.05 ± 0.06	**	ND	ND	-
C 22:4n-6	0.00 ± 0.00	0.01 ± 0.02	NS	0.17 ± 0.16	0.27 ± 0.07	**
C 22:5 n-3	ND	ND	-	0.34 ± 0.18	0.14 ± 0.09	***
C 22:6n-3	ND	ND	-	0.15 ± 0.17	0.07 ± 0.08	NS
∑ PUFA	**12.05 ± 2.56**	**27.42 ± 5.68**	***	**12.93 ± 4.42**	**21.07 ± 4.75**	***
unknown	2.75 ± 0.87	0.69 ± 0.23	***	2.88 ± 1.18	1.65 ± 0.54	***
MUFA/SFA	0.83 ± 0.13	0.90 ± 0.16	NS	1.24 ± 0.11	1.17 ± 0.09	NS
PUFA/SFA	0.26 ± 0.08	0.76 ± 0.31	***	0.35 ± 0.15	0.61 ± 0.19	***
∑ n6/ ∑ n3	9.11 ± 0.43	26.20 ± 2.02	***	9.82 ± 1.20	27.01 ± 6.57	***
18:2 n6/18:3n3	8.86 ± 0.28	25.47 ± 1.68	***	14.22 ± 2.38	30.61 ± 3.66	***

BT = beef tallow; SO = sunflower oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
NS: $P > .05$; *$P < .05$; **$P < .01$; ***$P < .001$.

Table 6
Fatty acid composition of erythrocytes, and liver

	Erythrocytes		Signi-ficance	Liver		Signi-ficance
	BT	SO		BT	SO	
Analysed fatty acids (g/100g methylester)						
C 14:0	0.41 ± 0.04	0.30 ± 0.04	***	0.56 ± 0.27	0.40 ± 0.37	NS
C 16:0	26.90 ± 1.70	28.00 ± 1.70	NS	15.28 ± 2.14	13.41 ± 3.05	*
C 17:0	1.57 ± 0.18	1.42 ± 0.18	***	0.98 ± 0.30	0.75 ± 0.29	**
C 18:0	16.20 ± 0.78	18.26 ± 0.78	***	21.13 ± 4.43	22.06 ± 4.67	NS
C 20:0	0.68 ± 0.09	0.70 ± 0.09	NS	ND	ND	-
C 22:0	2.96 ± 0.35	2.83 ± 0.35	NS	ND	ND	-
C 24:0	6.28 ± 0.65	6.81 ± 0.65	*	ND	ND	-
∑ SFA	**55.29 ± 2.71**	**58.38 ± 2.71**	**	**38.01 ± 2.76**	**36.65 ± 1.86**	NS
C 16:1	0.57 ± 0.07	0.09 ± 0.07	***	1.04 ± 0.36	0.58 ± 0.53	**
C 17:1	0.45 ± 0.06	0.15 ± 0.06	***	0.38 ± 0.08	0.07 ± 0.15	***
C 18:1 n-7	1.63 ± 0.13	1.10 ± 0.13	**	1.99 ± 0.24	1.36 ± 0.31	***
C 18:1 n-9	29.08 ± 2.85	25.81 ± 2.85	***	18.25 ± 4.32	14.98 ± 5.18	NS
C 20:1 n-9	1.17 ± 0.16	1.21 ± 0.16	NS	0.18 ± 0.11	0.25 ± 0.08	*
C 24:1	2.64 ± 0.33	2.67 ± 0.33	NS	ND	ND	-
∑ MUFA	**35.53 ± 2.66**	**31.03 ± 2.66**	***	**21.84 ± 4.80**	**17.24 ± 6.07**	*
C 18:2 n-6	6.09 ± 1.29	7.49 ± 1.29	***	15.94 ± 3.41	22.54 ± 2.49	***
C 18:3 n-3	ND	ND	-	0.93 ± 0.41	0.65 ± 0.42	NS
C 18:3 n-6	ND	ND	-	0.38 ± 0.17	0.42 ± 0.17	NS
C 20:2 n-6	ND	ND	-	0.38 ± 0.10	0.74 ± 0.22	***
C 20:3 n-6	ND	ND	-	0.27 ± 0.28	0.59 ± 0.24	NS
C 20:3n-3	ND	ND	-	ND	ND	-
C 20:4n-6	0.58 ± 0.21	0.48± 0.22	NS	13.37 ± 3.00	16.03 ± 4.61	NS
C 20:5n-3	ND	ND	-	0.82 ± 0.26	0.16 ± 0.12	***
C 22:3	0.13 ± 0.18	0.16 ± 0.18	-	0.03 ± 0.09	0.05 ± 0.09	NS
C 22:4n-6	ND	ND	-	0.65 ± 0.21	1.02 ± 0.29	***
C 22:5 n-3	ND	ND	-	2.17 ± 0.70	1.22 ± 0.23	***
C 22:6n-3	ND	ND	-	1.63 ± 0.44	1.06 ± 0.21	***
∑ PUFA	**6.81 ± 1.47**	**8.14 ± 1.47**	**	**36.85 ± 3.21**	**44.48 ± 5.51**	***
unknown	2.36 ± 1.53	2.45 ± 1.53	NS	3.30 ± 3.37	1.63 ± 4.61	***
MUFA/SFA	0.55 ± 0.08	0.57 ± 0.08	NS	0.58 ± 0.17	0.48 ± 0.19	NS
PUFA/SFA	0.18 ± 0.03	0.17 ± 0.03	NS	0.97 ± 0.12	1.21 ± 0.14	***
∑ n6/ ∑ n3	ND	ND	-	5.91 ± 1.93	13.62 ± 2.68	***
18:2 n6/18:3n3	ND	ND	-	19.85 ± 7.07	45.64 ± 0.12	***

BT = beef tallow; SO = sunflower oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
NS: $P > .05$; *$P < .05$; **$P < .01$; ***$P < .001$.

and meat quality when compared with supplementation of BT at the same level. The SO diet increased the incorporation of PUFA, mainly linoleic acid (C18:2 n-6), in the various animal tissues. Apparently, under the conditions of this study there was no impact of fatty acid composition of tissues on meat quality.

Acknowledgements

The authors gratefully acknowledge the excellent technical assistance of Mr. Suksan Pimchai, Mr. Adisak Auimla-or, Mr. Nimit Kaewmalun, Mr. Narongsak Nantharad, and Mr. Thanaphum Bunmee. Moreover, the authors would like to thank the Sakon Nakhon Agricultural Research and Training Center, Rajamangala University of Technology-Isan for providing facilities and Mahanakorn University of Technology for financial support.

References

AOAC. 1990. Association of Official Analytical Chemists, Washington DC.

Assmann G, P Cullen, F Jossa, B Lewis and M Mancini. Coronary heart disease: reducing the risk. A worldwide view. International Task Force for the Prevention of Coronary Heart Disease. *Circulation (1999) 100,* 1930-1938.

Bee, G., Gebert, S., & Messikommer, R. (2002). Effect of dietary energy supply and fat source on the fatty acid pattern of adipose and lean tissues and lipogenesis in the pig. *Journal of Animal Science, 80*, 1564-1574.

Bryhni, E. A., Kjos, N. P., Ofstad, R., & Hunt, M. (2002). Polyunsaturated fat and fish oil in diets for growing-finishing pigs: effect on fatty acid composition and meat, and sausage quality. *Meat Science, 62*, 1-8.

Cera, K. R., Mahan, D. C., & Reinhart, G. A. (1990). Evaluation of various extracted vegetable oils, roasted soybeans, medium-chain triglyceride and an animal-vegetable fat blend for postweaning swine. *Journal of Animal Science*, *68,* 2756-2765.

Enser, M., Richardson, R. I., Wood, J. D., Gill, B. P., & Sheard, P. R. (2000). Feeding linseed to increase the n-3 PUFA of pork: fatty acid composition of muscle, adipose tissue, liver and sausages. *Meat Science, 55*, 201-212.

Hartman, A. D., Costello, W. J., Libal, G. W., & Walhlstrom, R. C. (1985). Effect of sunflower seeds on performance, carcass quality, fatty acids and acceptability of pork. *Journal of Animal Science, 60*, 212-219.

Honikel, K. O. (1987). How to measure the water-holding capacity of meat? Recommendation of standardized methods. In P. V. Tarrangt, G. Eikelen-boom, & G. Monin, *Evaluation and control of meat quality in pigs* (pp. 129-142). Dordrecht, the Netherlands: Martinus Nijhoff.

Horwitz, W. (1975). Official Methods of Analysis of Official Analytical Chemist. Benjamin Franklin Station: Association of Official Analytical Chemist, 12 ed.

Jorgensen, H., Jensen, S. K., & Eggum, B. O. (1996). The influence of rapeseed oil on digestibility, energy metabolism and tissue fatty acid composition in pigs. *Acta Agriculturae Scandinavica. Section A, Animal science, 43*, 65-75.

Leyton, J., Drury, P. J., & Crawford, M. A. (1987). Differential oxidation of saturated and unsaturated fatty acids in vivo in the rat. *The British Journal of Nutrition, 57*, 383-393.

Metcalfe, L. D., Schmitz, A. A., & Pelka, J. R. (1966). Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. *Analytical chemistry, 38*, 514-515.

Miller, M. F., Shackelford, S. D., Hayden, K. D., & Reagan, J. O. (1990). Determination of the alteration in fatty acid profiles, sensory characteristics and carcass traits of swine fed elevated levels of monounsaturated fats in the diet. *Journal of Animal Science, 68*, 1624-1631.

Monin, G., Larzul, C., Le Roy, P., Culioli, J., Mourot, J., Rousset-Akrim, S., Talmant, A., Touraille, C., & Sellier, P. (1999). Effects of the halothane genotype and slaughter weight on texture of pork. *Journal of Animal Science, 77*, 408-415.

Nakai, H., Saito, F., Ikeda, T., Ando, S., & Komatsu, A. (1975). Standard models of pork colour. *Bulletin of the National Institute of Animal Industry (Chiba, Japan, 29*, 69–75.

Nelson, G. J. (1975). Isolation and purification of lipids from animal tissues, In E. G. Perkins (ed), *Analysis of lipids and lipoproteins* (pp.1-22). USA: The Americal Oil Chemists Society, Champaign, Ill.

Nguyen, L. Q., Nuijens, M. C. G. A., Everts, H., Salden, N., & Beynen, A. C. (2003). Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. *Meat Science, 65*, 1399-1406

Nuernberg, K., Fischer, K., Nuernberg, G., Kuechenmeister, U., Klosowska, D., Eliminowska-Wenda, G., Fiedler, I., & Ender, K. (2005). Effects of dietary olive and linseed oil on lipid composition, meat quality, sensory characteristics and muscle structure in pigs. *Meat Science, 70*, 63-74.

Pfeiffer, H., & Falkenberg, H. (1972). Masse am Lendenspiegel zur objektiven Ermittlung der Schlachtkörper Zusammensetzung beim Schwein. *Tierzucht, 26*, 466-467.

Rhee, K.S., Davidson, T.L., Knabe, D.A., Cross, H.R., Ziprin, Y.A. & Rhee, K. (1988). Effect of dietary high-oleic sunflower oil on pork carcass traits and fatty acid profiles of raw tissues. *Meat Science, 24*, 249-260.

Scheeder, M., Gläser, K. R., Eichenberger, B., & Wenk, C. (2000). Influence of different fats in pig feed on fatty acids composition of phospholipids and physical meat quality characteristics. *European Journal of Lipid Science and Technology, 102*, 391-401.

Sheard, P. R., Enser, M., Wood, J. D., Nute, G. R., Gill, B. P., & Richardson, R. I. (2000). Shelf life and quality of pork and pork products with a raised n-3 PUFA. *Meat Science, 55*, 213-221.

Smith, D. R., Knabe, D. A., & Smith, S. B. (1996). Depression of lipogenesis in swine adipose tissue by specific dietary fatty acids. *Journal of Animal Science, 74*, 975-983.

SPSS. (1999). SPSS for Windows, SPSS Inc., Chicago, IL, USA.

Sripromma, J. (1984). Einfluss der Fütterungsintensität und der Körpermasse auf die Schlachtkörper Qualität beim Schwein. *Ph.D. Thesis, Hohemheim University, Stuttgard, Germany*.

Van Oeckel, M. J., Casteels, M., Warnants, N., Damme, L. V., & Boucque, C. V. (1996). Omega-3 fatty acids in pig nutrition: Implications for the intrinsic and sensory quality of the meat. *Meat Science, 44*, 55-63.

West, R .L., & Myer, R. O. (1987). Carcass and meat quality characteristics and backfat fatty acid composition of swine as affected by the consumption of

peanuts remaining in the field after harvest. *Journal of Animal Science, 65*, 475-480.

Wiseman, J., & Agunbiade, J. A. (1998). The influence of changes in dietary fat and oils on fatty acid profiles of carcass fat in finishing pigs. *Livestock Production Science, 54*, 217-227.

Wood, J. D., Richardson, R. I., Nute, G. R., Fisher, A. V., Campo, M. M., Kasapidou, E., Sheard, P. R., & Enser, M. (2003). Effects of fatty acids on meat quality: a review. *Meat Science, 66*, 21-32.

CHAPTER 3

Digestion and deposition of individual fatty acids in growing-finishing pigs fed diets containing either beef tallow or sunflower oil

J. Mitchaothai[a,*], H. Everts[c], C. Yuangklang[b], S. Wittayakun[b], K. Vasupen[b], S. Wongsuthavas[b], P. Srenanul[b], R. Hovenier[c] and A. C. Beynen[c]

[a]Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand
[b]Sakon Nakhon Agricultural Research and Training Center, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand
[c]Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands

Journal of Animal Physiology and Animal Nutrition, in press.

ABSTRACT

The apparent digestibility and deposition in carcass of individual dietary fatty acids were determined in growing-finishing pigs fed diets containing either beef tallow or sunflower oil. The beef tallow was rich in saturated fatty acids and the sunflower oil had a high content of polyunsaturated fatty acids. A total of 39 barrows was used; the mean initial body weight was 31 kg. The experimental diets contained 5 % (w/w) of the variable fat source and were fed *ad libitum*. The dietary fat type had no effect ($p > 0.05$) on growth performance, even though the apparent digestibilities of crude fat and crude protein were higher ($p < 0.05$) in the animals fed sunflower oil. The pigs fed the sunflower oil diet showed higher apparent digestibilities ($p < 0.05$) of the sum of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), but had a lower digestibility ($p < 0.05$) of stearic acid (C18:0). The intakes of individual digestible fatty acids were derived from feed intake data, fatty acid contents of the diets and the digestibility of individual fatty acids. For the entire feeding period of 13 weeks, the ratio of deposition in carcass to intake of digestible fatty acids was increased ($p < 0.05$) for palmitic acid (C16:0) and C18:0 in the pigs fed sunflower oil, but the ratios for oleic acid (C18:1 n-9) and linoleic acid (C18:2 n-6) were decreased ($p < 0.001$). In the pigs fed sunflower oil instead of beef tallow, the deposition:intake ratio was raised for the SFA ($p < 0.001$), but diminished for the MUFA ($p < 0.05$) . The calculated minimum *de novo* synthesis of SFA was increased ($p < 0.05$) and that of MUFA decreased ($p < 0.05$) in the pigs fed sunflower oil. It is concluded that the feeding of a diet with sunflower oil instead of beef tallow improved apparent digestibility of SFA, MUFA and PUFA, increased the deposition:digestible intake ratio for SFA, but lowered that for MUFA and PUFA.

Keywords: Pig; Dietary fat type; Digestibility; Fatty acid deposition; Fatty acid metabolism

INTRODUCTION

In relation to the development of cardiovascular disease in humans, there are concerns about the use of animal fats as feed ingredients for production animals, causing a shift towards the use of vegetable oils (Van Oeckel et al., 1996). Thus, the effects on production animals of dietary inclusion of vegetable oils, instead of animal fat, are of interest. In general, pigs digest vegetable oils rich in unsaturated fatty acids more efficiently than they do animal fats rich in saturated fatty acids (Cera et al., 1988, , 1990; Jorgensen et al., 1996). The fatty acid composition of subcutaneous and intramuscular fat of pigs reflects the fatty composition of the diet (Nguyen et al., 2003). In particular, the percentage in the dietary fat of linoleic acid (C18:2 n-6) and alpha-linolenic acid (C18:3 n-3) is directly related to the percentage of these fatty acids in the adipose tissue (Nguyen et al., 2003). This direct relationship may relate to the fact that linoleic and alpha-linolenic acid are essential fatty acids that cannot be synthesized in the body of pigs. Thus, the production of swine meat rich in linoleic acid can be achieved by feeding a diet rich in this polyunsaturated fatty acid.

In the body there is metabolism of fatty acids. Upon digestion, dietary fatty acids may be oxidized or deposited in tissues. Except for the two essential polyunsaturated fatty acids, there is *de novo* synthesis of fatty acids from acetyl-CoA derived from carbohydrate and/or protein breakdown (Acheson et al., 1988). The complex metabolism of fatty acids implies that the deposition in the carcass of individual dietary fatty acids may not be directly related to their intake. The aim of this study was to gain insight into the interactions between metabolism and intake of individual fatty acids in growing-finishing swine. As a measure, we used for individual fatty acids the ratio of deposition in carcass to the amount digested. It was hypothesized that the ratios would be influenced by the type of fat in the diet. In the light of the increased interest in the use of vegetable oils for manufacturing animal feeds we compared the feeding of beef tallow with that of sunflower oil in growing-finishing pigs.

MATERIALS AND METHODS

Animals, diets and feeding

Thirty-nine castrated, male pigs (Landrace × Large White × Duroc crossbred) with average body weight of 31 kg were used (Mitchaothai et al., 2007). The study was conducted at the Sakon Nakhon Agricultural Research and Training Center, Rajamangala University of Technology-Isan, Thailand. Three pigs with an average body weight of 31 kg were selected to be killed for baseline measurements. Then, the remaining pigs were allotted to one of the two dietary treatments on the basis of body weight and were housed in 12 pens with 3 pigs each (6 pens per treatment). The pigs were allowed *ad libitum* access to feed and water throughout the experiment. Feed consumption was recorded twice a day. There were two experimental diets containing either beef tallow (Montee slaughter house, Phangkhon, Sakon Nakhon, Thailand) or sunflower oil (Aro®, Siam Makro Ltd., Bangkapi, Bangkok, Thailand) (Table 1). The other ingredients were purchased locally. To determine macronutrient digestibility, the pigs were kept individually in metabolism cages during weeks 4 and 10 after the start of the experiment. The pigs were weighed weekly until the end of the experiment. All pigs were slaughtered at an average body weight of 96 kg in a commercial slaughterhouse.

Table 1. Ingredient and analysed composition of the diets on as-fed basis

	Beef tallow diet	Sunflower oil diet
Raw materials (%)		
Cassava chips	45.53	45.53
Soybean meal (44% CP)	34.00	34.00
Extruded soy beans	7.00	7.00
Beef tallow	5.00	-
Sunflower oil	-	5.00
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Di-calcium phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Premix*	0.25	0.25
Analysed macronutrients (%)		
Dry matter	86.5	86.3
Crude protein	19.3	19.2
Crude fat	6.8	6.7
Dietary fiber	4.7	4.9
Ash	7.7	8.2

*1 kilogram of vitamin and mineral premix contained: 325,000 IU vitamin A; 75,000 IU vitamin D_3; 75 IU vitamin E; 1 mg vitamin B_{12}; 80 mg vitamin K_3; 300 mg riboflavin; 1,200 mg niacinamide; 540 mg pantothenic acid; 6,000 mg choline chloride; 4,700 mg Fe; 6,500 mg Zn; 4,500 mg Mn; 20 mg Co; 1,400 mg Cu and 45 mg I.

Collection and pre-treatment of samples

The three pigs for baseline measurements were killed by an intravenous overdose of sodium pentobarbital (200 mg/kg of body weight). Pigs were scalded, the head was removed, and the carcass was split into halves along the median plane. Both sides of the pig carcass, head, visceral organs (minus gastrointestinal contents), blood and perirenal fat were weighed, wrapped in a polyethylene bag to prevent evaporative moisture losses, and then frozen at –20 ^{o}C for at least 1 week before analysis. Upon removal from the freezer, each sample was weighed and sawed in cross-sectional slices (2.54 cm) along its length (Shields et al., 1983). The sawed carcass samples were dried at 60 ^{o}C for 1 week in a forced-hot air oven and were then ground once through a 0.2-cm plate and twice through a 0.1-cm plate by a hammer mill.

The 36 pigs in the feeding trial were sacrificed at 13 weeks after the start. Carcass measurements of loin eye area (LEA,cm^2) and back fat depth at the 10th rib (FD10R) were done as described previously (Wagner et al., 1999). Samples of back fat at midline between the 3rd and 4th rib, right inguinal subcutaneous fat, retroperitoneal fat, loin, liver, and erythrocytes were collected to determine fatty acid composition.

Chemical analyses

The diet and faeces samples were dried at 60 ^{o}C for 72 h in a forced-hot air oven and were then analysed for crude protein, crude fiber, and ash (AOAC, 1990). The dried meat, liver, and whole carcass samples were analysed for moisture and fat (AOAC, 1990). The fat extracted from meat was defined as intramuscular fat (IMF).

Total fat in the fresh and dried samples (diets, faeces, adipose tissues, loin, and liver) were extracted as described previously (Horwitz, 1975). Each sample was added to a flask and 2 ml of ethanol was added. Subsequently, 10 ml of HCl (8 mol/L) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80 ^{o}C for 30-40 min. The tubes were cooled down, 10 ml of ethanol (96%, w/w) and 25 ml of petroleum ether (boiling point between 40 and 60 ^{o}C) were added and the tube was shaken for one min. The fat-containing upper layer was decanted into a 150-ml round-bottom flask. The extraction procedure was repeated twice with 15 ml of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath of 40 ^{o}C. The round-bottom flasks with the lipids were dried overnight at 60 ^{o}C and the total lipids were measured gravimetrically. Total lipids were saponified and methylated according to the procedure of Metcalfe et al. (1966) followed by gas chromatography for determination of fatty acid composition (Javadi et al., 2004).

Estimation of carcass fat content

The total lean mass and the total fat mass in the whole body were estimated by the using the following equations (Schinckel et al., 2001). Fat-free, total lean mass (kg) = 5.00 + [0.434 × carcass weight (kg)] + [0.168 × LEA (cm^2)] + [(-3.38) × FD10R (cm)]. Total fat mass (kg) = (-10.7) + [0.395 × carcass weight (kg)] + [(-0.150) × LEA (cm^2)] + [4.49 × FD10R (cm)]. Thus, intramuscular fat mass = fat-free, lean mass (kg) × [fraction IMF × (100 - fraction IMF)$^{-1}$]. Based on literature data (Enser et al., 2000; Irie and Sakimoto, 1992; Otten et al., 1993), it can be concluded that the fatty acid profiles of adipose tissue from different sites do not differ much. Thus, the average percentage of fatty acids from different sites of adipose tissue can be considered to be representative for the whole body mass of adipose tissue. The amount of fat in blood is negligible because dried blood meal only contains 1.6% fat (NRC, 1998) and therefore was not taken into account. Thus, the mass (kg) of adipose tissue equals total fat mass (kg) in whole body – fat mass (kg) in muscle (IMF) – fat mass (kg) in liver.

Calculation of digestible fatty acid intake, fatty acid deposition and minimum de novo synthesis

The total digestible fatty acid intake was calculated as fatty acid intake (kg/13 weeks) × average apparent fatty acid digestibility (fraction of intake). The deposition of fatty acids was calculated with the following formula. Fatty acid deposition (kg/13 weeks) = carcass content of fatty acid at the end of the study – carcass content of fatty acid at the start of the study. Carcass content of fatty acid at the end (kg) was calculated as the sum of adipose tissue mass (kg) × fraction of adipose fatty acid, IMF mass (kg) × fraction of IMF fatty acid and fat mass in liver (kg) × fraction of liver fatty acid. The ratio of fatty acid deposition and digestible fatty acid intake was calculated for selected individual fatty acids and groups of fatty acids. Absorbed fatty acids can be deposited in the body, oxidized to generate energy or transformed into other fatty acids. In addition, various fatty acids can be synthesized *de novo* in the body. In this study, it was not possible to quantify fatty acid oxidation, transformation and synthesis, but fatty acid absorption and deposition could be determined. By subtracting the intake of digestible fatty acids from the amount deposited, an estimate of minimum *de novo* synthesis was obtained.

Statistical analyses

The effect of dietary fat type was evaluated for statistical significance by the Student's *t* test (SPSS, 1999). The results are expressed as means ± SD. There were 6 experimental units per treatment for the measurement of growth performance, digestible fatty acid intake, fatty deposition, the ratio of digestible intake:deposition, and minimum *de novo* synthesis. All experimental pigs were kept individually in metabolism cages during collecting period for macronutrient digestibility determination, therefore there were 18 experimental units used for analysis of macronutrient and fatty acid digestibility according to the following model:

$$Y_{ijk} = \mu + D_i + P_j + e_{ijk}$$

where μ is the overall mean; D_i is dietary treatment; P_j is period of the sample collection; and e_{ijk} is the error term.

RESULTS

Dietary fatty acid composition

The fatty composition of the experimental diets is shown in Table 2. The fat component of the beef tallow diet contained approximately 53 % total saturated fatty acids (SFA), whereas that of the sunflower oil diet contained approximately 51 % total polyunsaturated fatty acids (PUFA).

Table 2. Analysed fatty acid composition of the experimental diets

	Beef tallow diet	Sunflower oil diet
Fatty acid, g methylester/100 g methylesters		
C 14:0 (Myristic acid)	1.48	0.05
C 15:0 (Pentadecanoic acid)	0.51	0.06
C 15:1 (Pentadecenoic acid)	0.37	0.37
C 16:0 (Palmitic acid)	19.23	9.26
C 16:1 (Palmitoleic acid)	0.68	0.04
C 17:0 (Margaric acid)	1.59	0.05
C 17:1 (Heptadecenoic acid)	0.28	0.00
C 18:0 (Stearic acid)	29.48	4.34
C 18:1 n-9 (Oleic acid)	21.91	31.52
C 18:1 n-7 (Vaccenic acid)	4.71	1.11
C 18:2 n-6 (Linoleic acid)	11.65	49.07
C 18:3 n-6 (γ-linolenic acid)	0.32	0.00
C 18:3 n-3 (α-linolenic acid)	1.63	1.42
C 20:0 (Arachidic acid)	0.44	0.34
C 20:1 n-9 (Eicosenoic acid)	0.48	0.39
C 20:5 n-3 (Eicosapentaenoic acid)	0.05	0.05
C 22:0 (Behenic acid)	0.02	0.77
C 22:1 n-9 (Erucic acid)	0.00	0.11
C 24:0 (Lignoceric acid)	0.10	0.33
Unidentified fatty acids	4.75	0.57
Σ Saturated fatty acids (SFA)	52.92	15.27
Σ Monounsaturated fatty acids (MUFA)	28.49	33.60
Σ Polyunsaturated fatty acids (PUFA)	13.65	50.54

Σ SFA = C8:0 + C10:0 + C14:0 + C15:0 + C16:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C14:1 + C15:1 + C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:3 n-6 + C18:4 n-3 + C20:5 n-3

Growth performance and nutrient digestibility

The initial body weight of the pigs to be fed the diets containing either beef tallow or sunflower oil was 30.9 ± 3.4 kg and 31.1 ± 3.1 kg, respectively. The final body weight of pigs fed beef tallow diet or sunflower oil was 95.6 ± 6.1 kg and 96.5 ± 8.4 kg, respectively. The average daily gain (ADG) and the feed:gain ratio for the pigs given the diet with beef tallow diet or sunflower oil diet were 0.790 ± 0.06 kg/day and 0.809 ± 0.07 kg/day, respectively. There was no diet effect ($p > 0.05$) on feed intake, growth (ADG), feed conversion ratio (feed:gain) and final body weight.

The pigs fed the sunflower oil diet had significantly higher apparent digestibilities of dry matter (DM) ($p < 0.01$), organic matter (OM) ($p < 0.01$), crude protein ($p < 0.05$) and crude fat ($p < 0.001$) than those fed the beef tallow diet (Table 3) with an effect of time ($p < 0.05$) on crude fat digestibility.

Table 4 shows the apparent digestibility values for selected individual fatty acids and groups of fatty acids. The digestibility was calculated only for fatty acids with a dietary content higher than 0.04 g/100 g of the total fat so that the impact of faecal excretion of endogenous fatty acids would not be overwhelming. The palmitic acid (C16:0) digestibility for the sunflower oil diet was greater ($p < 0.001$) than that for the beef tallow diet, but for the sunflower oil diet the stearic acid (C18:0) digestibility was lower ($p < 0.001$). The pigs fed the sunflower oil diet displayed a significantly greater digestibility ($p < 0.001$) of oleic acid (C18:1 n-9) than those fed the beef tallow diet. The apparent digestibilities of linoleic acid (C18:2 n-6) and alpha-linolenic acid (C18:3 n-3) were greater ($p < 0.001$) for the sunflower oil diet with an effect of time ($p < 0.001$) (Table 4). The digestibilities of SFA, monounsaturated fatty acids (MUFA), and PUFA for the sunflower oil diet were significantly higher ($p < 0.001$) than those for the beef tallow diet.

Table 3. Effect of dietary fat type on apparent macronutrient digestibility.

	Beef tallow diet	Sunflower oil diet	Time	Diet
Apparent digestibility, % of intakes				
Dry matter	89.7 ± 2.2	91.3 ± 2.4	0.504	0.003
Organic matter	91.3 ± 1.8	92.8 ± 2.1	0.270	0.003
Crude fat	76.7 ± 6.5	82.3 ± 5.7	0.047	0.000
Crude protein	89.0 ± 2.5	90.6 ± 3.6	0.056	0.036

Means ± SD for 18 pigs per experimental diet.

Table 4. Effect of dietary fat type on apparent fatty acid digestibility

	Beef tallow diet	Sunflower oil diet	Time	Diet
Apparent digestibility, % of intakes				
C 16:0 (Palmitic acid)	75.2 ± 6.2	85.6 ± 4.7	<0.001	<0.001
C 18:0 (Stearic acid)	58.7 ± 11.1	39.8 ± 16.0	0.190	<0.001
C 18:1 n-9 (Oleic acid)	94.7 ± 1.4	96.3 ± 1.7	<0.001	<0.001
C 18:2 n-6 (Linoleic acid)	94.6 ± 1.7	96.3 ± 2.4	<0.001	0.001
C 18:3 n-3 (α-linolenic acid)	94.8 ± 2.4	97.0 ± 2.1	<0.001	<0.001
Σ Saturated fatty acids (SFA)	71.0 ± 7.7	77.5 ± 6.5	<0.001	<0.001
Σ Monounsaturated fatty acids (MUFA)	91.7 ± 3.3	97.5 ± 1.9	<0.001	<0.001
Σ Polyunsaturated fatty acids (PUFA)	94.7 ± 2.0	96.6 ± 2.2	<0.001	<0.001

Means ± SD for 18 pigs per experimental diet.

Σ SFA = C8:0 + C10:0 + C14:0 + C15:0 + C16:0 + C18:0 + C20:0 + C22:0 + C24:0

Σ MUFA = C15:1 + C16:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9

Σ PUFA = C18:2 n-6 + C18:3 n-3

Digestible fatty acid intake and deposition

The intake of digestible palmitic and stearic acid of the pigs fed the beef tallow diet was greater ($p < 0.001$) than that of the pigs fed the sunflower oil diet, whereas the pigs fed the sunflower oil diet had a higher ($p < 0.001$) intake of digestible oleic and linoleic acid (Table 5). There was no difference ($p > 0.05$) for the intake of digestible alpha-linolenic acid. The pigs fed the sunflower oil diet had a greater intake of total digestible fatty acids (FA) ($p < 0.05$), MUFA ($p < 0.05$) and PUFA ($p < 0.001$), but had a lower intake ($p < 0.001$) of digestible SFA. When the fatty acids were pooled according to their structural similarities, the sunflower oil diet was found to provide more digestible n-9 MUFA and n-6 PUFA ($p < 0.001$), whereas the amount of digestible n-3 PUFA was similar to ($p > 0.05$) that provided by the beef tallow diet.

The analysed whole body of the pigs at baseline contained an average total fat mass of 4.05 kg. The calculated total fat mass in the carcass at the end of the experiment was 22.82 ± 4.02 kg and 23.42 ± 4.15 kg, n = 18 ($p = 0.696$) for the pigs fed the beef tallow and sunflower oil diet, respectively. The pigs fed beef tallow diet had deposited more ($p < 0.05$) stearic acid, but less ($p < 0.001$) linoleic acid when compared with the pigs fed the sunflower oil diet (Table 5). There was no difference ($p > 0.05$) between both diets in the deposition of palmitic acid, oleic acid and alpha-linolenic acid. The pigs fed the sunflower oil diet did deposit more PUFA ($p < 0.001$) than those fed beef tallow diet, but no significant difference ($p > 0.05$) was seen for the deposition of total FA, SFA and MUFA. There was more deposition ($p < 0.001$) of n-6 PUFA in the pigs fed the sunflower oil diet, but that of the n-9 MUFA and n-3 PUFA was similar ($p > 0.05$) as in the pigs fed the beef tallow diet.

The deposition:intake ratios for palmitic and stearic acid in the pigs fed the sunflower oil diet were greater ($p < 0.01$) than those in their counterparts fed the beef tallow diet. The ratio for stearic acid was extremely high for the pigs fed the diet containing sunflower oil (Table 5). However, the pigs fed sunflower oil had a lower deposition:intake ratio ($p < 0.001$) for both oleic and linoleic acid. There was no diet effect ($p > 0.05$) on the deposition:intake ratio for alpha-linolenic acid, the ratio for the two groups of pigs being less than one. There were greater ratios ($p < 0.01$) for total FA, MUFA, PUFA, n-6 PUFA and n-9 MUFA in pigs fed the beef tallow diet, but the ratio of SFA was higher ($p < 0.01$) in pigs fed the sunflower oil diet.

Minimum de novo synthesis

Based on the calculation of minimum *de novo* synthesis, the pigs fed the diet containing sunflower oil had a greater synthesis ($p < 0.05$) of SFA, but lower synthesis ($p < 0.05$) of MUFA when compared with their counterparts fed beef tallow (Table 6).

DISCUSSION

The main fatty acids in the diet containing beef tallow diet were stearic and palmitic acid, whereas the sunflower oil diet was very rich in linoleic acid, corroborating our previous study (Mitchaothai et al., 2007). The amounts of oleic acid in the two diets was not much different. Thus, the beef tallow diet is representative for diets rich in saturated fatty acids, whereas the sunflower oil diet may represent diets rich in n-6 polyunsaturated fatty acids. This could imply that the two dietary treatments are a reference for other diets with similar fatty acid compositions.

Table 5. Effect of dietary fat type on digestible fatty acid intake, fatty acid deposition and the deposition:intake ratio during the whole feeding period

Fatty acid	Beef tallow diet	Sunflower oil diet	*p* value
Digestible fatty acid intake (kg/13 weeks)			
C 16:0 (Palmitic acid)	2.24 ± 0.21	1.24 ± 0.16	<0.001
C 18:0 (Stearic acid)	2.69 ± 0.37	0.21 ± 0.05	<0.001
C 18:1 n-9 (Oleic acid)	3.22 ± 0.28	4.59 ± 0.62	<0.001
C 18:2 n-6 (Linoleic acid)	1.76 ± 0.16	7.14 ± 0.98	<0.001
C 18:3 n-3 (α-linolenic acid)	0.24 ± 0.02	0.21 ± 0.03	0.059
Σ Fatty acids (FA)	11.93 ± 1.20	14.47 ± 1.98	0.023
Σ Saturated fatty acids (SFA)	5.26 ± 0.61	1.70 ± 0.21	<0.001
Σ Monounsaturated fatty acids (MUFA)	3.91 ± 0.36	4.65 ± 0.63	0.031
Σ Polyunsaturated fatty acids (PUFA)	2.00 ± 0.18	7.35 ± 1.00	<0.001
Σ n-9 Monounsaturated fatty acids	3.29 ± 0.29	4.64 ± 0.63	<0.001
Σ n-6 Polyunsaturated fatty acids	1.76 ± 0.16	7.14 ± 0.98	<0.001
Σ n-3 Polyunsaturated fatty acids	0.24 ± 0.02	0.21 ± 0.03	0.059
Total fat content at the start (kg)	4.05 ± 0.80	4.05 ± 0.80	-
Total fat content at the end (kg)	22.83 ± 4.02	23.42 ± 4.15	0.696
Fatty acid deposition (kg/13 weeks)			
C 16:0 (Palmitic acid)	4.55 ± 0.82	3.75 ± 0.73	0.106
C 18:0 (Stearic acid)	3.08 ± 0.55	2.39 ± 0.48	0.045
C 18:1 n-9 (Oleic acid)	6.63 ± 1.32	5.87 ± 0.98	0.284
C 18:2 n-6 (Linoleic acid)	1.81 ± 0.28	4.69 ± 0.86	<0.001
C 18:3 n-3 (α-linolenic acid)	0.19 ± 0.03	0.17 ± 0.03	0.195
Σ Fatty acids (FA)	18.53 ± 3.21	18.24 ± 3.09	0.877
Σ Saturated fatty acids (SFA)	7.97 ± 1.43	6.39 ± 1.26	0.070
Σ Monounsaturated fatty acids (MUFA)	7.71 ± 1.53	6.51 ± 1.06	0.144
Σ Polyunsaturated fatty acids (PUFA)	2.10 ± 0.33	5.11 ± 0.94	<0.001
Σ n-9 Monounsaturated fatty acids	6.79 ± 1.35	6.01 ± 0.99	0.284
Σ n-6 Polyunsaturated fatty acids	1.93 ± 0.30	4.96 ± 0.90	<0.001
Σ n-3 Polyunsaturated fatty acids	0.18 ± 0.03	0.19 ± 0.04	0.173
Deposition : intake ratio			
C 16:0 (Palmitic acid)	2.03 ± 0.35	3.06 ± 0.60	0.005
C 18:0 (Stearic acid)	1.16 ± 0.25	11.90 ± 3.96	<0.001
C 18:1 n-9 (Oleic acid)	2.05 ± 0.32	1.29 ± 0.18	<0.001
C 18:2 n-6 (Linoleic acid)	1.03 ± 0.16	0.69 ± 0.09	<0.001
C 18:3 n-3 (α-linolenic acid)	0.80 ± 0.10	0.79 ± 0.14	0.972
Σ Fatty acids (FA)	1.55 ± 0.23	1.27 ± 0.19	0.041
Σ Saturated fatty acids (SFA)	1.53 ± 0.29	3.80 ± 0.78	<0.001
Σ Monounsaturated fatty acids (MUFA)	1.97 ± 0.30	1.41 ± 0.20	0.003
Σ Polyunsaturated fatty acids (PUFA)	1.05 ± 0.16	0.69 ± 0.09	<0.001
Σ n-9 Monounsaturated fatty acids	2.06 ± 0.32	1.30 ± 0.18	<0.001
Σ n-6 Polyunsaturated fatty acids	1.10 ± 0.17	0.70 ± 0.09	<0.001
Σ n-3 Polyunsaturated fatty acids	0.74 ± 0.08	0.71 ± 0.16	0.685

Means ± SD for 6 experimental units per diet; each experimental unit consisted of a pen with three pigs each. The fatty acid content of the pigs at the start as follows (kg): C16:0 (0.86), C18:0 (0.33), C18:1 n-9 (1.56), C18:2 n-6 (0.56), C18:3 n-3 (0.05), FA (3.85), SFA (1.26), MUFA (1.26), PUFA (0.66), total n-9 (1.58), total n-6 (0.59), and total n-3 (0.07).

Σ FA = Σ SFA + Σ MUFA + Σ PUFA

Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0

Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9

Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:3 n-6 + C20:5n-3

Σ n-9 = C20:1 n-9 + C22:1 n-9

Σ n-6 = C18:2 n-6 + C18:3 n-6

Σ n-3 = C18:3 n-3 + C20:5n-3

Table 6. Effect of dietary fat type on minimum *de novo* synthesis of fatty acids during the whole feeding period

Fatty acid	Beef tallow diet	Sunflower oil diet	*p* value
Minimum synthesis (kg/13 weeks)			
Saturated fatty acids (SFA)	2.71 ± 1.44	4.70 ± 1.22	0.028
Monounsaturated fatty acids (MUFA)	3.81 ± 1.32	1.86 ± 0.92	0.014

Means ± SD for 6 experimental units per diet; each experimental unit consisted of a pen with three pigs each.
Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9

The pigs fed either the beef tallow or sunflower oil diet showed no difference in growth performance. This observation agrees with previous investigations (Cera et al., 1990; Smith et al., 1996; Wiseman and Agunbiade, 1998). Thus, the vegetable sunflower oil can be substituted for the animal fat, beef tallow, without any adverse effect on growth performance. However, the crude fat and protein components of the sunflower oil diet had a higher apparent digestibility than had these components of the beef tallow diet. The higher digestibility of oils rich in linoleic acid, as opposed to fats rich in saturated fatty acids, is well known (Cera et al., 1989; Li et al., 1990). Lessire and Leclereq (1982) found in poultry that fat digestibility was increased with higher degrees of unsaturation in the C18 fatty acids. This could be the result of more micelles with unsaturated instead of saturated fatty acids (Stahly, 1984). More micelle formation could explain the higher fat digestibility in the pigs fed the diet with sunflower oil. It is difficult to see why dietary sunflower oil versus beef tallow would raise apparent protein digestibility, but similar observations have been reported earlier (Li et al., 1990). The increase in apparent digestibility of fat and protein in pigs fed the diet containing sunflower oil diet explains the observed higher digestibilities of dry matter and organic matter. The higher digestibility of the sunflower oil diet is in accordance with the observed tendencies towards, higher group mean final body weight and lower feed:gain ratio in the pigs fed sunflower oil. The differences did not reach statistical significance, which may relate to low statistical power.

The fat type had marked effects on the apparent digestibility of individual fatty acids. The palmitic, oleic, linoleic and alpha-linolenic acids in sunflower oil were digested more efficiently than these same fatty acids, but present in beef tallow. On the other hand, the stearic acid in sunflower oil was less well digested than that in beef tallow. A combination of different factors may be responsible for the observed diet-induced differences in apparent digestibility of identical fatty acids. As mentioned above, the total digestibility of sunflower oil was greater than that of beef tallow, which may relate to enhanced micelle formation after feeding sunflower oil. An improved micelle formation may favourably influence the digestion of all fatty acids in the diet. The position of a given fatty acid in the triacylglycerol molecule also plays a role. Fatty acids at the 2 position of glycerol in triacylglycerol molecules are better digested than those at the 1,3 position (Bracco, 1994; Innis and Dyer, 1997; Nelson and Innis, 1999). During digestion, the pancreatic lipase action specifically removes fatty acids at the 1,3 position while the resulting monoacylglycerol molecule is efficiently incorporated into micelles (Lien, 1994), leading to preferential absorption of fatty acids at the 2 position of the glycerol backbone of triacylglycerols. The intake level of a given individual fatty acid and its faecal excretion of endogenous origin will also affect the calculated apparent digestibility. A low intake level in combination

with a high endogenous excretion will by itself lead to a low apparent digestibility. When comparing the digestibilities of palmitic and stearic acid for the sunflower oil and beef tallow diet, the values for the sunflower oil diet may biased to lower values because their intake levels were lower on the sunflower oil diet. On the other hand, the apparent digestibility for linoleic acid on the sunflower oil diet may be biased towards a higher value.

The calculated intake of digestible fatty acids reflects the amount in the diet and feed intake, combined with the measured apparent digestibility. The deposition in the body of fatty acids was calculated as based on the fatty acid composition of the total body fat that was gained during the entire feeding period. As would be expected, the pigs fed the sunflower oil diet deposited more linoleic acid and those fed beef tallow had deposited more stearic acid in their whole body. Similar data have been shown for mice (Javadi et al., 2004) and goats (Yeom et al., 2005). The increased deposition of palmitic acid in the pigs fed the beef tallow diet did not reach statistical significance.

To obtain clues as to preferential deposition of fatty acids, the ratio of deposition of a given fatty acid or group of fatty acids to the intake of digestible fatty acid or group of fatty acids was calculated. The deposition:intake ratio for fatty acid reflects the net amount synthesized and/or the proportion of the digested fatty acid not oxidized. A deposition:intake ratio > 1 would point at net *de novo* synthesis, whereas a ratio < 1 would indicate net oxidation. The low deposition:intake ratio for linoleic acid in the pigs fed sunflower oil is consistent with the well-known preferential oxidation of linoleic acid (Cunnane and Anderson, 1997; Jones et al., 1985; Yeom et al., 2005) and the fact that linoleic acid cannot be synthesized by pigs (Azain, 2000; Nguyen et al., 2005). The deposition:intake ratio for the essential polyunsaturated fatty acids, linoleic and alpha-linolenic acid, cannot be higher than 1. Indeed, the ratios for alpha-linolenic acid in both groups and the ratio for linoleic acid in the pigs fed the sunflower oil diet were below 1. The pigs fed beef tallow had a group mean deposition:intake ratio for linoleic acid that was just above 1, but was not significantly higher than 1. The extremely high deposition:intake ratio for stearic acid in pigs fed the sunflower oil diet is explained by a relatively low intake and high net synthesis of this fatty acid.

The pigs fed sunflower oil instead of beef tallow had a higher deposition:intake ratio for SFA, but lower ratios for MUFA and PUFA. The diet effect on the ratios for SFA and MUFA can be explained by the calculated minimum synthesis of these groups of fatty acids. Feeding the diet containing sunflower oil stimulated the synthesis of SFA, but depressed that of MUFA. This might point at *de novo* fatty acid synthesis being regulated to balance the amount of saturated and unsaturated fatty acids in the body because the intake of linoleic acid was very high in the pigs fed the diet with sunflower oil.

In conclusion, the inclusion of sunflower oil instead of beef tallow in the diet for growing-finishing pigs had no negative effect growth performance, but rather improved the digestibility of crude fat and protein. Feeding the diet with sunflower oil produced a markedly increased deposition of linoleic acid in the whole body. For groups of fatty acids, the ratio of deposition in the whole body to the intake of digestible fatty acids was calculated. It then became clear that the type of dietary fat had marked, specific effects on the synthesis and oxidation of fatty acids. Thus, not only the composition of dietary fat, but also synthesis and oxidation, determine the fatty acid pattern of deposited tissues in a concerted way.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the excellent technical assistance of Suksan Pimpilai, Adisak Aiumla-or, Nimit Kaewmalun, Narongsak Nantarad, and Thanaphum Bunmee. We also thank the Sakon Nakhon Agricultural Research and Training Center, Rajamangala University of Technology-Isan for use of its facilities supports and the Mahanakorn University of Technology for financial support.

LITERATURE CITED

Acheson, K.J.; Schutz, Y.; Bessard, T.; Anantharaman, K., Flatt, J. P.; Jequier, E., 1988: Glycogen storage capacity and de novo lipogenesis during massive carbohydrate overfeeding in man. *American Journal of Clinical Nutrition* **48**, 240-247.

AOAC, 1990: Association of official analysis chemists, Washington DC.

Azain, M. J., 2000: *Fat in swine nutrition*, *In:* Swine nutrition, 2edn, CRC press, New York, USA.

Bracco, U., 1994: Effect of triglyceride structure on fat absorption. *American Journal of Clinical Nutrition* **60**, 1002S-1009S.

Cera, K. R.; Mahan; D. C.; Reinhart, G. A., 1988: Weekly digestibilities of diets supplemented with corn oil, lard or tallow by weanling swine. *Journal of Animal Science* **66**, 1430-1437.

Cera, K. R.; Mahan, D. C.; Reinhart, G. A., 1989: Apparent fat digestibilities and performance responses of postweaning swine fed diets supplemented with coconut oil, corn oil or tallow. *Journal of Animal Science* **67**, 2040.

Cera, K. R.; Mahan, D. C.; Reinhart, G. A., 1990: Evaluation of various extracted vegetable oils, roasted soybeans, medium-chain triglyceride and an animal-vegetable fat blend for postweaning swine. *Journal of Animal Science* **68**, 2756-2765.

Cunnane, S. C.; Anderson, M. J., 1997: The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. *Journal of Nutrition* **127**, 146-152.

Enser, M.; Richardson, R. I.; Wood, J. D.; Gill, B. P.; Sheard, P. R., 2000. Feeding linseed to increase the n-3 PUFA of pork: fatty acid composition of muscle, adipose tissue, liver and sausages. *Meat Science* **55**, 201-212.

Horwitz, W., 1975: *Official Methods of Analysis of Official Analytical Chemist*. *In*: Official Methods of Analysis of Official Analytical Chemist, Benjamin Franklin Station, USA

Innis, S. M.; Dyer, R., 1997: Dietary triacylglycerols with palmitic acid (16:0) in the 2-position increase 16:0 in the 2-position of plasma and chylomicron triacylglycerols, but reduce phospholipid arachidonic and docosahexaenoic acids, and alter cholesteryl ester metabolism in formula-Fed piglets. *Journal of Nutrition* **127**, 1311-1319.

Irie, M.; Sakimoto, M., 1992: Fat characteristics of pigs fed fish oil containing eicosapentaenoic and docosahexaenoic acids. *Journal of Animal Science* **70**, 470-477.

Javadi, M.; Everts, H.; Hovenier, R.; Kocsis, S.; Lankhorst, A. E.; Lemmens, A. G.; Schonewille, J. T.; Terpstra, A. H.; Beynen, A. C., 2004: The effect of six different C18 fatty acids on body fat and energy metabolism in mice. *British Journal of Nutrition* **92**, 391-399.

Jones, P. J. H.; Pencharz, P. B.; Clandinin, M. T., 1985: Whole body oxidation of dietary fatty acids: implications for energy utilization. *American journal of clinical nutrition* **42**, 769-777.

Jorgensen, H.; Jensen, S. K.; Eggum, B. O., 1996: The influence of rapeseed oil on digestibility, energy metabolism and tissue fatty acid composition in pigs. *Acta Agriculturae Scandinavica Section A-Animal Science* **43**, 65-75.

Lessire, M.; Leclereq B., 1982: Metabolisable energy value of fats in chicks and adult cockerels. *Animal Feed Science and Technology* **7**, 365-374

Li, D. F.; Thaler, R. C.; Nelssen, J. L.; Harmon, D. L.; Allee, G. L.; Weeden, T. L., 1990: Effect of fat sources and combinations on starter pig performance, nutrient digestibility and intestinal morphology. *Journal of Animal Science* **68**, 3694-3704.

Lien, E. L., 1994: The role of fatty acid composition and positional distribution in fat absorption in infants. *Journal of Pediatric* **125**, S62-S68.

Metcalfe, L. D.; Schmitz, A. A.; Pelka, J. R., 1966. Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. *Analytical Chemistry.* **38**, 514-515.

Mitchaothai, J.; Yuangklang, Y.; Wittayakun, S.; Vasupen, K.; Wongsutthavas, S.; Srenanul, P.; Hovenier, R.; Everts, H.; Beynen, A. C., 2007: Effect of dietary fat type on meat quality and fatty acid composition of various tissues in growing-finishing swine. *Meat Science* **76**, 95-101

Van Oeckel, M. J.; Casteels, M.; Warnants, N.; Van Damme, L.; Boucque, C. V., 1996: Omega-3 fatty acids in pig nutrition: Implications for the intrinsic and sensory quality of the meat. *Meat Science* **44**, 55-63.

Nelson, C. M.; Innis, S. M., 1999: Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. *American Journal of Clinical Nutrition* **70**, 62-69.

Nguyen, L. Q.; Everts, H.; Beynen, A. C., 2005: Intake of essential fatty acids by growing-finishing pigs kept on smallholdings in central Vietnam. *Tropical Animal Health and Production* **37**, 65-76.

Nguyen, L. Q.; Nuijens, M. C. G. A.; Everts, H.; Salden, N.; Beynen, A. C., 2003: Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. *Meat Science* **65**, 1399-1406.

NRC, 1998: *Nutrient requirements of swine,* 10edn, National Academy Press, Washington, D. C., USA.

Otten, W.; Wirth, C.; Iaizzo, P. A.; Eichinger, H. M., 1993: A high omega 3 fatty acid diet alters fatty acid composition of heart, liver, kidney, adipose tissue and skeletal muscle in swine. *Annuals of Nutrition and Metabolism* **37**, 134-141.

Schinckel, A. P.; Wagner, J. R.; Forrest, J. C.; Einstein, M. E., 2001: Evaluation of alternative measures of pork carcass composition. *Journal of Animal Science* **79**, 1093-1119.

Shields, R. G., Jr.; Mahan, D. C.; Graham, P. L., 1983: Changes in swine body composition from birth to 145 kg. *Journal of Animal Science* **57**, 43-54.

Smith, D. R.; Knabe, D. A.; Smith, S. B., 1996: Depression of lipogenesis in swine adipose tissue by specific dietary fatty acids. *Journal of Animal Science* **74**, 975-83.

SPSS, 1999: SPSS for Windows, SPSS Inc., Chicago, IL, USA.

Stahly, 1984: *Use of fats in diets for growing pigs*, *In:* Fats in Animal Nutrition, Butterworths, London, UK.

Wagner, J. R.; Schinckel, A. P.; Chen, W.; Forrest, J. C.; Coe, B. L., 1999: Analysis of body composition changes of swine during growth and development. *Journal of Animal Science* **77**, 1442-1466.

Wiseman, J.; Agunbiade, J. A., 1998: The influence of changes in dietary fat and oils on fatty acid profiles of carcass fat in finishing pigs. *Livestock Production Science* **54**, 217-227.

Yeom, K.-H.; Schonewille, J. T.; Everts, H.; Lee, K.-W.; van Trierum, G.; Beynen, A. C., 2005: Growth performance and body composition of goat kids fed milk replacers with increasing levels of linoleic acid. *Journal of Animal Physiology and Animal Nutrition* **89**, 29-34.

CHAPTER 4

Meat quality, digestibility and deposition of fatty acids in growing-finishing pigs fed restricted, iso-energetic amounts of diets containing either beef tallow or sunflower oil

J. Mitchaothai†§, H. Everts†, C. Yuangklang‡, S. Wittayakun‡, K. Vasupen‡, S. Wongsuthavas‡, P. Srenanul‡, R. Hovenier†, and A.C. Beynen†

§Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand

‡Faculty of Natural Resources, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand

†Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, P.O. Box 50.152, 3508 TD, Utrecht, The Netherlands

Asian-Australian Journal of Animal Science, in press.

ABSTRACT

The influence of dietary beef tallow (BT) versus sunflower oil (SO) on meat quality and apparent digestibility and deposition of individual fatty acids in whole carcass was investigated by using 39 castrated male growing pigs. The pigs were fed a diet containing either 5% (wt/wt) BT or 4.5% (wt/wt) SO. The diets contained equal amounts of metabolizable energy in the form of the variable fats and were fed on an iso-energetic, restricted basis. As would be expected crude fat in the SO diet was better digested ($P < 0.001$) than that in the BT diet. The dietary fat type had no effect ($P > 0.05$) on growth performance. Physical properties of carcass and meat quality were not affected ($P > 0.05$) by the type of fat in the diet. The pigs fed the BT diet showed a lower ($P < 0.001$) apparent digestibilities for palmitic and linoleic acid, but that of oleic and α-linolenic acid was not affected ($P > 0.05$). The ratio of deposition in carcass to intake of digestible fatty acids for the whole feeding period was increased ($P < 0.01$) for oleic and linoleic acid in pigs fed the SO diet. The pigs fed the SO diet instead of the BT diet had a lowered ($P < 0.05$) deposition:intake ratio of MUFA. The calculated minimum *de novo* synthesis of SFA was increased for the SO diet, but that of MUFA was not different ($P > 0.05$). In conclusion, the iso-energetic replacement of BT by SO had a marked impact of the fatty acid composition of tissues, but did not affect carcass and meat quality traits in spite of the marked increase in the deposition of linoleic acid in the adipose tissues, loin muscle, and whole body. After calculating the deposition:digestible intake ratio for individual fatty acids, it became clear that the type of dietary fat had marked, specific effects on the synthesis and oxidation of fatty acids.

Keywords: deposition, digestibility, fat type, fatty acid, meat quality, pigs

INTRODUCTION

There is great interest in the use of vegetable oils instead of animal fat as dietary fat source for pig production. Vegetable oils are generally rich in PUFA, which will be reflected in the pig meat (Nguyen et al., 2003), thus yielding healthy meat for human consumption, but their technical quality may be diminished due to high susceptibility to oxidation and low consistency (Rey et al., 2001; Ramirez et al., 2004; Guo et al., 2006). In addition, there is increasing concern among the general public about the use of animal feedstuffs, including animal fat, for formulating pig diets. Thus, the stage is set for further collecting data on the effects of vegetable versus animal fats in pig diets. In general, vegetable oils are better digested by pigs than are animal fats (Cera et al., 1988; Cera et al., 1989). The influence of replacement of dietary animal fat by vegetable oil on meat quality of pigs has been described (Wiseman and Agunbiade, 1998; Wood et al., 2004; Mitchaothai et al., 2007). However, the various studies on meat quality can be criticized because the pigs had free access to diets containing equal inclusion percentages of either animal of vegetable fat. The unrestricted feed intake and difference in digestibility between the experimental fats interferes with the interpretation of the results. In this study we used diets containing iso-energetic amounts of either vegetable oil or animal fat, and the diets were fed on a restricted basis to growing-finishing pigs so that their energy intake was identical. Sunflower oil (SO) and beef tallow (BT) were used as representatives of vegetable and animal fat source. In the pigs we either measured directly or calculated the quality of meat, fatty acid composition of tissues, digestibility and deposition of individual fatty acids, the ratio of deposition to digested amount of fatty acids and minimum, whole body de novo fatty acid synthesis.

MATERIALS AND METHODS

Animals, Diets and Feeding

Thirty-nine castrated-male pigs, Landrace × Large White × Duroc crossbred, were used in the current study. Three pigs with an average BW of 31.0 kg were selected to be slaughtered for baseline measurements. Then, the remaining pigs with an average 30.39 ± 2.32 kg BW were allotted to one of the two dietary treatments on the basis of BW and were housed in individual cages. There were two experimental diets with either beef tallow (BT) or sunflower oil (SO) as shown in Table 1. The earlier measured (Mitchaothai et al., *in press*) difference in digestibility between BT and SO was taken into account so that the two diets would contain iso-energetic amounts of added fat. Table 1 shows the ingredient and nutrient compositions of the experimental diets. The analyzed fatty composition of the diets is illustrated in Table 2.

The pigs were fed twice a day at an energy intake level of 80% of the *ad libitum* intake determined earlier (Mitchaothai et al., *in press*). Iso-energetic amounts of the two experimental diets were supplied. There were no feed leftovers. The pigs had free access to water throughout the experiment. The pigs were weighed weekly until the end of the experiment which lasted 98 days. All pigs were slaughtered at an average body weight of 100.74 ± 4.64 kg in a commercial slaughterhouse.

Collection and Pre-treatment of Samples

Table 1. Composition of the diets (as-fed basis)

Item	BT	SO
Raw materials (g)		
Cassava chips	45.53	45.53
Soybean meal (44% CP)	34.00	34.00
Extruded soy beans	7.00	7.00
Beef tallow (BT)	5.00	-
Sunflower oil (SO)	-	4.50
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Di-calcium phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Premix	0.25	0.25
Total	**100.00**	**99.50**
Analyzed nutrients (%)		
Dry matter	88.63	88.05
Crude protein	20.94	21.55
Crude fat	7.34	7.31
Crude fiber	6.65	6.25
Ash	8.37	8.36
Calculated ME (kJ/kg)	13,815.94	13,862.03

BT = beef tallow; SO = sunflower oil

The three pigs for baseline measurements were killed by an intravenous overdose of sodium pentobarbital (200 mg/kg of BW) (Nembutal®, Ceva Animale, La Ballastiere, France). Pigs were scalded, the head was removed, and the carcass was split into halves along the median plane. Both sides of the pig carcass, head, visceral organs (minus gastrointestinal contents), blood and leaf fat were weighed, wrapped in a polyethylene bag to prevent evaporative moisture losses, and then frozen at -20°C for at least 1 week before analysis. Upon removal from the freezer, each sample was weighed and sawn in cross-sectional slices (2.54 cm) along its length (Shields et al., 1983). The sawn carcass and samples containing bone were dried at 60°C for 1 week in a forced-hot air oven and were then ground three times.

The 36 pigs in the feeding trial were sacrificed at 14 weeks after the commencement. Carcass measurements (loin eye area at 10th rib, LEA (cm^2), and back fat depth at 10th rib, FD10R) were done as described previously (Wagner et al., 1999). Samples of back fat of midline between 3rd and 4th rib, right inguinal subcutaneous fat, retroperitoneal fat, and loin were collected to determine fatty acid composition.

Carcass Traits and Meat Quality

Hot carcass weight was measured after removing all internal organs including kidney. The thickness of back fat was determined according to standard methods described previously (Sripromma, 1984). Fat-lean ratio was expressed according to the LSQ (Lenden-Speck-Quotient) system of Pfeiffer & Falkenberg (1972). Briefly, the LSQ is calculated as (B1 + B2)/(2 × B3) where B1 = back fat thickness at the front base of gluteus muscle, B2 = back fat thickness on top of gluteus muscle (at the thinnest part of back fat) and B3 = shortest distance from the front base of gluteus

muscle to the dorsal border of the spinal cord. The right *M. longissimus* was used for meat quality assessing. The colour score of *M. longissimus thoracic* was measured by using the 6-point Japanese pork colour scale (JPCS) (Nakai et al., 1975) and by determining the CIELAB colour co-ordinates (colour L*, a*, b*) in triplicate with a HunterLab (Colour flex®) device after a 30-min blooming time (D65 light source, 10° standard observer, 45°/0° geometry, 1 in. light surface, white standard; Hunter, Reston, VA). Duplicate 3 g diced samples were taken to determine water-holding capacity (WHC) using the expressible moisture test of Goerl et al. (1995) with modification of duration and pressure force. Samples were placed in the centre of Whatmam no. 2 filter paper and pressed between glass sheets (15 cm × 15 cm × 8 mm) under 1 kg/90 cm^2 pressure for 20 min. The resulting meat ring and expressed juice ring were subsequently measured using a digital planimeter (Placom KP-90N, Topcon®, Topcon Instruments (Thailand), Bangkok, Thailand). The amount of expressible moisture was recorded as the ratio of the juice area to the muscle area. Intramuscular fat content was determined after Soxhlet extraction using hexane on the meat slices. The pH-value of muscle was determined using the Meat pH meter (Model HI99163, Hanna Instruments, Portugal) at 45 min (pH_1) and 24 hr (pH_U) after slaughter. Drip loss was assessed as the proportionate weight loss of a slice of muscle (175 to 185 g with thickness of 2.54 cm) that had been suspended in a plastic bag for 24 h at 2°C (Honikel, 1987). Right *M. longissimus lumborum* (loin) was collected and stored at -20°C until analyzing the remaining meat quality parameters. Pork chops were weighed before and after cooking to determine percentage of cooking loss. The pork chops were put in a plastic bag and then cooked for 40 min in a water bath with constant temperature of 70°C. After the chops had cooled to room temperature (25°C), five 1.27-cm-diameter cores from each chop were removed with a cylindrical core parallel to the muscle fiber orientation. Cores were sheared perpendicular to the muscle fiber orientation using a Warner-Bratzler shear V-blade attached to an Texture Analyzer (Stable Micro System Ltd., Surrey, England) fitted with a 10-g compression load cell with a crosshead speed of 900 mm/min. Peak force values of cores sheared through the centre were used to determine the mechanical tenderness of the sample. Sarcomere length was assessed (in four replicate) by taking the slice from the central part of small *M. longissimus thoracic* (3 to 4 g) according to a method described previously (Monin et al., 1999). The diet and meat samples were dried at 60°C for 72 hr in a forced-hot air oven and then were analyzed for crude protein, crude fiber, and ash (AOAC, 1990). The dried meat samples were quantified for the percentage of moisture and fat contents according to standard chemical analyses (AOAC, 1990) and then the percentage of fat was calculated and expressed as loin intramuscular fat.

Estimation of Carcass Fat Content

The total lean mass and the total fat mass in the whole body were estimated by the using the following equations (Schinckel et al., 2001): Fat-free, total lean mass (kg) = 5.00 + [0.434 × carcass weight (kg)] + [0.168 × LEA (cm^2)] + [(-3.38) × FD10R (cm)]; Total fat mass (kg) = (-10.7) + [0.395 × carcass weight (kg)] + [(-0.150) × LEA (cm^2)] + [4.49 × FD10R (cm)]. Thus, intramuscular fat mass = fat-free, lean mass (kg) / 100 × [fraction IMF × (100 - fraction IMF)$^{-1}$]. Based on literature data (Irie and Sakimoto, 1992; Otten et al., 1993; Enser et al., 2000), it can be concluded that the fatty acid profiles of adipose tissue from different sites do not differ much. Thus, the average percentage of fatty acids from different sites of adipose

tissue can be considered to be representative for the whole body mass of adipose tissue. The amount of fat in blood is negligible and therefore was not taken into account. Thus, the mass (kg) of adipose tissue equals total fat mass (kg) in whole body - fat mass (kg) in muscle - fat mass (kg) in liver.

Chemical Analyses

The diet and feces samples were dried at 60°C for 72 hr in a forced-hot air oven and were then analyzed for crude protein, crude fiber, and ash (AOAC, 1990). The dried meat, liver, and whole carcass samples were analyzed for moisture and fat (AOAC, 1990). The fat extracted from meat was defined as intramuscular fat (IMF).

Total fat in the fresh and dried samples (diets, feces, adipose tissues, and loin) were extracted as described previously (Horwitz, 1975). Each sample was added to a flask and 2 ml of ethanol was added. Subsequently, 10 ml of HCl (8 mol/L) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80°C for 30 to 40 min. The tubes were cooled down, 10 ml of ethanol (96%, wt/wt) and 25 ml of petroleum ether (boiling point between 40 and 60°C) were added and the tube was shaken for one min. The fat-containing upper layer was decanted into a 150-ml round-bottom flask. The extraction procedure was repeated twice with 15 ml of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath of 40°C. The round-bottom flasks with the lipids were dried overnight at 60°C and the total lipids were measured gravimetrically. Total lipids were saponified and methylated according to the procedure of Metcalfe et al. (1966) followed by gas chromatography for determination of fatty acid composition (Javadi et al., 2004).

Calculation of Digestible Fatty Acid Intake, Fatty Acid Deposition and Minimum De novo Synthesis

The total digestible fatty acid intake was calculated as fatty acid intake (kg/14 weeks) × apparent fatty acid digestibility (fraction of intake). The deposition of fatty acids was calculated with the following formula: Fatty acid deposition (kg/14 weeks) = carcass content of fatty acid at the end of the study–carcass content of fatty acid at the start of the study. Carcass content of fatty acid at the end (kg) was calculated as the sum of adipose tissue mass (kg) × fraction of adipose fatty acid, IMF mass (kg) × fraction of IMF fatty acid and fat mass in liver (kg) × fraction of liver fatty acid. The ratio of fatty acid deposition and digestible fatty acid intake was calculated for selected individual fatty acids and groups of fatty acids. Minimum *de novo* fatty acid synthesis was calculated as fatty deposition minus digestible fatty acid intake.

Statistical Analyses

The effect of dietary fat type was evaluated for statistical significance ($P < 0.05$) by the Student's *t* test (SPSS, 1999). Results are expressed as means ± SD.

RESULTS

Dietary Fatty Acid Composition

The fatty acid composition of the experimental diets is shown in Table 2. The fat component of the BT diet contained approximately 51% total saturated fatty acids (SFA), whereas that of the SO diet contained approximately 42% total polyunsaturated fatty acids (PUFA). The amounts of total mono-unsaturated fatty acids (MUFA) were approximately 29% and 32% for the BT and SO diets, respectively.

Table 2. Analyzed fatty acid composition of the experimental diets

Item	BT	SO
Fatty acids, g methylester/100 g methylesters		
C 10:0	0.13	0.13
C 14:0	1.34	0.26
C 15:0	0.42	0.09
C 16:0	21.69	14.21
C 17:0	1.32	0.17
C 18:0	24.72	5.85
C 20:0	0.42	0.35
C 22:0	0.42	0.82
C 24:0	0.28	0.45
C 16:1	0.62	0.14
C 17:1	0.21	0.00
C 18:1 n-9	23.24	30.45
C 18:1 n-7	3.92	1.22
C 20:1 n-9	0.58	0.51
C 22:1 n-9	0.00	0.08
C 18:2 n-6	12.37	40.47
C 18:3 n-6	0.24	0.00
C 18:3 n-3	1.50	1.35
C 20:5 n-3	0.11	0.11
Unidentified	6.48	3.35
Σ SFA	50.72	22.33
Σ MUFA	28.57	32.40
Σ PUFA	14.22	41.92

BT = beef tallow; SO = sunflower oil

SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids

Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0

Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9

Σ PUFA = C18:2 n-6 + C18:3 n-6 + C18:3 n-3 + C20:5 n-3

Growth Performance, Meat Quality, and Fatty Acid Composition

There was no diet effect ($P > 0.05$) on feed intake, growth (average daily gain), feed conversion ratio (feed:gain) and final BW (Table 3). The feed:gain ratio in pigs fed the SO diet tended ($P = 0.082$) to be lowered.

Meat Quality

Carcass traits and meat quality characteristics, excepted for the colour score, did not differ ($P > 0.05$) for the two dietary fat sources (Table 3). The pig fed the SO diet had a significantly higher ($P < 0.05$) colour score than did those fed the BT diet,

but the colour difference could not be detected by technical measurement of the colour coordinates.

Table 3. Effect of fat type on animal performance, carcass traits, and meat quality

Item	BT	SO	*P*-value
Growth performance			
Initial BW, kg	30.44 ± 1.79	30.33 ± 2.81	0.888
Final BW, kg	99.74 ± 5.52	101.81 ± 3.32	0.198
ADFI, kg/d	2.07 ± 0.05	2.04 ± 0.06	0.137
ADG, kg/d	0.735 ± 0.06	0.756 ± 0.04	0.231
Feed : gain	2.79 ± 0.25	2.66 ± 0.14	0.082
Carcass quality			
Hot carcass, kg	78.64 ± 4.44	79.93 ± 2.47	0.307
Back fat thickness, cm	2.24 ± 0.30	2.22 ± 0.38	0.892
Fat-lean ratio (LSQ)	0.21 ± 0.06	0.21 ± 0.06	0.887
Meat quality (loin)			
Colour[1]	3.21 ± 0.47	3.57 ± 0.46	0.036
Colour **L*** value	35.19 ± 2.21	34.80 ± 1.60	0.565
Colour **a*** value	9.82 ± 2.87	10.26 ± 2.88	0.660
Colour **b*** value	11.11 ± 1.08	11.55 ± 1.13	0.260
pH_1	6.03 ± 0.27	6.03 ± 0.30	0.982
pH_U	5.76 ± 0.38	5.71 ± 0.40	0.707
Drip loss, %	1.02 ± 0.42	1.15 ± 0.48	0.264
Cooking loss, %	25.03 ± 2.73	23.85 ± 2.34	0.312
WHC, %	3.29 ± 0.96	2.90 ± 0.55	0.301
Shear force, N	64.37 ± 12.56	60.90 ± 9.82	0.204
Sarcomere length, μm	1.86 ± 0.12	1.90 ± 0.15	0.554
IMF, % wet weight	2.39 ± 0.72	2.41 ± 0.59	0.928

Means ± SD for 18 pigs per experimental diet
BT = beef tallow; SO = sunflower oil
WHC = water holding capacity
IMF = intramuscular fat
[1]Japanese colour score: 1 = pale, pinkish grey, 6 = dark, purplish red.

Fatty Acid Composition of Tissues

Fatty acid patterns of adipose tissues and loin are documented in Table 4 and 5. The relative concentrations of C14:0, C16:0, C17:0, and C18:0 after feeding the BT diet were higher ($P < 0.001$) for all three sites of adipose tissues, but in loin muscle there only was a significantly lower ($P < 0.001$) percentage of C17:0. There was no treatment difference ($P > 0.05$) for the levels of C16:0 and C18:0 in loin. The pigs fed the BT diet showed higher ($P < 0.05$) tissue concentrations of MUFA, of which C18:1 n-9 is the major component. The concentrations of C18:2 n-6 (linoleic acid) and C20:2 n-6 (eicosadienoic acid) in adipose tissues and loin were higher ($P < 0.001$) in the pigs fed the SO diet. On the other hand, there were higher ($P < 0.05$) levels of C18:3 n-3 (α-linolenic acid) in the retroperitoneal fat and loin of the pigs fed the BT diet. No treatment effect ($P > 0.05$) was seen for the concentration of α-linolenic acid in subcutaneous and back fat. The content of SFA in adipose tissues was higher ($P < 0.001$) for the pigs fed the BT diet, but the SFA content of loin was unchanged ($P >$

0.05). In both adipose tissues and loin of the pigs fed BT, the levels of MUFA were higher ($P < 0.01$) and those of PUFA were lower ($P < 0.01$). In the pigs fed the BT diet, the ratio of MUFA/SFA was higher ($P < 0.001$) in adipose tissues, but it was lower ($P < 0.01$) in loin. The ratios of PUFA/SFA, n-6/n-3 and C18:2 n-6/C18:3 n-3 in both adipose tissues and loin were higher ($P < 0.05$) for the pigs fed the SO diet.

Table 4. Fatty acid composition of subcutaneous and back fat

Item	Subcutaneous fat		Signi-ficance	Back fat		Signi-ficance
	BT	SO		BT	SO	
Analyzed fatty acids (g/100g methylester)						
C 14:0	1.86 ± 0.12	1.26 ± 0.13	***	1.59 ± 0.14	0.99 ± 0.08	***
C 16:0	25.39 ± 1.41	21.28 ± 1.78	***	22.54 ± 0.96	18.29 ± 1.10	***
C 17:0	0.87 ± 0.18	0.40 ± 0.07	***	0.85 ± 0.13	0.38 ± 0.06	***
C 18:0	17.63 ± 1.64	13.55 ± 2.19	***	14.02 ± 1.73	10.08 ± 1.13	***
C 20:0	0.19 ± 0.06	0.20 ± 0.04	NS	0.12 ± 0.09	0.17 ± 0.05	NS
C 16:1	0.51 ± 0.33	0.33 ± 0.06	*	0.44 ± 0.04	0.37 ± 0.06	***
C 17:1	0.45 ± 0.07	0.18 ± 0.03	***	0.53 ± 0.06	0.21 ± 0.03	***
C 18:1 n-7	2.52 ± 0.18	1.32 ± 0.24	***	2.80 ± 0.14	1.59 ± 0.14	***
C 18:1 n-9	32.64 ± 2.01	30.13 ± 2.31	**	37.17 ± 2.42	34.25 ± 1.70	***
C 20:1 n-9	0.67 ± 0.10	0.57 ± 0.07	**	0.85 ± 0.07	0.77 ± 0.08	**
C 22:1 n-9	0.00 ± 0.00	0.01 ± 0.03	NS	ND	ND	-
C 18:2 n-6	12.11 ± 2.13	27.09 ± 4.03	***	13.24 ± 1.31	28.66 ± 2.91	***
C 18:3 n-3	1.16 ± 0.21	1.12 ± 0.16	NS	1.21 ± 0.13	1.14 ± 0.12	NS
C 20:2 n-6	0.35 ± 0.07	0.81 ± 0.07	***	0.48 ± 0.06	1.13 ± 0.12	***
C 20:3 n-6	0.00 ± 0.00	0.02 ± 0.04	NS	ND	ND	-
C 20:3 n-3	0.02 ± 0.05	0.00 ± 0.00	NS	0.01 ± 0.03	0.01 ± 0.04	NS
C 20:4 n-6	0.10 ± 0.09	0.27 ± 0.05	***	0.11 ± 0.09	0.27 ± 0.04	***
C 20:5 n-3	ND	ND	-	ND	ND	-
C 22:4 n-6	ND	ND	-	ND	ND	-
C 22:5 n-3	ND	ND	-	ND	ND	-
unknown	3.35 ± 0.46	1.45 ± 0.37	***	3.90 ± 0.67	1.66 ± 0.49	***
SFA	45.94 ± 2.75	36.68 ± 3.83	***	39.11 ± 2.55	29.91 ± 2.07	***
MUFA	36.79 ± 2.08	32.54 ± 2.61	***	41.78 ± 2.54	37.20 ± 1.86	***
PUFA	13.74 ± 2.44	29.31 ± 4.25	***	15.05 ± 1.51	31.22 ± 3.09	***
MUFA/SFA	0.80 ± 0.09	0.90 ± 0.14	*	1.07 ± 0.11	1.25 ± 0.11	***
PUFA/SFA	0.30 ± 0.06	0.82 ± 0.20	***	0.39 ± 0.05	1.05 ± 0.18	***
n6/n3	10.64 ± 0.77	25.31 ± 0.99	***	11.33 ± 0.54	26.19 ± 1.08	***
18:2n6/18:3n3	10.43 ± 0.61	24.31 ± 0.93	***	10.91 ± 0.45	25.15 ± 0.80	***

Means ± SD for 18 pigs per experimental diet
BT = beef tallow; SO = sunflower oil; ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
NS: $P > 0.05$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C20:2 n-6 + C20:3 n-6 + C20:3 n-3 + C20:4 n-6 + C20:5 n-3 + C22:4 n-6 + C22:5 n-3

Table 5. Fatty acid composition of retroperitoneal fat and loin intramuscular fat

Item	Retroperitoneal fat		Signi-	Loin		Signi-
	BT	SO	ficance	BT	SO	ficance
Analyzed fatty acids (g/100g methylester)						
C 14:0	1.80 ± 0.24	1.26 ± 0.23	***	1.65 ± 0.21	1.51 ± 0.21	NS
C 16:0	24.31 ± 1.34	20.76 ± 1.78	***	24.99 ± 1.22	24.99 ± 1.39	NS
C 17:0	0.83 ± 0.18	0.43 ± 0.15	***	0.51 ± 0.09	0.34 ± 0.08	***
C 18:0	16.55 ± 1.81	13.00 ± 1.55	***	12.65 ± 0.91	12.52 ± 0.68	NS
C 20:0	0.12 ± 0.08	0.13 ± 0.09	NS	0.05 ± 0.08	0.11 ± 0.10	NS
C 16:1	0.41 ± 0.06	0.35 ± 0.05	**	2.84 ± 0.33	2.35 ± 0.36	***
C 17:1	0.43 ± 0.10	0.20 ± 0.10	***	0.42 ± 0.07	0.23 ± 0.09	***
C 18:1 n-7	2.41 ± 0.41	1.46 ± 0.25	***	3.64 ± 0.25	2.97 ± 0.28	***
C 18:1 n-9	32.49 ± 1.30	31.00 ± 0.92	***	37.77 ± 1.87	35.86 ± 2.41	*
C 20:1 n-9	0.65 ± 0.09	0.60 ± 0.07	NS	0.85 ± 0.12	0.80 ± 0.12	NS
C 22:1 n-9	ND	ND	-	0.10 ± 0.10	0.23 ± 0.13	**
C 18:2 n-6	14.72 ± 4.84	26.28 ± 4.62	***	9.89 ± 2.33	13.69 ± 3.42	***
C 18:3 n-3	1.25 ± 0.12	1.11 ± 0.13	**	0.46 ± 0.19	0.34 ± 0.13	*
C 20:2 n-6	0.43 ± 0.15	0.89 ± 0.21	***	0.27 ± 0.09	0.47 ± 0.12	***
C 20:3 n-6	ND	ND	-	0.11 ± 0.15	0.11 ± 0.12	NS
C 20:3n-3	0.01 ± 0.03	0.00 ± 0.00	NS	ND	ND	-
C 20:4n-6	0.16 ± 0.08	0.27 ± 0.04	***	1.03 ± 0.76	1.08 ± 0.66	NS
C 20:5n-3	ND	ND	-	0.01 ± 0.05	0.00 ± 0.00	NS
C 22:4n-6	ND	ND	-	0.06 ± 0.11	0.09 ± 0.13	NS
C 22:5 n-3	ND	ND	-	0.11 ± 0.19	0.02 ± 0.07	NS
unknown	3.44 ± 0.89	2.27 ± 2.38	NS	2.51 ± 0.65	2.26 ± 1.03	NS
SFA	43.61 ± 3.17	35.58 ± 3.30	***	39.85 ± 2.08	39.47 ± 1.97	NS
MUFA	36.39 ± 1.73	33.60 ± 0.98	***	45.61 ± 2.19	42.44 ± 2.87	**
PUFA	16.56 ± 5.06	28.55 ± 4.85	***	11.95 ± 3.56	15.79 ± 4.37	**
MUFA/SFA	0.84 ± 0.05	0.95 ± 0.10	***	1.15 ± 0.06	1.08 ± 0.07	**
PUFA/SFA	0.39 ± 0.17	0.82 ± 0.21	***	0.30 ± 0.10	0.41 ± 0.13	*
n6/n3	12.28 ± 4.26	24.80 ± 4.05	***	21.35 ± 9.46	44.03 ± 8.54	***
18:2n6/18:3n3	11.91 ± 4.34	23.76 ± 3.89	***	22.19 ± 8.12	40.94 ± 9.30	***

Means ± SD for 18 pigs per experimental diet
BT = beef tallow; SO = sunflower oil; ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
NS: $P > 0.05$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C20:2 n-6 + C20:3 n-6 + C20:3 n-3 + C20:4 n-6 + C20:5 n-3 + C22:4 n-6 + C22:5 n-3

Nutrient Digestibility

In the first and second period of the experiment, the pigs fed the SO diet had a significantly higher ($P < 0.01$) apparent digestibility of crude fat than those fed the BT

diet, but there was no significant dietary fat effect ($P > 0.05$) on the digestibility of dry matter (DM) and crude protein (CP) (Table 6). The digestibility of DM, CP, and crude fat trended to be increased with age, irrespective of dietary treatment.

Table 6. Effect of dietary fat type on apparent macronutrient and fatty acid digestibility

Item	BT	SO	*P*-value
Apparent digestibility, % of intake at 4 weeks			
Dry matter	87.22 ± 3.13	87.13 ± 2.10	0.920
Crude fat	69.69 ± 9.45	77.46 ± 5.72	0.006
Crude protein	87.49 ± 6.08	86.57 ± 4.69	0.619
Apparent digestibility, % of intake at 10 weeks			
Dry matter	87.74 ± 2.93	88.19 ± 3.46	0.678
Crude fat	73.68 ± 8.88	81.56 ± 6.53	0.006
Crude protein	90.17 ± 2.33	88.95 ± 3.34	0.229
Apparent digestibility, % of intake at 4 weeks			
C 16:0	66.32 ± 7.93	73.93 ± 8.19	0.009
C 18:0	54.43 ± 12.26	-37.86 ± 39.35	<0.001
C 18:1 n-9	94.45 ± 1.65	94.34 ± 1.40	0.825
C 18:2 n-6	94.58 ± 1.84	97.40 ± 0.74	<0.001
C 18:3 n-3	94.60 ± 1.86	93.97 ± 1.57	0.292
Σ SFA	49.07 ± 13.12	-20.51 ± 20.16	<0.001
Σ MUFA	90.11 ± 2.77	90.28 ± 3.51	0.882
Σ PUFA	94.59 ± 1.80	95.68 ± 1.14	0.039
Apparent digestibility, % of intake at 10 weeks			
C 16:0	58.70 ± 9.52	76.70 ± 7.06	<0.001
C 18:0	42.84 ± 12.51	-50.78 ± 42.39	<0.001
C 18:1 n-9	94.01 ± 1.67	94.33 ± 2.34	0.651
C 18:2 n-6	95.65 ± 1.28	98.06 ± 0.86	<0.001
C 18:3 n-3	96.41 ± 2.21	96.26 ± 2.43	0.855
Σ SFA	47.12 ± 11.09	-20.20 ± 34.34	<0.001
Σ MUFA	91.68 ± 3.17	89.37 ± 6.71	0.270
Σ PUFA	96.03 ± 1.70	97.37 ± 1.39	0.019

Means ± SD for 18 pigs per experimental diet
BT = beef tallow; SO = sunflower oil
Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C16:1 + C18:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3

Table 6 also shows the apparent digestibility for selected individual fatty acids and groups of fatty acids. The palmitic acid (C16:0) digestibility for the SO diet was greater ($P < 0.01$) than that for the BT diet. The digestibility of C18:0 for the SO diet was calculated to be negative which may be explained by a combination of low intake and fecal excretion due to bacterial synthesis. The apparent digestibility of oleic (C18:1 n-9) was not affected ($P > 0.05$) by diet. The pigs fed the SO diet displayed a significantly greater ($P < 0.001$) digestibility of linoleic acid (C18:2 n-6), but the digestibility of α-linolenic acid (C18:3 n-3) did not differ ($P > 0.05$) between both diets. The digestibility of PUFA was higher ($P > 0.05$) for the pigs fed the SO diet, but that of MUFA did not differ ($P > 0.05$) between the experimental diets.

Table 7. Effect of dietary fat type on digestible fatty acid intake, fatty acid deposition and the deposition:intake ratio during the whole feeding period

Fatty acids	BT	SO	*P*-value
Digestible fatty acid intake (kg/14 weeks)			
C 16:0	1.89 ± 0.23	1.44 ± 0.12	<0.001
C 18:0	1.68 ± 0.39	-0.37 ± 0.29	<0.001
C 18:1 n-9	3.06 ± 0.16	3.89 ± 0.35	<0.001
C 18:2 n-6	1.64 ± 0.09	5.36 ± 0.49	<0.001
C 18:3 n-3	0.20 ± 0.01	0.17 ± 0.02	<0.001
Σ FA	10.03 ± 0.89	10.69 ± 0.83	0.031
Σ SFA	3.88 ± 0.67	1.11 ± 0.37	<0.001
Σ MUFA	3.63 ± 0.19	3.93 ± 0.36	0.006
Σ PUFA	1.87 ± 0.10	5.54 ± 0.51	<0.001
Σ n-9	3.13 ± 0.16	3.95 ± 0.35	<0.001
Σ n-6	1.66 ± 0.09	5.36 ± 0.49	<0.001
Σ n-3	0.21 ± 0.01	0.18 ± 0.02	<0.001
Fatty acid deposition (kg/14 weeks)			
C 16:0	3.57 ± 0.62	3.29 ± 0.39	0.186
C 18:0	2.49 ± 0.44	2.11 ± 0.34	0.022
C 18:1 n-9	4.81 ± 0.91	4.81 ± 0.51	0.999
C 18:2 n-6	1.70 ± 0.18	4.27 ± 0.65	<0.001
C 18:3 n-3	0.14 ± 0.03	0.14 ± 0.02	0.768
Σ FA	14.36 ± 2.22	15.77 ± 1.42	0.072
Σ SFA	6.49 ± 1.07	5.67 ± 0.75	0.036
Σ MUFA	5.40 ± 1.00	5.19 ± 0.56	0.516
Σ PUFA	1.94 ± 0.23	4.63 ± 0.70	<0.001
Σ n-9	4.92 ± 0.93	4.92 ± 0.53	0.432
Σ n-6	1.79 ± 0.20	4.49 ± 0.68	<0.001
Σ n-3	0.14 ± 0.03	0.14 ± 0.03	0.735
Deposition : intake ratio			
C 16:0	1.99 ± 0.49	2.29 ± 0.27	0.076
C 18:0	1.70 ± 0.67	-7.43 ± 5.20	<0.001
C 18:1 n-9	1.56 ± 0.30	1.23 ± 0.10	0.004
C 18:2 n-6	1.03 ± 0.13	0.80 ± 0.14	<0.001
C 18:3 n-3	0.72 ± 0.15	0.81 ± 0.17	0.154
Σ FA	1.43 ± 0.28	1.97 ± 1.34	0.184
Σ SFA	1.60 ± 0.47	4.04 ± 2.59	0.016
Σ MUFA	1.50 ± 0.26	1.22 ± 0.37	0.035
Σ PUFA	0.88 ± 0.13	0.80 ± 0.15	0.203
Σ n-9	1.12 ± 0.16	1.44 ± 0.23	<0.001
Σ n-6	1.04 ± 0.13	0.80 ± 0.14	<0.001
Σ n-3	0.72 ± 0.15	0.81 ± 0.17	0.154

Means ± SD for 18 pigs per experimental diet.
BT = beef tallow; SO = sunflower oil
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3

Digestible Fatty Acid Intake and Deposition

The intake of digestible palmitic, stearic, and α-linolenic acid of the pigs fed the BT diet was greater ($P < 0.001$) than that of the pigs fed the SO diet, whereas the pigs fed the SO diet had higher ($P < 0.001$) intakes of digestible oleic and linoleic acid (Table 7). The pigs fed the SO diet had a greater ($P < 0.05$) intake of total digestible fatty acids (FA), MUFA and PUFA, but had a lower ($P < 0.001$) intake of digestible SFA. When the fatty acids were pooled according to their structural similarities, the SO diet was found to provide more digestible n-9 MUFA and n-6 PUFA ($P < 0.001$), whereas the amount of digestible n-3 PUFA was lower ($P < 0.001$) than that provided by the BT diet.

The whole body of the pigs at baseline contained an average total fat mass of 4.1 ± 0.8 kg. The calculated total fat mass in the carcass at the end of the experiment was 18.99 ± 3.01 and 19.19 ± 1.55, n = 18 (p = 0.819) for the pigs fed the BT and SO diet, respectively. The pigs fed BT diet had deposited more stearic acid ($P < 0.05$), but less ($P < 0.001$) linoleic acid when compared with the pigs fed the SO diet (Table 7). There was no difference ($P > 0.05$) between both diets in the deposition of palmitic acid, oleic acid and α-linolenic acid. The pigs fed the SO diet showed a trend towards higher ($P = 0.072$) deposition of total fatty acids with unaltered deposition of MUFA ($P > 0.05$). SFA deposition in the pigs fed the SO diet was less ($P < 0.05$) than that in those fed the BT diet, whereas PUFA deposition was higher ($P < 0.001$). There was more deposition ($P < 0.001$) of n-6 PUFA in the pigs fed the SO diet, but deposition of the n-9 MUFA and n-3 PUFA was similar ($P > 0.05$) to that in the pigs fed the BT diet.

The deposition:intake ratios for palmitic acid tended ($P = 0.076$) to be higher in the pigs fed the SO diet when compared with the pigs fed the BT diet. The calculated deposition:intake ratio for stearic acid in pigs fed the SO diet was negative because of the negative apparent digestibility and thus negative digestible intake of stearic acid. This would imply that the stearic acid deposited in body of the pigs fed the SO diet originated from *de novo* synthesis. The deposition: intake ratios for oleic and linoleic acid in the pigs fed the SO diet were lower ($P < 0.01$) than those in the pigs fed the BT diet. There was no diet effect ($P > 0.05$) on the deposition:intake ratio for α-linolenic acid, the ratio for the two groups of pigs being less than one. Dietary treatment did not influence ($P > 0.05$) the deposition:intake ratios for total FA, PUFA and n-3 PUFA, but the ratio for SFA and n-9 was higher ($P > 0.05$) in pigs fed the SO diet, whereas the ratios for MUFA and n-6 PUFA were lower ($P < 0.05$).

Table 8. Effect of dietary fat type on minimum *de novo* synthesis of fatty acids during the whole feeding period

Fatty acids	BT	SO	*P*-value
Minimum synthesis (kg/14 weeks)			
SFA	2.75 ± 1.57	4.64 ± 0.79	0.002
MUFA	1.76 ± 1.02	1.26 ± 0.45	0.141
SFA/(SFA+MUFA)	0.60 ± 0.12	0.79 ± 0.05	<0.001

Means ± SD for 18 pigs per experimental diet.
BT = beef tallow; SO = sunflower oil
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-9 + C20:1 n-9

Minimum de novo synthesis

The pigs fed the diet containing SO had a greater ($P < 0.01$) synthesis of SFA, but no difference ($P > 0.05$) was observed for the synthesis of MUFA when compared to their counterparts fed the diet with BT. The synthesis ratio of SFA:(SFA+MUFA) was greater ($P < 0.001$) in the pigs fed the diet containing SO (Table 8).

DISCUSSION

As far as we know, this is the first study in which growing-finishing pigs were fed on a restricted basis diets containing iso-energetic amounts of either BT or SO to determine the effect of vegetable fat on meat quality. The influence of replacement of dietary animal fat by vegetable oil on meat quality of pigs has been described by various investigations (Wiseman and Agunbiade, 1998; Miller et al., 1990; Wood et al., 2004; Mitchaothai et al., 2007). However, the various studies on meat quality can be criticized because the pigs had free access to diets containing equal inclusion percentages of either animal of vegetable fat. The well-known difference in digestibility between animal fat and vegetable oil (Cera et al., 1989) interferes with the interpretation of the results. Under those conditions, the dietary content of metabolizable energy from vegetable fat is greater than that from animal fat. As a consequence, the pigs fed the diet with vegetable fat would consume a diet containing less protein and carbohydrates, when expressed as a percentage of dietary metabolizable energy. This in turn affects de-novo fatty acid synthesis and thus the fatty acid composition of tissues and thereby meat quality. A similar reasoning would hold for a difference in feed intake only. The diets and feeding regimen used in this study should allow a proper comparison of feeding either SO or BT as fat source.

The main fatty acids in the diet containing BT diet were stearic, palmitic, and oleic acid, whereas the SO diet was very rich in linoleic acid. The difference in oleic acid content between the two diets was relatively small. Thus, the BT diet can be considered representative for diets rich in saturated fatty acids, whereas the SO diet may represent diets rich in n-6 polyunsaturated fatty acids. The diets were identical in terms of percentages of metabolizable energy provided by the macronutrients. The two experimental diets were fed on an iso-energetic basis. Indeed, the pigs fed the BT or SO diet showed no difference in growth performance. It may thus be concluded that in the present experiment the only dietary variable was the amount of dietary energy in the form of either BT or SO.

Carcass quality traits did not differ for the two dietary fat sources. This outcome agrees with reports published previously (Miller et al., 1990; Bee et al., 2002; Nuernberg et al., 2005; Mitchaothai et al., 2007). Meat quality characteristics traits, except for the Japanese colour score, were not statistically different between the BT and SO treatments. These results agree with those of Scheeder et al. (2000) who found that feeding 7% pork fat, 4.95% olive oil or 3.17% soybean oil to growing-finishing pigs did not affect pH, cooking losses, texture, or colour of pork. The higher Japanese colour score of the pork in the SO treatment may relate to the somewhat higher redness (colour **a*** value). It might imply that meat colour assessment by eye is more sensitive than that done technically. In keeping with the present results, Miller et al. (1990) also found no difference in drip losses , cooking losses, shear force, and marbling between pigs fed either animal fat or sunflower oil. The sarcomere length of

pork of both dietary treatments (1.86 to 1.90 μm) was in the normal range (1.50 to 2.0 μm) for longissimus muscle (Wheeler et al., 2000). The intramuscular fat or marbling of loin was not statistically different between both treatments which are in accordance with the results of Nuernberg et al. (2005) when the effect of olive and linseed oil were compared. On the basis of the results for carcass and meat quality, it may be concluded that supplementation of SO instead of BT to the diet did not alter the quality of pig meat.

The relative concentrations of myristic (C14:0), palmitic (C16:0), margaric (C17:0), and stearic (C18:0) in adipose tissues were higher after feeding the BT diet. This is explained by the larger fractions of these fatty acids in the BT diet. However, in loin of the pigs fed the BT diet the incorporation of palmitic and stearic acid was not increased. Apparently, there are differences in the efficiency of deposition in different tissues for the various fatty acids. Both adipose tissues and loin of the pigs fed the BT diet instead of the SO diet contained a higher oleic fraction, even though the amount of this fatty acid in the BT diet was smaller than in the SO diet. This could relate to diet-induced differences in hydrogenation in liver and preferential utilization for energy generation of oleic acid. The relative concentration of linoleic acid in adipose tissues and loin of the pigs fed the SO diet was greater than that of the pigs fed the BT diet, but the concentration of α-linolenic acid was lower. The higher and lower incorporation of linoleic acid and α-linolenic acid in pigs fed the SO can be explained by the higher and lower intakes of these essential fatty acids.

The crude fat component of the SO diet had a higher apparent digestibility than had this component of the BT diet, but there were no differences in the apparent digestibilities of dry matter (DM) and crude protein (CP) between both diets. In a previous study (Mitchaothai et al., *in press*) we found higher apparent digestibilities of DM, CP, and crude fiber (CF) when growing-finishing pigs were fed a diet containing SO instead of BT. In our previous study, the pigs had *ad libitum* access to the experimental diets. It is difficult to see why ad libitum feeding instead of restricted feeding would increase the digestibility of DM and CP in a diet containing SO. The superior digestibility of oils rich in linoleic acid, as opposed to fats rich in saturated fatty acids, is well known (Cera et al., 1989; Li et al., 1990) and probably relates to a more efficient incorporation of polyunsaturated fatty acids into micelles (Garrett and Young, 1975). As mentioned above, the higher fat digestibility of the SO was taken into account when formulating the diets. Furthermore, the BT and SO diets were fed on an iso-energetic basis. This explains why growth performance of the pigs fed either the SO or BT diet was similar.

Feeding the two fat sources was associated with marked differences in the apparent digestibility of individual fatty acids. Palmitic and linoleic acid in the SO diet were digested more efficiently than these fatty acids in the BT diet. On the other hand, stearic acid in the SO diet was less well digested than stearic acid in the BT diet. There were no diet effects on the digestibilities of oleic and α-linolenic acid. A combination of different factors may be responsible for the observed diet-induced differences in apparent digestibility of identical fatty acids. As mentioned above, the total fat digestibility of SO was greater than that of BT, which may be associated with enhanced micelle formation after feeding the SO diet. An improved micelle formation may favourably influence the digestion of all fatty acids in the diet. The position of a given fatty acid in the triacylglycerol molecule also plays a role. Fatty acids at the 2 position of glycerol in triacylglycerol molecules are better digested than those at the 1,3 position (Bracco, 1994; Innis and Dyer, 1997; Nelson and Innis, 1999; Scheeder et al., 2000; Scheeder et al., 2003). During digestion, the pancreatic lipase specifically

removes fatty acids at the 1,3 position while the resulting monoacylglycerol molecule is efficiently incorporated into micelles (Lien, 1994), leading to preferential absorption of fatty acids at the 2 position of the glycerol backbone of triacylglycerols. The intake level of a given individual fatty acid and its fecal excretion of endogenous origin will also affect the calculated apparent digestibility. A low intake level in combination with a high endogenous excretion will by itself lead to a low apparent digestibility. When comparing the digestibilities of palmitic and stearic acid for the SO and BT diet, the values for the SO diet may be biased towards lower values because the intake levels were lower. This may hold especially for stearic acid, which yielded a negative apparent digestibility for the SO treatment. On the other hand, the apparent digestibility of linoleic acid for the SO diet may have been biased towards a higher value.

The calculated intake of digestible fatty acids reflects the amount in the diet and feed intake, combined with the measured apparent digestibility. The deposition in the body of fatty acids was calculated as based on the fatty acid composition of the total body fat that was gained during the entire feeding period. As would be expected, the pigs fed the SO diet deposited more linoleic acid and those fed BT had deposited more stearic acid in their whole body. Similar data have been shown for mice (Javadi et al., 2004) and goats (Yeom et al., 2005). To obtain clues as to preferential deposition of fatty acids, the ratio of deposition of a given fatty acid or group of fatty acids to the intake of digestible fatty acid or group of fatty acids was calculated. The deposition:intake ratio for fatty acid reflects the net amount synthesized and/or the proportion of the digested fatty acid not oxidized. A deposition:intake ratio > 1 would point at net *de novo* synthesis, whereas a ratio < 1 would indicate net oxidation. The low deposition:intake ratio for linoleic acid in the pigs fed SO is consistent with the well-known preferential oxidation of linoleic acid (Jones et al., 1985; Cunnane and Anderson, 1997; Yeom et al., 2005) and the fact that linoleic acid cannot be synthesized by pigs (Azain, 2000; Nguyen et al., 2005). The deposition:intake ratio for the essential polyunsaturated fatty acids, linoleic and α-linolenic acid, cannot be higher than 1. Indeed, the ratios for α-linolenic acid were below and so was the ratio for linoleic acid in the pigs fed the SO diet. The pigs fed BT had a group mean deposition:intake ratio for linoleic acid that was just above 1, but was not significantly higher than 1. The negative deposition:intake ratio for stearic acid in pigs fed the SO is a consequence of the negative apparent digestibility that was calculated.

The pigs fed SO instead of BT had a higher deposition:intake ratio for SFA, but lower ratio for MUFA and no difference for the ratio of PUFA. The diet effect on the ratios for SFA and MUFA can be explained by the calculated minimum synthesis of these groups of fatty acids. Feeding the diet containing SO stimulated the synthesis of SFA, but tended to depress that of MUFA. The higher synthesis ratio for SFA:(SFA+MUFA) in pigs fed the SO diet indicates that there was selective synthesis of SFA in the pigs fed the SO diet. This might point at *de novo* fatty acid synthesis being regulated to balance the amount of saturated and unsaturated fatty acids in the body because the intake of linoleic acid was very high in the pigs fed the diet with SO.

In conclusion, the iso-energetic replacement of BT by SO had a marked impact of the fatty acid composition of tissues, but did not affect carcass and meat quality traits. Feeding the diet with SO produced a markedly increased deposition of linoleic acid in the adipose tissues, loin muscle, and whole body. After calculating the deposition:digestible intake ratio for individual fatty acids, it became clear that the

type of dietary fat had marked, specific effects on the synthesis and oxidation of fatty acids.

ACKNOWLEDGEMENTS

The authors gratefully thank Kittipong Chai-ya-chet, Naphaporn Mungmart, and Yupha Leethong for their excellent assistance, the Faculty of Natural Resources, Rajamangala University of Technology-Isan for providing facilities and Mahanakorn University of Technology for funding.

LITERATURE CITED

AOAC. 1990. Association of official analysis chemists, Washington DC.

Azain, M.J. 2000. Fat in swine nutrition. 2nd ed. CRC press, New York, USA.

Bee, G., S. Gebert, and R. Messikommer. 2002. Effect of dietary energy supply and fat source on the fatty acid pattern of adipose and lean tissues and lipogenesis in the pig. J. Anim. Sci. 80:1564-1574.

Bracco, U. 1994. Effect of triglyceride structure on fat absorption. Am. J. Clin. Nutr. 60:1002S-1009S.

Cera, K.R., D.C. Mahan, and G.A. Reinhart. 1988. Weekly digestibilities of diets supplemented with corn oil, lard or tallow by weanling swine. J. Anim. Sci. 66:1430-1437.

Cera, K.R., D.C. Mahan, and G.A. Reinhart. 1989. Apparent fat digestibilities and performance responses of postweaning swine fed diets supplemented with coconut oil, corn oil or tallow. J. Anim. Sci. 67:2040-2047.

Cunnane, S.C., and M.J. Anderson. 1997. The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. J. Nutr. 127:146-152.

Enser, M., R.I. Richardson, J.D. Wood, B.P. Gill, and P.R. Sheard. 2000. Feeding linseed to increase the n-3 PUFA of pork: fatty acid composition of muscle, adipose tissue, liver and sausages. Meat Sci. 55:201-212.

Garrett, R.L., and J. Young. 1975. Effect of micelle formation on the absorption of neutral fat and fatty acids by the chicken. J. Nutr. 105:827-838.

Goerl, K.F., S.J. Eilert, R.W. Mandigo, H.Y. Chen, and P.S. Miller. 1995. Pork characteristics as affected by two populations of swine and six crude protein levels. J. Anim. Sci. 73:3621-3626.

Guo, Q., B.T. Richert, J.R. Burgess, D.M. Webel, D.E. Orr, M. Blair, G.E. Fitzner, D.D. Hall, A.L. Grant, and D.E. Gerrard. 2006. Effects of dietary vitamin E and fat supplementation on pork quality. J. Anim. Sci. 84:3089-3099.

Honikel, K.O. 1987. How to measure the water-holding capacity of meat? Recommendation of standardized methods, Page 129 in Evaluation and control of meat quality in pigs. Martinus Nijhoff Publishers, The Netherlands, Dordrecht.

Horwitz, W. 1975. Official Methods of Analysis of Official Analytical Chemist, Page 225. Official Methods of Analysis of Official Analytical Chemist, 12 ed. Benjamin Franklin Station, DC.

Innis, S.M., and R. Dyer. 1997. Dietary triacylglycerols with palmitic acid (16:0) in the 2-position increase 16:0 in the 2-position of plasma and chylomicron triacylglycerols, but reduce phospholipid arachidonic and docosahexaenoic acids, and alter cholesteryl ester metabolism in formula-Fed piglets. J. Nutr. 127:1311-1319.

Irie, M., and M. Sakimoto. 1992. Fat characteristics of pigs fed fish oil containing eicosapentaenoic and docosahexaenoic acids. J. Anim. Sci. 70:470-477.

Javadi, M., H. Everts, R. Hovenier, S. Kocsis, A.E. Lankhorst, A.G. Lemmens, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2004. The effect of six different C18 fatty acids on body fat and energy metabolism in mice. Br. J. Nutr. 92:391-399.

Jones, P.J.H., P.B. Pencharz, and M.T. Clandinin. 1985. Whole body oxidation of dietary fatty acids: implications for energy utilization. Am. J. Clin. Nutr. 42:769-777.

Li, D.F., R.C. Thaler, J.L. Nelssen, D.L. Harmon, G.L. Allee, and T.L. Weeden. 1990. Effect of fat sources and combinations on starter pig performance, nutrient digestibility and intestinal morphology. J. Anim. Sci. 68:3694-3704.

Lien, E.L. 1994. The role of fatty acid composition and positional distribution in fat absorption in infants. J. Pediatr. 125:S62-S68.

Metcalfe, L.D., A.A. Schmitz, and J.R. Pelka. 1966. Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. Anal. Chem. 38:514-515.

Miller, M.F., S.D. Shackelford, K.D. Hayden, and J.O. Reagan. 1990. Determination of the alteration in fatty acid profiles, sensory characteristics and carcass traits of swine fed elevated levels of monounsaturated fats in the diet. J. Anim. Sci. 68:1624-1631.

Mitchaothai, J., C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsutthavas, P. Srenanul, R. Hovenier, H. Everts, and A.C. Beynen. 2007. Effect of dietary fat type on meat quality and fatty acid composition of various tissues in growing–finishing swine. Meat Sci. 76:95-101.

Mitchaothai, J., H. Everts, C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsuthavas, P. Srenanul, R. Hovenier, and A.C. Beynen. Digestion and deposition of individual fatty acids in growing-finishing pigs fed diets containing either beef tallow of sunflower oil. Journal of Animal Physiology and Animal Nutrition. *(In press).*

Monin, G., C. Larzul, P. Le Roy, J. Culioli, J. Mourot, S. Rousset-Akrim, A. Talmant, C. Touraille, and P. Sellier. 1999. Effects of the halothane genotype and slaughter weight on texture of pork. J. Anim. Sci. 77:408-415.

Nakai, H., F. Saito, T. Ikeda, S. Ando, and A. Komatsu. 1975. Standard models of pork colour. Bull. Natl. Inst. Anim. Ind. 29:69.

Nelson, C.M., and S.M. Innis. 1999. Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. Am. J. Clin. Nutr. 70:62-69.

Nguyen, L.Q., H. Everts, and A.C. Beynen. 2005. Intake of essential fatty acids by growing-finishing pigs kept on smallholdings in central Vietnam. Trop. Anim. Health Prod. 37:65-76.

Nguyen, L.Q., M.C.G.A. Nuijens, H. Everts, N. Salden, and A.C. Beynen. 2003. Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. Meat Sci. 65:1399-1406.

Nuernberg, K., K. Fischer, G. Nuernberg, U. Kuechenmeister, D. Klosowska, G. Eliminowska-Wenda, I. Fiedler, and K. Ender. 2005. Effects of dietary olive and linseed oil on lipid composition, meat quality, sensory characteristics and muscle structure in pigs. Meat Sci. 70:63-74.

Otten, W., C. Wirth, P.A. Iaizzo, and H.M. Eichinger. 1993. A high omega 3 fatty acid diet alters fatty acid composition of heart, liver, kidney, adipose tissue and skeletal muscle in swine. Ann. Nutr. Metab. 37:134-141.

Pfeiffer, H., and H. Falkenberg. 1972. Masse am Lendenspiegel zur objektiven Ermittlung der SchlachtkOrper Zusammensetzung beim Schwein. Tierzucht 26:466-467.

Ramirez, M.R., M. Estevez, D. Morcuende, and R. Cava. 2004. Effect of the type of frying culinary fat on volatile compounds isolated in fried pork loin chops by using SPME-GC-MS. J. Agric. Food. Chem. 52:7637-7643.

Rey, A.I., J.P. Kerry, P.B. Lynch, C.J. Lopez-Bote, D.J. Buckley, and P.A. Morrissey. 2001. Effect of dietary oils and alpha-tocopheryl acetate supplementation on lipid (TBARS) and cholesterol oxidation in cooked pork. J. Anim. Sci. 79:1201-1208.

Scheeder, M., K.R. Gläser, B. Eichenberger, and C. Wenk. 2000. Influence of different fats in pig feed on fatty acids composition of phospholipids and physical meat quality characteristics. Eur. J. Lipid Sci. Technol. 102:391-401.

Scheeder, M., D. Gumy, R. Messikommer, C. Wenk, and P. Lambelet. 2003. Effect of PUFA at sn-2 position in dietary triacylglycerols on the fatty acid composition of adipose tissues in non-ruminant farm animals. Eur. J. Lipid Sci. Technol. 105:74-82.

Schinckel, A.P., J.R. Wagner, J.C. Forrest, and M.E. Einstein. 2001. Evaluation of alternative measures of pork carcass composition. J. Anim. Sci. 79:1093-1119.

Shields, R.G., Jr., D.C. Mahan, and P.L. Graham. 1983. Changes in swine body composition from birth to 145 kg. J. Anim. Sci. 57:43-54.

SPSS, 1999. SPSS for Windows, SPSS Inc., Chicago, IL, USA

Sripromma, J. 1984. Einfluss der Fütterungsintensität und der Körpermasse auf die Schlachtkörper Qualität beim Schwein. Ph.D. Thesis, Hohemheim University, Stuttgard, Germany.

Wagner, J.R., A.P. Schinckel, W. Chen, J.C. Forrest, and B.L. Coe. 1999. Analysis of body composition changes of swine during growth and development. J. Anim. Sci. 77:1442-1466.

Wheeler, T.L., S.D. Shackelford, and M. Koohmaraie. 2000. Variation in proteolysis, sarcomere length, collagen content, and tenderness among major pork muscles. J. Anim. Sci. 78:958-965.

Wiseman, J., and J.A. Agunbiade. 1998. The influence of changes in dietary fat and oils on fatty acid profiles of carcass fat in finishing pigs. Livest. Prod. Sci. 54:217-227.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: a review. Meat Sci. 66:21-32.

Yeom, K.-H., J.T. Schonewille, H. Everts, K.-W. Lee, G. van Trierum, and A.C. Beynen. 2005. Growth performance and body composition of goat kids fed milk replacers with increasing levels of linoleic acid. J. Anim. Physiol. Anim. Nutr. 89:29-34.

CHAPTER 5

Fatty acid metabolism in Thai indigenous pigs fed diets with low or high linoleic acid : α-linolenic acid ratio

J. Mitchaothai†§, H. Everts†, C. Yuangklang‡, S. Wittayakun‡, K. Vasupen‡, S. Wongsuthavas‡, P. Srenanul‡, R. Hovenier†, and A. C. Beynen†

§Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand

‡Faculty of Natural Resources, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand

†Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, P.O. Box 50.152, 3508 TD, Utrecht, The Netherlands

To be submitted

ABSTRACT

Twenty six Thai indigenous Kadon pigs (13 males and 13 females), with an average BW of 10.51 ± 1.40 kg were used to investigate the effect of the dietary linoleic acid (LA) : α-linolenic acid (ALA) ratio on meat quality, fatty acid digestibility and deposition, body composition and energy balance. The pigs were either fed a diet low in ALA, containing 4.5 % (w/w) soy bean oil + 0.5 % linseed oil, or a diet high in ALA, containing 0.5 % soya bean oil + 4.5 %linseed oil. The pigs were kept in individual cages and fed *ad libitum* for 35 days. In selected tissues and whole carcass of the pigs fed the low-ALA diet, more linoleic acid was deposited, whereas the pigs fed the high-ALA diet had deposited more α-linolenic acid. There were no diet effects on growth performance, digestibility of gross energy, crude fat, and fatty acids, and on energy balance. Pigs fed the high-ALA diet showed a higher ALA digestibility. The percentage of fat in the whole body of the pigs fed the high-ALA diet was lower than in that of their counterparts fed the low-ALA diet. There was a tendency for the pigs fed the high-ALA diet to have a lower body-fat content and lower fat deposition. The total absorption and deposition of LA during the entire feeding period were higher for the low-ALA diet. Total absorption and deposition of ALA were higher for the high-ALA group. The incorporation rate of LA into the whole body was higher than the rate of ALA. LA and ALA were preferentially oxidized or transformed compared to other fatty acids. The calculated minimum *de novo* synthesis of MUFA tended to be decreased for the high-ALA diet, but that of SFA was not affected by dietary treatment. In conclusion, the dietary levels of LA and ALA had no effect no growth performance, macronutrient digestibility and energy balance. LA deposition in tissues and the whole body was increased after feeding the high-LA diet and ALA deposition was increased after feeding the high- ALA diet, but the rate of ALA deposition was less than that of LA.

Keywords: deposition, digestibility, linoleic acid, α-linolenic acid, indigenous pigs

INTRODUCTION

Consumers increasingly prefer animal products containing high levels of polyunsaturated fatty acids because of the assumed health benefits. Linoleic (C18:2 n-6; LA) and α-linolenic (C18:3 n-3; ALA) acid are essential fatty acids for pigs as they cannot be synthesized in the body of pigs. The dietary concentrations of LA and ALA are directly associated with the concentrations of these fatty acids in the adipose tissues and intramuscular fat of pigs (Nguyen et al., 2003). So far the relationship between the quantitative intake of LA and ALA and the deposition of these fatty acids in the whole body of pigs has not been described.

ALA is more preferentially oxidized than is LA (Cunnane and Anderson, 1997). The preferential oxidation of ALA should result in less deposition of ALA in the body and it should affect energy expenditure or storage in body. Whole carcass analysis for fatty acid composition and energy balance has been performed in various animal species (Crespo and Esteve-Garcia, 2002a; Javadi et al., 2007a; Yeom et al., 2005). However, whole carcass analysis in pigs usually concerns proximate analysis (Shields et al., 1983; Wagner et al., 1999), rather than fatty acid composition. In this study, we used Thai indigenous Kadon pigs for whole carcass analysis, because these pigs are relatively small. It is assumed that fatty acid and energy metabolism of the Kadon pig is similar to that of the commercial pigs. Using the indigenous pigs we tested the hypothesis that feeding a diet low LA:ALA ratio versus highLA:ALA ratio would alter the whole body fatty acid metabolism and deposition and energy expenditure.

MATERIALS AND METHODS

Animals, Diets and Feeding

Twenty-six male and female Thai indigenous Kadon pigs were used. The BW was 10.51 ± 1.40 kg. Three males and three females were selected to be slaughtered for baseline measurements. Then, the remaining 20 pigs were allotted to one of the two dietary treatments on the basis of body weight and sex and were housed in individual cages. The pigs were allowed *ad libitum* access to feed and water throughout the experiment. Feed consumption was recorded twice a day and individual body weight of the pigs was measured weekly. There were two experimental diets either low in ALA (4.5 % soybean oil and 0.5% linseed oil) or high in ALA (0.5 % soybean oil & 4.5% linseed oil) as shown in Table 1. Throughout the entire experiment, faeces and urine samples of all pigs were collected to determine digestibility and balance of macronutrients and gross energy. A 10% solution of sulphuric acid (H_2SO_4) was used to prevent ammonia evaporation during urine collection and the urine samples were stored at 5 ^oC until analysis. All pigs were slaughtered at an average body weight of 20.62 ± 3.02 kg. Ingredient and macronutrient composition of the experimental diets are shown in Table 1. The fatty acid composition of the diets is documented in Table 2.

Collection and Pre-treatment of Samples

The six pigs (3 males and 3 females) for baseline measurements were killed by an intravenous injection of sodium pentobarbital (200 mg/kg of body weight). The pigs were scalded, blood was collected and weighed, the head was removed, and the

carcass was split into halves along the median plane according to the procedure previously reported by Shields et al. (1983). At this moment a few small samples (1-2 g) of adipose tissue and loin were taken and stored at – 20 ^{o}C until analysis. This procedure was modified by drying compartment samples and both sides of the sawed carcass at 60 ^{o}C for 1 week in a forced-hot air oven with placing the frozen materials on a mesh with an underneath tray to collect the dripping oil. The dried whole carcass samples derived from each pig were pooled and then weighed. The dripping oil samples were pooled for each pig and weighed in the same way as the dried whole carcass samples. The dried samples were individually ground once through a 2 mm screen and twice through a 1 mm screen by a hammer mill. The 20 pigs in the feeding trial were sacrificed at 5 weeks after the commencement. Then, whole carcass sample was prepared in the same way as for the baseline pigs.

Table 1.
Composition of the experimental diets (as-fed basis) and analysed macronutrients

	Low ALA	High ALA
Raw materials (%)		
Cassava chips	45.53	45.53
Soybean meal (40% CP)	40.00	40.00
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Di-calcium phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Soya bean oil (SO)	4.50	0.50
Linseed oil (LO)	0.50	4.50
Premix	0.25	0.25
Analysed macro nutrients (%)		
Dry matter	88.59	88.77
Crude protein	18.20	18.21
Crude fat	7.04	7.05
Crude fiber	5.02	4.74
Ash	12.44	12.08
Calculated ME (MJ/kg)	13.93	13.96

Chemical Analyses and Fatty Acid Composition Determination

The samples of diets and faeces were dried at 60 ^{o}C for 72 h in a forced-hot air oven. The dried diets, faeces, and whole carcass samples were analysed for moisture, energy, crude protein, crude fat, and ash (AOAC, 1990). The urine samples were also analysed to quantify the content of energy and nitrogen (AOAC, 1990).

The fresh loin and the dried samples of diets, faeces, and whole carcasses samples were extracted as described previously (Horwitz, 1975). Each sample was added to a flask and 2 ml of ethanol was added. Subsequently, 10 ml of HCl (8 mol/L) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80 ^{o}C for 30-40 min. The tubes were cooled down, 10 ml of ethanol (96%, w/w) and 25 ml of petroleum ether (boiling point between 40 and 60 ^{o}C) were added and the tube was shaken for one min. The fat-containing upper layer was decanted into a 150-ml round-bottom flask. The extraction procedure was repeated twice with 15 ml

of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath of 40 ^oC. The round-bottom flasks with the lipids were dried overnight at 60 ^oC and the total lipids were measured gravimetrically. Total lipids, the small samples of adipose tissue and the dripping oil were saponified and methylated according to the procedure of Metcalfe et al. (1966) followed by gas chromatography for determination of fatty acid composition (Javadi et al., 2004).

Determination of Body Composition and Energy Balance

The body weight and composition of the baseline pigs showed no effect of sex, hence the average energy or nutrient content in whole body of the baseline pigs was assumed to be the similar fraction in the whole body of the experimental pigs at the start. All types of samples including the dripping oil were determined with a bomb calorimeter (Parr® Oxygen Bomb Calorimeter 1341 EE adiabatic, Parr Instrument Company, Moline, Illinois, USA). As a thermo-chemical standard, benzoic acid was used (Asian Pacific Specialty Chemical Limited, NSW, Australia). The total amount of energy that was lost as heat (heat production or energy expenditure) was calculated with the formula: Energy lost as heat = energy in food – energy in faeces and urine – energy stored in body. Energy stored in the body was determined as total energy at the end of the 35 d feeding period minus the energy in the body at the beginning of the experiment (= individual body weight × mean energy content). The same procedure was used to calculate the water, protein, fat and ash retention.

Relationship between dietary fatty acid and adipose tissue

The regression formulas for the relations between dietary linoleic and α-linolenic acids (g / MJ ME) and adipose tissue linoleic and α-linolenic acids (% of total fat) were built to compare with the previous results (Nguyen et al., 2003). However, in the present study, we have investigated two levels of LA and ALA. Due to this design it was not appropriate to do a regression analysis between the fraction of stored fatty acids and the fatty acid content of the diet. In the current experiment, the incorporation in absolute amounts was quantified by the use of the comparative slaughter technique. Instead of the regression analysis, the slope between fatty acid deposited in the body and digestible fatty acid intake (Δ g fatty acid incorporated / Δ g fatty acid digested) could be calculated.

Calculation of Digestible Fatty Acid Intake, Fatty Acid Deposition and Minimum De novo Synthesis

The total digestible fatty acid intake was calculated as fatty acid intake (g/35 days) × 0.95 × apparent fatty acid digestibility (fraction of intake). The deposition of fatty acids was calculated with the following formula: Fatty acid deposition (g/35 days) = individual carcass content of fatty acid at the end of the study – individual carcass content of fatty acid at the start (= BW × 0.95 × mean fatty acid composition of the baseline pigs) of the study. Carcass content of fatty acid at the end (g) was calculated as the whole carcass fat mass (g) × 0.95 × fraction of fatty acid in whole carcass. The ratio of fatty acid deposition and digestible fatty acid intake was calculated for selected individual fatty acids and groups of fatty acids. The quantity of fatty acids oxidized, transformed and synthesized could not be measured. Therefore,

minimum *de novo* fatty acid synthesis was calculated as fatty acid deposition minus digestible fatty acid intake.

Statistical Analyses

The effect of α-linolenic acid : linoleic acid ratio in the diets was evaluated for statistical significance by the Student's *t* test (SPSS, 1999) and SAS programme (SAS, 1996). Results are expressed as means ± SD.

RESULTS

Dietary Fatty Acid Composition

The fatty composition of the experimental diets is illustrated in Table 2. The fat component of the low-ALA and high-ALA diets contained similar patterns of total saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA). The major difference between both experimental diets was a high LA fraction (46 % of total fatty acids) in the low-ALA diet and a high ALA fraction (31% of total fatty acids) in the high-ALA diet.

Fatty Acid Composition of Adipose Tissues and Loin Muscle

Fatty acid pattern of fat in adipose tissues and loin are demonstrated in Table 3. After feeding the experimental diets, the relative concentration of SFA (mainly C16:0 and C18:0) in adipose tissues and loin was not different between the low-ALA and the high-ALA diet. The relative concentration of MUFA (main component C18:1 n-9) of the investigated tissues also was not different between two experimental diets. As to the PUFA concentration in both adipose tissues and loin muscle, the LA concentration in the pigs fed the low-ALA diet was higher ($p < 0.001$) than in those fed the high-ALA diet, whereas the concentration of ALA in the pigs fed the high-ALA diet was higher ($p < 0.001$) than in those fed the low-ALA diet. The relative concentrations of the other longer chain unsaturated fatty acids tended to higher for the n-6 PUFAs in the pigs fed the low-ALA diet and tended to be higher for the n-3 PUFAs in the pigs fed the high-ALA diet, even though the fraction of the longer chain n-3 PUFAs was quite low. When considering the groups of fatty acids, the concentrations of SFA, MUFA, and PUFA, including the ratios of MUFA/SFA and PUFA/SFA, showed no difference between the two dietary treatments. Conversely, the ratios of n-6: n-3 PUFA and C18:2 n-6: C18:3 n-3 in both adipose tissues and loin of the pigs fed the low-ALA diet ranged between 8 and 10 and in the pigs fed the high ALA-diet it ranged between 1 and 2. The ratios for the low-ALA treatment were significantly higher ($p < 0.001$) than those for the high-ALA treatment.

Growth Performance and Nutrient Digestibility

The type of dietary fat had no effect on feed intake, growth (average daily gain), feed conversion ratio (feed:gain) and final BW (Table 4). There was neither a diet effect on the apparent faecal digestibility of crude fat and gross energy (Table 4) nor on the digestibility of individual or grouped fatty acids (Table 5), except for higher ALA and n-3 PUFA digestibility in the pigs fed the high-ALA diet.

Table 2.
Fatty acid composition of the experimental diets

	Low ALA	High ALA
Fatty acids, g methylester/100 g methylesters		
C 10:0	0.16	0.16
C 14:0	0.19	0.12
C 16:0	15.95	11.80
C 17:0	0.17	0.15
C 18:0	4.42	4.29
C 20:0	0.35	0.39
C 22:0	0.28	0.28
C 24:0	0.17	0.17
C 16:1	0.12	0.08
C 17:1	0.03	0.00
C 18:1 n-9	21.13	21.59
C 18:1 n-7	0.51	0.51
C 20:1 n-9	0.14	0.14
C 18:2 n-6	46.15	26.96
C 18:3 n-3	6.92	30.96
C 18:3 n-6	0.89	0.15
C 20:3 n-3	0.12	0.03
C 20:3 n-6	0.02	0.02
C 20:5 n-3	0.16	0.16
C 22:6 n-3	0.08	0.08
unknown	2.00	1.94
Σ SFA	21.70	17.37
Σ MUFA	21.94	22.33
Σ PUFA	54.35	58.36

SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids

Σ SFA = C10:0 + C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0

Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9

Σ PUFA = C18:2 n-6 + C18:3 n-6 + C18:3 n-3 + C18:4 n-3 + C20:2 n-6 + C20:3 n-6 + C20:4 n-6 + C20:3 n-3 + C20:5 n-3 + C22:2 n-6 + C22:4 n-6 + C22:3 + C22:5 n-3 + C22:6 n-3

Body Composition and Energy Balance

Table 4 shows that the deposition of macronutrients (water, crude protein and ash) in whole body of the pigs was not different between the dietary treatments, except that the percentage of crude fat in the pigs fed the higher-ALA diet was lower than in those fed the low-ALA diet. Moreover, the absolute amount of deposition of crude fat for the high-ALA treatment tended to be lower (p = 0.1195) than for the low- ALA treatment.

Energy intake, storage, expenditure, and losses with faeces and urine showed no difference between the two dietary treatments in terms of absolute values or as percentage of energy intake. Table 4 indicates that the final amount of energy in the whole body as derived from measurement by bomb calorimeter was close to that calculated from the equation used by Javadi et al. (2004).

Table 3.
Fatty acid composition of adipose tissue and loin intramuscular fat

	Adipose tissue		Signi-ficance	Loin		Signi-ficance
	Low ALA	High ALA		Low ALA	High ALA	
Analysed fatty acids (g/100g methylester)						
C 14:0	1.32± 0.29	1.39± 0.22	NS	1.37± 0.24	1.37± 0.23	NS
C 16:0	22.82± 1.95	22.44± 0.66	NS	24.03± 1.16	23.38± 0.79	NS
C 17:0	0.28± 0.08	0.28± 0.07	NS	0.31± 0.09	0.30± 0.05	NS
C 18:0	13.35± 1.52	12.64± 1.10	NS	12.84± 1.40	12.90± 1.34	NS
C 20:0	0.23± 0.09	0.25± 0.03	NS	0.22± 0.08	0.26± 0.04	NS
C 16:1	1.34± 0.31	1.49± 0.16	NS	1.75± 0.44	1.70± 0.25	NS
C 18:1 n-9	33.03± 2.29	33.88± 2.51	NS	33.84± 2.30	34.70± 2.10	NS
C 18:1 n-7	1.96± 0.14	1.94± 0.14	NS	2.50± 0.27	2.28± 0.23	NS
C 20:1 n-9	0.70± 0.14	0.69± 0.10	NS	0.76± 0.10	0.75± 0.10	NS
C 18:2 n-6	20.19± 3.72	13.74± 1.45	<0.001	16.78± 2.48	11.98± 1.19	<0.001
C 18:3 n-3	2.25± 0.45	8.56± 1.98	<0.001	1.68± 0.29	6.79± 1.56	<0.001
C 20:2 n-6	0.76± 0.13	0.52± 0.05	<0.001	0.61± 0.10	0.44± 0.05	<0.001
C 20:4 n-6	0.21± 0.08	0.19± 0.03	NS	0.74± 0.40	0.47± 0.17	NS
C 20:3 n-3	0.25± 0.08	1.05± 0.16	<0.001	0.21± 0.03	0.90± 0.14	<0.001
C 22:5 n-3	0.04± 0.05	0.20± 0.05	<0.001	0.16± 0.13	0.35± 0.08	<0.001
unidentified	1.04± 0.28	0.50± 0.12	<0.001	1.63± 0.47	0.87± 0.31	<0.001
SFA	38.00± 3.03	36.99± 1.31	NS	38.77± 1.85	38.21± 1.73	NS
MUFA	37.19± 2.53	38.18± 2.50	NS	39.24± 2.57	39.73± 2.10	NS
PUFA	23.78± 4.29	24.33± 3.15	NS	20.36± 3.11	21.19± 2.54	NS
MUFA/SFA	0.98± 0.09	1.03± 0.06	NS	1.01± 0.07	1.04± 0.08	NS
PUFA/SFA	0.64± 0.16	0.66± 0.10	NS	0.53± 0.10	0.56± 0.09	NS
n-6/n-3	8.42± 0.84	1.51± 0.27	<0.001	9.04± 0.90	1.61± 0.31	<0.001
18:2 n-6/18:3 n-3	9.03± 0.79	1.66± 0.32	<0.001	10.12± 1.15	1.83± 0.37	<0.001

Means ± SD for 10 pigs per experimental diet
NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid

Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-6 + C18:3 n-3 + C20:3 n-6 + C20:3 n-3 + C20:5 n-3 + C22:6 n-3

Relationship between dietary and adipose tissue fatty acids

The relationships between dietary and adipose fatty acids concerning LA and ALA are given in the Table 6 and illustrated in Figure 1. The slope of the regression for LA was higher than that for ALA. The ratio of Δ g LA deposited:Δ g digestible LA intake was 0.488 and that of Δ ALA deposition:Δ digestible ALA intake was 0.358.

Digestible Fatty Acid Intake and Deposition

Table 4.
Body composition and energy balance in Thai native pigs fed either a low or high ALA-diet for 35 d.

	Low ALA	High ALA	*P* value
Food intake (kg dry food/35 d)	24.80 ± 2.20	24.31 ± 1.46	0.5846
Feed conversion	2.48 ± 0.40	2.32 ± 0.46	0.4613
Initial body weight (kg)	10.41 ± 1.89	10.74 ± 1.28	0.6486
Final body weight (kg)	20.38 ± 2.41	20.86 ± 3.64	0.7329
Average daily gain (g)	290 ± 34	309 ± 59	0.4294
Apparent fat digestibility (%)	88.28 ± 3.64	88.89 ± 3.33	0.5397
Apparent gross energy digestibility (%)	91.02 ± 1.78	90.12 ± 1.57	0.2693
Body composition at the end			
Fat (kg)	2.95 ± 0.39	2.67 ± 0.34	0.1195
Water (kg)	13.09 ± 1.36	14.17 ± 1.97	0.1982
Protein (kg)	3.29 ± 0.46	3.32 ± 0.50	0.8917
Ash (kg)	0.70 ± 0.16	0.62 ± 0.12	0.2573
Recovery (%)	95.39 ± 1.75	95.35 ± 1.47	0.9590
Water : Protein ratio (kg/kg)	4.04 ± 0.67	4.30 ± 0.61	0.3932
Fat (%)	14.09 ± 1.98	12.31 ± 1.47	0.0472
Water (%)	62.29 ± 4.06	64.97 ± 2.64	0.1203
Protein (%)	15.68 ± 1.75	15.31 ± 1.76	0.6681
Ash (%)	3.33 ± 0.68	2.88 ± 0.58	0.1455
Change in body composition			
Weight gain (kg)	10.15 ± 1.19	10.80 ± 2.07	0.4294
Fat deposition (kg)	1.76 ± 0.41	1.47 ± 0.38	0.1327
Water deposition (kg)	5.64 ± 0.99	6.64 ± 1.70	0.1496
Protein deposition (kg)	1.80 ± 0.43	1.81 ± 0.46	0.9373
Ash deposition (kg)	0.39 ± 0.17	0.31 ± 0.11	0.2393
Energy balance (MJ)			
Intake	441.30 ± 39.20	422.07 ± 25.42	0.2376
Storage	109.04 ± 22.59	100.91 ± 16.84	0.4003
Expenditure	286.15 ± 34.71	268.22 ± 28.89	0.3844
In faeces	40.20 ± 11.94	41.92 ± 8.35	0.7289
In urine	5.91 ± 3.11	6.57 ± 3.07	0.6568
In faeces as fat	24.09 ± 4.37	26.57 ± 7.33	0.3992
In fat free faeces	16.11 ± 8.16	15.35 ± 5.27	0.8176
Energy in whole body (MJ)			
Initial body energy (MJ)	100.23 ± 12.34	101.20 ± 10.59	0.8596
Final body energy (MJ) (measured)[1]	209.27 ± 21.32	202.11 ± 15.30	0.4264
Final body energy (MJ) (calculated)[2]	204.90 ± 23.80	196.40 ± 20.53	0.4416
Percentage of energy intake			
Stored in the body	24.83 ± 5.57	24.04 ± 4.48	0.7454
Expended as heat	64.83 ± 5.39	64.49 ± 3.78	0.8789
Lost in faeces	8.98 ± 1.78	9.88 ± 1.57	0.2693
Lost in urine	1.36 ± 0.73	1.59 ± 0.80	0.5462
Lost in faeces as fat	5.43 ± 0.55	6.24 ± 1.47	0.1497
Lost in fat-free faeces	3.55 ± 1.44	3.64 ± 1.25	0.8893

Means ± SD for 10 pigs per experimental diet
[1]Measured with a bomb calorimeter
[2]Calculated on basis of the body composition, given 1 g of animal fat has a gross energy of 39.8 kJ and 1 g of protein has a gross energy of 23.7 kJ (Javadi et al., 2004)

Table 5.
Effect of ALA intake on apparent fatty acid digestibility

Fatty acid	Low ALA	High ALA	*p* value
C 16:0	82.70 ± 2.32	81.14 ± 3.24	0.258
C 18:0	38.29 ± 22.03	25.71 ± 33.40	0.360
C 18:1 n-9	93.57 ± 1.61	93.77 ± 1.45	0.786
C 18:2 n-6	97.77 ± 0.54	97.45 ± 0.38	0.169
C 18:3 n-3	97.90 ± 0.36	98.71 ± 0.20	0.000
Σ SFA	47.60 ± 6.75	40.02 ± 13.86	0.160
Σ MUFA	92.01 ± 10.99	95.27 ± 4.27	0.444
Σ PUFA	98.76 ± 0.37	97.10 ± 2.90	0.127
Σ n-6	98.57 ± 0.67	96.52 ± 3.16	0.113
Σ n-3	98.95 ± 0.18	99.35 ± 0.10	0.000

Means ± SD for 10 pigs per experimental diet
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid

Σ SFA = C10:0 + C14:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:3 n-6 + C20:5 n-3
Σ n-6 = C18:2 n-6 + C18:3 n-6
Σ n-3 = C18:3 n-3 + C20:5 n-3

The higher digestible intake of C16:0 (palmitic acid), LA, SFA, and n-6 PUFA by the low-ALA group and the higher digestible intake of ALA and n-3 PUFA by the high-ALA group were associated with a higher content of these fatty acids in the diets (Table 7). There was no treatment difference for the digestible intake of other individual and grouped fatty acids. The deposition of LA was higher in the pigs fed the low-ALA diet while the deposition of ALA was higher in the pigs fed the high-ALA diet (Table 7). When the ratio of individual fatty acid deposition:digestible intake was calculated, the ratios for LA, ALA, PUFA, n-6 PUFAs and n-3 PUFAs were lower than 1. The ratio of deposition:digestible intake for LA was lower in the pigs fed the high-ALA diet, but there was no diet effect on the deposition:digestible intake ratio for ALA, n-6 PUFA and n-3 PUFA. These ratios for all other individual and grouped fatty acids were not different between both dietary treatments (Table 7).

Minimum de novo synthesis

The pigs fed the low-ALA and high-ALA diet displayed no difference in the synthesis of SFA and MUFA. The ratio for the *de novo* synthesis of SFA:(SFA + MUFA) also was not different between the two dietary treatments (Table 8).

DISCUSSION

The low-ALA diet was rich in LA, whereas the high-ALA diet mainly contained ALA. The difference between the two diets in the contents of palmitic acid was small; it was approximately 4% higher for the low-ALA diet. The contents of the other fatty acids were similar for the two experimental diets. Thus, the low-ALA diet is representative for diets rich in LA, whereas the high-ALA diet may represent diets rich in ALA.

Table 6. The regression formulas for the relations between LA or ALA in the diet and that in the adipose tissue

Origin of equation	Data source	Fatty acid	R^2	Intercept	S.E.	F probability	Slope	S.E.	F probability
Nguyen et al. (2003)	Literature + trial	C18:2 n-6	0.750	7.437	0.737	<0.001	5.577	0.378	<0.001
		C18:3 n-3	0.682	1.414	0.318	<0.001	3.603	0.340	<0.001
This study	Trial	C18:2 n-6	0.568	4.739	2.479	0.072	6.175	1.211	<0.001
		C18:3 n-3	0.845	0.199	0.468	0.675	3.939	0.385	<0.001

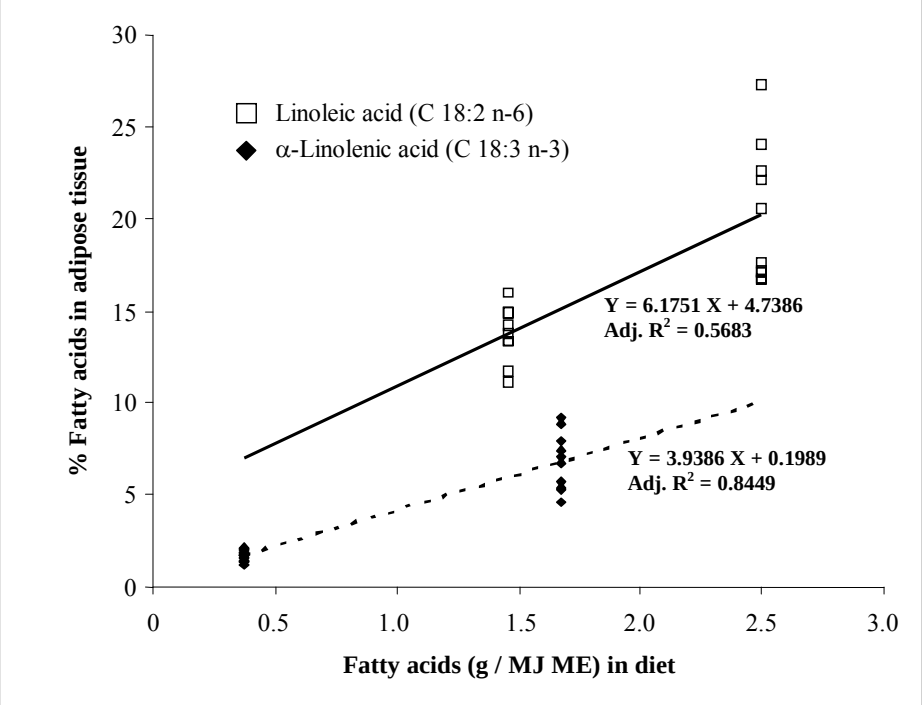

Figure 1. Relationship between polyunsaturated fatty acid intake and group mean content in adipose tissue of the corresponding fatty acid for pigs in the present feeding trial.

The relative concentrations of palmitic (C16:0), stearic (C18:0), and oleic (C18:1 n-9) in adipose tissues and loin muscle were similar for the two diet groups. This may be explained by the similar fractions of these fatty acids in the experimental diets and/or by compensation through *de novo* synthesis. Obviously, both adipose tissues and loin muscle of the pigs fed the low-ALA diet contained a higher LA fraction, whereas these tissues in the pigs fed the high-ALA diet contained a high ALA acid fraction. This agrees with previous investigations reporting that the fatty acid composition in pig's tissues is associated with the fatty acid profile of the obtained diet (Kouba et al., 2003; Miller et al., 1990; Wood et al., 2004). The grouped fatty acids (SFA, MUFA, and PUFA) and the ratios of MUFA:SFA and PUFA:SFA in adipose tissues and loin did not differ between the two dietary treatments. However, the ratios of n-6:n-3 PUFA and C18:2 n-6:C18:3 n-3 for the pigs fed the low-ALA diet were higher than those for the pigs fed the high-ALA diet. These results indicate that the levels of n-6 PUFAs and n-3 PUFAs were increased by elevating the dietary levels of dietary LA and ALA, respectively. As to the composition of animal products in relation to human health, the recommended value for the n-6:n-3 PUFA ratio is less than 4 (Wood et al., 2004). Thus, the results of the current study indicate that the ratios of n-6:n-3 PUFA and C18:2 n-6:C18:3 n-3 in pork can be improved by dietary supplementation of oils rich in ALA.

The pigs fed either the low-ALA or high-ALA diet showed no difference in growth performance. This observation agrees with earlier work (Kouba et al., 2003). It can be concluded that feeding ALA versus LA has no effect on growth performance. There was no diet effect on the digestibility of crude fat, gross energy, individual and grouped fatty acids, except for the higher digestibility of ALA and n-3 PUFAs in the pigs fed the high-ALA diet. It has been known that an improved micelle

Table 7. Effect of ALA intake on digestible fatty acid intake, fatty acid deposition and the deposition:intake ratio during the whole feeding period

Fatty acid	Low ALA	High ALA	*p* value
Digestible fatty acid intake (g/35 days)			
C 16:0	246.55 ± 17.23	175.39 ± 7.93	0.000
C 18:0	31.912 ± 17.20	19.51 ± 26.99	0.262
C 18:1 n-9	369.65 ± 27.40	371.13 ± 18.34	0.895
C 18:2 n-6	844.43 ± 71.79	481.98 ± 27.61	0.000
C 18:3 n-3	12.82 ± 11.18	560.64 ± 33.48	0.000
Σ FA	1,676.65 ± 126.06	1,660.54 ± 40.88	0.771
Σ SFA	295.53 ± 14.93	210.43 ± 36.43	0.000
Σ MUFA	375.64 ± 27.27	379.72 ± 13.40	0.745
Σ PUFA	1,005.48 ± 87.32	1,070.39 ± 41.97	0.126
Σ n-9	352.15 ± 49.31	344.41 ± 56.13	0.796
Σ n-6	870.61 ± 75.35	493.83 ± 18.86	0.000
Σ n-3	134.87 ± 11.98	576.56 ± 23.09	0.000
Fatty acid deposition (g/35 days)			
C 16:0	391.51 ± 119.86	304.91 ± 81.90	0.092
C 18:0	291.92 ± 74.19	234.72 ± 51.35	0.075
C 18:1 n-9	519.55 ± 145.08	427.63 ± 153.55	0.210
C 18:2 n-6	305.72 ± 69.35	115.00 ± 56.41	0.000
C 18:3 n-3	51.98 ± 9.96	210.76 ± 58.89	0.000
Σ FA	1,756.94 ± 280.33	1,549.38 ± 249.10	0.189
Σ SFA	786.77 ± 151.74	633.54 ± 105.88	0.062
Σ MUFA	596.62 ± 121.25	523.14 ± 126.27	0.308
Σ PUFA	373.55 ± 84.74	392.70 ± 114.60	0.736
Σ n-9	575.37 ± 108.11	505.94 ± 121.53	0.299
Σ n-6	312.16 ± 73.97	137.74 ± 52.98	0.001
Σ n-3	61.39 ± 11.53	254.96 ± 69.01	0.000
Deposition : intake ratio			
C 16:0	1.59 ± 0.47	1.73 ± 0.43	0.504
C 18:0	8.63 ± 2.33	7.53 ± 1.62	0.314
C 18:1 n-9	1.40 ± 0.37	1.14 ± 0.37	0.155
C 18:2 n-6	0.36 ± 0.09	0.24 ± 0.12	0.019
C 18:3 n-3	0.41 ± 0.09	0.38 ± 0.11	0.468
Σ FA	1.05 ± 0.17	0.93 ± 0.15	0.221
Σ SFA	2.67 ± 0.53	3.17 ± 1.21	0.342
Σ MUFA	1.59 ± 0.32	1.37 ± 0.30	0.233
Σ PUFA	0.37 ± 0.09	0.37 ± 0.11	0.893
Σ n-9	1.54 ± 0.23	1.33 ± 0.29	0.210
Σ n-6	0.36 ± 0.09	0.28 ± 0.11	0.178
Σ n-3	0.46 ± 0.10	0.44 ± 0.12	0.807

Means ± SD for 10 pigs per experimental diet
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid

Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C17:1 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:3 n-6 + C20:3 n-3 + C20:5 n-3 + C22:6 n-3
Σ n-9 = C18:1 n-9 + C20:1 n-9
Σ n-6 = C18:2 n-6 + C18:3 n-6
Σ n-3 = C18:3 n-3 + C20:3 n-3 + C20:5 n-3 + C22:6 n-3

Table 8.
Effect of dietary ALA level on minimum *de novo* synthesis of fatty acids during the whole feeding period

Fatty acid	Low ALA	High ALA	*p* value
Minimum synthesis (g/35 days)			
SFA	491.24 ± 152.09	423.11 ± 127.90	0.406
MUFA	220.98 ± 118.50	143.42 ± 118.49	0.264
SFA/(SFA+MUFA)	0.70 ± 0.09	0.78 ± 0.11	0.184

Means ± SD for 10 pigs per experimental diet
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C18:1 n-9

formation may favourably influence the digestion of all fatty acids in the diet and that the position of a given fatty acid in the triacylglycerol molecule also affects digestibility (Bracco, 1994; Innis and Dyer, 1997; Nelson and Innis, 1999; Scheeder et al., 2000; Scheeder et al., 2003). It is possible that a different position of ALA in the triacylglycerol molecules of soya bean oil and linseed oil had influenced ALA digestibility. In addition, for the two diets the apparent faecal digestibility of ALA could be affected differently by of microbial hydrogenation in the large bowel. This would affect ALA excretion with faeces and thus apparent digestibility.

The current study has demonstrated that there was no differential effect of LA and ALA level on body composition, except for the fraction of crude fat. There was a trend towards a lower total amount of fat in the whole body after feeding the high-ALA diet. These results are similar to those reported for in mice fed diets with 10% LA or 10% ALA (Javadi et al., 2007a). The lower fat content in the whole body might be explained by the lower rate of ALA incorporation into adipose tissue and muscle when compared with that of LA (Nguyen et al., 2003). The lower incorporation rate may be explained by the increased β-oxidation both in mitochondria and peroxisomes after feeding a high-ALA diet to rats (Cunnane and Anderson, 1997; Kabir and Ide, 1996). However, there was no effect of dietary treatment on energy balance. Cha and Jones (1996) found that an insufficient energy intake level can cause an increase in the oxidation rate and deposition pattern of fatty acids. The pigs in the current study ingested sufficient energy so that the lower fat content in the whole body after feeding the high-ALA diet should be a specific effect of the high ALA intake.

The calculated intake of digestible fatty acids is based on the amount of fatty acids ingested and the measured apparent digestibility. The deposition of fatty acids in the body fat was based on the increase of fatty acids in the total body fat during the entire feeding period. As would be expected, the pigs fed the high-ALA diet deposited more ALA and those fed the low-ALA diet had deposited more LA in their whole body. Similar data have been shown for mice (Javadi et al., 2004) and goats (Yeom et al., 2005). To determine the preferential deposition of fatty acids, the ratio of deposition of a given fatty acid or group of fatty acids to the intake of digestible fatty acid or group of fatty acids was calculated. The deposition:intake ratio for fatty acids reflects the net amount synthesized and/or the proportion of the digested fatty acid not oxidized. A deposition:intake ratio > 1 would point at net *de novo* synthesis, whereas a ratio < 1 would indicate net oxidation. The ratios of deposition:digestible intake for palmitic acid, stearic acid, oleic acid, SFA and MUFA were higher than 1 for both dietary treatments, pointing at *de novo* synthesis of these fatty acids. The calculated

deposition:intake ratio < 1 for LA and ALA for diets must have been the result of the well-known preferential oxidation of LA and ALA (Cunnane and Anderson, 1997; Jones et al., 1985; Yeom et al., 2005). However, the deposition:intake ratio for LA on the high-ALA diet was lower ($p<0.05$) than that on the low-ALA diet, which is in disagreement with previous investigations (Cunnane and Anderson, 1997; Ide et al., 1996; Jones et al., 1985; Yeom et al., 2005) indicating that ALA is more preferentially oxidized and less stored than is LA. This discrepancy may be explained by the fact that the ratio of fatty acid deposition:digestible fatty acid was calculated for a given point of time.

The mathematical relationships between the intake of LA and ALA and their relative percentages in adipose tissue indicate that LA has a higher rate of deposition than does ALA (Nguyen et al., 2003). In the current study, the slopes for the relationships between LA and ALA contents in the diet and their fractions in the body fat were close to those reported by Nguyen et al. (2003). The absolute amounts of fatty acids deposited in whole body will reflect the actual amount and rate of fatty incorporation into the whole body. The calculated ratio of the Δ g deposited and Δ g digestible intake for LA was higher than that for ALA. This would explain the lower fat content in the whole body on the high-ALA diet. It would also explain the discrepancy with the earlier work (Cunnane and Anderson, 1997; Ide et al., 1996; Jones et al., 1985; Yeom et al., 2005) mentioned above. Additionally, the observation implies a similar metabolism and incorporation of fatty acids in Thai indigenous pigs and commercial bred pigs. The similar ratio of deposition:digestible intake for n-6 PUFA and n-3 PUFA would again point to similar fat digestibility of the two oils of plant origin that were used to formulated the experimental diets. Furthermore, hydrogenation of PUFA by microbes in the large intestine may have increased the digestibility of PUFA and decreased the digestibility of SFA and MUFA. Further research is needed to quantify the ileal digestibility for individual and grouped fatty acids.

The diet effect on the deposition:intake ratios for SFA and MUFA can be explained by the calculated minimum synthesis of these groups of fatty acids. Feeding the diet with low or high ALA content had no effect on synthesis of SFA. The similar synthesis ratio for SFA:(SFA + MUFA) in pigs fed either diet points at selective synthesis of SFA and MUFA. This might relate to *de novo* fatty acid synthesis being regulated to balance the amount of saturated and unsaturated fatty acids in the body because the intake of PUFA was very high in the current study.

In conclusion, the feeding of diets with low or high ratio of LA:ALA had a marked impact of the fatty acid composition of tissues. Feeding the diet with high ratio of LA:ALA produced a markedly increased deposition of LA adipose tissues, loin muscle, and whole body, whereas a marked deposition of ALA was found after feeding the diet with the low ratio of LA:ALA. However, the incorporation rate in the pig body for LA was higher than that for ALA. The different intake levels of LA and ALA had no effect on energy expenditure, but the high-ALA diet lowered the body fat fraction and tended to lower the fat content and fat deposition in the whole body.

ACKNOWLEDGEMENTS

The authors thank Wilaiwan Lorthong, Jiraporn Phonphan, Nattawut Suphan and Wasuwat Pimpa for their excellent assistance. This research work was supported by Faculty of Natural Resources, Rajamangala University of Technology Isan in the

form of providing the facilities and by Mahanakorn University of Technology in the form of funding.

LITERATURE CITED

AOAC. 1990. Association of official analysis chemists, Washington DC.

Bracco, U. 1994. Effect of triglyceride structure on fat absorption. Am J Clin Nutr 60:1002S-1009S.

Cha, M.C., and P.J.H. Jones. 1996. Tissue fatty acid deposition is influenced by an interaction of dietary oil source and energy intake level in rats. Nutritional Biochemistry 7:650-658.

Crespo, N., and E. Esteve-Garcia. 2002. Dietary polyunsaturated fatty acids decrease fat deposition in separable fat depots but not in the remainder carcass. Poult Sci 81:512-8.

Cunnane, S.C., and M.J. Anderson. 1997. The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. Journal of Nutrition 127:146-152.

Goerl, K.F., S.J. Eilert, R.W. Mandigo, H.Y. Chen, and P.S. Miller. 1995. Pork characteristics as affected by two populations of swine and six crude protein levels. J Anim Sci 73:3621-6.

Horwitz, W. 1975. Official Methods of Analysis of Official Analytical Chemist, p. 225 Official Methods of Analysis of Official Analytical Chemist, 12 ed. Benjamin Franklin Station, DC.

Ide, T., M. Murata, and M. Sugano. 1996. Stimulation of the activities of hepatic fatty acid oxidation enzymes by dietary fat rich in alpha-linolenic acid in rats. J Lipid Res 37:448-63.

Innis, S.M., and R. Dyer. 1997. Dietary triacylglycerols with palmitic acid (16:0) in the 2-position increase 16:0 in the 2-position of plasma and chylomicron triacylglycerols, but reduce phospholipid arachidonic and docosahexaenoic acids, and alter cholesteryl ester metabolism in formula-Fed piglets. J Nutr 127:1311-9.

Javadi, M., M.J. Geelen, A.G. Lemmens, A. Lankhorst, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2007. The influence of dietary linoleic and alpha-linolenic acid on body composition and the activities of key enzymes of hepatic lipogenesis and fatty acid oxidation in mice. J Anim Physiol Anim Nutr (Berl) 91:11-8.

Javadi, M., H. Everts, R. Hovenier, S. Kocsis, A.E. Lankhorst, A.G. Lemmens, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2004. The effect of six different C18 fatty acids on body fat and energy metabolism in mice. Br J Nutr 92:391-9.

Jones, P.J.H., P.B. Pencharz, and M.T. Clandinin. 1985. Whole body oxidation of dietary fatty acids: implications for energy utilization. American journal of clinical nutrition 42:769-777.

Kabir, Y., and T. Ide. 1996. Activity of hepatic fatty acid oxidation enzymes in rats fed alpha-linolenic acid. Biochim Biophys Acta 1304:105-19.

Kouba, M., M. Enser, F.M. Whittington, G.R. Nute, and J.D. Wood. 2003. Effect of a high-linolenic acid diet on lipogenic enzyme activities, fatty acid composition, and meat quality in the growing pig. J Anim Sci 81:1967-79.

Metcalfe, L.D., A.A. Schmitz, and J.R. Pelka. 1966. Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. Anal. Chem. 38:514-515.

Miller, M.F., S.D. Shackelford, K.D. Hayden, and J.O. Reagan. 1990. Determination of the alteration in fatty acid profiles, sensory characteristics and carcass traits of swine fed elevated levels of monounsaturated fats in the diet. J Anim Sci 68:1624-31.

Nelson, C.M., and S.M. Innis. 1999. Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. Am J Clin Nutr 70:62-9.

Nguyen, L.Q., M.C.G.A. Nuijens, H. Everts, N. Salden, and A.C. Beynen. 2003. Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. Meat Science 65:1399-1406.

Pfeiffer, H., and H. Falkenberg. 1972. Masse am Lendenspiegel zur objektiven Ermittlung der SchlachtkOrper Zusammensetzung beim Schwein. Tierzucht 26:466-467.

SAS. 1996. User's guide; Statistics Statistical analysis system inst., Cary, NC, USA.

Scheeder, M., K.R. Gläser, B. Eichenberger, and C. Wenk. 2000. Influence of different fats in pig feed on fatty acids composition of phospholipids and physical meat quality characteristics. European Journal of Lipid Science and Technology 102:391-401.

Scheeder, M., D. Gumy, R. Messikommer, C. Wenk, and P. Lambelet. 2003. Effect of PUFA at sn-2 position in dietary triacylglycerols on the fatty acid composition of adipose tissues in non-ruminant farm animals. Eur. J. Lipid Sci. Technol. 105:74-82.

Shields, R.G., Jr., D.C. Mahan, and P.L. Graham. 1983. Changes in swine body composition from birth to 145 kg. J. Anim. Sci. 57:43-54.

SPSS. 1999. SPSS for Windows, SPSS Inc., Chicago, IL, USA.

Wagner, J.R., A.P. Schinckel, W. Chen, J.C. Forrest, and B.L. Coe. 1999. Analysis of body composition changes of swine during growth and development. J Anim Sci 77:1442-66.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: a review. Meat Science 66:21-32.

Yeom, K.-H., J.T. Schonewille, H. Everts, K.-W. Lee, G. van Trierum, and A.C. Beynen. 2005. Growth performance and body composition of goat kids fed milk replacers with increasing levels of linoleic acid. Journal of Animal Physiology and Animal Nutrition 89:29-34.

CHAPTER 6

Effect of background fat source in the diet on α-linolenic acid metabolism and deposition in Thai indigenous pigs

J. Mitchaothai†§, H. Everts†, C. Yuangklang‡, S. Wittayakun‡, K. Vasupen‡, S. Wongsuthavas‡, P. Srenanul‡, R. Hovenier†, and A. C. Beynen†

§Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand

‡Faculty of Natural Resources, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand

†Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, P.O. Box 50.152, 3508 TD, Utrecht, The Netherlands

To be submitted

ABSTRACT

Eighteen Thai indigenous pigs (9 males and 9 females) with an average BW of 7.32 ± 1.22 kg were used to investigate the interaction between dietary fat source and α-linolenic acid (ALA). The main hypothesis tested was that a dietary background rich in linoleic acid (LA) instead of low in LA, would increase the oxidation of ALA and thus lower the deposition of ALA in the whole body. For the two diets, fatty acid digestibility, body composition, energy expenditure and incorporation of fatty acids in the whole body were measured. The pigs were fed either a diet containing beef tallow (BT) and linseed oil (5 % (w/w) beef tallow + 1 % linseed oil) or sunflower oil (SO) and linseed oil (5 % sunflower oil + 1 % linseed oil). The two diets had similar, high levels of ALA as provided by the linseed oil. The pigs were kept in individual cages and fed *ad libitum* for 35 days. The pigs fed the SO diet deposited more linoleic acid (LA) ($p < 0.05$) in adipose tissue and loin muscle. There were no differences ($p > 0.05$) in growth performance, body composition and energy balance, except for the lower ($p < 0.05$) amount of energy excreted with faeces as fat in pigs fed the SO diet. The digestibilities of gross energy, stearic acid and ALA acid were not different ($p > 0.05$) for the two treatments, but the digestibility of crude fat, palmitic acid and LA acid was greater ($p < 0.05$) for the SO diet. Feeding the SO diet instead of the BT diet increased ($p < 0.01$) the deposition of LA, but decreased ($p < 0.05$) the deposition of ALA in the whole body. The pigs fed the SO diet had a markedly higher ($p < 0.05$) ratio of deposition:intake for stearic acid, a greater ($p < 0.05$) oxidation of ALA, but there was no effect ($p > 0.05$) on the oxidation of LA. The minimum rate of *de novo* synthesis for SFA tended to be higher ($p = 0.076$) and the ratio of this synthesis for SFA:(SFA+MUFA) was significantly higher ($p < 0.05$) for the SO diet. It is concluded that feeding pigs with a diet rich in LA may increase the digestion and deposition of LA, elevates *de novo* synthesis of SFA and stimulates the oxidation of ALA, without affecting growth performance and energy expenditure.

Keywords: deposition, digestibility, beef tallow, sunflower oil, α-linolenic acid, indigenous pigs

INTRODUCTION

Our previous studies (Mitchaothai et al., *in press;* Mitchaothai et al., *submitted*) have shown that pigs fed a diet containing sunflower oil (SO) instead of beef tallow (BT) had higher rates of oxidation for linoleic acid (LA), but there was no difference for α-linolenic acid (ALA). This finding was unexpected as based on earlier investigations (Cunnane and Anderson, 1997; Ide et al., 1996; Jones et al., 1985), reporting that ALA is more preferentially oxidised than is LA. The possible explanation for this discrepancy could be the low level of ALA in both the BT and SO diet. Pigs only need a relatively small amount of ALA, which is an essential fatty acid. ALA is required as a precursor for the production of eicosanoids and for the deposition in body membranes. In terms of energy metabolism, the efficiency of the conversion of glucose into fatty acids for esterification in body triacylglycerols is less than of dietary fatty acids being incorporated into body triacylglycerols (Newsholme and Leech, 1986). Dietary polyunsaturated fatty acids (PUFA) instead of saturated fatty acids (SFA) are preferentially oxidized (Beynen and Katan, 1985), resulting in carbohydrates being shifted from the oxidative into the lipogenic pathway. Hence, increasing the dietary PUFA level at the expense of SFA would increase heat expenditure due to higher fat oxidation. It could thus be hypothesized that a high level of ALA in a diet rich in SO would result in higher energy expenditure and more oxidation of ALA than a high level of ALA in a diet rich in BT. In the present study, this hypothesis was tested.

MATERIALS AND METHODS

Animals, Diets and Feeding

Eighteen male and female pigs, Thai indigenous Kadon pigs, with an average BW of 7.32 ± 1.22 kg were used. Three male and three female were selected to be slaughtered for baseline measurements. Then, the remaining 12 pigs were allotted to one of the two dietary treatments on the basis of body weight and sex and were housed in individual cages. The pigs were allowed *ad libitum* access to feed and water throughout the experiment for 35 days. Feed consumption was recorded twice a day and individual body weight of the pigs was measured weekly. There were two experimental diets with either beef tallow (5%) and linseed oil (1%) (BT diet) or sunflower oil (5%) and linseed oil (1%) (SO) as shown in Table 1. Throughout the entire experiment, the faeces and urine samples of all pigs were collected for determining digestibility and balance of the macronutrients and gross energy. A 10% solution of sulphuric acid (H_2SO_4) was used to prevent ammonia evaporation during urine collection and the urine samples then were stored at 5 ^oC until analysis. All pigs were slaughtered at an average BW of 17.34 ± 2.06 kg. Ingredient and macronutrient composition of the experimental diets are shown in Table 1. The fatty acid composition of the diets is documented in Table 2.

Collection and Pre-treatment of Samples

The six pigs (3 males and 3 females) for baseline measurements were killed by an intravenous injection of sodium pentobarbital (200 mg/kg of body weight). The pigs were scalded, blood was collected and weighed, the head was removed, and the carcass was split into halves along the median plane according to the procedure

previously reported by Shields et al. (1983). At this moment a few small samples (1-2 g) of adipose tissue and loin were taken and stored at – 20 ^{o}C until analysis. The procedure according to Shields et al. (1983) was modified by drying compartment samples and both sides of the sawed carcass at 60 ^{o}C for 1 week in a forced-hot air oven with placing the frozen materials on a mesh with an underneath tray to collect the dripping oil. The dried whole carcass samples derived from each pig were pooled and then weighed. The dripping oil samples were pooled for each pig and weighed in the same way as the dried whole carcass samples. The dried samples were individually ground once through a 2 mm screen and twice through a 1 mm screen by a hammer mill. The 12 pigs in the feeding trial were sacrificed at 5 weeks after the commencement. Then, whole carcass sample was prepared in the same way as for the baseline pigs.

Table 1. Composition of the diet (as-fed basis)

Item	BT	SO
Raw materials (g/100g)		
Cassava chips	45.53	45.53
Soybean meal (44% CP)	40.00	40.00
Beef tallow	5.00	-
Sunflower oil	-	5.00
Linseed oil	1.00	1.00
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Dicalcuim phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Premix	0.25	0.25
Total	100.00	100.00
Analysed nutrients (%)		
Dry matter	87.23	87.62
Crude protein	19.63	20.06
Crude fat	7.24	7.09
Ash	6.91	7.64
Analysed GE (MJ/kg)	17.26	17.04
Calculated ME (MJ/kg)	14.12	14.07

BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil

Chemical Analyses and Fatty Acid Composition Determination

The samples of diets and faeces were dried at 60 ^{o}C for 72 h in a forced-hot air oven. The dried diets, faeces, and whole carcass samples were analysed for moisture, gross energy, crude protein, crude fat, and ash (AOAC, 1990). The urine samples were also analysed to quantify the content of energy and nitrogen (AOAC, 1990).

The fresh loin and the dried samples of diets, faeces, and whole carcasses were extracted as described previously (Horwitz, 1975). Each sample was added to a flask and 2 ml of ethanol was added. Subsequently, 10 ml of HCl (8 mol/L) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80 ^{o}C for 30-40 min. The tubes were cooled down, 10 ml of ethanol (96%, w/w) and 25 ml of petroleum ether (boiling point between 40 and 60 ^{o}C) were added and the tube was shaken for one min. The fat-containing upper layer was decanted into a 150-ml round-

bottom flask. The extraction procedure was repeated twice with 15 ml of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath of 40 ^{o}C. The round-bottom flasks with the lipids were dried overnight at 60 ^{o}C and the total lipids were measured gravimetrically. Total lipids, the small samples of adipose tissue and the dripping oil were saponified and methylated according to the procedure of Metcalfe et al. (1966) followed by gas chromatography for determination of fatty acid composition (Javadi et al., 2004).

Determination of Body Composition and Energy Balance

The body weight and composition of the baseline pigs showed no effect of sex, hence the average energy or nutrient content in whole body of the baseline pigs was assumed to be the similar fraction in whole body of the experimental pigs at the start. All types of samples including the dripping oil were determined with a bomb calorimeter (Parr® Oxygen Bomb Calorimeter 1341 EE adiabatic, Parr Instrument Company, Moline, Illinois, USA). As a thermo-chemical standard, benzoic acid was used (Asian Pacific Specialty Chemical Limited, NSW, Australia). The total amount of energy that was lost as heat (heat production or energy expenditure) was calculated with the formula: Energy lost as heat = energy in food – energy in faeces and urine – energy stored in body. Energy stored in the body was determined as total energy at the end of the 35 d feeding period minus the energy in the body at the beginning of the experiment (= individual body weight × mean energy content of the baseline pigs). The same procedure was used to calculate the water, protein, fat and ash retention.

Calculation of Digestible Fatty Acid Intake, Fatty Acid Deposition and Minimum de novo Synthesis

The total digestible fatty acid intake was calculated as fatty acid intake (g/35 days) × 0.95 × apparent fatty acid digestibility (fraction of intake). The deposition of fatty acids was calculated with the following formula: Fatty acid deposition (g/35 days) = individual carcass content of fatty acid at the end of the study – individual carcass content of fatty acid at the start of the study (= Whole carcass fat mass (g) × 0.95 × mean fatty acid composition of the baseline pigs). Carcass content of fatty acid at the end (g) was calculated as the whole carcass fat mass (g) × 0.95 × fraction of fatty acid in whole carcass. The ratio of fatty acid deposition and digestible fatty acid intake was calculated for selected individual fatty acids and groups of fatty acids. The quantity of fatty acids oxidized, transformed and synthesized could not be measured. Therefore, minimum *de novo* fatty acid synthesis was calculated as fatty deposition minus digestible fatty acid intake.

Statistical Analyses

The effect of fat source in the diets was evaluated for statistical significance by the Student's *t* test (SPSS, 1999). Results are expressed as means ± SD.

RESULTS

Dietary Fatty Acid Composition

Table 2. Fatty acid composition of the experimental diets

Item	BT	SO
Fatty acids, g methylester/100 g methylesters		
C 14:0	1.30	0.08
C 15:0	0.42	0.04
C 16:0	18.71	10.18
C 17:0	1.34	0.02
C 18:0	25.54	4.02
C 20:0	0.38	0.29
C 22:0	0.15	0.64
C 16:1	0.60	0.05
C 18:1 n-9	21.72	29.94
C 18:1 n-7	3.97	0.89
C 20:1 n-9	0.43	0.35
C 18:2 n-6	12.58	44.61
C 18:3 n-6	0.29	0.01
C 18:3 n-3	7.85	7.68
Unknown	4.33	0.75
∑ SFA	47.89	15.33
∑ MUFA	26.96	31.33
∑ PUFA	20.82	52.59

BT= Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids
Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C14:1 + C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-6 + C18:3 n-3

In Table 2, the fatty acid composition of both experimental diets is shown. The BT diet was rich in SFA (48%), mainly consisting of palmitic acid (C16:0) and stearic acid (C18:0), whereas the SO diet contained a high level of LA (47%). The SO diet contained about 4% of monounsaturated fatty acids (MUFA). The ALA level was similar; the BT contained 7.9% and the SO diet 7.7%.

Fatty Acid Composition of Adipose Tissues and Loin Muscle

The fatty acid patterns of fat in adipose tissue and loin are shown in Table 3. After feeding the experimental diets, the relative concentration of SFA (mainly palmitic and stearic acid) in the adipose tissue was higher ($p < 0.05$) for the BT diet than for the SO diet, but no difference between both diets was found for the SFA content in loin muscle ($p > 0.05$). There was a higher ($p < 0.05$) relative concentration of MUFA in both adipose tissue and loin muscle for the pigs fed the BT diet when compared those fed the SO diet. As to the PUFA concentration in adipose tissue and loin muscle, the LA concentration in the pigs fed the SO diet was higher ($p < 0.001$) than that in the pigs fed BT diet whereas the concentration of ALA in loin muscle for the pigs fed the SO diet was lower ($p < 0.05$). The ALA content of adipose tissue for pigs fed the SO diet tended to be lower ($p = 0.055$) than for the pigs fed the BT diet. The ratios of MUFA:SFA in adipose and loin were not different ($p > 0.05$) for the two dietary treatments, whereas the ratios of PUFA:SFA, n-6 PUFA:n-3 PUFA and C18:2 n-6:C18:3 n-3 in both adipose tissue and loin of the pigs fed the BT diet were lower (*p*

< 0.01) than those for pigs fed the SO diet. The ratios of n-6:/ n-3 PUFA and C18:2 n-6 : C18:3 n-3 in both adipose tissue and lion muscle for the pigs fed the BT diet ranged between 3 and 4 and for the pigs fed the SO diet between 7 and 10.

Table 3 Fatty acid composition of adipose tissues fat and loin intramuscular fat

	Adipose			Loin		
	BT	SO	*P* value	BT	SO	*P* value
Analysed fatty acids, g methylester/100 g methylesters						
C 14:0	1.78 ± 0.13	1.33 ± 0.12	0.001	1.56± 0.18	1.34± 0.20	0.136
C 16:0	24.25 ± 1.44	21.46 ± 0.81	0.008	24.71± 1.48	23.67± 1.69	0.368
C 17:0	0.81 ± 0.18	0.46 ± 0.14	0.011	0.71± 0.11	0.51± 0.15	0.069
C 18:0	14.94 ± 1.40	12.41 ± 1.15	0.020	15.00± 2.14	14.01± 1.70	0.462
C 16:1	1.94 ± 0.45	1.24 ± 0.19	0.015	2.04± 0.51	1.35± 0.18	0.026
C 17:1	0.55 ± 0.13	0.28 ± 0.10	0.012	0.53± 0.12	0.29± 0.09	0.014
C 18:1 n-9	34.78 ± 2.57	31.72 ± 0.60	0.095	36.33± 2.08	32.87± 1.19	0.016
C 18:1 n-7	2.73 ± 0.46	1.85 ± 0.30	0.010	2.91± 0.59	2.09± 0.21	0.022
C 20:1 n-9	0.62 ± 0.07	0.69 ± 0.08	0.220	0.82± 0.08	0.82± 0.07	0.961
C 18:2 n-6	11.01 ± 4.29	23.36 ± 1.45	0.000	9.01± 2.55	17.27± 1.94	0.001
C 18:3 n-3	3.60 ± 0.45	3.05 ± 0.26	0.055	2.30± 0.33	1.75± 0.22	0.018
C 20:2 n-6	0.31 ± 0.18	0.86 ± 0.06	0.000	0.30± 0.14	0.68± 0.07	0.001
C 20:4n-6	0.13 ± 0.10	0.23 ± 0.03	0.077	0.65± 0.27	0.80± 0.46	0.603
C 20:3n-3	0.33 ± 0.03	0.37 ± 0.03	0.132	0.25± 0.04	0.25± 0.02	0.848
C 22:5 n-3	0.24 ± 0.05	0.09 ± 0.05	0.002	0.31± 0.22	0.18± 0.12	0.270
unknown	1.27 ± 0.38	0.35 ± 0.03	0.017	2.01± 0.44	1.62± 0.41	0.230
∑ SFA	42.08 ± 2.46	35.88 ± 1.71	0.003	42.21± 3.55	39.75± 3.60	0.340
∑ MUFA	40.63 ± 3.55	35.79 ± 0.94	0.021	42.63± 3.25	37.43± 1.45	0.014
∑ PUFA	16.02 ± 4.53	27.98 ± 1.67	0.001	12.87± 1.95	20.96± 2.69	0.002
∑ MUFA/∑ SFA	0.97 ± 0.10	1.00 ± 0.06	0.572	1.02± 0.16	0.95± 0.10	0.452
∑ PUFA/∑ SFA	0.38 ± 0.12	0.78 ± 0.08	0.000	0.31± 0.05	0.54± 0.11	0.007
∑ n-6/∑ n-3	2.54 ± 1.07	6.98 ± 0.36	0.000	3.72± 1.95	8.73± 1.04	0.002
18:2 n-6/18:3 n-3	3.08 ± 1.17	7.68 ± 0.36	0.000	4.12± 1.92	9.95± 1.15	0.001

Means ± SD for 6 pigs per experimental diet
BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C17:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:4 n-3 + C20:3 n-6 + C20:4 n-6 + C20:3 n-3 + C20:5 n-3

Growth Performance and Nutrient Digestibility

The dietary fatty acid composition had no effect ($p > 0.05$) on feed intake, growth (average daily gain), feed conversion ratio (feed:gain) and final BW (Table 4). There was no diet effect ($p > 0.05$) on the apparent faecal digestibility of gross energy, but the pigs fed the SO diet had a higher ($p < 0.01$) apparent faecal digestibility of crude fat (Table 4). The apparent faecal digestibilities for palmitic acid, linoleic acid and SFA in the pigs fed the SO diet were greater ($p < 0.01$) than in their counterparts given the BT, but there were no diet differences ($p > 0.05$) for the apparent faecal digestibilities of stearic acid, oleic acid, ALA, MUFA and PUFA (Table 5).

Table 4. Body composition and energy balance in Thai native pigs fed the experimental diets for 35 days

	BT	SO	*P* value
Food intake (kg dry food/35 d)	24.39 ± 3.40	24.41 ± 3.00	0.9942
Feed conversion	2.46 ± 0.22	2.47 ± 0.16	0.9690
Initial body weight (kg)	7.51 ± 1.48	7.19 ± 1.10	0.6902
Final body weight (kg)	17.40 ± 2.14	17.13 ± 2.17	0.8342
Average daily gain (g)	282.57 ± 25.08	284.08 ± 45.47	0.9429
Apparent fat digestibility (%)	74.11 ± 5.10	87.19 ± 3.99	0.0018
Apparent gross energy digestibility (%)	86.44 ± 2.71	87.14 ± 1.61	0.6263
Body composition at the end			
Fat (kg)	2.22 ± 0.74	2.58 ± 0.65	0.4117
Water (kg)	11.00 ± 1.48	10.46 ± 1.18	0.5197
Protein (kg)	3.22 ± 0.50	3.15 ± 0.42	0.8103
Ash (kg)	0.62 ± 0.08	0.60 ± 0.10	0.7191
Recovery (%)	98.09 ± 1.03	98.16 ± 0.92	0.8944
Water : Protein ratio (kg/kg)	3.50 ± 0.80	3.38 ± 0.20	0.7489
Fat (%)	12.65 ± 3.13	14.93 ± 2.35	0.2099
Water (%)	63.29 ± 6.16	61.16 ± 1.95	0.4912
Protein (%)	18.59 ± 2.99	18.38 ± 0.74	0.8846
Ash (%)	3.56 ± 0.39	3.49 ± 0.45	0.7914
Change in body composition			
Weight gain (kg)	9.82 ± 0.78	9.86 ± 1.60	0.9516
Fat deposition(kg)	1.40 ± 0.63	1.79 ± 0.56	0.2974
Water deposition(kg)	5.84 ± 1.04	5.52 ± 0.97	0.6078
Protein deposition(kg)	2.18 ± 0.46	2.16 ± 0.34	0.9272
Ash deposition(kg)	0.40 ± 0.06	0.39 ± 0.08	0.8161
Energy balance (MJ)			
Intake	366.39 ± 51.33	396.07 ± 43.55	0.3249
Storage	73.75 ± 38.09	91.52 ± 30.75	0.4155
Expenditure	237.93 ± 34.67	247.07 ± 20.76	0.6175
In faeces	48.88 ± 6.83	50.54 ± 5.02	0.6588
In urine	5.84 ± 1.72	6.49 ± 1.39	0.2716
In faeces as fat	14.93 ± 2.31	10.05 ± 1.84	0.0022
In fat free faeces	33.95 ± 5.50	40.48 ± 4.09	0.0390
Energy in whole body (MJ)			
Initial body energy (MJ)	75.93 ± 14.94	72.65 ± 11.13	0.6902
Final body energy (MJ) (measured)[1]	167.63 ± 48.78	181.53 ± 42.96	0.6230
Final body energy (MJ) (calculated)[2]	168.22 ± 42.48	180.93 ± 34.18	0.5963
Percentage of energy intake			
Stored in the body	19.55 ± 8.45	22.71 ± 5.76	0.4930
Expended as heat	65.27 ± 8.21	62.66 ± 4.77	0.5475
Lost in faeces	13.56 ± 2.71	12.86 ± 1.61	0.6263
Lost in urine	1.63 ± 0.58	1.76 ± 0.39	0.6551
Lost in faeces as fat	4.10 ± 0.60	2.55 ± 0.51	0.0007
Lost in fat-free faeces	9.46 ± 2.22	10.31 ± 1.33	0.4240

Means ± SD for 6 pigs per experimental diet
BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
[1]Measured with a bomb calorimeter
[2]Calculated on basis of the body composition, given 1 g of animal fat has a gross energy of 39.8 kJ and 1 g of protein has a gross energy of 23.7 kJ (Javadi et al., 2004)

Table 5
Effect of dietary fat source on apparent fatty acid digestibility

Fatty acid	BT	SO	*p* value
C 16:0	49.03 ± 5.48	81.92 ± 4.60	0.000
C 18:0	46.25 ± 5.16	40.65 ± 16.47	0.460
C 18:1 n-9	90.44 ± 5.43	90.90 ± 3.74	0.860
C 18:2 n-6	95.35 ± 0.73	96.77 ± 1.41	0.002
C 18:3 n-3	97.53 ± 0.20	97.60 ± 1.30	0.874
Σ SFA	47.42 ± 4.76	67.48 ± 10.00	0.002
Σ MUFA	90.18 ± 5.31	90.55 ± 3.53	0.881
Σ PUFA	96.18 ± 0.52	96.89 ± 1.39	0.303

Means ± SD for 6 pigs per experimental diet
BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C16:0 + C18:0 + C20:0
Σ MUFA = C16:1 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-6

Body Composition and Energy Balance

Table 4 shows that the macronutrient composition and deposition (water, crude fat, crude protein and ash) in the whole body was not different ($p > 0.05$) between the experimental diets, when expressed either as absolute amount or as percentage. Energy intake, storage, expenditure, and losses in faeces and urine expressed either as absolute amount or percentage of intake showed no diet difference ($p > 0.05$). However, the energy losses with faeces as fat and as fat-free material for the BT treatment were higher ($p < 0.05$) than those for the SO treatment. However, when the losses were expressed as percentage of energy intake, only the fat energy loss with faeces was higher ($p < 0.001$) for the BT diet. The final amount of energy in the whole body as derived from measurements with the bomb calorimeter was close to that as calculated using the equation given by Javadi et al. (2004) (Table 4).

Digestible Fatty Acid Intake and Deposition

When the intake of digestible fatty acids is compared between two experimental diets, the intake on the BT higher ($p < 0.001$) for stearic acid and SFA and lower for oleic acid ($p < 0.05$), LA ($p < 0.001$), PUFA ($p < 0.001$), n-9 MUFA (p < 0.01) and n-6 PUFA ($p < 0.001$), whereas there was no difference ($p > 0.05$) for palmitic acid, ALA, MUFA and n-3 PUFA (Table 6). The pigs fed the BT diet showed no differences ($p > 0.05$) for the deposition of palmitic acid, stearic acid, oleic acid, SFA, MUFA and n-9 MUFA, but the deposition of LA, PUFA and n-6 PUFA was lower ($p < 0.01$) and that of ALA was higher ($p < 0.05$) with a tendency towards a higher ($p = 0.058$) deposition of n-3 PUFA when compared with the pigs fed the SO diet (Table 6). The calculated the ratio of fatty acid deposition and digestible fatty acid intake showed that the pigs fed the BT diet had a lower ratio for palmitic acid ($p < 0.05$), stearic acid ($p < 0.05$) and SFA ($p < 0.001$) and a higher ($p < 0.05$) ratio for ALA and n-3 PUFA, but there were no differences ($p > 0.05$) for the ratios for oleic

acid, LA, MUFA, PUFA, n-9 MUFA and n-6 PUFA when compared with the pigs fed the SO diet (Table 6).

Table 6. Effect of dietary fat source on digestible fatty acid intake, fatty acid deposition and the deposition:intake ratio during the whole feeding period

Fatty acid	BT	SO	*p* value
Digestible fatty acid intake (g/35 days)			
C 16:0	141.41± 25.75	121.35± 24.80	0.189
C 18:0	182.32± 36.16	30.60± 7.53	0.000
C 18:1 n-9	301.51± 34.18	394.15± 66.66	0.016
C 18:2 n-6	185.00± 27.11	626.31± 112.64	0.000
C 18:3 n-3	117.95± 16.59	108.70± 19.54	0.400
∑ FA	1,046.22± 165.37	1,361.32± 185.93	0.026
∑ SFA	356.06± 72.60	165.69± 27.63	0.000
∑ MUFA	373.08± 52.01	416.82± 58.91	0.264
∑ PUFA	317.08± 45.41	778.80± 106.25	0.000
∑ n-9	314.06± 36.94	419.11± 53.45	0.009
∑ n-6	194.20± 28.83	662.80± 90.30	0.000
∑ n-3	122.88± 16.58	116.01± 15.96	0.529
Fatty acid deposition (g/35 days)			
C 16:0	316.19± 158.56	394.05± 154.49	0.400
C 18:0	203.92± 115.55	235.10± 83.90	0.583
C 18:1 n-9	563.28± 309.31	587.62± 263.02	0.882
C 18:2 n-6	86.13± 44.10	267.96± 135.75	0.007
C 18:3 n-3	28.34± 11.20	15.68± 6.56	0.025
∑ FA	1,475.87± 575.86	1,798.55± 587.54	0.416
∑ SFA	638.82± 250.52	727.34± 243.24	0.593
∑ MUFA	705.95± 323.70	734.69± 250.32	0.878
∑ PUFA	131.10± 35.37	336.52± 122.27	0.008
∑ n-9	661.59± 311.14	708.98± 237.81	0.791
∑ n-6	100.59± 35.15	317.73± 117.95	0.005
∑ n-3	30.51± 11.26	18.79± 5.69	0.058
Deposition : intake ratio			
C 16:0	2.16± 0.80	3.16± 0.73	0.041
C 18:0	1.20± 0.45	8.60± 3.63	0.010
C 18:1 n-9	1.81± 0.78	1.45± 0.47	0.319
C 18:2 n-6	0.45± 0.19	0.42± 0.17	0.733
C 18:3 n-3	0.24± 0.10	0.14± 0.05	0.030
∑ FA	1.38± 0.34	1.21± 0.30	0.391
∑ SFA	1.75± 0.43	4.44± 1.07	0.000
∑ MUFA	1.84± 0.56	1.58± 0.43	0.392
∑ PUFA	0.41± 0.08	0.37± 0.15	0.694
∑ n-9	2.05± 0.71	1.50± 0.47	0.134
∑ n-6	0.51± 0.12	0.42± 0.17	0.336
∑ n-3	0.25± 0.11	0.14± 0.05	0.027

Means ± SD for 6 pigs per experimental diet
BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0
Σ MUFA = C16:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C18:3 n-6 + C20:5 n-3
Σ n-9 = C18:1 n-9 + C20:1 n-9
Σ n-6 = C18:2 n-6 + C18:3 n-6
Σ n-3 = C18:3 n-3 + C20:5 n-3

Minimum de novo synthesis

For the entire feeding period, minimum *de novo* synthesis was calculated and found to show a tendency towards higher synthesis ($p = 0.076$) of SFA for the pigs on the SO diet, but there was no difference ($p > 0.05$) for the synthesis of MUFA. The calculated ratio for the *de novo* synthesis of SFA:(SFA + MUFA) was higher ($p < 0.05$) for the pigs fed the SO diet (Table 7).

Table 7.
Effect of dietary fat source on minimum *de novo* synthesis of fatty acids during the whole feeding period

Fatty acid	BT	SO	*p* value
Minimum synthesis (g/35 days)			
SFA	282.76± 188.32	561.65± 225.66	0.076
MUFA	332.87± 272.70	317.86± 201.42	0.922
SFA/(SFA+MUFA)	0.47± 0.10	0.65± 0.08	0.012

Means ± SD for 6 pigs per experimental diet
BT = Beef tallow + Linseed oil; SO = Sunflower oil + Linseed oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C14:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0
Σ MUFA = C16:1 + C18:1 n-7 + C18:1 n-9 + C20:1 n-9

DISCUSSION

The pigs fed the BT diet had a higher SFA content in adipose tissue than did the pigs fed the SO diet. This is caused by the different fatty acid patterns of the diets and *de novo* fatty acid synthesis. However, there was no diet difference for the SFA content of loin muscle, which may be explained by the fact that loin muscle is mainly comprised of structural lipids which can hardly be changed by the type of dietary fat. The higher MUFA fraction in adipose tissue and loin muscle of pigs fed the BT diet, which contained less oleic acid and MUFA, could be caused by a higher rate of desaturation or *de novo* synthesis. Consumption of the SO diet increased the PUFA content both in adipose tissue and loin muscle, which is the result of the higher content of LA and PUFA in the SO diet. In keeping with our hypothesis as mentioned above, there was a lower group mean lower content of ALA in loin muscle and adipose tissue on the SO versus BT diet even though the two diets had a similar ALA content. This observation would be explained by ALA being preferentially incorporated into muscle rather than into adipose tissue (Nguyen et al., 2003) and by ALA being preferentially oxidised (Cunnane and Anderson, 1997; Kabir and Ide, 1996). The higher ratios of PUFA:SFA, n-6 PUFA:n-3 PUFA and LA:ALA in adipose tissue and loin muscle of the pigs fed the SO diet are explained by the higher LA content of the SO diet.

The pigs fed either the BT diet or the SO diet displayed no difference in growth performance, although the digestibility of crude fat for the pigs fed the SO diet was higher than that for those fed the BT diet. The higher digestibility of palmitic and linoleic acid could relate to their position in the triacylglycerol molecule, fatty acids at the 2 position of the glycerol backbone of triacylglycerols being more efficiently absorbed than those at the 1,3 position (Bracco, 1994; Innis and Dyer, 1997; Nelson and Innis, 1999). In addition, saturated fatty acids located at the 1,3 position of

triacylglycerols will be hydrolysed by pancreatic lipase and subsequently could form insoluble calcium soaps that are excreted with faeces (Mu and Porsgaard, 2005). The high variation of stearic acid digestibility on the SO diet would be explained by microbial hydrogenation in the large intestine, transforming C18 unsaturated fatty acids to C18 saturated and/or monounsaturated fatty acids.

The current study demonstrates that the type of dietary fat had no effect on macronutrient composition and deposition (crude fat, water, crude protein and ash) in the whole body, when expressed either as absolute amount or as percentage of the whole body. The different dietary fat sources (beef tallow versus sunflower oil) in combination with similar ALA acid level had no influence on the intake, storage, expenditure and excretion of energy. Thus, the hypothesis that energy expenditure would be higher on the SO diet has to be rejected. However, Javadi et al. (2004) have found that different dietary fatty acids may differently affect energy expenditure in mice. On the other hand, the present study shows that there was higher minimum *de novo* fatty acid synthesis for the pigs fed the SO diet which would increase energy storage from *de novo* synthesis, subsequently resulting in an unchanged energy expenditure. The energy excreted with faeces as fat, expressed as either as absolute amount or as percentage of energy intake, was lower when the pigs were fed the SO diet. This is explained by the lower fraction of fat recovered in faeces as caused by the higher fat digestibility in these pigs.

Measurement of the apparent faecal digestibility for individual fatty acids and the intake of individual fatty acids made possible the calculation of digestible fatty acid intake. For the entire feeding period (35 days), the deposition of fatty acids in whole body fat was measured as based on the fatty acid composition of the total body before and after the experiment. As would be expected, the pigs fed the BT diet had ingested more stearic acid, less linoleic acid, but the same amount of ALA. The pigs fed the BT diet deposited in the body less LA, more ALA and an unchanged amount of stearic acid. Consequently, the calculated ratio of fatty acid deposition:digestible fatty acid intake for SO diet showed a ratio for stearic acid (8.60) that was much higher than 1, pointing at a very high rate of *de novo* synthesis of stearic acid. On the other hand, feeding pigs on the SO diet displayed ratios less than 1 for LA and ALA, indicating preferential oxidation of these fatty acids. There was no diet effect on the ratio for LA. The apparent difference in oxidation of ALA and LA may be the result of competition of these fatty acids for the enzymes involved in β-oxidation, these enzymes having a greater preference for n-3 fatty acids. When combining all results of fatty acid composition and deposition in the whole body, it would follow that feeding pigs on a LA-rich diet and a high level of ALA increases the deposition of LA and considerably elevates *de novo* synthesis of fatty acids. Furthermore, ALA was more preferentially oxidized when feeding the pigs on a diet rich in LA, which was predicted on the basis of previous investigations (Cunnane and Anderson, 1997; Ide et al., 1996; Jones et al., 1985).

The diet effect on the deposition:intake ratios for SFA and MUFA can be explained by the calculated minimum synthesis of these groups of fatty acids. Feeding the SO diet tended to increase the *de novo* synthesis of SFA, without affecting the *de novo* synthesis for MUFA. This would lead to a higher ratio of SFA:(SFA + MUFA) in the pigs receiving the SO diet. The higher *de novo* synthesis for SFA on the SO diet would increase energy retention in the body and may reduce energy expenditure as calculated from energy intake minus energy storage and excreted energy.

In summary, feeding pigs with a diet rich in LA and high level of ALA markedly increased the content of LA in adipose and loin muscle and increased the

digestibility of crude fat and LA, but did not influence the deposition and expenditure of energy when compared with feeding pigs on a diet rich in SFA and a high level of ALA. The measurements for fatty acid oxidation and deposition in the whole body indicated a higher *de novo* synthesis of fatty acids and preferential oxidation of ALA when the pigs were fed the diet rich in SFA and high in ALA.

ACKNOWLEDGEMENTS

The authors gratefully thank Chai-yod Phudtha and Sitthichai Phangsri for their excellent assistance. This study was supported by the Faculty of Natural Resources, Rajamangala University of Technology Isan by providing research facilities and by Mahanakorn University of Technology by providing financial support.

LITERATURE CITED

AOAC. 1990. Association of official analysis chemists, Washington DC.

Beynen, A.C., and M.B. Katan. 1985. Why do polyunsaturated fatty acids lower serum cholesterol? Am J Clin Nutr 42:560-3.

Bracco, U. 1994. Effect of triglyceride structure on fat absorption. Am J Clin Nutr 60:1002S-1009S.

Cunnane, S.C., and M.J. Anderson. 1997. The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. Journal of Nutrition 127:146-152.

Horwitz, W. 1975. Official Methods of Analysis of Official Analytical Chemist, p. 225 Official Methods of Analysis of Official Analytical Chemist, 12 ed. Benjamin Franklin Station, DC.

Ide, T., M. Murata, and M. Sugano. 1996. Stimulation of the activities of hepatic fatty acid oxidation enzymes by dietary fat rich in alpha-linolenic acid in rats. J Lipid Res 37:448-63.

Innis, S.M., and R. Dyer. 1997. Dietary triacylglycerols with palmitic acid (16:0) in the 2-position increase 16:0 in the 2-position of plasma and chylomicron triacylglycerols, but reduce phospholipid arachidonic and docosahexaenoic acids, and alter cholesteryl ester metabolism in formula-Fed piglets. J Nutr 127:1311-9.

Javadi, M., H. Everts, R. Hovenier, S. Kocsis, A.E. Lankhorst, A.G. Lemmens, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2004. The effect of six different C18 fatty acids on body fat and energy metabolism in mice. Br J Nutr 92:391-9.

Jones, P.J.H., P.B. Pencharz, and M.T. Clandinin. 1985. Whole body oxidation of dietary fatty acids: implications for energy utilization. American journal of clinical nutrition 42:769-777.

Kabir, Y., and T. Ide. 1996. Activity of hepatic fatty acid oxidation enzymes in rats fed alpha-linolenic acid. Biochim Biophys Acta 1304:105-19.

Metcalfe, L.D., A.A. Schmitz, and J.R. Pelka. 1966. Rapid preparation of fatty acid esters from lipids for gas chromatographic analysis. Anal. Chem. 38:514-515.

Mitchaothai, J., H. Everts, C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsuthavas, P. Srenanul, and A.C. Beynen. Meat quality, digestibility and deposition of fatty acids in growing-finishing pigs fed restricted, iso-energetic

amounts of diets containing either beef tallow or sunflower oil. Asian-Australian Journal of Animal Science, *Submitted*.

Mitchaothai, J., H. Everts, C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsuthavas, P. Srenanul, R. Hovenier, and A.C. Beynen. Digestion and deposition of individual fatty acids in growing-finishing pigs fed diets containing either beef tallow of sunflower oil. Journal of Animal Physiology and Animal Nutrition, *in press*.

Mu, H., and T. Porsgaard. 2005. The metabolism of structured triacylglycerols. Prog Lipid Res 44:430-48.

Nelson, C.M., and S.M. Innis. 1999. Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. Am J Clin Nutr 70:62-9.

Newsholme, E.A., and A. Leech. 1986. Biochemistry for the Medical Sciences. Reprint and correction ed. John Wiley & Sons, New York.

Nguyen, L.Q., M.C.G.A. Nuijens, H. Everts, N. Salden, and A.C. Beynen. 2003. Mathematicl relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. Meat Science 65:1399-1406.

Shields, R.G., Jr., D.C. Mahan, and P.L. Graham. 1983. Changes in swine body composition from birth to 145 kg. J. Anim. Sci. 57:43-54.

SPSS. 1999. SPSS for Windows, SPSS Inc., Chicago, IL, USA.

CHAPTER 7

Ileal digestibility of fatty acids and fatty acid metabolism in in Thai indigenous pigs fed diets containing either sunflower oil or beef tallow

J. Mitchaothai†§, H. Everts†, C. Yuangklang‡, S. Wittayakun‡, K. Vasupen‡, S. Wongsuthavas‡, P. Srenanul‡, R. Hovenier†, and A. C. Beynen†

§Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Nong-Chok, Bangkok 10530, Thailand

‡Faculty of Natural Resources, Rajamangala University of Technology-Isan, Phangkhon, Sakon Nakhon 47160, Thailand

†Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University, P.O. Box 50.152, 3508 TD, Utrecht, The Netherlands

To be submitted

ABSTRACT

The apparent ileal digestibility of fatty acids was measured in Thai indigenous pigs fed diets containing either 5 % (w/w) of beef tallow (BT) or sunflower oil (SO). In addition, the deposition in the whole body of macronutrients, individual fatty acids and energy were determined. Six cannulated pigs were fed a restricted amount of each diet and were subjected to a 2 × 2 Latin square design with 3 replicates and two periods of 15 days each. Another 18 pigs fed restricted amounts of each diet were used to determine whole body composition. The SO diet was found to increase ($p < 0.05$) the apparent ileal digestibilities of palmic, stearic, linoleic and saturated fatty acids and tended ($p = 0.627$) to increase the digestibility of polyunsaturated fatty acids (PUFA). The type of dietary fat had no effect ($p > 0.05$) on growth performance, proximate body composition and energy expenditure, even though the apparent digestibility of crude fat was higher ($p < 0.05$) and fat energy excreted with faeces was lower ($p < 0.05$) in the pigs fed the SO diet. The measured apparent ileal digestibilities were taken into account to calculation the amounts of digestible fatty acids ingested. Feeding the SO diet instead of the BT diet produced a higher ($p < 0.05$) deposition of linoleic acid (LA), oleic acid and PUFAs, but a lower ($p < 0.05$) deposition of stearic acid and saturated fatty acids (SFA). For the entire feeding period of 49 days, the ratio of deposition to intake of digestible fatty acids was increased ($p < 0.05$) for palmitic acid (C16:0) and stearic acid (C18:0) in the pigs fed the SO diet, but the ratio for α-linolenic acid (ALA) was decreased while the ratio for LA tended to be decreased. For both diets, the ratios of deposition:digestible intake for LA, ALA, PUFA, n-6 PUFA and n-3 PUFA were below 1, but the ratios for palmitic acid, stearic acid, oleic acid were above 1. The calculated minimum *de novo* synthesis of SFA was increased ($p < 0.05$) in pigs fed the SO diet, while that of MUFA was unchanged. It is concluded that feeding of a diet containing SO instead of BT may improved the apparent ileal digestibility of fatty acids, but may decrease the deposition:intake ratio for ALA in combination with a tendency towards decrease in the ratio for LA and without an effect on proximate body composition and energy expenditure.

Keywords: Deposition, Ileal digestibility, Beef tallow, Sunflower oil, Energy expenditure, Indigenous pigs

INTRODUCTION

Previous studies (Mitchaothai et al., *Chapter 5;* Nguyen et al., 2003) have shown that the incorporation rate of α-linolenic acid (ALA) into adipose tissue is lower than that of linoleic acid (LA). This may be explained by ALA being preferentially oxidized when compared with LA which in turn is oxidized more rapidly than are saturated fatty acids (Cunnane and Anderson, 1997; Jones et al., 1985). Studies in chickens have demonstrated that the feeding of diets rich in polyunsaturated fatty acids (PUFA) can lower abdominal fat (Crespo and Esteve-Garcia, 2002a; Crespo and Esteve-Garcia, 2002b; Sanz et al., 2000b). This observation may be explained by the high rate of oxidation of PUFA. Carbohydrates can be metabolized into fatty acids which can be stored in adipose tissue, but the energetic efficiency for the deposition of *de novo* synthesized fatty acids is less than the efficiency of the deposition in adipose tissue of dietary fatty acids (Kirchgessner and Muller, 1998). When planning the present study we hypothesized that pigs fed a diet rich in fed PUFA instead of saturated fatty acids (SFA) would have higher energy expenditure and fatty acid oxidation. The oxidation of individual fatty acids can be estimated from the calculated ratio of deposition in whole body:digestible intake for fatty acids. Ideally, for such calculations, the ileal digestibility of individual fatty acid should be known. The published values for the apparent ileal digestibility of fatty acids in pigs are quite variable (Jørgensen et al., 1997; Li et al., 1990), indicating that the values may depend on diet and animal characteristics. Thus, in this trial the apparent ileal digestibility of fatty acids was measured so that the subsequent calculations would be valid for the diets and species and category of the pigs used.

MATERIALS AND METHODS

Experiment 1. Determination of ileal and total-tract digestibility of fatty acids

This experiment was designed to determine the ileal digestibility of macronutrients, including individual fatty acids.

Animals and surgical procedures

Three female and three male Thai indigenous Kadon pigs were anaesthetized with xylazine (2 mg/kg i.m.; Xylaz®, Farvet Laboratories, The Netherlands) and sodium pentobarbitone (5 mg/kg with maximum dose of 10 mg/kg, i.v.; Nembutal®, Ceva Animale, France). All pigs were fitted with a simple T-piece cannula made from silicone at 10 - 15 cm anterior to the ileocaecal junction according to the surgical procedure described by Donkoh et al. (1994). The cannulated pigs regained consciousness 6 – 8 h after surgery. The pigs were kept individually in smooth-walled metabolism cages. Average BW of the pigs was 18.10 ± 1.60 kg at the commencement of the experimental period.

Design, diets and feeding

The experimental design used was a Latin square design (2 × 2) with 3 replicates. Each experimental period consisted of 7 days of adaptation, followed by 5 days for total collection, one day of collection of ileal digesta, 1 day of rest and a second day of collection of ileal digesta. The two experimental diets are given in

Table 1. The diets had an identical base composition, but they contained 5% of either beef tallow (BT) or sunflower oil (SO). Titanium dioxide (TiO_2) was included in both diets at 1 g/kg feed to measure ileal digestibility. The pigs were fed at a level of 4% DM of the experimental diets relative to the average BW; water was freely available. The experimental diets were provided for each pig by splitting them into three equal portions and were fed at 08:00, 16:00, and 24:00 h.

Table 1. Composition of the diets (as-fed basis)

Item	Beef tallow diet	Sunflower oil diet
Raw materials (g/100 g)		
Cassava chips	45.43	45.43
Soybean meal (44% CP)	34.00	34.00
Extruded soy beans	7.00	7.00
Molasses	4.00	4.00
Calcium carbonate	1.00	1.00
Di-calcium phosphate	2.70	2.70
NaCl	0.35	0.35
DL-Methionine	0.17	0.17
Premix	0.25	0.25
TiO_2 (Marker)	0.10	0.10
Beef tallow	5.00	-
Sunflower oil	-	5.00
Total	100.00	100.00
Analysed nutrients (%)		
Dry matter	89.15	88.29
Crude protein	20.61	21.44
Crude fat	7.43	7.21
Ash	7.81	7.52
Calculated ME (MJ/kg)	13.82	13.97

Digesta and faeces collection and chemical analysis

Faeces were quantitatively collected for 24 hours on five consecutive days by the use of plastic bags. The bags were weighed and then stored at – 20 ^{o}C pending analysis. Ileal digesta were collected according to the digesta collection method described by Jørgensen et al. (1997). All ileal digesta samples were weighed and stored at – 20 ^{o}C.

The samples of faeces and ileal digesta were freeze-dried, pooled for each day, pig and period and then ground through a 1-mm mesh screen. All samples were then analysed for crude protein, crude fat, and ash (AOAC, 1990). The concentration of titanium dioxide in feed, ileal digesta and faeces were quantified by emission spectrophotometry as the procedure described by Myers et al. (2004).

Digestibility calculations and statistical analysis

Apparent ileal (AID_M) and faecal digestibility (AFD_M) measured by the dietary marker method were calculated according to the equations [1] and [2], respectively.

$$AID_{Ma} = 100 - \{100 \times [(I_a \times D_{Ti}) / (I_{Ti} \times D_a)]\} \quad [1]$$

$$AFD_{Ma} = 100 - \{100 \times [(F_a \times D_{Ti}) / (F_{Ti} \times D_a)]\} \quad [2]$$

where I_a = concentration of nutrient a in ileal digesta (g/kg), I_{Ti} = concentration of TiO_2 in ileal digesta (g/kg), D_a = concentration of nutrient a in feed (g/kg), D_{Ti} = concentration of TiO_2 in feed (g/kg), F_a = concentration of nutrient a in faeces (g/kg), and F_{Ti} = concentration of TiO_2 in faeces (g/kg).

To compare with the dietary marker method, apparent faecal digestibility (AFD_{Total}) measured by the total collection method was calculated according to equation [3].

$$AFD_{Total} = [(DFI_a - DFE_a)/(DFI_a)] \times 100 \quad [3]$$

where DFI_a = daily feed intake of nutrient a (g), DFE_a = daily faecal excretion of nutrient a (g).

Correlation between the dietary marker and the total collection method for the apparent faecal digestibility for macronutrients, selected fatty acids and grouped fatty acids were evaluated by Bivariate correlation in SPSS programme (SPSS, 1999) and the difference between apparent ileal and faecal digestibility was evaluated by the paired-sample Student's *t*-test with the SPSS programme (SPSS, 1999). The difference between the two dietary treatments for the macronutrients, selected fatty acids and grouped fatty acids were evaluated with the use of ANOVA according to a 2 × 2 Latin square in SAS programme (SAS, 1996). The level of statistical significance was preset at $p < 0.05$.

Experiment 2. Determination of fatty acid digestion and deposition, body composition and energy balance

This experiment was designed to determine the apparent faecal digestibility of macronutrients and fatty acids, the balance of macronutrients and fatty acids and the deposition of fatty acids in whole body.

Animals, Diets and Feeding

Eighteen male and female Thai indigenous Kadon pigs were used; the average BW was 11.94 ± 1.83 kg. Three males and three females were selected to be slaughtered for baseline measurements. Then, the remaining 12 pigs were allotted to one of the two dietary treatments on the basis of body weight and sex and were housed in individual cages. The pigs were fed a restricted amount of the experimental diets and were given 4% of DM relative the initial BW. Drinking water was freely available. The experimental diets contained either 5.0 % of BT or SO (Table 1); the amount of cassava chips was increased to a level of 45.53 % because the marker (TiO_2) was not added to the diets. Throughout the entire 49 days of the experiment, the faeces and urine samples of all pigs were collected to determine digestibility and balance of macronutrients and gross energy. A 10% solution of sulphuric acid (H_2SO_4) was used to prevent ammonia evaporation during urine collection and the urine samples were stored at 5 ^oC until analysis. All pigs were slaughtered at an average BW of 25.07 ± 1.45 kg. Ingredients and macronutrient composition of the experimental diets are shown in Table 1. The fatty acid composition of the diets is given in Table 2.

Collection and Pre-treatment of Samples

The six pigs (3 male and 3 female) for baseline measurements were killed by an intravenous injection of sodium pentobarbital (200 mg/kg of body weight). The

pigs were scalded, blood was collected and weighed, the head was removed, and the carcass was split into halves along the median plane according to the procedure previously reported by (Shields et al., 1983). This procedure was modified by drying compartment samples and both sides of the sawed carcass at 60 ^{o}C for 1 week in a forced-hot air oven with placing the frozen materials on a mesh with an underneath tray to collect the dripping oil. The dried whole carcass samples derived from each pig were pooled and then weighed. The dripping oil samples were pooled and weighed in the same way as the dried whole carcass samples, therefore there were the dried and oil samples for each pig. The dried samples were individually ground once through a 2 mm screen and twice through a 1 mm screen by a hammer mill. The 12 pigs in the feeding trial were sacrificed at 7 weeks after the commencement. Then, whole carcass sample was prepared in the same way as for the baseline pigs.

Chemical Analyses and Fatty Acid Determination

The samples of diets and faeces were dried at 60 ^{o}C for 72 h in a forced-hot air oven. The dried diets, faeces, and whole carcass samples were analysed for moisture, energy, crude protein, crude fat, and ash (AOAC, 1990). The urine samples were also analysed to quantify the content of energy and nitrogen (AOAC, 1990).

The dried samples of diets, faeces, and whole carcasses samples were extracted as described previously (Horwitz, 1975). Each sample was added to a flask and 2 ml of ethanol was added. Subsequently, 10 ml of HCl (8 mol/L) was added; the contents were gently mixed, and the flask was placed in a water-bath of 80 ^{o}C for 30-40 min. The tubes were cooled down, 10 ml of ethanol (96%, w/w) and 25 ml of petroleum ether (boiling point between 40 and 60 ^{o}C) were added and the tube was shaken for one min. The fat-containing upper layer was decanted into a 150-ml round-bottom flask. The extraction procedure was repeated twice with 15 ml of diethyl ether and 15 ml of petroleum ether and the lipid extract was evaporated to dryness under N_2 in a water-bath of 40 ^{o}C. The round-bottom flasks with the lipids were dried overnight at 60 ^{o}C and the total lipids were measured gravimetrically. Total lipids and the dripping oil were saponified and methylated according to the procedure of Metcalfe et al. (1966) followed by gas chromatography for determination of fatty acid composition (Javadi et al., 2004).

Determination of Body Composition and Energy Balance

The body weight and composition of the baseline pigs showed no effect of sex, hence the average energy or nutrient content in whole body of the baseline pigs was assumed to be similar to the fraction in the whole body of the experimental pigs at the start. All types of samples including the dripping oil were determined with a bomb calorimeter (Parr® Oxygen Bomb Calorimeter 1341 EE adiabatic, Parr Instrument Company, Moline, Illinois, USA). As a thermo-chemical standard, benzoic acid was used (Asian Pacific Specialty Chemical Limited, NSW, Australia). The total amount of energy that was lost as heat (heat production or energy expenditure) was calculated with the formula: Energy lost as heat = energy in food – energy in faeces and urine – energy stored in body. Energy stored in the body was determined as total energy at the end of the 49 d feeding period minus the energy in the body at the beginning of the experiment (= individual body weight × mean energy content). The same procedure was used to calculate the water, protein, fat and ash retention.

Table 2. Analysed fatty acid composition of the experimental diets

Item	Beef tallow diet	Sunflower oil diet
Fatty acids, g methylester/100g methylesters		
C 10:0	0.05	0.01
C 14:0	3.47	0.14
C 14:1	0.27	0.00
C 15:0	0.81	0.05
C 16:0	27.90	13.71
C 16:1	1.66	0.01
C 17:0	1.61	0.45
C 17:1	0.45	0.05
C 18:0	24.52	6.78
C 18:1n-9	25.31	38.51
C 18:1n-7	-	-
C 18:2n-6	10.18	35.80
C 18:3n-6	-	-
C 18:3n-3	2.08	2.30
C 18:4n-3	-	-
C 20:0	0.45	0.50
C 20:1n-9	0.02	0.09
C 20:2n-6	0.04	0.10
C 20:3n-7	-	-
C 20:4n-6	-	0.01
C 20:3n-3	0.01	0.03
C 20:5n-3	0.32	0.04
C 22:0	0.19	-
C 22:1n-9	-	0.05
C 22:2n-6	-	0.05
C 22:4n-6	-	-
C 22:3	-	-
C 22:5 n-3	-	-
C 24:0	-	0.14
C 22:6n-3	-	-
Unidentified	0.66	1.18
Σ SFA	59.00	21.78
Σ MUFA	27.71	38.71
Σ PUFA	12.63	38.32

SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids

Σ SFA = C10:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C14:1 + C16:1 + C17:1 + C18:1 n-9 + C20:1 n-9 + C22:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-6 + C18:3 n-3 + C20:2 n-6 + C20:3 n-3 + C20:5 n-3 + C22:2 n-6

Calculation of Ileal Digestible Fatty Acid Intake, Fatty Acid Deposition and Minimum De novo Synthesis

The ileal digestible fatty acid intake was calculated as fatty acid intake (g/49 days) × 0.95 × apparent ileal fatty acid digestibility (fraction of intake). The deposition of fatty acids was calculated with the following formula: Fatty acid deposition (g/49 days) = individual carcass content of fatty acid at the end of the study – individual carcass content of fatty acid at the start of the study (= BW × 0.95 × mean fatty acid composition of the baseline pigs). Carcass content of fatty acid at the end (g) was calculated as the whole carcass fat mass (g) × 0.95 × fraction of fatty acid in whole carcass. The ratio of fatty acid deposition and ileal digestible fatty acid intake was calculated for selected individual fatty acids and groups of fatty acids. The quantity of fatty acids oxidized, transformed and synthesized could not be measured. Therefore, minimum *de novo* fatty acid synthesis was calculated as fatty deposition minus ileal digestible fatty acid intake.

Statistical Analyses

The effect of fat type in the diets was evaluated for statistical significance by the Student's *t* test (SPSS, 1999) and SAS programme (SAS, 1996) at the significant level of $p < 0.05$. Results are expressed as means ± SD.

RESULTS

Experiment 1

Dietary Fatty Acid Composition

The fatty composition of the experimental diets is shown in Table 2. The fat component of the BT diet contained approximately 59 % SFA, whereas that of the SO diet contained approximately 38 % PUFA.

Digestibility of macronutrients and fatty acids

One pig died before the end of the first period. The necropsy and histopathology findings indicated that mycoplasma pneumonia infection was the cause of death. Thus, there were 5 pigs for analysis in the Experiment 1. There was no relation ($p > 0.05$) between the apparent faecal dry matter digestibility as measured by dietary marker method and total-collection method, but the apparent faecal digestibility of crude fat and crude protein as measured by the dietary marker method was significantly correlated ($p < 0.05$) to those measured by the total-collection method (Table 3). There were high correlations ($p < 0.01$) between both measurement methods for the apparent digestibility of palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1 n-9), LA, ALA, SFA, mono-unsaturated fatty acid (MUFA) and PUFA (Table 3). All apparent faecal digestibilities, except for stearic acid and SFA, measured by dietary marker method were higher than those obtained by the total-collection method.

When the apparent ileal and faecal digestibilities for macronutrients and fatty acids as determined by the dietary marker method were compared (Table 3), there were no differences ($p > 0.05$) for the digestibilities of oleic acid and MUFA. There was a trend towards lower ileal digestibility of crude fat ($p = 0.064$) and there were significantly lower ($p < 0.05$) ileal digestibilities for dry matter, crude protein,

palmitic acid, oleic acid, LA, ALA and PUFA, and significantly higher ($p < 0.05$) ileal digestibilities for stearic acid and SFA.

The apparent ileal digestibility of crude fat for the SO diet was higher ($p < 0.05$) than that for the BT diet, but there was no difference ($p > 0.05$) between both dietary treatments as to dry matter and crude protein.

The pigs fed the SO diet had higher ($p < 0.05$) apparent ileal digestibilities for palmitic acid, stearic acid, LA and SFA, and showed a trend ($p = 0.0627$) towards higher ileal digestibility of PUFA (Table 4). There were no differences ($p > 0.05$) between both diets for the apparent ileal digestibilities of oleic acid, ALA and MUFA. No difference ($p > 0.05$) between both treatments was found for the apparent digestibilities of oleic acid and MUFA (Table 4). The apparent faecal digestibilities for palmitic acid, LA, ALA and PUFA were higher ($p < 0.05$) in the pigs fed the SO diet, but that for stearic was lower ($p < 0.05$) and that for SFA tended to be lower ($p = 0.0743$) in the pigs fed the SO diet.

Experiment 2

Growth performance and body composition

There was no difference ($p > 0.05$) in average daily gain and feed conversion ratio between both experimental diets (Table 5).

Digestibility of macronutrients and fatty acids

The fatty acid composition of the diets were almost identical to that of the diets used in Experiment 1 (Table 1). The apparent faecal digestibilities for macronutrients (Table 5) and fatty acids (Table 6) in Experiment 2 were similar to that in Experiment 1 and the two experiments yielded similar diet effects, except for lack of diet effect ($p > 0.05$) on the apparent digestibility of palmitic acid in Experiment 2.

Body composition and energy balance

Table 5 illustrates that the absolute weight, percentage, and amounts of deposition of macronutrients (fat, water, crude protein, and ash) in the whole pig body were not different ($p > 0.05$) for the dietary treatments. Energy intake, storage, expenditure, losses in faeces and urine, and losses in faeces as fat-free matter showed no diet effect ($p > 0.05$) when expressed as absolute values or as percentage of energy intake. However, the losses with faeces as fat in the pigs fed the SO diet were higher ($p < 0.05$) than in those fed the BT diet, irrespective of being expressed as absolute amount or percentage of total energy intake. The amount of energy in the whole body at the end of the experiment as derived from measurement by bomb calorimetry was similar to that calculated using the equation given Javadi et al. (2004) (Table 5).

Ileal Digestible Fatty Acid Intake and Deposition

The ileal digestible intake for individual and grouped fatty acid in both experimental diets had no variation because there were no feed refusals. Thus the values were based on one amount of feed intake for all pigs and the mean of apparent ileal digestibility derived from the Experiment 1 (Table 7). There was no difference (*p*

> 0.05) in the deposition of palmitic acid for both dietary treatments. The pigs fed the BT diet had a higher deposition of stearic acid and SFA ($p < 0.05$) and tended to have a higher deposition of ALA ($p = 0.0798$) and n-3 PUFA ($p = 0.0884$) (Table 7). The depositions of oleic acid, LA, MUFA, PUFA, n-9 MUFA and n-6 PUFA were higher in the pigs fed the SO diet. The calculated ratios of fatty deposition: ileal digestible intake for oleic acid, MUFA, PUFA, n-9 MUFA, n-3 PUFA and total fatty acids were not different for the two diet groups ($p > 0.05$). The pigs fed the SO diet showed higher ($p < 0.05$) ratios for palmitic acid, stearic acid and SFA, whereas the pigs fed the BT diet had higher ratio for ALA ($p < 0.05$) and a tendency towards a higher ratio for LA ($p = 0.0957$) and n-6 PUFA ($p = 0.0945$) (Table 7). The ratios for LA, ALA and PUFA were less than 1.

Minimum de novo synthesis

The pigs fed the SO diet showed a higher ($p < 0.01$) minimum *de novo* synthesis of SFA, but no difference ($p > 0.05$) as to the synthesis of MUFA (Table 8). The ratio for the minimum *de novo* synthesis of SFA:(SFA + MUFA) in the pigs fed the SO diet was higher ($p < 0.05$) than in the pigs fed the BT diet.

DISCUSSION

The high correlation between the dietary marker method and the total-collection method for the apparent faecal digestibilities of macronutrients and fatty acids indicate that the dietary marker can be used to assess the apparent faecal digestibility. This would enhance the usefulness of the data from various previous investigations (Donkoh et al., 1994; Houdijk et al., 1999; Jagger et al., 1992; Jørgensen et al., 1997; Myers et al., 2004). From these investigations, various details concerning feeding interval, sample collection and marker quantification were integrated in the present protocol. The faecal digestibility of dry matter and crude protein was higher than the apparent ileal digestibility which can be explained by the activity of microbes in the large intestine as has been described (Li et al., 1990; Lindberg and Cortova, 1995; Pettersson and Lindberg, 1997). It was also described that faecal digestibility of crude fat would be lower, whereas in this study an opposite trend was found. This discrepancy could be the result of a difference in endogenous losses and/or an effect of fat type, resulting in different digestibilities for individual fatty acids. Jørgensen et al. (2000) reported a higher faecal than ileal digestibility of crude fat for diets containing fish oil and rapeseed oil diet. Additionally, the age of the pigs might have an influence as Houdijk et al. (1999) reported a higher faecal crude fat digestion for growing pigs, but a lower faecal crude fat digestion in weanling pigs.

The effect of fat type on the digestibility of macronutrients and fatty acids can be compared at the levels of the terminal ileum and faeces. The pigs fed the SO diet had a higher crude fat digestibility both at the end of ileum and at the faecal level which may be caused by the high PUFA content of the diet. PUFA are better digestible than are SFA, which were abundant in the BT diet. The absence of a diet effect on the digestibility of dry matter and crude protein at the levels of the end of the ileum and faeces probably relates to the similar macronutrient composition of the two diets. In comparison with data published by Jørgensen et al. (2000), the apparent ileal and faecal digestibility for selected and grouped fatty acids were higher in the current study. This was probably due to the difference in fat sources between the two studies. However, the apparent ileal digestibility for fatty acids in the current study was close

Table 3. Correlation of measured faecal digestibility of macronutrients and fatty acids between dietary marker and total collection methods and comparison between ileal and faecal digestibility measured by dietary marker method

Item	Method		Pearson correlation (r)	P value	Position of measurement		P value
	Dietary marker	Total collection			Ileal digestibility	Faecal digestibility	
DM	88.86 ± 0.75	86.24 ± 1.29	-0.316	0.374	76.69 ± 2.62	88.86 ± 0.75	0.000
EE	81.11 ± 5.13	76.86 ± 5.73	0.925	0.000	79.13 ± 5.69	81.11 ± 5.13	0.064
CP	88.75 ± 1.19	86.03 ± 2.22	0.636	0.048	84.79 ± 2.47	88.75 ± 1.19	0.000
C 16:0	77.54 ± 4.11	72.39 ± 4.55	0.783	0.007	73.43 ± 6.17	77.54 ± 4.11	0.036
C 18:0	42.38 ± 20.68	27.66 ± 27.69	0.961	0.000	66.91 ± 10.57	42.38 ± 20.68	0.014
C 18:1 n-9	88.76 ± 2.41	86.19 ± 2.89	0.800	0.005	86.83 ± 2.39	88.76 ± 2.41	0.143
C 18:2 n-6	95.45 ± 1.29	94.46 ± 2.23	0.966	0.000	83.21 ± 4.21	95.45 ± 1.29	0.000
C 18:3 n-3	94.54 ± 3.10	93.29 ± 3.69	0.989	0.000	87.31 ± 3.69	94.54 ± 3.10	0.000
∑ SFA	59.96 ± 9.41	50.03 ± 12.91	0.936	0.000	70.17 ± 8.16	59.96 ± 9.41	0.028
∑ MUFA	88.76 ± 2.41	86.19 ± 2.88	0.800	0.005	86.83 ± 2.39	88.76 ± 2.41	0.143
∑ PUFA	94.99 ± 2.47	93.87 ± 2.93	0.832	0.003	85.26 ± 3.49	94.99 ± 2.47	0.000

SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids

Σ SFA = C16:0 + C18:0

Σ MUFA =C18:1 n-9

Σ PUFA = C18:2 n-6 + C18:3 n-3

Table 4. Effect of dietary fat type on ileal and faecal digestibility of macronutrients and fatty acids by the use of dietary marker

Item	BT	SO	P value
Ileal digestibility, % of intake			
Dry matter	76.25 ± 3.17	77.13 ± 2.24	0.4810
Crude fat	74.66 ± 3.50	83.60 ± 3.26	0.0055
Crude protein	84.17 ± 2.93	85.41 ± 2.05	0.3282
Faecal digestibility, % of intake			
Dry matter	88.48 ± 0.62	89.24 ± 0.70	0.1717
Crude fat	76.59 ± 2.20	85.63 ± 1.84	0.0016
Crude protein	88.51 ± 1.54	88.99 ± 0.83	0.1811
Ileal digestibility, % of intake			
C 16:0	70.74 ± 3.92	76.12 ± 7.22	0.0367
C 18:0	59.35 ± 6.37	74.46 ± 8.27	0.0002
C 18:1 n-9	87.21 ± 2.47	86.45 ± 2.53	0.6441
C 18:2 n-6	80.11 ± 2.79	86.32 ± 2.82	0.0223
C 18:3 n-3	86.00 ± 3.85	88.62 ± 3.38	0.3790
Σ SFA	65.09 ± 4.93	75.29 ± 7.73	0.0048
Σ MUFA	87.21 ± 2.47	86.45 ± 2.53	0.6441
Σ PUFA	83.05 ± 3.28	87.47 ± 2.11	0.0627
Faecal digestibility, % of intake			
C 16:0	74.22 ± 2.26	80.86 ± 2.31	0.0029
C 18:0	57.28 ± 4.28	27.47 ± 19.71	0.0465
C 18:1 n-9	88.10 ± 1.53	89.42 ± 3.11	0.4235
C 18:2 n-6	93.94 ± 1.38	96.96 ± 0.83	0.0377
C 18:3 n-3	91.75 ± 1.45	97.32 ± 0.38	0.0058
Σ SFA	65.75 ± 3.21	54.16 ± 10.24	0.0743
Σ MUFA	88.10 ± 1.53	89.42 ± 3.11	0.4235
Σ PUFA	92.84 ± 1.40	97.14 ± 0.42	0.0107

BT = beef tallow; SO = sunflower oil
SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids

Σ SFA = C10:0 + C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C18:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C20:3 n-3 + C20:5 n-3

to the values reported by Li et al. (1990). The higher ileal digestibility for palmitic acid, stearic acid and ALA acid for the SO diet could relate to a difference in the position of these fatty acids in the triacylglycerol molecule. Fatty acids at the 2 position of glycerol backbone of triacylglycerol molecules are better digested than those at the 1,3 position (Bracco, 1994; Innis and Dyer, 1997; Nelson and Innis, 1999). On the other hand, fatty acids released from the 1,3 position of triacylglycerols may lead to the less formation of indigestible calcium soaps (Mu and Porsgaard, 2005). For both dietary treatments, the apparent faecal digestibilities for palmitic acid, oleic acid, LA, ALA, MUFA and PUFA were increased when compared with the apparent ileal digestibility of these fatty acids. This could be a consequence of fatty acids that were digested in the small intestine or were modified by microbes in the

large intestine. On the other hand, the faecal digestibility for stearic acid was dramatically decreased, resulting in a very lower digestibility of SFA for the SO diet. However there was little decrease in stearic acid faecal digestibility and the increase in palmitic acid faecal digestibility for the BT diet led to a slightly higher feacal digestibility for this dietary treatment. The decrease in the faecal digestibility of stearic acid and the higher faecal digestibility for LA and ALA for the SO oil diet could be the result of microbial hydrogenation in the large intestine, leading to the conversion of LA and ALA into stearic acid. This would then explain the lower faecal digestibility for stearic acid and the higher faecal digestibility for LA and ALA for the BT diet, but the amount of LA in the BT diet was low and thus had only a small effect on the faecal digestibility of SFA.

In the experiment 2, it was found that the proximate body composition was not influenced by dietary fat type, irrespective of whether it was expressed as absolute amounts or fractions of fat, water, protein and ash. These results agree with a previous report for broiler chickens (Wongsuthavas et al., 2007). Similarly, energy expenditure, either expressed as absolute amount or as percentage of intake, was not significantly related to the high amounts of LA or PUFA in the SO diet. This lack of diet effect is at variance with a previous report on a study with mice (Javadi et al., 2004). Possibly, the method used to determine energy expenditure was not sufficiently sensitive.

The absence of a diet effect on growth performance agrees with the lack of diet effect on macronutrient deposition and the amount of energy stored in the body. However, the pigs fed the SO diet digested fat more efficiently than those fed the BT diet. This may be explained by the lower absolute energy content and the lower percentage of intake relative to the fat fraction in faeces.

The apparent faecal digestibilities of dry matter, crude fat and crude protein in Experiment 2 were similar to those found in Experiment 1. In addition, the apparent faecal digestibility results for individual and grouped fatty acids in experiment 2 were also comparable to those seen in Experiment 1. Hence, it would be assumed that the apparent ileal digestibilities for macronutrients and fatty acids in Experiment 1 may be used to calculate intakes of digestible fatty acids in Experiment 2.

As would be expected on the basis of the fatty acid composition of the diets, the pigs fed the SO diet stored less stearic acid and SFA, but stored more oleic acid, LA, MUFA, PUFA, n-9 MUFA and n-6 PUFA which is explained by the fractions of these fatty acids in the diet, except for ALA and n-3 PUFA which tended to be deposited less than for the BT diet which contained less ALA.

To determine the preferential deposition of fatty acids, the ratio of deposition of a given fatty acid or group of fatty acids to the intake of ileal digestible fatty acid or group of fatty acids was calculated. The deposition:intake ratio for fatty acid reflects the net amount synthesized and/or the proportion of the digested fatty acid not oxidized. A deposition:intake ratio > 1 would point at net *de novo* synthesis, whereas a ratio < 1 would indicate net oxidation and/or metabolism in the eicosanoid pathway. The ratio of deposition: ileal digestible intake for palmitic acid, stearic acid, oleic acid, SFA and MUFA were higher than 1 for both dietary treatments indicating significant *de novo* synthesis of these fatty acids, whereas the ratio for LA and ALA was less than 1 for the two diets, which must have resulted from the well-known preferential oxidation of LA and ALA, when compared with SFA (Cunnane and Anderson, 1997; Jones et al., 1985; Yeom et al., 2005). Additionally, the deposition:intake ratio for ALA and LA for the SO diet was lower, or tended to be lower, than that for the BT diet. This may be explained by the fact that the SO diet provided more LA and ALA for energy expenditure. It should be note that the ratio

Table 5. Body composition and energy balance in Thai indigenous pigs fed the experimental diets for 49 days

Item	BT	SO	P value
Food intake (kg dry food/49 d)	30.77	30.47	-
Feed conversion	2.92 ± 0.21	2.75 ± 0.17	0.1724
Initial body weight (kg)	12.68 ± 1.73	12.98 ± 0.77	0.7103
Final body weight (kg)	24.57 ± 1.66	25.57 ± 1.14	0.2547
Average daily gain (g/day)	243 ± 18.72	257 ± 16.35	0.1900
Apparent dry matter digestibility (%)	88.90 ± 2.37	89.08 ± 1.93	0.8867
Apparent fat digestibility (%)	78.78 ± 4.07	84.36 ± 3.32	0.0272
Apparent crude protein digestibility (%)	89.68 ± 2.11	89.88 ± 1.87	0.8667
Apparent gross energy digestibility (%)	91.78 ± 2.12	91.84 ± 1.75	0.9603
Body composition			
Fat (kg)	2.77 ± 0.25	2.95 ± 0.18	0.1912
Water (kg)	14.67 ± 0.93	14.92 ± 0.95	0.6520
Protein (kg)	6.02 ± 0.56	6.35 ± 0.58	0.3260
Ash (kg)	0.52 ± 0.05	0.54 ± 0.05	0.4251
Recovery (%)	97.61 ± 1.55	96.84 ± 2.15	0.4961
Water : Protein ratio (kg/kg)	2.45 ± 0.19	2.36 ± 0.22	0.4743
Fat (%)	11.26 ± 0.50	11.54 ± 0.73	0.4573
Water (%)	59.76 ± 2.07	58.37 ± 2.83	0.3561
Protein (%)	24.47 ± 1.31	24.82 ± 1.37	0.6635
Ash (%)	2.12 ± 0.33	2.11 ± 0.15	0.9770
Change in body composition			
Weight gain (kg)	11.88 ± 0.92	12.58 ± 0.80	0.1900
Fat deposition(kg)	1.36 ± 0.10	1.51 ± 0.20	0.1448
Water deposition(kg)	6.43 ± 0.66	6.48 ± 0.95	0.9069
Protein deposition(kg)	3.42 ± 0.47	3.69 ± 0.47	0.3313
Ash deposition(kg)	0.23 ± 0.08	0.25 ± 0.04	0.6587
Energy balance (MJ)			
Intake	513.16 -	502.95 -	-
Storage	127.31 ± 61.59	139.32 ± 53.62	0.7261
Expenditure	335.79 ± 57.47	315.10 ± 56.23	0.5469
In faeces	42.17 ± 10.89	41.04 ± 8.78	0.8477
In urine	7.89 ± 0.85	7.48 ± 1.00	0.4613
In faeces as fat	16.87 ± 3.24	12.53 ± 2.55	0.0287
In fat free faeces	25.31 ± 8.00	28.52 ± 6.47	0.4631
Energy in whole body (MJ)			
Initial body energy (MJ)	131.64 ± 18.01	134.76 ± 8.00	0.7103
Final body energy (MJ) (measured)[1]	260.05 ± 69.11	275.29 ± 51.28	0.6745
Final body energy (MJ) (calculated)[2]	259.14 ± 23.88	276.75 ± 18.98	0.1891
Percentage of energy intake			
Stored in the body	24.81 ± 12.00	27.70 ± 10.66	0.6684
Expended as heat	65.44 ± 11.20	62.65 ± 11.18	0.6756
Lost in faeces	8.22 ± 2.12	8.16 ± 1.75	0.9603
Lost in urine	1.54 ± 0.17	1.49 ± 0.20	0.6428
Lost in faeces as fat	3.29 ± 0.63	2.49 ± 0.51	0.0379
Lost in fat-free faeces	4.93 ± 1.56	5.67 ± 1.29	0.3926

Means ± SD for 6 pigs per experimental diet
BT = beef tallow; SO = sunflower oil
[1]Measured with a bomb calorimeter
[2]Calculated on basis of the body composition, given 1 g of animal fat has a gross energy of 39.8 kJ and 1 g of protein has a gross energy of 23.7 kJ (Javadi et al., 2004)

Table 6. Effect of dietary fat type on faecal digestibility of fatty acids

Item	BT	SO	P value
C 16:0	69.37± 3.96	73.18± 7.99	0.3281
C 18:0	55.45± 5.23	27.79± 12.56	0.0018
C 18:1 n-9	86.51± 3.36	89.66± 3.42	0.1389
C 18:2 n-6	93.34± 1.83	96.92± 0.91	0.0033
C 18:3 n-3	93.39± 3.29	98.34± 0.74	0.0132
Σ SFA	60.58± 5.00	49.35± 13.54	0.1026
Σ MUFA	86.51± 3.36	89.66± 3.42	0.1389
Σ PUFA	93.36± 2.45	97.63± 0.60	0.0070

BT = beef tallow; SO = sunflower oil
SFA = saturated fatty acids; MUFA = mono-unsaturated fatty acids; PUFA = poly-unsaturated fatty acids
Σ SFA = C10:0 + C14:0 + C16:0 + C17:0 + C18:0 + C20:0
Σ MUFA = C17:1 + C18:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3

of ALA for both dietary treatments was lower than the ratio of LA which implies that ALA is more preferentially oxidized than is LA. Furthermore, the lower ratio of deposition:intake for ALA agrees with earlier reports of a lower rate of incorporation into adipose tissue and muscle for ALA than for LA (Mitchaothai et al., *Chapter 5*; Nguyen et al., 2003).

The pigs fed the SO diet instead of the BT diet had a higher ratio of deposition:intake for SFA, but there was no diet effect on the ratios for MUFA and PUFA. The diet effect on the ratios for SFA and MUFA can be explained by the calculated minimum synthesis of these groups of fatty acids. Feeding the diet containing SO stimulated the synthesis of SFA. The higher synthesis ratio for SFA:(SFA + MUFA) in pigs fed the SO diet indicates that there was selective synthesis of SFA in the pigs fed the sunflower oil diet and it might be the result of *de novo* fatty acid synthesis from carbohydrate to balance the amount of saturated and unsaturated fatty acids in the body because of the very high intake of LA by the pigs fed the SO diet. The higher *de novo* synthesis would result in increased energy deposition for the pigs fed the SO diet so that there would be no difference in energy expenditure between the two dietary treatments.

In conclusion, the dietary TiO_2 marker can be efficiently used to determine the apparent ileal digestibility for macronutrients and fatty acids. The apparent digestibility for macronutrients and fatty acids was higher than the apparent faecal digestibility. Inclusion of SO instead of BT in the diet raised the apparent ileal digestibility of palmitic acid, stearic acid and LA, without an effect on proximate body composition and energy expenditure. Feeding the diet with SO produced a marked increase in LA deposition in whole body. The ileal digestibility of individual fatty acids was used to calculate fatty acid incorporation into the body. The type of dietary fat had marked effects on oxidation and synthesis of fatty acids and the lack of effect on energy expenditure might be the result of balancing by the changes in *de novo* synthesis.

Table 7. Effect of dietary fat source on digestible fatty acid intake, fatty acid deposition and the deposition:intake ratio during the whole feeding period

Fatty acid	BT	SO	P value
Digestible ileal fatty acid intake (g/49 days)			
C 16:0	428.72	202.32	-
C 18:0	316.14	83.95	-
C 18:1 n-9	479.46	700.57	-
C 18:2 n-6	177.15	598.26	-
C 18:3 n-3	38.86	41.26	-
∑ FA	1,601.39	1,644.58	-
∑ SFA	856.32	301.85	-
∑ MUFA	521.91	701.75	-
∑ PUFA	223.16	640.98	-
n-9	479.46	700.57	-
n-6	177.15	598.26	-
n-3	46.02	42.72	-
Fatty acid deposition (g/49 days)			
C 16:0	452.14± 46.74	389.53± 92.94	0.1817
C 18:0	494.88± 61.41	374.27± 54.37	0.0049
C 18:1 n-9	610.93± 125.94	802.65± 101.73	0.0165
C 18:2 n-6	145.58± 34.18	391.59± 54.80	0.0000
C 18:3 n-3	24.37± 5.66	18.52± 4.62	0.0798
∑ FA	1,831.31± 136.64	2,034.73± 227.53	0.0964
∑ SFA	1,032.96± 51.62	800.71± 160.87	0.0150
∑ MUFA	703.54± 87.02	846.61± 88.31	0.0326
∑ PUFA	171.11± 37.87	410.96± 59.30	0.0000
n-9	610.93± 125.94	802.65± 101.73	0.0165
n-6	146.92± 35.32	392.73± 54.20	0.0000
n-3	25.53± 6.40	19.36± 4.67	0.0884
Deposition : intake ratio			
C 16:0	1.05± 0.11	1.93± 0.46	0.0049
C 18:0	1.57± 0.19	4.46± 0.65	0.0000
C 18:1 n-9	1.27± 0.26	1.15± 0.15	0.3257
C 18:2 n-6	0.82± 0.19	0.65± 0.09	0.0957
C 18:3 n-3	0.63± 0.15	0.45± 0.11	0.0405
∑ FA	1.14± 0.09	1.24± 0.14	0.1944
∑ SFA	121± 0.06	2.65± 0.53	0.0011
∑ MUFA	1.35± 0.17	1.21± 0.13	0.1701
∑ PUFA	0.77± 0.17	0.64± 0.09	0.1513
n-9	1.27± 0.26	1.15± 0.15	0.3257
n-6	0.83± 0.20	0.66± 0.09	0.0945
n-3	0.55± 0.14	0.45± 0.11	0.1916

Means ± SD for 6 pigs per experimental diet; BT = beef tallow; SO = sunflower oil
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C18:1 n-9
Σ PUFA = C18:2 n-6 + C18:3 n-3 + C20:2 n-6 + C20:4 n-6 + C20:3 n-3 + C22:5 n-3
Σ n-9 = C18:1 n-9
Σ n-6 = C18:2 n-6 + C20:2 n-6 + C20:4 n-6
Σ n-3 = C18:3 n-3 + C20:3 n-3 + C22:5 n-3

Table 8. Effect of dietary LA and ALA on minimum *de novo* synthesis of fatty acids during the whole feeding period

	BT	SO	*p* value
Minimum synthesis (g/49 days)			
SFA	176.64 ± 51.62	498.86 ± 160.87	0.003
MUFA	181.64 ± 87.02	144.86 ± 88.31	0.526
SFA/(SFA+MUFA)	0.52 ± 0.18	0.79 ± 0.14	0.037

Means ± SD for 6 pigs per experimental diet
BT = beef tallow; SO = sunflower oil
ND = not detectable; NS = not significant
SFA = Saturated fatty acid, MUFA = Monounsaturated fatty acid, PUFA = Polyunsaturated fatty acid
Σ SFA = C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0
Σ MUFA = C18:1 n-9

ACKNOWLEDGEMENTS

The authors gratefully thank Chai-yod Phudtha and Sitthichai Phangsri for their excellent assistance. This research work was supported by the Faculty of Natural Resources, Rajamangala University of Technology Isan by providing research facilities and by Mahanakorn University of Technology by providing funding.

LITERATURE CITED

AOAC. 1990. Association of official analysis chemists, Washington DC.

Arjmandi, B.H., D.A. Kahn, Shanil-Juma, M.L. Drum, Sreevidya-Venkatesh, E. Sohn, L. Wei, and R. Derman. 1998. Whole flaxseed consumption lowers serum LDL-cholesterol and lipoprotein(a) concentrations in postmenopausal women. Nutrition Research 18:1203-1214.

Asghar, A., J.I. Gray, A.M. Booren, E.A. Gomma, M.M. Abouzied, and E.R. Miller. 1991. Effects of supranutritional dietary vitamin E levels on subcellular disposition of α–tocopherol in the muscle and on pork quality. Journal of the Science of Food and Agriculture 57:31-41.

Bazinet, R.P., E.G. McMillan, R. Seebaransingh, A.M. Hayes, and S.C. Cunnane. 2003. Whole-body beta-oxidation of 18:2omega6 and 18:3omega3 in the pig varies markedly with weaning strategy and dietary 18:3omega3. J Lipid Res 44:314-9.

Beynen, A.C., and M.B. Katan. 1985. Why do polyunsaturated fatty acids lower serum cholesterol? Am J Clin Nutr 42:560-3.

Bondi, A.A., and D. Drori. 1987. Animal nutrition. English ed. Wiley ;, Chichester New York,.

Bracco, U. 1994. Effect of triglyceride structure on fat absorption. Am J Clin Nutr 60:1002S-1009S.

Crespo, N., and E. Esteve-Garcia. 2002a. Dietary polyunsaturated fatty acids decrease fat deposition in separable fat depots but not in the remainder carcass. Poult Sci 81:512-8.

Crespo, N., and E. Esteve-Garcia. 2002b. Dietary linseed oil produces lower abdominal fat deposition but higher de novo fatty acid synthesis in broiler chickens. Poult Sci 81:1555-62.

Cunnane, S.C., and M.J. Anderson. 1997. The majority of dietary linoleate in growing rats is b-oxidized or stored in visceral fat. Journal of Nutrition 127:146-152.

Donkoh, A., P.J. Moughan, and W.C. Smith. 1994. Comparison of the slaughter method and simple T-piece cannulation of the terminal ileum for determining ileal amino acid digestibility in meat and bone meal for the growing pig. Animal Feed Science and Technology 49:43-56.

Doreau, M., and Y. Chilliard. 1997. Digestion and metabolism of dietary fat in farm animals. Br J Nutr 78 Suppl 1:S15-35.

Gandemer, G. 1999. Lipids and meat quality: lipolysis, oxidation, maillard reaction and flavour. Science des Aliments 19:439-458.

Garg, M.L., and R. Blake. 1997. Cholesterol dynamics in dynamics in rats fed diets containing either canola oil or sunflower oil. Nutrition Research 17:485-492.

Goyens, P.L., and R.P. Mensink. 2006. Effects of alpha-linolenic acid versus those of EPA/DHA on cardiovascular risk markers in healthy elderly subjects. Eur J Clin Nutr 60:978-84.

Horwitz, W. 1975. Official Methods of Analysis of Official Analytical Chemist, p. 225 Official Methods of Analysis of Official Analytical Chemist, 12 ed. Benjamin Franklin Station, DC.

Houdijk, J.G., M.W. Bosch, S. Tamminga, M.W. Verstegen, E.B. Berenpas, and H. Knoop. 1999. Apparent ileal and total-tract nutrient digestion by pigs as affected by dietary nondigestible oligosaccharides. J Anim Sci 77:148-58.

Ide, T., M. Murata, and M. Sugano. 1996. Stimulation of the activities of hepatic fatty acid oxidation enzymes by dietary fat rich in alpha-linolenic acid in rats. J Lipid Res 37:448-63.

Innis, S.M. 2007. Dietary (n-3) fatty acids and brain development. J Nutr 137:855-9.

Innis, S.M., and R. Dyer. 1997. Dietary triacylglycerols with palmitic acid (16:0) in the 2-position increase 16:0 in the 2-position of plasma and chylomicron triacylglycerols, but reduce phospholipid arachidonic and docosahexaenoic acids, and alter cholesteryl ester metabolism in formula-Fed piglets. J Nutr 127:1311-9.

Jagger, S., J. Wiseman, D.J. Cole, and J. Craigon. 1992. Evaluation of inert markers for the determination of ileal and faecal apparent digestibility values in the pig. Br J Nutr 68:729-39.

Jakobsen, K. 1999. Dietary modifications of animal fats: status and future perspectives. Lipid-Fett 101:475-483.

Javadi, M., M.J. Geelen, A.G. Lemmens, A. Lankhorst, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2007a. The influence of dietary linoleic and alpha-linolenic acid on body composition and the activities of key enzymes of hepatic lipogenesis and fatty acid oxidation in mice. J Anim Physiol Anim Nutr (Berl) 91:11-8.

Javadi, M., M.J.H. Geelen, H. Everts, R. Hovenier, S. Javadi, H. Kappert, and A.C. Beynen. 2007b. Effect of dietary conjugated linoleic acid on body composition and energy balance in broiler chickens. British Journal of Nutrition *in press*.

Javadi, M., H. Everts, R. Hovenier, S. Kocsis, A.E. Lankhorst, A.G. Lemmens, J.T. Schonewille, A.H. Terpstra, and A.C. Beynen. 2004. The effect of six different C18 fatty acids on body fat and energy metabolism in mice. Br J Nutr 92:391-9.

Jones, P.J.H., P.B. Pencharz, and M.T. Clandinin. 1985. Whole body oxidation of dietary fatty acids: implications for energy utilization. American journal of clinical nutrition 42:769-777.

Jørgensen, H., J.E. Lindberg, and C. Andersson. 1997. Diurnal variation in the composition of ileal digesta and the ileal digestibilities of nutrients in growing pigs. J. Sci. Food Agric. 74:244-250.

Jørgensen, H., V.M. Gabert, M.S. Hedemann, and S.K. Jensen. 2000. Digestion of fat does not differ in growing pigs fed diets containing fish oil, rapeseed oil or coconut oil. J Nutr 130:852-7.

Kabir, Y., and T. Ide. 1996. Activity of hepatic fatty acid oxidation enzymes in rats fed alpha-linolenic acid. Biochim Biophys Acta 1304:105-19.

Kirchgessner, M., and H.L. Muller. 1998. Comparison of excessive dietary carbohydrate and fat on energy balance. Journal of Animal Physiology and Animal Nutrition 80:18-30.

Kloareg, M., L. Le Bellego, J. Mourot, J. Noblet, and J. van Milgen. 2005. Deposition of dietary fatty acids and of de novo synthesised fatty acids in growing pigs: effects of high ambient temperature and feeding restriction. Br J Nutr 93:803-11.

Kouba, M., M. Enser, F.M. Whittington, G.R. Nute, and J.D. Wood. 2003. Effect of a high-linolenic acid diet on lipogenic enzyme activities, fatty acid composition, and meat quality in the growing pig. J Anim Sci 81:1967-79.

Leyton, J., P.J. Drury, and M.A. Crawford. 1987. Differential oxidation of saturated and unsaturated fatty acids in vivo in the rat. Br J Nutr 57:383-93.

Li, D.F., R.C. Thaler, J.L. Nelssen, D.L. Harmon, G.L. Allee, and T.L. Weeden. 1990. Effect of fat sources and combinations on starter pig performance, nutrient digestibility and intestinal morphology. J Anim Sci 68:3694-704.

Lindberg, J.E., and Z. Cortova. 1995. The effect of increasing inclusion of lucerne leafmeal in a barley-based diet on the partition of digestion and on nutrient utilization in pigs. Animal Feed Science and Technology 56:11-20.

Maynard, L.A. 1979. Animal nutrition. 7th ed. McGraw-Hill, New York ; Sydney.

Miller, M.F., S.D. Shackelford, K.D. Hayden, and J.O. Reagan. 1990. Determination of the alteration in fatty acid profiles, sensory characteristics and carcass traits of swine fed elevated levels of monounsaturated fats in the diet. J Anim Sci 68:1624-31.

Mitchaothai, J., H. Everts, C. Yuangklang, S. Wittayakun, K. Vasupen, S. Wongsuthavas, P. Srenanul, R. Hovenier, and A.C. Beynen. Fatty acid metabolism in Thai indigenous pigs fed diets with low or high linoleic acid : α-linolenic acid ratio. *(Chapter 5).*

Mu, H., and T. Porsgaard. 2005. The metabolism of structured triacylglycerols. Prog Lipid Res 44:430-48.

Myer, R.O., J.H. Brendemuhl, F.W. Leak, and J.B. Hess. 2004. Evaluation of a rendered poultry mortality-soybean meal product as a supplemental protein source for pig diets. J Anim Sci 82:1071-8.

Myers, W.D., P.A. Ludden, V. Nayigihugu, and B.W. Hess. 2004. Technical note: a procedure for the preparation and quantitative analysis of samples for titanium dioxide. J Anim Sci 82:179-83.

Napier, J.A. 2007. The production of unusual Fatty acids in transgenic plants. Annu Rev Plant Biol 58:295-319.

Nelson, C.M., and S.M. Innis. 1999. Plasma lipoprotein fatty acids are altered by the positional distribution of fatty acids in infant formula triacylglycerols and human milk. Am J Clin Nutr 70:62-9.

Newsholme, E.A., and A. Leech. 1986. Biochemistry for the Medical Sciences. Reprint and correction ed. John Wiley & Sons, New York.

Nguyen, L.Q., M.C.G.A. Nuijens, H. Everts, N. Salden, and A.C. Beynen. 2003. Mathematicl relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. Meat Science 65:1399-1406.

Nicolosi, R.J., T.A. Wilson, G. Handelman, T. Foxall, J.F. Keaney, and J.A. Vita. 2002. Decreased aortic early atherosclerosis in hypercholesterolemic hamsters fed oleic acid-rich TriSun oil compared to linoleic acid-rich sunflower oil. J Nutr Biochem 13:392-402.

Okuno, M., K. Kajiwara, S. Imai, T. Kobayashi, N. Honma, T. Maki, K. Suruga, T. Goda, S. Takase, Y. Muto, and H. Moriwaki. 1997. Perilla oil prevents the excessive growth of visceral adipose tissue in rats by down-regulating adipocyte differentiation. J Nutr 127:1752-7.

Peddie, J., W.A. Dewar, A.B. Gilbert, and D. Waddington. 1982. The use of titanium dioxide for determining apparent digestibility in mature domestic fowls (*Gallus domesticus*). The Journal of Agricultural Science 99:233-236.

Pettersson, A., and J.E. Lindberg. 1997. Ileal and total tract digestibility in pigs of naked and hulled barley with different starch composition. Animal Feed Science and Technology 66:97-109.

Raes, K., S. De smet, and D. Demeyer. 2004. Effect of dietary fatyy acids on incorporation of long chain polyunsaturated fatty acids and conjugated linolenic acid in lamb, beef and pork meat: a review. Animal Feed Science and Technology 113:199-221.

Reaven, P., S. Parthasarathy, B.J. Grasse, E. Miller, D. Steinberg, and J.L. Witztum. 1993. Effects of oleate-rich and linoleate-rich diets on the susceptibility of low density lipoprotein to oxidative modification in mildly hypercholesterolemic subjects. J Clin Invest 91:668-76.

Reaven, P., S. Parthasarathy, B.J. Grasse, E. Miller, F. Almazan, F.H. Mattson, J.C. Khoo, D. Steinberg, and J.L. Witztum. 1991. Feasibility of using an oleate-rich diet to reduce the susceptibility of low-density lipoprotein to oxidative modification in humans. Am J Clin Nutr 54:701-6.

Sanz, M., A. Flores, and C.J. Lopez-Bote. 2000a. The metabolic use of energy from dietary fat in broilers is affected by fatty acid saturation. Br Poult Sci 41:61-8.

Sanz, M., C.J. Lopez-Bote, D. Menoyo, and J.M. Bautista. 2000b. Abdominal fat deposition and fatty acid synthesis are lower and beta-oxidation is higher in broiler chickens fed diets containing unsaturated rather than saturated fat. J Nutr 130:3034-7.

SAS. 1996. User's guide; Statistics Statistical analysis system inst., Cary, NC, USA.

Scheeder, M., K.R. Gla¨ser, B. Eichenberger, and C. Wenk. 2000. Influence of different fats in pig feed on fatty acids composition of phospholipids and physical meat quality characteristics. European Journal of Lipid Science and Technology 102:391-401.

Scheeder, M., D. Gumy, R. Messikommer, C. Wenk, and P. Lambelet. 2003. Effect of PUFA at sn-2 position in dietary triacylglycerols on the fatty acid composition of adipose tissues in non-ruminant farm animals. Eur. J. Lipid Sci. Technol. 105:74-82.

Seiquer, I., M. Manas, E. Martinez-Victoria, M.C. Ballesta, and J. Mataix. 1995. The influence of dietary fat source (sunflower oil or olive oil) on LDL composition and serum lipid levels in miniature swine (Sus scrofa). Comp Biochem Physiol B Biochem Mol Biol 111:163-9.

Shields, R.G., Jr., D.C. Mahan, and P.L. Graham. 1983. Changes in swine body composition from birth to 145 kg. J. Anim. Sci. 57:43-54.

SPSS. 1999. SPSS for Windows, SPSS Inc., Chicago, IL, USA.

Stubbs, R.J., and B.J. Tolkamp. 2006. Control of energy balance in relation to energy intake and energy expenditure in animals and man: an ecological perspective. Br J Nutr 95:657-76.

Titgemeyer, E.C., C.K. Armendariz, D.J. Bindel, R.H. Greenwood, and C.A. Loest. 2001. Evaluation of titanium dioxide as a digestibility marker for cattle. J Anim Sci 79:1059-63.

Wagner, J.R., A.P. Schinckel, W. Chen, J.C. Forrest, and B.L. Coe. 1999. Analysis of body composition changes of swine during growth and development. J Anim Sci 77:1442-66.

Wongsuthavas, S., C. Yuangklang, K. Vasupen, J. Mitchaothai, and A.C. Beynen. 2007. Energy expenditure by broiler chickens fed diets containing various blends of beef tallow and soybean oil. Ph.D. Thesis, Utrecht University, Utrecht, The Netherlands.

Wood, J.D., R.C.D. Jones, J.A. Bayntun, and E. Dransfield. 1985. Backfat quality in boars and borrow at 90 kg live weight. Animal Production 40:481-487.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2003. Effects of fatty acids on meat quality: a review. Meat Science 66:21-32.

Wood, J.D., R.I. Richardson, G.R. Nute, A.V. Fisher, M.M. Campo, E. Kasapidou, P.R. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: a review. Meat Science 66:21-32.

Yeom, K.-H., J.T. Schonewille, H. Everts, K.-W. Lee, G. van Trierum, and A.C. beynen. 2005. Growth performance and body composition of goat kids fed milk replacers with increasing levels of linoleic acid. Journal of Animal Physiology and Animal Nutrition 89:29-34.

Yin, Y.-L., R. Hunan, H. Zhong, C.-M. Chen, and H. Dai. 1991. Influence of different cannulation techniques on the pre-caecal digestibility of protein, amino acids and cell wall constituents from diets, containing different protein meals, in pigs. Anirmal Feed Science and Technology 35:271-281.

Yu, S., J. Derr, T.D. Etherton, and P.M. Kris-Etherton. 1995. Plasma cholesterol-predictive equations demonstrate that stearic acid is neutral and monounsaturated fatty acids are hypocholesterolemic. Am J Clin Nutr 61:1129-39.

Zollner, N., and F. Tato. 1992. Fatty acid composition of the diet: impact on serum lipids and atherosclerosis. Clin Investig 70:968-1009.

CHAPTER 8

General conclusions

This thesis describes the effects of dietary substitution of vegetable oil for animal fat on meat quality and fatty acid metabolism in growing-finishing pigs of a commercial breed. To study metabolism at the level of the whole body, indigenous Thai Kadon pigs were used as a model. Many vegetable oils contain a high proportion of polyunsaturated fatty acids (PUFA) including the essential fatty acids linoleic acid (LA) and α-linolenic acids (ALA), albeit that the concentration of the latter generally is much lower. Animal fats generally are rich in saturated fatty acids (SFA). In the present studies the LA-rich oils, sunflower oil (SO) and soybean oil, were used and also linseed oil which is rich in ALA. As animal fat, beef tallow (BT) was added to the diet of the pigs.

There is an increasing tendency that consumers are only interested in products of animal origin if these products can be considered healthy for them. Supplementing swine diets with vegetable oils can increase the PUFA level of pork meat. Increased intakes of PUFA may contribute to reducing the risk of cardiovascular disease in man. However, adverse effects can occur: the firmness of the meat may decrease and maximum storage time may become less. Therefore, the impact of dietary fat type on meat and carcass quality and on fatty acid composition of adipose tissue and muscle were investigated. Digestibility and incorporation of fatty acids in the whole body, calculated *de novo* fatty acid synthesis and energy metabolism were studied in order to explain the observations in metabolic terms.

Chapter 1 sets the stage for the studies in this PhD thesis by reviewing the relevant basics. The literature review revealed that there have been few studies on *de novo* synthesis, oxidation of fatty acids and energy expenditure at the level of the whole body of pigs. In addition, the supplementation of swine diets with SO has been investigated scarcely. The studies in this thesis attempted to focus on metabolism of individual fatty acids in relation to meat quality, whole body composition, energy expenditure and growth performance. Concerning the metabolism of individual or groups of fatty acids, aspects that were measured directly or calculated indirectly were faecal and ileal digestibility, *de novo* fatty acid synthesis, fatty acid oxidation and fat acid incorporation into various tissues and the whole body.

The key conclusions from the various studies in this thesis may be formulated as follows:

1. The feeding of diets with 5% SO instead of BT to growing-finishing swine increased deposition of PUFA (mainly LA) into tissues, but had no effect on the physical properties of pork.

Feeding growing-finishing pigs on diets containing either SO or BT (Chapter 2) had no effect on the carcass traits, back fat thickness and fat-lean ratio (LSQ). Likewise, there was no diet effect on the pork quality characteristics, colour, pH, drip and cooking loss, tenderness (shear force), sarcomere length and percentage of intramuscular fat. As would be expected, dietary SO increased the incorporation of PUFA, especially LA, into adipose tissues and loin muscle. It is concluded that the supplementation of swine diets with vegetable oils rich in LA may not have an adverse impact on pork quality. High LA contents in pork are considered to be beneficial for consumers.

2. When growing-finishing pigs were fed a diet rich in LA, the digestion of SFA, except for stearic acid, monounsaturated fatty acids (MUFA) and PUFA (LA and

ALA) was increased. The ratio of deposition: digestible intake for SFA was increased by the feeding of a LA-rich diet, but the ratios for MUFA and PUFA were decreased.

Chapter 3 describes that pigs fed on a diet with SO instead of BT showed a higher digestibility of palmitic acid, oleic acid, LA and ALA. This could be the result of a higher proportion of these fatty acids bound at the *sn*2-position of the glycerol backbone of the triacylglycerols in SO. The PUFA and LA levels in the whole body were markedly increased in the pigs fed the SO diet. However, the SO diet induced a higher deposition: digestible intake ratio for SFA (especially stearic acid) whereas this ratio for MUFA and PUFA was lower. The increased minimum *de novo* synthesis for SFA in pigs fed the SO diet could point at an attempt to balance the ratio of SFA:PUFA in the body because of the higher incorporation of LA.

3. Feeding the pigs iso-energetic amounts of diets containing either SO or BT did not affect the diet effects on digestion and deposition of fatty acids when compared to *ad libitum* feeding, but it tended to increase the efficiency of energy utilization.

For the study described in Chapter 4, the digestibility of crude fat as measured in the previous experiment (Chapter 3) was taken into account to formulate an iso-energetic exchange of dietary fat sources. It was hypothesized that the response of the pigs could be altered when they obtained equal amounts of digestible fat, but of different sources. The results collected were similar to those of the previous experiment, except for a negative digestibility of stearic acid and a tendency towards a better energetic efficiency on the SO. It was speculated microbial hydrogenation in the large bowel had caused the negative value for the digestibility for stearic acid.

4. When comparing the metabolism of ALA and LA, it was found that the rate of incorporation into tissues was lesser for ALA than for LA.

Literature data point to ALA being more preferentially oxidized than LA. It was hypothesized that ALA would be incorporated into tissues less efficiently than LA. The preferential oxidation of ALA would imply that on a high-ALA diet more glucose would be converted into fatty acids, this process not being energetically efficient and thus increasing energy expenditure. In Thai indigenous pigs fed either a high-LA or high-ALA diet, the digestion of individual fatty acids was measured as well as the deposition and metabolism of fatty acids, and energy expenditure at the level of the whole body (Chapter 5). A higher digestibility of ALA, a lower total body fat fraction, and increased ALA contents in adipose tissue, muscle and whole body were observed in pigs fed the high-ALA diet. However, no effect of high ALA versus high LA intake on energy expenditure or on *de novo* fatty acid synthesis was seen. Irrespective of the composition of the diet, ALA was found to have a lower rate of incorporation into the body than did LA. This observation supports the idea of a ALA being preferentially oxidized when compared to LA.

5. The fat background of the diet influenced ALA incorporation into tissues: SO as fat background diminished ALA incorporation when compared to BT as fat background.

In the form of linseed oil, ALA was added to the diets containing identical amounts of either SO or BT and were fed to Thai indigenous pigs (Chapter 6). The

hypothesis was that the diet rich in LA would increase the oxidation of ALA and raise energy expenditure. The SO diet rich in LA increased the digestion and deposition of LA and lowered the level of ALA in adipose tissue, muscle and whole body. The SO diet increased *de novo* synthesis of stearic acid and SFA synthesis, and stimulated ALA oxidation. The SO and BT diets rich in ALA did not differently influence growth performance and energy expenditure. It is suggested that extra *de novo* synthesis of SFA compensates for increased ALA oxidation.

6. Apparent ileal digestibility of fatty acids is lower than the apparent faecal digestibility, except for stearic acid. Feeding pigs with dietary SO instead of BT increased the digestibility of fat and fatty acids both at ileal and faecal level, and also raised the oxidation of LA and ALA and *de novo* synthesis of SFA, but left unchanged energy expenditure and body composition.

The data in Chapter 7 show that the apparent ileal digestibility of fatty acids is lower than the apparent faecal digestibility, except for stearic acid. The aberrant characteristic of stearic acid may be explained by microbial hydrogenation of unsaturated C18-fatty acids in the large intestine. The digestion of fatty acids was better for the SO diet than for the BT diet, except for stearic acid. In this study, any effect of microbial hydrogenation on the calculated values for the synthesis or oxidation of fatty acids was avoided because the ileal fatty acid digestibility was used for the calculations. This study confirms the data in other chapters, showing that feeding the SO diet increased the deposition of LA and raised *de novo* synthesis of SFA. Again, evidence was obtained that ALA is more preferentially oxidized than is LA.

Taken together the results of all studies in this thesis, it is clear that dietary SO rich in LA increased LA deposition in the body of pigs without any adverse effect on the physical properties of pork meat. The digestibility of plant oils rich in PUFA is higher than that of animal fat. Between experiments there were differences in the values for the deposition: intake ratio for individual fatty acids, which relates to the different conditions of each experiment and most likely the difference in breeds of pigs used. In any event, the results of all experiments pointed in the same direction. The essential fatty acids, LA and ALA, are preferentially oxidized compared to other fatty acids and high intakes of LA and ALA stimulated the *de novo* synthesis of SFA. ALA oxidation was generally found to be preferred over the oxidation of LA.

Summary

This thesis focuses on the replacement of animal fat by vegetable oil in the diet for growing-finishing pigs. Generally, but not exclusively, fats of animal origin contain higher proportions of saturated fatty acids (SFA) than vegetable oils that are commonly rich in polyunsaturated fatty acids (PUFA). There is concern among the consumers of pork meat as to the use of fats of animal origin in the diet of pigs. High intakes of animal fats by pigs leads to pork meat rich in SFA, which in turn increases the risk of cardiovascular disease in man. Consequently, consumers tend to prefer animal products derived from animals fed on diets containing oils of plant origin. The PUFA concentrations in pork meat can be raised by feeding pigs diets rich in PUFA, but the impact on physical meat quality was not known. From biochemical studies it follows that PUFAs are preferentially oxidised when compared to SFA. Thus, it could be suggested that the inclusion of vegetable oils into the diets for pigs would alter the metabolism of fatty acids and energy.

The main question that was addressed in this thesis is as follows: What is the effect of feeding diets containing either sunflower oil (SO) or beef tallow (BT) on meat quality and fatty acid and energy metabolism? Variables measured were the incorporation of fatty acids into adipose tissue, meat and the whole body, the oxidation and *de novo* fatty acid synthesis of fatty acids and energy expenditure. The feeding of diets with SO instead of BT did not affect growth performance, but markedly increased the level of linoleic acid (LA) in adipose tissues and pork meat without any adverse effect on the physical aspects of pork quality. The digestibility of crude fat and fatty acids was higher for the SO diets, except for the digestibility of stearic acid. The SO diets raised the oxidation of LA and the *de novo* synthesis of SFA synthesis.

To study fatty acid metabolism at the level of the whole body, the relatively small, Thai indigenous pigs were used as a model. In these pigs the metabolism of LA and ALA was compared after feeding diets with a low or high LA:ALA ratio. Adipose tissue and pork of the pigs fed on the high-ALA diet were enriched with ALA. The ratios of deposition: digestible intake for LA and ALA were lower than 1, indicating preferential oxidation of these fatty acids, which was associated with increased *de novo* synthesis of SFA synthesis in keeping with the studies in commercial pigs. For both the high-ALA and high-LA diet, the rate of ALA incorporation into the whole body was lesser than for LA, which is in line with preferential oxidation of ALA over LA. In a subsequent study, ALA metabolism was studied in pigs fed ALA-rich diets with identical levels of either SO or BT as fat background. It was found that the digestion and deposition of LA, *de novo* synthesis of SFA synthesis and ALA oxidation were higher, whereas ALA deposition was lower for the SO diet. It is concluded that the fat background of the diet can affect metabolism of supplemental fatty acids. In order to assess the digestibility of fatty acids more accurately, cannulated pigs were used to determine apparent ileal digestibility instead of faecal digestibility of fatty acids. The apparent ileal digestibility of fatty acids was found to be lower than apparent feacal digestibility, except for that of stearic acid. The aberrant outcome for stearic acid indicates that C18 unsaturated fatty acids are escaping from ileal digestion and are hydrogenated and transformed into C18 saturated fatty acids by microbes in the large intestine. Energy expenditure and proximate body composition were not influenced by the type of fat in the diet.

Samenvatting

Dit proefschrift richt zich op de vervanging van dierlijk vet door plantaardig vet in het voer voor vleesvarkens. In het algemeen bevatten dierlijke vetten een hoog aandeel verzadigde vetzuren en zijn plantaardige vetten vaak rijk aan meervoudig onverzadigde vetzuren. Het gebruik van dierlijk vet in het voer van vleesvarkens verontrust de consumenten van varkensvlees. Immers, een hoge opname van verzadigd vet door vleesvarkens leidt tot varkensvlees met een hoog gehalte aan verzadigde vetzuren. Verzadigde vetzuren worden als risicofactor gezien voor het ontstaan van hart- en vaatziekten bij de mens. Daarom zal de consument een voorkeur hebben voor producten die afkomstig zijn van dieren die gevoerd zijn met plantaardig vet in plaats van dierlijk vet. Door de vleesvarkens te voeren met een voer dat rijk is aan meervoudig onverzadigde vetzuren kan de concentratie aan meervoudig onverzadigde vetzuren in varkensvlees verhoogd worden, maar welke effect dat heeft op de kwaliteit van het vlees is onvoldoende bekend. Uit biochemisch onderzoek is bekend dat MOV preferentieel worden geoxideerd ten opzichte van verzadigde vetzuren. Het gebruik van plantaardige vetten in het voer voor vleesvarkens kan daarom ook gevolgen hebben voor het metabolisme van de vetzuren en de energiestofwisseling van de varkens.

De belangrijkste vraag die in dit proefschrift aan de orde komt is: Wat is het effect van het voeren van vleesvarkensvoer met zonnebloemolie (ZO) of met rundvet (RV) op de vleeskwaliteit, de vetzuurstofwisseling en het energiemetabolisme? In de proeven werd de mate van inbouw van vetzuren in vetweefsel en in het vlees en het totale dier gemeten. Daarnaast werd de maximale oxidatie en minimale *de novo* synthese van vetzuren berekend en werd de totale warmteafgifte berekend.

Het voeren van vleesvarkensvoer met ZO in plaats van RV had geen effect op de groei van de dieren, maar verhoogde wel het gehalte aan linolzuur in vetweefsel en vlees zonder negatief effect op de vleeskwaliteit. De verteerbaarheid van ruw vet en van de vetzuren was hoger bij de voeders met ZO, behalve de verteerbaarheid van stearinezuur. Verder verhoogde een voer met ZO de oxidatie van linolzuur en de *de novo* synthese van verzadigde vetzuren.

Voor het onderzoek naar het vetzuurmetabolisme op dierniveau werden relatief kleine, inheemse Thaise varkens als model gebruikt. Hierbij werd een vleesvarkensvoer met een lage verhouding tussen linolzuur en α-linoleenzuur vergeleken met een voer met een hoge verhouding. Het voer met veel α-linoleenzuur verhoogde het gehalte aan α-linoleenzuur in het vetweefsel en in het varkensvlees. De verhouding tussen aanzet en opgenomen verteerbaar vetzuur was lager dan 1 voor zowel linolzuur als α-linoleenzuur. Dit wijst er op dat deze vetzuren mogelijk preferentieel worden verbrand. Tegelijkertijd nam de *de novo* synthese van verzadigde vetzuren toe, net als in de proeven met commerciële varkens. Zowel op het voer met de hoge als de lage verhouding tussen linolzuur en α-linoleenzuur was de mate van inbouw van α-linoleenzuur lager dan die van linolzuur, hetgeen in lijn is met de preferentiële oxidatie van α-linoleenzuur ten opzichte van linolzuur.

In een andere proef werd het α-linoleenzuur metabolisme bestudeerd bij varkens die een α-linoleenzuurrijk voer kregen met of ZO of RV als achtergrond. De verteerbaarheid, de mate van inbouw van linolzuur, de *de novo* synthese van verzadigde vetzuren en de verbranding van α-linoleenzuur waren hoger op het voer met ZO dan op het voer met RV. Verder bleek dat de inbouw van α-linoleenzuur op

het voer met ZO lager was dan op het voer met RV. Het is duidelijk dat de keuze van het type voervet een duidelijk effect kan hebben op het metabolisme van het gesupplementeerde vetzuur.

Om een beter beeld te krijgen van de verteerbaarheid van de vetzuren is de ileale verteerbaarheid in plaats van de fecale verteerbaarheid gemeten bij gecanuleerde varkens. De schijnbare ileale verteerbaarheid van de langketen vetzuren was lager dan de fecale verteerbaarheid, behalve voor stearinezuur. Deze afwijkende uitkomst voor stearinezuur duidt er op dat onverzadigde vetzuren met 18 koolstofatomen die niet in de dunne darm worden geabsorbeerd door de microflora in de dikke darm worden gehydrogeneerd en uiteindelijk als stearinezuur worden uitgescheiden via de feces.

De warmteproductie en de lichaamssamenstelling van de varkens werd overigens niet beïnvloed door het type vet in het vleesvarkensvoer.

บทสรุป

วิทยานิพนธ์เล่มนี้มุ่งเน้นศึกษาการทดแทนไขมันสัตว์ด้วยน้ำมันจากพืชในอาหารสำหรับสุกรรุ่น-ขุน โดยส่วนใหญ่แล้ว ไขมันที่ได้จากสัตว์มีสัดส่วนของกรดไขมันอิ่มตัวสูงกว่าน้ำมันที่ได้จากพืชซึ่งมักอุดมไปด้วยกรดไขมันไม่อิ่มตัวเชิงซ้อน ปัจจุบันผู้บริโภคหันมาให้ความสำคัญและคำนึงถึงคุณภาพของเนื้อสุกรจากสุกรที่ได้รับ อาหารที่เสริมด้วยไขมันจากสัตว์ เนื่องจากสุกรที่ได้รับไขมันจากสัตว์เป็นปริมาณมากจะทำให้ปริมาณของกรดไขมันอิ่มตัวในเนื้อสุกรมีมากตามไปด้วย ซึ่งส่งผลให้ผู้บริโภคมีความเสี่ยงของการป่วยด้วยโรคในระบบไหลเวียนโลหิตและหัวใจ (cardiovascular disease) ผลที่ตามมา คือ ผู้บริโภคมีแนวโน้มหันมาเลือกบริโภคผลิตภัณฑ์จากสัตว์ที่ได้รับอาหารที่มีน้ำมันจากพืชประกอบอยู่มากขึ้น การให้อาหารสุกรด้วยอาหารที่อุดมไปด้วยกรดไขมันไม่อิ่มตัวเชิงซ้อน สามารถเพิ่มความเข้มข้นของกรดไขมันไม่อิ่มตัวเชิงซ้อนในเนื้อสุกรได้ แต่อย่างไรก็ตามผลกระทบที่ตามมาต่อคุณภาพของเนื้อยังไม่ทราบ นอกจากนี้การศึกษาทางชีวเคมี พบว่ากรดไขมันไม่อิ่มตัวเชิงซ้อนมักถูกออกซิไดซ์ (oxidised) ได้มากกว่ากรดไขมันอิ่มตัว ดังนั้นการเติมน้ำมันจากพืชลงในอาหารสำหรับสุกรอาจสามารถเปลี่ยนแปลงกระบวนการเมทาบอลิซึม (metabolism) ของกรดไขมันและพลังงานได้

โจทย์ปัญหาหลักในวิทยานิพนธ์เล่มนี้ คือ เมื่อสุกรได้รับอาหารที่เสริมด้วยน้ำมันทานตะวันหรือไขวัว จะมีผลกระทบตามมาต่อคุณภาพเนื้อ และกระบวนการเมทาบอลิซึมของกรดไขมันและพลังงานเป็นอย่างไร? ดังนั้นตัวแปรที่ใช้วัดจึงประกอบด้วย การสะสมของกรดไขมันในเนื้อเยื่อไขมัน เนื้อ และ ร่างกาย, กระบวนการออกซิเดชั่น (oxidation)และการสังเคราะห์ใหม่ (*de novo* synthesis) ของกรดไขมัน, และการเผาผลาญพลังงาน (energy expenditure) ซึ่งผลการทดลองพบว่าการให้อาหารที่เสริมด้วยน้ำมันทานตะวันทดแทนไขวัวแก่สุกร ไม่มีผลต่อประสิทธิภาพการเจริญเติบโต แต่เพิ่มระดับของกรดลิโนเลอิก (linoleic acid; LA) สูงขึ้นอย่างเด่นชัดในเนื้อเยื่อไขมันและเนื้อสุกร ความสามารถในการย่อยได้ (digestibility) ของไขมันรวมและกรดไขมันในสุกรกลุ่มที่ได้รับอาหารเสริมน้ำมันทานตะวันมีค่าสูงกว่ากลุ่มที่เสริมด้วยไขวัว ยกเว้นกรดสเตียริก (stearic) ซึ่งให้ผลตรงกันข้าม นอกจากนี้สุกรที่ได้รับอาหารที่เสริมด้วยน้ำมันทานตะวันสามารถเพิ่มกระบวนการออกซิเดชั่นของกรดลิโนเลอิกให้มากขึ้นและทำให้การสังเคราะห์กรดไขมันอิ่มตัวขึ้นใหม่เพิ่มสูงขึ้น

การศึกษากระบวนการเมทาบอลิซึมของกรดไขมันในระดับทั้งตัวสัตว์ จำเป็นต้องใช้สุกรพันธุ์พื้นเมืองของไทย (Thai indigenous pigs) เป็นแบบจำลองในการศึกษา เนื่องจากสุกรพันธุ์นี้มีขนาดตัวค่อนข้างเล็ก โดยเริ่มจากการเปรียบเทียบกระบวนการเมทาบอลิซึมของกรดลิโนเลอิกและกรดอัลฟ่าลิโนเลนิก (α-linolenic acid; ALA) หลังจากสุกรได้รับอาหารที่มีอัตราส่วน (ratio) ของกรดลิโนเลอิกต่อกรดอัลฟ่าลิโนเลนิกในระดับต่ำหรือสูง ซึ่งพบว่า เนื้อเยื่อไขมันและเนื้อของสุกรในกลุ่มที่ได้รับอาหารที่มีกรดอัลฟ่าลิโนเลนิกในระดับสูงจะอุดมไปด้วยกรดอัลฟ่าลิโนเลนิก อัตราส่วนของการสะสมต่อปริมาณการย่อยนำไปใช้ประโยชน์ (ratios of deposition : digestible intake) ของกรดลิโนเลอิกและอัลฟ่าลิโนเลนิกมีค่าต่ำกว่า ๑ ซึ่งบ่งชี้ว่ากรดไขมันทั้งสองชนิดนี้มักถูกเลือกนำไปใช้ในกระบวนการออกซิเดชั่น โดยมีความสัมพันธ์กับการเพิ่มสูงขึ้นของการสังเคราะห์กรดไขมันอิ่มตัวขึ้นใหม่ โดยผลที่ได้รับนี้สอดรับกับการศึกษาก่อนหน้านี้ในสุกรพันธุ์ที่ผลิตในเชิงพานิชย์ (commercial pigs) สุกรทั้งสองกลุ่มที่ได้รับกรดลิโนเลอิกและอัลฟ่าลิโนเลนิกในระดับสูง มีอัตราการสะสมของกรดอัลฟ่าลิโนเลนิกในร่างกายทั้งตัวต่ำกว่ากรดลิโนเลนิก ซึ่งสอดรับกับการถูกเลือกใช้เพื่อกระบวนการออกซิเดชั่นของกรดอัลฟ่าลิโนเลนิกที่มากกว่ากรดลิโนเลอิก ในการศึกษาถัดมา ได้ศึกษากระบวนการเมทาบอลิซึมของกรดอัลฟ่าลิโนเลนิกในสุกรที่ได้รับอาหารที่อุดมด้วยกรดลิโนเลนิกและเสริมด้วย

ACKNOWLEDGEMENTS

I appreciate it very much to have this opportunity to express my thankfulness to all people who contributed to the completion of this thesis.

First of all, my gratefulness goes to the support of three institutes: Mahanakorn University of Technology for providing financial support, the Faculty of Natural Resources, Rajamangala University of Technology Isan, Sakon Nakhon Campus for providing research facilities and the Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University for providing analytical support. The collaboration of the three institutes made it possible to be successful in my Ph.D. studies.

I definitely offer my sincere gratitude to my supervisor, Prof. Dr. Ir. Anton C. Beynen, who made me proud and happy to be working under his supervision. I was always supported by his optimistic phrase both in English ("no problem, don't worry) and in Thai ("mai mee pan ha") which gave me energy to continue and finish my work. He is a really professional scientist and I have gained invaluable research experience from him. I will follow your scientific guidance and the ways of your scientific thinking. Thank you very much for fulfilling my knowledge.

My utmost gratitude goes to Dr. Henk Everts, my co-supervisor, who opened my scientific points of view. It required all of my efforts to give him scientific answers even though I was usually wrong. However, I am very glad to have been able to discuss with him. I got insight of how to do scientific thinking and I will cherish the way of his thinking. This thesis could not have been completed without his constructive corrections and advice.

I deeply thank Dr. Chalermpon Yuangklang, my co-supervisor in Thailand, for his excellent support, hospitality and understanding during my Ph.D. programme. He was always right along me and he entirely got rid of all problems during my experimental work. My special thanks for giving me a chance to participate in his nutrition research group. Asst. Prof. Dr. Suntorn Wittayakun is thanked for his kindness and support.

I am very pleased to deeply thank Asst. Prof. Col. Dr. Udom Tipayamontri, the former dean of the Faculty of Veterinary Medicine, Assoc. Prof. Dr. Theera Rukkwamsuk, and Dr. Prasit Damrongsuntornchai for their encouragement to pursue both the master and doctoral degree. I also wish to extend my thanks to Assoc. Prof. Maj. Gen. Taweesak Buranawut and Assoc. Prof. Dr. Piboon Yutisri for their continuous support. For taking care of me, my thanks also go to Dr. Manich Tungtrakun, the director of Sakon Nakhon Agricultural and Training Centre (former name) and Ajarn Sutein Namwong, the dean of the Faculty of Natural Resources (current name), Rajamangala University of Technology Isan, Sakon Nakhon campus.

I am grateful to Dr. Thomas Schonewille, Dr. Math Geelen, Dr. David van Doorn, and Dr Esther Plantinga for their guidelines and assistance during my stay in the Netherlands.

I would like to express my special thanks to Robert Hovenier for his excellent laboratory analysis of fat and fatty acids. I could not prepare this thesis without his contribution. Not only assisting me in the laboratory, but also his kindness for taking me to visit the most interesting places in the Netherlands, including the football match in the Amsterdam Arena.

My thanks also go to Inez Lemmens and Xandra Fielmich-Bouman at the Department of Laboratory Animal Science, for their excellent laboratory analysis. I also thank to Hugo Wouterse and Jan van der Kuilen for their friendship.

I really thank my Thai colleagues, Kraisit Vasupen and Sasiphan Wongsuthavas for their hospitality and kind helping, especially at the beginning of my first experiment. I have enjoyed working with them. I also bring my special thanks to Paiwan Srenanul who was willing to help with laboratory analyses in Sakon Nakhon. It was my pleasure to share experiences and talking with the other Thai colleagues, Adisorn Yawongsa, Wandee Rungrattanaubol and Chaiwat Jarassaeng. I have enjoyed talking with my foreign colleagues, Du Thanh Hang and Mohammed Elmusharaf including Jason Tsai who is a nice and friendly guy from the Department of Farm Animal Health.

I would like to thank all staff members at the Faculty of Veterinary Medicine, Mahanakorn University of Technology for their support and willingness to assist me during my Ph.D. study, especially my colleagues in the Department of Clinic for Swine.

I would like to thank my Dutch friends, Saskia van der Drift and Marc Zevenbergen for their hospitality, friendship, and for helping me to survive in the Dutch environment and culture. Special thanks for all happiness, trips and meetings together, for the delicious Thai foods in Dutch style, and for their funny Thai words ("come", "horse", "dog" in Thai). I also thank Susanne Eisenberg for her friendship and academic help.

I also like to thank all staff member at the Department of Animal Science and Food Science and Technology, Faculty of Natural Resources, Rajamangala University of Technology Isan, Sakon Nakhon Campus for their kindness and help. Special thanks to Achara Lukkananukool for transforming raw data into electronic form. My thanks are also extended to all of my students, Suksan, Adisak, Nimit, Narongsak, Thanaphum, Kittipong, Yupha, Naphaporn, Nattawut, Wasuwat, Jiraporn, Wilaiwan, Chaiyod, and Sitthichai for their assistance and taking care of the animals.

I specially thank all of the Thai students in Utrecht for making my life enjoyable and colourful: Narin, Pichai, Chaidate, Prapansak, Ulsulee, Thanakarn, Niwat, Jitkamol, Surasak, Theerawat, Niyada, Thanawat, Narisara, Sietse Koopmans, Manote, Jadee, Chatchote, Chaweng, Paramee, Supranee, Montarop, Ekkamol, Nareupol, Kamolsiri, Sawaluck, Chalika, Duangnapa, Sansanee, Gib, May, doctor Song, doctor Naeng and all the others who I have not mentioned; my apologies if I did. I also thank the Thai Students Association in the Netherlands (TSAN) for giving me the opportunity to meet other Thai people and communities in different places and cities.

All of the animals that were used in my experiments are acknowledged for their contribution to scientific innovative knowledge and human wellbeing.

From my soul, my sincere and deepest thanks are towards my parents and four elder sisters (Jae Pui, Jae Nuan, Jae Ruay, and Jae Yao) who form my eternal support both physically and spiritually.

CURRICULUM VITAE

Jamlong Mitchaothai was born on July 29th, 1973 in Angthong, Thailand. He received his elementary education from Wat Saladin school, and primary and secondary education from Wisetchaichan Tantiwitthayaphoom school in Angthong. After he passed the special quota examination for excellent students, he started his study in the Faculty of Veterinary Medicine, Kasetsart University, Bangkok, Thailand in 1991 and received the degree of Doctor of Veterinary Medicine (DVM) in 1997. After graduation, he became a lecturer staff member in the Department of Clinic for Swine, Faculty of Veterinary Medicine, Mahanakorn University of Technology, Bangkok, Thailand. In September 2002, he was granted permission and support from Mahanakorn University of Technology to study for a Master degree at the Department of Farm Animal Health, Faculty of Veterinary Medicine, Utrecht University, The Netherlands; he received the degree of Master of Science (Veterinary Epidemiology and Economics) in March 2004. In the same month and year, he received a fellowship and permission from Mahanakorn University of Technology to pursue a Ph.D. study at Utrecht University. He chose to study at the Department of Nutrition, Faculty of Veterinary Medicine, Utrecht University under the supervision of Professor Dr. Ir. A. C. Beynen in the form of a sandwich Ph.D. programme through collaboration with the Sakon Nakon Agricultural Research and Training Centre, Rajamangala Institute of Technology (Faculty of Natural Resources, Rajamangala University of Technology Isan; current name), Phang Khon city, Sakon Nakhon, Thailand. His Ph.D. research work focused on the influence of dietary fat source on meat quality, body composition and energy balance in pigs. He will defend his Ph.D. thesis in public and obtain his Ph.D. degree on November 8th, 2007. Subsequently, he will return to Bangkok where he is appointed as a lecturer at the Department of Clinic for Swine, Faculty of Veterinary Medicine, Bangkok, Thailand.

List of publications (not included in this thesis)

First author

Mitchaothai, J.; Yuangklang, C.; Wittayakun, S.; Wasupen, K.; Wongsutthavas, S.; Srenanul, P.; Beynen, A. C., The effect of dietary fat source (beef tallow or sunflower oil) on fatty acid composition of main tissues and plasma lipid levels in growing-finishing pigs. Proceedings of the British Society of Animal Science 2006, York, UK, 27 – 29 March 2006: 69.

Mitchaothai, J.; Yuangklang, C.; Everts, H.; Vasupen, K.; Wongsutthavas, S.; Srenanul, P.; Beynen, A. C., Effect of dietary alpha-linolenic acid and linoleic acid level on growth performance, carcass and meat characteristics of Thai indigenous growing pigs. Proceedings of the British Society of Animal Science 2007, Southport, UK, 2 – 4 April 2007: 19.

Mitchaothai, J.; Yuangklang, C.; Wittayakun, S.; Vasupen, K.; Wongsuthavas, S.; Srenanul, P.; Beynen, A. C., Effect of dietary fat type on nutrient digestibility and carcass quality in growing finishing swine. AUNP symposium: New dietary strategies to improve animal health and food safety. In: Proceedings of Integrating Livestock-Crop Systems to Meet the Challenges of Globalisation. vol 2. AHAT/BSAS International Conference. Khon Kaen, Thailand, 14 – 18 November 2005: Vol 3:80-84.

Mitchaothai, J.; Yuangklang, C.; Wittayakun, S.; Vasupen, K.; Wongsuthavas, S.; Srinanaun, P.; Beynen, A. C., Effect of dietary fat type on growth performance, nutrient digestibility, and Ca – P absorption in growing – finishing swine. In: Proceedings of Integrating Livestock-Crop Systems to Meet the Challenges of Globalisation. vol 2. AHAT/BSAS International Conference. Khon Kaen, Thailand, 14 – 18 November 2005: Vol 2:T73.

Mitchaothai, J.; Yuangklang, C.; Wittayakun, S.; Vasupen, K.; Wongsuthavas, S.; Srenanul, P.; Beynen, A. C., Mathematical modeling between pork colour and pork and carcass qualities in commercial finishing pigs. Proceeding of the 31st congress on science and technology of Thailand, Suranaree University of Technology, Nakhon Ratchasima, Thailand, 18 - 20 October 2005: F0043.

Mitchaothai, J.; van Leengoed, L. A. M. G.; van der Meer, F. J. U. M.; Cruijsen, A.; Heesterbeek, J. A. P., Comparative testing and immune response for PRRSV in vaccinated sows. Kasetsart Journal. Supplement Issue, Vol 39, number 5: 23-29.

Mitchaothai, J.; Yuangklang, C.; Wittayakun, S.; Vasupen, K.; Wongsuthavas, S.; Srinanuan, P.; Beynen, A.C., 2006. Effect of fat type and calcium level supplementary on growth performance, fat digestibility, and Ca & P absorption and balance in growing-finishing pigs. Kasetsart Journal: Natural Science 40 (5): 298-303.

Mitchaothai, J.; Wiengchareon, J.; Chaiworaporn., J.; Srenanuan, P.; Yuangklang, C., 2005. Balantidiasis in Growing Pigs: A Case report. Khon Kaen University Vet Journal, Thailand, 15:166-174. (in Thai)

Co-author

Srenanul, P.; Yuangklang, C.; Vasupen, K.; Wongsutthavas, W.; **Mitchaothai, J.**; Wachirapakorn, C.; Beynen, A. C., Effect of trace mineral supplementation on feed intake, milk yield and milk composition in dairy cows. In: Proceeding of the XII th AAAP congress.18-22th September BEXCO, Busan, Korea. CO1-PP-019.

Vasupen, K.; Yuangklang, C.; Wongsuthavas, C.; **Mitchaothai, J.**; Srenanual, P.; Beynen, A. C., Study on Carcass and Pork Characteristics of Kadon Pigs. In Proceedings of The First Joint PSU-UNS International Conference on BioScience ; Food, Agriculture, and Environment. Songkhla, Thailand,17 – 18 August, 2006. 238-242.

Vasupen, K.; Yuangklang, C.; Sarnklong, C.; Wongsuttawas, S.; Wittayakun, S.; Srinanul, P.; **Mitchoathai, J.**; Beynen, A. C., Effect of carbohydrate sources on nutrient digestibility and nitrogen utilization of native growing pigs. In: Proceedings of Integrating Livestock-Crop Systems to Meet the Challenges of Globalisation. vol 2. AHAT/BSAS International Conference. Khon Kaen, Thailand. 14 – 18 November 2005, Vol 2 : T172.

Vasupen, K.; Yuangklang, C.; Sarnklong, C.; Wongsuthavas, S.; **Mitchaothai, J.**; Srenanul, P., Effects of total mixed ration and fermented total mixed ration on voluntary feed intake, digestion nutrients digestibility, and milk production in lactating dairy cows. Proceedings of the 44th Kasetsart University Annual Conference, Kasetsart University, Bangkok, Thailand, 30 January 2 February, 2006 Subject: Animals, Veterinary Medicine: 167-176.

Vasupen, K.; Wongsuttawas, S.; **Mitchoathai, J.**; Srinanaun, P.; Yuangklang, C., 2006. Effects of total mixed ration and fermented total mixed ration on feed intake, nutrients digestibility, milk and milk composition in dairy cattle. In: Proceeding of the XII th AAAP congress.18-22th September BEXCO, Busan, Korea. CO1-PP-101.

Wongsuthavas, S.; Yuangklang, C.; **Mitchaothai, J.**; Vasupen, K.; Beynen, A. C., 2007. Energy balance of beef tallow and soya bean oil in broiler chickens. Proceedings of the British Society of Animal Science 2007, Southport, UK, 2 – 4 April 2007: 240.

Wongsuthavas, S.; Yuangklang, C.; Vasupen, K.; **Mitchaothai, J.**; Srenanul P.; Beynen, A. C., Effect of saturated fatty acid and unsaturated fatty acid ratio in broiler ration on growth performance and fat deposition. Proceeding of the 31st congress on science and technology of Thailand, Suranaree University of Technology, Nakhon Ratchasima, Thailand, 18 - 20 October 2005: F0041.

Wongsuthavas, S.; Yuangklang C.; Wittayakun, S.; Vasupen, K.; **Mitchoathai, J.**; Srinanul, P.; Beynen, A. C., Effect of saturated to unsaturated fatty acids ratios on fat digestibility and growth performance in broiler chickens. In: Proceedings of Integrating Livestock-Crop Systems to Meet the Challenges of Globalisation. vol 2. AHAT/BSAS International Conference. Khon Kaen, Thailand. 14 – 18 November 2005, Vol 2 : T162.

Wongsuthavas, S.; Yuangklang, C.; Vasupen, K.; **Mitchaothai**, **J.**; Srenanual, P.; Wittayakun, S.; Beynen, A. C., Eeffect of fat types on energy utilization, body deposition and plasma lipids in broiler diets. In: Proceeding of the XII th AAAP congress.18-22th September BEXCO, Busan, Korea. CO4-OP-04.

Yuangklang, C.; Vasupen, K.; Wongsutthavas, S.; Srenanul, P.; **Mitchaothai, J.**, Effect of fat sources and sodium fumarate supplementation on nitrogen utilization in goats. Proceedings of the British Society of Animal Science 2006, York, UK, 27 – 29 March 2006: 156.

Yuangklang, C.; Vasupen, K.; Srenanul, P.; Wongsuthavas, S.; **Mitchaothai, J.**; Kongwaha, K., 2007. Effect of substitution of soybean meal by dried tomato pomace on feed intake, rumen fermentation and nitrogen utilization in goats. Proceedings of the British Society of Animal Science 2007, Southport, UK, 2 – 4 April 2007: 240.

Yuangklang, C.; Vasupen, K; Wittayakun, S.; Wongsutthavas, S.; **Mitchaothai, J.**; Srenanaul, P.; Ngamsen, N.; Kongweha, K., Effects of sodium fumarate supplementation on voluntary feed intake, rumen fermentation, nutrient digestibility and blood metabolites in beef heifers. Proceeding of the 31st congress on science and technology of Thailand, Suranaree University of Technology, Nakhon Ratchasima, Thailand, 18 - 20 October 2005: F0053.

Yuangklang, C.; Vasupen, K.; Srenanul, P.; Wongsutthavas, S.; **Mitchoathai, J.**; Kongweha, K., Effect of Trace Element Supplementation on Growth Performance and Carcass Characteristics of Dairy Steers. The PSU-UNS International Conference on BioScience "Agriculture, Food and Environment", Songkhla, Thailand,17 – 18 August, 2006. 71-76.

Yuangklang, C.; Vasupen, K.; Wittayakun, S.; Sarnklong, C.; Wongsuttawas, S.; **Mitchoathai, J.**; Srinanul, P., Effect of substitution of soybean meal by dried tomato pomace on feed intake, digestibility, blood metabolites and milk production in lactation cows. AUNP symposium: New dietary strategies to improve animal health and food safety. In proceeding of AHAT-BSAS international conference, Khonkaen, Thailand, 14 – 18 November 2005: Vol 3:112-115.

Yuangklang, C.; Vasupen, K.; Srenanul, P.; Wongsutthavas, S.; **Mitchaothai, J.**, Effect of utilization of dried tomato pomace as roughage source on feed intake, rumen fermentation and blood metabolites in beef cattle. Proceedings of the 44th Kasetsart University Annual Conference, Kasetsart University, Bangkok, Thailand, 30 January 2 February, 2006 Subject: Animals, Veterinary Medicine: 158-166.

Yuangklang, C; Vasupen, K.; Srenanul, P.; Wongsutthavas, S.; **Mitchaothai, J.**, Effects of supplemental trace element (GT1000) on growth performance of dairy bulls. Proceedings of the 44th Kasetsart University Annual Conference, Kasetsart University, Bangkok, Thailand, 30 January 2 February, 2006 Subject: Animals, Veterinary Medicine: 150-157

Yuanklang, C.; Vasupen, K.; Srenanul, P.; Wongsutthavas, S.; **Mitchaothai, J.**; Phadankaew, S.; Yuwabuth, N., Effect of type and level of fat on feed intake and milk production in dairy cows. In: Proceeding of the XII th AAAP congress.18-22th September BEXCO, Busan, Korea. CO1-OP-16.

Yuanklang, C.; Vasupen, K.; Srenanul, P.; Wongsutthavas, S.; **Mitchaothai, J.**; Phawasiri, S.; Wuttisarn, K., Effect of protein source and fat types on feed intake and milk production in dairy cows. In: Proceeding of the XII th AAAP congress.18-22th September BEXCO, Busan, Korea. CO1-PP-105.