

## ABSTRACT

PRICE, KATHRYN LEIGH. Improving Fat Utilization by the Weaned pig: Effect of Diet Physical Form, Fatty Acid Chain length, and Emulsification. (Under the direction of Jack Odle.)

Previous research indicates that dietary fat utilization by the newly-weaned pig is low, while fat digestive capabilities prior to weaning are very high. Sow milk contains approximately 40% fat, whereas nursery diets are rarely formulated to contain more than 5%. The aim of this experiment was to determine if emulsification (plus or minus Tween-80), physical form of the diet (liquid vs dry) or fatty acid chain length (medium (MCT) vs long chain triglyceride (LCT)) effect fat utilization by the newly weaned pig. Two replicates were performed in the summer of 2006 at the NCSU Swine Educational Unit. Pigs (N= 96) were weaned at  $20 \pm 0.30$  d of age ( $6.8 \pm 0.04$  kg) and fed one of eight dietary treatments for 14 d according to a 2x2x2 factorial design. The MCT fat contained primarily C8 and C10 fatty acids while the LCT fat was supplied by choice white grease. Each fat was spray dried with or without the inclusion of Tween-80 emulsifier (at 2% w/w) and comprised 12% of the final diets. Diets were otherwise formulated to exceed NRC nutrient requirements. Liquid diets were reconstituted with water to 13% dry matter and were offered ad libitum via milk-replacer feeders (Kane Manufacturing).

Diet physical form greatly accelerated piglet growth ( $P < 0.05$ ), with liquid-fed pigs (0.49 kg/d) out gaining dry-fed pigs (0.340 kg/d) by 44%. Triglyceride chain length also impacted growth ( $P < 0.05$ ), with pigs fed LCT outperforming MCT-fed pigs by 23%. Effects of emulsifier were not detected ( $P > 0.1$ ) in growth performance. Accelerated growth was accompanied by elevated feed intake which was 17 % greater for liquid-fed than for dry-fed

pigs and was 21% greater for pigs fed LCT vs MCT. Accordingly, gain:feed was improved by 29% in liquid-fed pigs ( $P < 0.05$ ).

A comparison of two digestibility markers, C 36 alkane and Co-EDTA, was examined in this trial. The use of Co-EDTA resulted in a statistically ( $P < 0.05$ ) greater digestibility over the C 36 marker. However, it is unclear as to which marker is the more accurate when trying to determine fat digestibility. C 36 was used to determine all digestibility data and diet physical form had no effect ( $P > 0.1$ ) on digestibility. The digestibility of the fat was higher ( $P < 0.05$ ) in the MCT fat over the LCT fat (98.42% vs 93.39%, respectively). Emulsification increased the digestibility of the LCT fats and this effect was more evident in the long chain saturated fatty acids ( $P < 0.05$ ).

Ileal and Jejunal morphology showed increases in the villus height as the pigs aged ( $P < 0.05$ ). Emulsification of the fat resulted in an increased Ileal villus height ( $P < 0.05$ ) in all diets except for the liquid MCT fed pigs that were 14days post weaning.

Plasma ketone body concentrations were significantly ( $P < 0.05$ ) greater in the MCT fed pigs and interaction between chain length and diet physical form was observed. Pigs consuming the dry MCT diet had a higher plasma ketone body concentration followed by liquid MCT, liquid MCT, and finally dry LCT fed pigs.

Collectively, we infer that feeding liquid diets containing emulsified long chain triglycerides can increase growth performance for the entire weaning period.

Improving Fat Utilization by the Weaned pig: Effect of Diet Physical Form,  
Fatty Acid Chain length, and Emulsification

by  
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## **BIOGRAPHY**

Kathryn Leigh Price was born on February 23, 1983 in High Point, North Carolina to Allen and Leigh Price of Madison, North Carolina. Kathryn was an active member of the local 4-H club in Rockingham County where she gained her appreciation for animals. She graduated from McMichael High School in Madison, North Carolina in 2001. Kathryn attended North Carolina State University and completed her Bachelors of Science degrees in Animal Science and Poultry Science, with concentrations in Feed Mill Management and Agricultural Business Management in December, 2005. During this time, she discovered her interest in swine research while working as a lab research assistant. After graduation, Kathryn began working on her Masters of Science degree in Animal Science at NCSU under the direction of Dr. Jack Odle. She plans to attend Virginia Tech University to pursue a PhD under the direction of Dr. Jeffery Escobar.

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## CHAPTER I: LITERATURE REVIEW

### Introduction

Research has shown that the newly weaned pig is less capable of utilizing fat than a suckling pig. Typical weanling pig diets include fat at 3-6% of the diet to aid in the pelleting process (Maxwell and Carter 2001) and to increase the energy density of the diet (Stahly 1984). On the other hand, sow's milk contains roughly 40% fat (DeMan et al., 1963). The literature is inconsistent on the value of added fat in nursery diets. There have been several experiments that show no improvement or negative effects on growth performance when increasing the level of fat in the diet (Peo et al., 1957, Eusebio et al., 1957); however, there are also studies showing an increase in growth performance when using a diet high in fat (Oliver et al., 2005, Allee, et al., 1971 and Sewell and Miller 1965).

According to the NAHMS 2006 survey a total of 13.8% of suckling pigs and 9.8% of weaned pigs died due to starvation. Therefore, in an attempt to increase survivability research into the supplementation of medium-chain triglycerides (MCT) in weanling pig diets has shown that MCTs are more readily oxidized and absorbed by the young pig (Odle 1997). Both in vivo and in vitro analyses using rats have shown that the hydrolysis of MCT occurs at a much quicker rate than the hydrolysis of long-chain triglycerides (LCT, Greenberger et al., 1966).

The suckling pig is capable of 97% digestibility of fat from sow milk (Lucas and Lodge 1961) while fat in the weanling pig diet is approximately 85% digestible (Cera et al., 1988, Hamilton and McDonald). The digestibility of fat increases over time and medium chain fats have been shown to be more digestible than a long chain fat source. Dietary fats high in saturated fatty acids are more digestible than fats high in unsaturated fatty acids (Cera et al., 1989).

The use of added emulsifying agents in weanling pig diets is one method researchers are investigating in attempt to increase the digestibility of LCT diets. However, the literature is inconsistent as to which emulsifier and the amount needed to improve growth performance. Tween-80 has been shown to be a more effective emulsifier at improving plasma concentrations of medium-chain fatty acids when compared to soy-lecithin or gum-arabic (Wieland et al., 1993). The use of either lysolecithin or lecithin increased digestibility for the first 10 days after weaning when added at 0.02%, while piglet performance was improved when lysolecithin was added at 0.01% of the diet (Xing et al., 2004). However, in another study a decrease in digestibility was observed when lysolecithin was included in the diet and lecithin tended to increase the digestibility when either emulsifier was included at 5%, 10% or 30% of the fat (Jones et al., 1992). Growth performance decreased when lecithin was included at a level greater than 5% and when any level of lysolecithin was used. In older pigs (between 11 and 35 days post weaning) ADG and ADFI increased when either emulsifier was included above 5% in the diet, resulting in similar feed efficiencies among treatments (Jones et al., 1992).

Conventional weaning programs transfer weaned piglets on to a dry starter diet but the abrupt change can result in a loss of weight and/or a decrease in feed intake. Research has shown that feeding weaned piglets a liquid diet can improve growth performance by as much as 53% over sow reared pigs decreasing the time it takes a pig to reach market weight, 110kg, by 10 days (Harrell et al., 1993). Another study comparing sow reared piglets, piglets fed a dry starter diet and piglets fed a milk replacer diet resulted in a higher ADFI, ADG and BW in the pigs fed a milk replacer diet (Zijlstra et al., 1996).

Methods to improve fat utilization by examining diet physical form (liquid vs dry), fatty acid chain length (MCT vs LCT) and emulsification (added Tween-80 or no Tween-80) were investigated during this research trial and will be discussed in this review of the literature.

### **Production aspects of weaning pigs**

The weaning period in the swine industry can vary in duration and independent of the age at weaning, a decrease in growth performance is observed following weaning. According to the NAHMS 2006 Swine Report, 69.4% of swine farms wean pigs between 16 and 20 days of age. Over the past decade the average weaning age has decreased by 10 days (NAHMS, 2000). Actual piglet weaning ages have fluctuated between 10 and 35 days of age over the past decade (Odle and Harrell, 2003). The weaning age of piglets has been decreasing in an attempt to increase profits and to reduce the amount of diseases transferred from the dam to the piglet (Maxwell and Carter, 2001). A decrease in the weaning age allows for more litters per sow per year, therefore, increasing profits (Odle and Harrell, 2003). Regardless of the age of weaning a decrease in piglet growth performance occurs. This decrease lasts for an average of 3 days post weaning and is thought to be due to the high stress at weaning and changes in the diet (Kim et al., 2001, and Maxwell and Carter 2001). Weaning is a stressful time for piglets because not only are they being removed from their dam and litter mates but they are being handled by humans to be weighed and sorted into their new pens. Once in their new pens the piglets generally will not begin consuming feed for several hours to several days post weaning (Brooks et al., 2001). In a study by Brooks et al. (2001) it was shown that approximately 50% of weaned piglets do not consume their first meal until 24 hours post weaning and 10% of piglets still have not eaten 48 hours after weaning.

There are major dietary changes at weaning that the piglet must adjust to with the main change being in diet physical form. Prior to weaning the piglet is on an all milk diet and at weaning the form of the diet changes to a dry pellet. The main sources of protein, energy and carbohydrate change as well. The protein source changes from casein or whey protein source to a soy or corn protein source. The main energy source becomes carbohydrates instead of fat and the main carbohydrate source transitions from being lactose to starches (corn). Another major change in the diets of weaned pigs is the amount of fat included in the diet. Sow milk has as much as 40% fat on a dry matter basis, however; weaning diets rarely include more than 6% fat. Fat inclusion in swine nursery diets is typically added to aid in the pelleting process. However, in addition to improvements in pellet quality, a greater fat content in the diet increases the energy density of the diet. Adding more fat to weaning pig diets is thought to aid newly weaned piglets especially immediately after weaning when they are consuming little to no feed (Stahly 1984). As will be discussed later in this review, the digestive capabilities of the newly weaned pig may be insufficient to adequately digest the standard dry weaning diet and are not thought to be fully functioning until the pig is 4 weeks of age (Maxwell and Carter 2001).

Wolter et al. (2002) concluded that suckling pigs given access to milk replacer grew faster and had fewer preweaning deaths than sow reared piglets. During the postweaning (up to 14kg) and finishing (65-110kg) phases no detectable effect was observed between pigs fed a milk replacer diet or a dry starter diet. However, a significant increase in body weight was observed during the grower (25-65kg) phase in pigs that had been fed a milk replacer diet during the weaning period. Even though differences were not observed during the weaning and finishing phases the growth performance improvements during the grower phase resulted in a decrease in the time it took pigs to reach market weight by 3 days (Wolter et al., 2002).

New research attributes the decrease in growth performance at weaning to aggression in piglets. Aggression in weaned piglets is thought to be caused by the piglets creating new hierarchies once they have been arranged with their new pen mates. The more time the piglets spend fighting means they are not consuming feed. Therefore, this could contribute to the reduction in body weight seen immediately after weaning (Jensen et al., 2002). Studies by Newberry et al. (2000) and D'Eath et al. (2005) determined that piglets that were commingled with other litters during the suckling phase resulted in less fighting and the development of hierarchies quicker. A study by Weghe et al. (2006) examined the activity of piglets that were either commingled or not socialized prior to weaning and its effects prior to and after weaning. They concluded that immediately after weaning and until 4 hours post weaning that piglets not socialized showed more aggressive behavior than the commingled piglets (Weghe et al., 2006). They hypothesized that the piglets that were commingled during the suckling phase had already developed hierarchies resulting in less aggression during weaning.

#### *Diet Physical Form*

Conventional weaning programs transfer weaned piglets on to a dry starter diet but the abrupt change in diet appears to cause a loss of weight and/or a decrease in feed intake immediately after weaning. Research has shown that feeding weaned piglets a liquid diet can improve growth performance by as much as 53% over sow reared pigs, decreasing the time it takes a pig to reach market weight, 110kg, by 10 days (Harrell et al., 1993). Piglets fed a milk replacer diet had a higher ADFI, ADG and BW than pigs fed a dry starter diet or sow reared piglets (Zijlstra et al., 1996). In an experiment by Kim et al. (2001) liquid fed pigs outgrew dry fed pigs by 21% during the first 14 days post weaning (weaned at 11 days). Experimental diets were formulated to be identical, with diet physical form (liquid vs dry) being the only difference.

Providing newly weaned pigs with a readily available energy source is essential to improving both survivability and growth performance (NAHMS 2006 and Kim et al., 2001). Liquid fed pigs can increase their body weight gains in the first three days post weaning by four fold compared to dry fed pigs. Diet physical form had less of an impact once the pigs were older than 15 days post weaning but dry fed pigs never out performed liquid fed pigs. Pigs in this experiment reached market weight about 4 days earlier when fed a liquid diet compared to a dry diet (Kim et al., 2001). It has been suggested that both the piglet and the sow can limit the piglets ability to maximize their growth. The theory is that the sow is not stimulated enough to produce milk at her maximum therefore, piglet growth is limited. Weaning pigs too early, close to 10 days of age, generally results in poor growth performance which is mainly due to the diet being fed in a dry form (Odle & Harrell 2003).

#### *Value of added fat in nursery pig diets*

Fat in weanling pig diets is mainly added to help in the pelleting process and increase the energy density of the diet. However, research is inconsistent on the value of adding fat to nursery diets. Sow milk contains 40% fat (DeMan and Bowland 1963) which is 97% digestible (Lucas and Lodge 1961), however, typical weanling diets are only 3-6% (Maxwell and Carter 2001) fat which is between 85% and 93% digestible (Cera et al., 1988, Hamilton and McDonald ). Adjusting the source of fat and amount of fat included in nursery diets is one way researchers can attempt to maximize piglet growth performance at weaning.

Eusebio (1960) and Peo (1957) conducted several weanling pig trials examining the effect of increasing the level of fat included in the diet from 0 to as high as 38%. Eusebio et al. (1960) conducted 5 experiments on pigs weaned on average at 17.3 days onto stabilized lard, soybean oil or a coconut oil diet. In one of the experiments, piglets weaned onto the diet



containing soybean oil gained significantly less than pigs weaned onto the control diet (no fat) or the coconut oil diet. Piglets fed the control diet (no added fat) outperformed piglets weaned onto a diet containing dietary fat. Peo et al. (1957) conducted 4 trials with pigs weaned at 7.9 days of age onto a diet containing stabilized lard. Over the 9 experiments conducted by both Eusebio et al. (1970) and Peo et al. (1957) a decrease in total weight gain and feed efficiency was observed when the level of fat inclusion exceeded 5%. When fat was added between 0 and 5% growth performance was not significantly effected. Gain:Feed (G:F) was not significantly affected and in most cases a decrease in G:F was observed when higher levels of fat were included in the diet (Eusebio et al., 1960 and Peo et al., 1957).

Allee et al. (1971) conducted 3 research trials with pigs weaned at 5 weeks for 1 trial and weaned at 14 days for 2 trials. Dietary fat source used during this experiment was corn oil. Sewell and Miller (1965) conducted 3 trials with pigs weaned at 22.7 days of age onto either a diet containing corn oil, lard or beef tallow. Combined they observed a significant increase in gain to feed when increasing the amount of fat in the diet from 1% to as high as 13%. They reported a trend for a decrease in feed intake while body weights were unaffected by the added fat resulting in an improvement in G:F. Through their research they concluded that young pigs are capable of efficiently utilizing dietary fat (Allee et al., 1971 and Sewell and Miller 1965). Oliver et al. (2005) conducted a trial feeding liquid diets comparing a high fat, 25%, and a low fat, 2%, diet in pigs weaned at 10 days of age. In this experiment the level of fat (blend of lard and tallow) did not effect total weight gain; however, G:F was increased in pigs on the high fat diet (Oliver et al., 2005).

## **Fat digestion**

Chemical digestion of fat is generally thought to begin in the stomach; however, research shows that the breakdown of fat may actually start in the mouth. Chewing the food to break it into smaller particles and salivary secretions may start the breakdown of fat. Lingual lipase and gastric lipase are two enzymes secreted from the serous glands and the gastric mucosa, respectively, that are important in the hydrolysis of fat in the stomach. While lingual lipases are important in some species (Hamosh 1990) much lower levels are present in the pig (Dicklin et al., 2006.). The stomach serves as a warming mixer with cleaving abilities to help further break down the fat particles before they enter into the intestine. The previous two enzymes have a large range in pH (2.2 – 6.0) for optimal conditions during the digestion of fat. Although the important enzyme for fat breakdown (pancreatic lipase) comes from the pancreas, gastric lipase is thought to be responsible for 10-30% of the hydrolysis of triglycerides to smaller glyceride molecules (diglycerides, monoglycerides and free fatty acids) in young pigs on a milk based diet (Hamosh 1990). Prior to entering the intestine the digestion of either medium or long chain triglycerides is relatively similar, although MCFA can pass directly through the gastric mucosa (Hamosh 1990). Medium and short chain triglycerides can enter directly into the intestinal mucosal cells, while long chain triglycerides (LCT) must first be emulsified with the aid of bile and hydrolyzed in the lumen of the intestine. Hydrolysis of the LCT by pancreatic lipase and colipase will result in free fatty acids and a monoglyceride. Without colipase, pancreatic lipase would be ineffective due to the action of bile salts working to inactivate pancreatic lipase. The LCT digestion products are then formed into mixed-micelles (small in size and water soluble) by the action of bile salts. The micelle formation is what allows the digestion products to traverse through the unstirred water layer in the intestinal mucosal cells into the enterocyte. At this point

the LCT products are re-formed into long chain phospholipids and triglycerides and then made into a chylomicron. The chylomicron will then enter into the lymphatic system and travel through the heart to the liver where their fate is dependent on the nutritional state of the animal (oxidized to ketone body's or stored). On the other hand, MCT pass from the stomach straight through the intestine into the portal vein. Once in the blood stream MCT reach the liver quicker than an LCT due to the lymph system having a much slower flow rate. The portal vein system flows directly to the liver while the lymphatic blood flows through several organs prior to reaching the liver (Odle 1997 and Playoust et al., 1964 and Azain 2001). Finally a small amount of lipid digestion takes place in the large intestine when hydrogenation of unsaturated fatty acids occurs by the action of the microflora in the intestine (Azain 2001).

### **Factors affecting fat digestibility**

There are several factors that can affect the digestibility of fat, such as fatty acid chain length, the degree of saturation of the fat source, whether the fat is emulsified and the stage of development of the intestine. Increasing the digestibility of the fat source may increase fat utilization in the newly weaned pig. If the newly weaned pig is more efficient at digesting its diet one theory is that there will be less of a reduction in body weight just after weaning.

#### *Fatty acid chain length*

Some of the major differences between LCT and MCT are that MCTs are liquid at room temperature and contain fatty acid C6-C14 while LCT are normally solid at room temperature and are made up of fatty acids C16 and higher. Another major difference is that LCT can not be absorbed when bile salt and pancreatic lipase are not present or are present in low amounts while MCT can still be absorbed (Caliaria et al., 1996). Diets high in MCT can have palability issues and result in gastro-intestinal (GI) upsets whereas diets high in LCT do not have those issues

(Jekendrup and Aldred 2004). According to USDA (2000) survey a total of 12.5% of suckling pigs and 17.6% of weaned pigs died due to starvation (NAHMS, 2000). Therefore, research into the use of MCT supplementation in an attempt to increase survivability has shown that MCTs are more readily oxidized and absorbed by the young pig (Odle 1997).

Researchers have shown that there are differences in digestibility when it comes to chain length of the dietary fat source and when the pig transitions from sow's milk to a weanling diet. In a study by Greenberger et al. (1966) they were able to show by both in vivo and in vitro work using rats that the hydrolysis of MCT occurs at a much quicker rate than the hydrolysis of LCT and they also examined the impact bile salts have on hydrolysis of triglycerides. In their in vivo experiment they injected <sup>14</sup>C-labeled triolein (MCT) and tripalmitin (LCT) into healthy intestinal loops and examined how much of the radio-labeled lipid was hydrolyzed to fatty acid in 15 minutes. After the 15 minutes 92% of the MCT had been hydrolyzed while only 29% of the LCT had. They also conducted in vitro work to further prove that MCT are hydrolyzed quicker than LCT by incubating healthy intestinal juices with either the MCT or LCT source for 10 minutes. At the end of 10 minutes 73.5% of the MCT was hydrolyzed into fatty acids while only 14.5% of the LCT had been hydrolyzed in the same time period. A reduction in the breakdown of both triolein and tripalmitin in the intestine that had been previously irrigated to reduce the amount of pancreatic enzymes and bile salts prior to injecting the triglycerides was observed. The percentage of MCT hydrolyzed dropped from 92% to 2.9% while the percentage of LCT hydrolyzed dropped from 29% to 1%. Even though they observed a significant decrease in the amount of triglyceride hydrolyzed, the rate of MCT hydrolyzed was still greater than the amount of LCT hydrolyzed to fatty acids. The outcome of this experiment shows that bile salts

and pancreatic lipase are needed for efficient hydrolysis of triglycerides (Greenberger et al., 1966).

Cera et al. (1988) examined three different LCT fat sources: corn oil, tallow and lard, for their effects on digestibility in 21 day old weaned pigs when fed at 8% of the diet. The corn oil source (vegetable) was only 15% saturated fatty acids and 85% unsaturated fatty acids, while the lard and tallow fat sources (animal fat sources) were 35% saturated fatty acids and 65% unsaturated fatty acids. During the experiment they observed a higher apparent fat digestibility, in the corn oil fed pigs throughout the entire 4 week study compared to the tallow and lard groups. The digestibility of the tallow and lard group remained similar to each other throughout the entire trial. By the end of the 3<sup>rd</sup> week the digestibilities of the 2 animal fat sources had approached similar levels of digestibility as the vegetable fat source but they never reached the same digestibility. The digestibility of the corn oil increased from 79% at week 1 to 88.8% at week 4, lard increased from 68.1% to 84.9% and tallow increased from 64.8% to 82.5%. While on study, feed intake and body weight each week were similar among groups indicating that another factor may have affected growth performance (Cera et al., 1988).

Coconut oil is a commonly used source of MCT. An experiment by Cera et al. (1989), examined the digestibility of three different fat sources: coconut oil, tallow and corn oil, and their effect on piglet growth performance when fed at 8% of the diet. The coconut oil in the experiment contained approximately 60% MCT with dodecanoic, C12, making up half of the MCT in coconut oil (Cera et al., 1989). The coconut oil had a high proportion of saturated fatty acids, 73.6%, and only 25.6% of unsaturated fatty acids. The corn oil and tallow had similar values as previously noted. Tallow had the lowest digestibility throughout the entire trial and coconut oil was the highest with corn oil remaining in between the two fat sources. Over the 4

week period, the digestibility of each fat increased over time; coconut oil increased from 81.7% to 87.3%, corn oil went from 76.5% to 84.8% and tallow increased from 75.4% to 81.8% (Cera et al., 1989).

Cera et al. (1989) also examined the effect of feeding one fat source or mixing fat sources at a ratio of 1:1. The fat (included at 8% of the diet) sources used were coconut oil, corn oil, tallow, coconut:tallow, coconut:corn oil and a mixture of corn oil:tallow. Pigs fed the diet containing coconut oil had the greatest daily body weight gain throughout the entire trial. The pigs on the 1:1 mixture of tallow and corn oil performed the worst. The diets containing only tallow or corn oil had the lowest daily gain throughout the length of the trial (Cera et al., 1989).

Diets containing MCT do not always result in greater body weights as reported by Frobish et al. (1970). In a series of studies conducted by Frobish et al. (1970) the effect of experimental fat source on body weight gain in newly weaned piglets was examined. The experimental fat sources (butter, lard, coconut oil, and vegetable oil) used in the first study were included at 10% of the diet. In two additional experiments the same fat sources were used with the addition of methyl esters from corn oil and hydrolyzed vegetable and animal fat mixture. At the conclusion of all three studies, average body weight gains, after 15 days on trial, were not statistically different between fat sources. Piglets on the control (7.6 kg) diet had the greatest total gain followed by butter (6.7 kg), vegetable oil (6.2 kg), coconut oil (6.2 kg) and then lard (5.84 kg). However, after feeding the piglets for 10 days, fat digestibility was greatest for the hydrolyzed vegetable and animal fat mixture, 82%, followed by coconut oil (75.1%), methyl esters (72.1%), butter (68.9%), lard (67.7%), vegetable oil (65%), and then the control diet (23.8%). The digestibility of each fat source increased between day 10 and day 28. On day 28, the hydrolyzed vegetable and animal fat mixture had the highest fat digestibility at 89% followed

by butter (86.3%), lard (84.2%), coconut oil (83.8%), vegetable oil (82.4%), and the control (44.6%) diet had the lowest digestibility (Frobish et al., 1970).

In a review paper on medium chain triglycerides by Marten et al. (2006), they report that diets containing LCT had greater weight gains compared to those on a MCT diet. The diets were isoenergetic to prevent weight gain differences due to differing energy values among diets. Subjects on the MCT diet had less fat deposition than those on the LCT diet. They attribute some of the reduced weight gains to the thermogenic effect of MCT observed in rodents. In humans, greater expenditure of energy was observed after consuming a diet high in MCT and persisted for hours after the meal (Marten et al., 2006). An additional explanation for the lower weight gains in MCT fed rats could be due to MCT having a more satiating effect which would lead to lower feed intake (Marten et al., 2006).

#### *Degree of saturation*

The digestion of long chain fats is affected by the ratio of unsaturated fat (U) to saturated fat (S); whereas, the digestibility of medium and short chain fats are not as affected by the degree of saturation (Stahly 1984). A diet high in saturated fat is less digestible than one high in unsaturated fats due to a decrease in micelle formation. However, due to medium and short chain fats ability to easily form micelles they still have a high digestibility of 80-95% even though they are high in saturated fatty acids. The unsaturated fat to saturated fat (U/S) ratio can play a major role in the digestibility of various fat sources. While a fat with a U/S ratio of 1.5 or higher will be 85-92% digestible, a U/S ratio of 1.3 or less will result in a lower digestibility of 35-75%. Therefore, increasing the inclusion of unsaturated fats to a highly saturated diet could increase the overall digestibility of the fat (Stahly 1984).

According to a study by Ockner et al. (1972), long chain unsaturated fatty acids are absorbed in the proximal portion of the small intestines while long chain saturated fatty acids are more likely to be absorbed in the distal portion of the intestines. They also concluded that long chain saturated fatty acids require a greater surface area for absorption and they depend more on bile salts than do long chain unsaturated fatty acids. (Ockner et al., 1972)

### *Emulsification*

Dietary fat source is one of many variables being studied in an attempt to improve fat digestibility and growth performance of weanling pigs, but the use of added emulsifiers in weanling pig diets is becoming more popular. However, the literature is inconsistent as to which emulsifier and the amount needed to improve growth performance of the newly weaned pig. As shown by Cera et al. (1989) and Caliarra et al. (1996) digestion and utilization of fat is limited for the first couple of weeks after weaning and it is theorized that including emulsifiers in the diet may improve fat utilization. The addition of emulsifiers in the diet may assist the emulsification process performed by the body during fat digestion (Augur et al., 1947).

In an experiment involving suckling pigs, Wieland et al. (1993), determined that the rate of MCT hydrolysis to medium chain fatty acids (MCFA) was increased by using Tween-80 as an emulsifier. They examined the effects of added emulsifier or no emulsifier to MCT oil. The MCT oil contained two primary fatty acids, 75% octanoate (C8) and 25% decanoate (C10). Three different emulsifiers were tested in two trials, Tween-80, soy-lecithin and gum arabic. Pigs fed the Tween-80 treatment had a 20 fold increase in plasma C8 concentrations 2 hours post feeding compared to pigs fed the supplement with no added emulsifier. Pigs fed the diet containing gum-arabic had plasma C8 concentrations that were 3.5 times lower 2 hours post feeding than the pigs fed Tween-80. Plasma concentrations from Tween-80 fed pigs also



contained greater C10 concentrations both 1 and 2 hours post feeding compared to gum-arabic and non-emulsified fed pigs. In the second experiment, Tween-80 once again resulted in higher plasma concentrations compared to gum-arabic, lecithin or non-emulsified fed pigs. During the experiments conducted by Wieland et al. (1993), Tween-80 was more effective at increasing plasma concentrations. Therefore, suggesting that Tween-80 is a better emulsifier compared to soy-lecithin or gum-arabic (Wieland et al., 1993).

Xing et al. (2004) examined the effects of an added emulsifier (lysolecithin) on growth performance of weanling (21d) pigs over a 35 day trial. Their fat source for the trial was lard. Fat was added at either 0% or 5% of the diet and lysolecithin was added at 0.02% or 0.01% of the lard. The diet containing 5% lard and no added lysolecithin had the lowest final body weight and average daily gains (ADG) for the entire length of the trial. The pigs fed 0.01% lysolecithin + 5% lard were the heaviest at the end of the trial followed closely by the pigs fed the diet with no added fat. Pigs fed the 0.01% and 0.02% lysolecithin diets were 12.6% and 10.1% heavier at the end of the trial than the 5% lard + no emulsifier fed pigs. During the first half of the trial (d0-10), the 0.02% lysolecithin + 5% lard fed pigs had a higher fat digestibility of 75.9% while the 5% pigs fed the lard diet had a fat digestibility of 68.6%. The piglets fed the 0.01% lysolecithin + 5% lard diet had the lowest fat digestibility (64.6%). However, during the second half of the study the digestibility of the fat in the pigs fed 5% lard with no added lysolecithin and pigs fed the 0.02% lysolecithin + 5% lard were not significantly different (77.6 and 77.7, respectively). The 0.01% lysolecithin + 5% lard fed pigs had the lowest fat digestibility of 73.9%. Added emulsification increased digestibility for the first phase of the trial when added at 0.02% while piglet performance was improved for the entire length of the study when lysolecithin was added at 0.01% of the diet (Xing et al., 2004).

In a series of experiments by Jones et al. (1992) the effect of different emulsifiers on weanling pig growth were examined. In the first experiment, pigs were weaned at 17 days and placed on an experimental diet. The experimental fat was added at 10% of the diet. Emulsifiers were added at 10% of the total fat during the first half of the trial and at 5% of the total fat for the second half of the trial. Four fat sources were examined during this trial: soybean oil, coconut oil, tallow, and lard, while two emulsifying agents were studied: lecithin and lysolecithin. The digestibility of the soybean oil tended to increase when either emulsifying agent was included in the diet, however, a greater increase in digestibility was observed when lecithin was included in the diet as the emulsifier. The addition of an emulsifier did not affect the digestibility of the coconut oil. The greatest improvement in digestibility was observed when lecithin was added to the diet containing tallow. The digestibility increased from 80.9% in the diet with only tallow to 88.4% when lecithin was included in the diet. The digestibility of the fat increased slightly, by 3 units, when lysolecithin was included in the tallow diet. Lard was the only fat source that showed a decrease in digestibility when either emulsifier was added which is contrary to what Xing et al. (2004) observed. In a second experiment by Jones et al. (1992) piglets were weaned at 21 days of age onto experimental diets. Soybean oil and tallow were the two fat sources studied and fat was included at 10% and 5% of the diet during phases 1 and 2, respectively. Lecithin and lysolecithin were added to the tallow diets at 5%, 10% and 30% of the fat. A decrease in digestibility was observed when lysolecithin was included in the diet while lecithin tended to increase the digestibility of the fat. Fat digestibility increased by the 3 units when lecithin was included in the diet at any level. The addition of 5% lecithin tended to increase ADG for the entire 35 day trial. Growth performance decreased when lecithin was included in the diet at 5% or more. Digestibility also decreased when any level of lysolecithin was included

in the diet. Only during the last phase of the trial did ADG increase when lecithin or lysolecithin were included at levels above 5%. Pigs fed the diets with added emulsifiers tended to have greater ADG than those fed tallow but they also ate more causing the gain:feed ratios to be similar among those treatments (Jones et al., 1992). Gain:feed during of the pigs fed the tallow with added emulsifiers was around 0.71 overall (Jones et al., 1992) while Xing et al. (2004) reported a gain:feed ratio of 0.55 when lard was the experimental fat source and an emulsifier was included in the diet.

Overland et al. (1993) performed several experiments comparing the effects of lecithin and soy oil on growth performance and digestibility. Diets contained 0% or 2% of fat as lecithin or 0% or 6% soy oil for the first experiment. The digestibility of the fat was increased when lecithin or soy oil were added to the diet; however, lecithin did not improve the digestibility of the diets containing soy oil. In a second experiment studying the growth performance of lecithin and soy oil, ADG and ADFI were similar among treatment groups during the first week of weaning. The piglets on both 2% lecithin and 6% soy oil had a better gain:feed ratio for the second week of the trial and for the entire 35 day trial followed closely by the piglets fed 5% soy oil and no added lecithin. Piglets fed the diet containing 2% added lecithin and no soy oil performed better than those fed 0% lecithin or soy oil. Even though gain:feed was increased when the piglets were fed soy oil, lecithin or a mixture of both the final weight gains were not significantly affected (Overland et al., 1993).

In summary, addition of emulsifiers has been shown to have both positive and negative effects on both growth performance and digestibility. Continued research involving emulsifiers may prove beneficial in the effort to improve growth performance of newly weaned piglets.

### *Spray drying (encapsulation)*

Spray drying, encapsulation, of fat is a relatively new processing technique that involves encapsulating the fat in milk proteins (Keogh et al. 1999). The theory is that by spray drying the fat, the fats' physical structure may be altered which could aid in the digestion of the fat (Xing et al., 2004). Encapsulation of the fat also improves the handling and flow capabilities of the diet (Keogh et al., 1999). An increase in the digestion and absorption of fat may be achieved by spray drying the fat included in weanling pig diets (Xing et al., 2004). Although fat encapsulation did not affect final body weight, differences in ADG and G:F were observed. During the first two weeks of the trial ADG was slightly lower in pigs fed encapsulated fat; however, ADFI was significantly lower resulting in an increase in feed efficiency. During the second phase of the trial (d15-28) ADG and G:F was increased in the pigs fed the encapsulated fat. They concluded that spray drying fat may be beneficial in nursery pigs especially during the earlier stages of weaning. Xing et al. (2004) also examined the pellet quality of the various diets and were able to conclude that the encapsulated fat was not negatively altered during pelleting nor was pellet quality adversely affected, therefore, suggesting that spray drying fat may be good alternative to traditional methods of adding fat to weanling pig diets (Xing et al., 2004).

### **Intestinal Development**

#### *Digestive enzyme development*

In a study by Lindemann et al. (1986) they concluded that the major dietary changes experienced by weanling pigs is more likely to cause a fluctuation in digestive enzyme levels rather than chronological age. During their study, digestive enzymes, pancreatic lipase, amylase, chymotrypsin and trypsin levels steadily increased with age up to weaning at 4 weeks of age. One and two weeks post weaning, enzyme levels remained lower then they had been prior to

weaning. Gastric lipase was the only enzyme that did not appear to be affected by weaning the pigs onto a dry diet. Gastric lipase experienced the greatest increase in activity between 2 and 3 weeks of age which was prior to weaning (Lindemann et al., 1986). A similar increase in enzymes was observed in an experiment conducted by Corring et al. (1978). Pigs in this experiment were offered creep feed at 10 days of age and were weaned at 8 weeks of age which also coincided with the end of the trial. Creep feed intake levels did not begin to increase until about 4 weeks of age. Corring et al. (1978) opted to wean pigs at a later age so they could study the development of pancreatic lipase which has been shown to be much slower (low levels until 35 days after birth) at development than other digestive enzymes (Kitts et al., 1956). Pancreatic lipase activity steadily increased between birth and 6 weeks of age with the greatest increase occurring between weeks 3 and 4 after birth. Pancreatic lipase activity peaked 6 weeks after birth and at 8 weeks enzyme activity level had decreased. Amylase activity increased between birth and 6 weeks of age. At 8 weeks of age the activity level of amylase was not significantly different from the activity level at 6 weeks of age. On the other hand, trypsin and chymotrypsin decreased from birth until 4 and 3 weeks of age, respectively, when both enzyme activity levels began to increase. Lindemann et al. (1986) observed a decrease in chymotrypsin and trypsin activity when pigs were weaned at 4 weeks of age. It appears that major changes in diet affect digestive enzyme levels and a recovery period for the enzyme levels to rebound is present.

### *Intestinal morphology*

Along with differences in digestive enzyme development, differences in intestinal morphology are observed in newly weaned pig. The weaning period is a very stressful time for piglets; they must adapt to changes in diet physical form and to major changes in the diet composition. The main energy source of the diet changes from high levels of fat and low levels

of carbohydrates to a low fat and high carbohydrate diet. Lactose is the main carbohydrate in milk and at weaning starch becomes the main carbohydrate in weanling pig diets (Maxwell and Carter 2001).

Diet physical form has been shown to have a major impact on villus height in newly weaned pigs with the greatest impact on intestinal morphology being in the duodenum and with less of an effect in the ileum (Zijlstra et al., 1996). Zijlstra et al. (1996) examined villi heights in pigs weaned at 21 days of age onto a liquid or dry diet. Morphology samples were collected at both 18 and 25 days of age. A larger duodenal villus height was observed in liquid fed pigs compared to pigs left on the sow (900  $\mu\text{m}$  v 500  $\mu\text{m}$ , respectively). An even greater difference in duodenal villi heights was observed in liquid fed pigs compared to pigs fed a dry starter diet (900  $\mu\text{m}$  v 400  $\mu\text{m}$ , respectively). A decrease in duodenal villus height was present in both dry fed and sow suckled pigs when comparing measurements taken at days 18 and 25. Jejunal morphology measurements showed a villus height just over 1000  $\mu\text{m}$  in liquid fed pigs, 800  $\mu\text{m}$  in sow suckled pigs and 600  $\mu\text{m}$  in dry fed pigs. Jejunal villi heights in the dry fed pigs remained lower than day 18 measurements (Zijlstra et al., 1996). The results from the trial by Zijlstra et al. (1996) agree with Kelly et al. (1991) that villus heights are reduced for 3 days post weaning.

Kelly et al. (1991) examined the development of the digestive system in pigs that were weaned at 14 days. They determined that within the first 3 days of weaning there was approximately a 25% decrease in mucosa protein content. Within that same time frame they reported a decrease in villous height and crypt depth in weaned pigs compared to pigs left on the sow. The pigs that were not sow reared were force-fed a comparable amount of feed as the piglets left on the sow to eliminate any morphological changes in the intestinal mucosa due to a

lack of nutrition. Therefore, concluding that a reduction in feed intake at weaning causes a decrease in villus height (Kelly et al. 1991). However, Hampson (1986) hypothesized that an in vivo reduction in digestive capacity might explain why there is a low feed intake during weaning. Hampson determined that when there is a reduction in villi height there is also a decrease in amino acid transport by the villi (Hampson 1986).

Integrity of the intestinal mucosa is maintained via the epithelial cells which derive a portion of their energy from glucose (Mallet et al. 1986). Including glucose as the main energy source in weanling diets could result in increased feed intakes and enhance intestinal morphology. However, lactose is the main energy substrate in suckling pigs (Darragh & Moughan 1998) and liquid fed pig diets. A high lactose diet requires the enzyme lactase to cleave the lactose (occurs in the brush border membrane) to allow glucose and galactose to be absorbed by the enterocytes. Spreeuwenberg et al. (2001) was able to show that high lactose (lower protein) in the diet resulted in greater villus heights.

Spreeuwenberg et al. (2003) examined the effects of including glucose, lactose or starch as the main energy source and their effect on the morphology of the small intestine. They saw no significant differences in mucosal morphology between the diets. They observed a similar decrease in villus heights lasting for 3 days post weaning as Kelly et al. (1991). Although their results did not show one energy source being able to lessen the reduction in intestinal morphology at weaning they were able to conclude that the piglets fed the lactose diet seemed to recover more quickly than the piglets fed the glucose or starch diet (Spreeuwenberg et al., 2003).

## **Digestibility Marker**

### *Co-Edta vs C-36*

A good marker should closely follow the material of interest through the digestive tract without being absorbed. Chromic oxide and Cobalt-EDTA are two commonly used digestibility markers, however; they are not thought to closely follow fat during its digestion (Carlson and Bayley 1968). Liquid based diets require the use of a soluble marker such as Co-EDTA. Therefore, researchers began investigating the use of alkanes as fat digestibility markers due to their lipid like qualities. Alkanes are long chain hydrocarbons that are found naturally in plants and are indigestible in monogastrics (Choct & van Barneveld 1991). Mayes et al. (1986), studied C27 and C32, from plants, and reported an increase in the recovery of alkanes in the feces as the chain length of the alkane increased. However, as the length of the hydrocarbon exceeds C36 a decrease in fecal recovery has been observed in goats (Giraldez et al., 2006). The alkanes studied in the previous two experiments were fed in their natural plant form; however, feeding pig's the leaves and stems of plants is not a viable option. C36 and C34 are two commercially available alkanes that are water soluble making them ideal for use in pig fat digestibility experiments. A comparison between C36 and chromic oxide recovery in both ileal and fecal samples of pigs showed an almost perfect relationship between the two markers (Choct & van Barneveld). The results of this trial can pave the way for the use of C36 as a fat digestibility maker.



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CHAPTER II  
IMPROVING FAT UTILIZATION BY THE WEANED PIG: EFFECT OF DIET PHYSICAL  
FORM, FATTY ACID CHAIN LENGTH,  
AND EMULSIFICATION

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## INTRODUCTION

Researchers over the years have investigated many ways to improve post weaning growth performance in newly weaned piglets. Suckling pigs are capable of efficiently digesting milkfat, achieving digestibilities as high as 97% (Lucas and Lodge 1961), while fat digestibility by newly weaned pigs is approximately 85% (Cera et al., 1988, Hamilton and McDonald 1969). Typical weanling pig diets are formulated to contain a long-chain fat source which has been shown to be less digestible than medium chain fats (Cera et al., 1989). Additionally, fat in weanling pig diets is generally included at low levels (~5%) (Maxwell and Carter 2001) with little to no improvement observed in growth performance when included at higher levels (Peo et al. 1957, Allee et al. 1971).

Some methods investigated to improve fat utilization and growth performance of newly weanling pigs includes examining the effects of diet physical form, dietary fat source, added emulsifiers or including a spray dried fat. Liquid feeding of suckling piglets (Harrell et al., 1993) or newly weaned piglets (Zijlstra et al., 1996) has demonstrated substantial improvements in growth performance compared to sow-reared and dry-fed piglets, respectively. The use of MCT in a suckling pig diet results in a markedly greater rate of utilization compared to a diet containing LCT (Odle 1997); therefore, one could hypothesize that feeding a diet high in MCT would result in better growth performance immediately after weaning. Including an emulsifier in the diets of weanling pigs has had inconsistent results. Tween-80 has been shown to be a more effective emulsifier over soy-lecithin or gum-arabic (Wieland et al., 1993).

To our knowledge, this is the first study to utilize a complex (casein as protein source) diet (liquid or dry form) containing spray-dried fat (MCT or LCT) with or without an added

emulsifier in an attempt to improve fat utilization and growth performance of newly weaned pigs.

## **MATERIALS and METHODS**

### *Protocol*

All animal care and handling procedures were approved by the Institutional Animal Care and Use Committee of North Carolina State University. Ninety-six crossbred ([Landrace x Yorkshire] x [Duroc]) pigs were weaned at  $20 \pm 0.30$  d of age ( $6.8 \pm 0.04$  kg) and blocked according to body weight and ancestry. Piglets were assigned to one of eight treatment groups (Figure 1) to determine the effects of diet physical form, fatty acid chain length and emulsification on fat utilization in the young pig. Treatment groups were arranged in a 2 x 2 x 2 factorial design. Diets were fed in either a liquid or dry form with or without the inclusion of an emulsified or non emulsified fat (MCT or LCT). All pigs were housed within a conventional, raised-deck hot nursery (29.2°C) at the North Carolina State University Swine Educational Unit and arranged two pigs per pen (0.91 m by 1.52 m). Piglets received 14 h of light and 10 h of dark each day. The experiment was conducted in two replicates of 48 pigs each using 24 pens for each trial.

Diets were formulated (Table 1) to exceed pig nutrient requirements (NRC, 1998). Experimental fat source comprised 12% of the diets and was provided by either choice white grease (LCT; Milk Specialties, Inc., Dundee, IL) or Aldo-MCT (Lonza, Inc., Fairlawn, NJ). The analyzed fatty acid content of the MCT was predominantly octanoic and decanoic acids; whereas the LCT contained a mixture of long-chain fatty acids (Table 2). For treatments containing emulsifier, Tween 80 (Sigma Chemical Co., St. Louis MO) was added at 2% of the diet, displacing the fat source. We estimated this level of emulsifier to be sufficient to completely



encapsulate the supplemental fat with a coarse emulsion particle size of approximately 5000Å. Pigs were given ad libitum access to food and water. The experimental diets were fed either in dry form (mash) or were mixed with water to form the liquid diet. Liquid diets were reconstituted with water to 13% dry matter and fed in liquid feeders (Kane Manufacturing Company Inc., Des Moines, IA). Tetracosane, C36, was added to the diets as the main digestibility marker (Spectrum, Gardena, CA, Catalog # T2241-ZZ). Co-EDTA (Sigma Aldrich St. Louis, MO) was also added to a portion of the diets in order to serve as a comparison to the C36 marker. Co-EDTA was prepared by the procedure described by Uden et al. (1980). The C36 was blended with the fat sources and emulsifier prior to spray drying and the Co-EDTA was incorporated as the dry ingredients were mixed.

The growth trial was two weeks in duration and body weights were recorded on days 0, 2, 4, 6, 7, 10, 13, and 14. Pigs fed the liquid diet were fed each morning while dry-fed pigs were fed as needed. Feed amounts and feed weigh back amounts were recorded at each feeding. Liquid feeders were cleaned each morning while dry feeders were cleaned on piglet weigh days.

#### *Sample Collections & Laboratory Analyses*

One pig per pen was euthanized at day 7 while the second pig was euthanized at day 14 of the trial. On each euthanasia day body weights, blood samples, bile, ileal digesta, jejunal digesta, ileal morphology and jejunal morphology samples were collected.

Blood was collected in a heparinized vacuum tube and kept on ice until it was centrifuged (same day as collection) and the plasma was stored at -80°C. Plasma ketone body analysis (Kientsch-Engel 1982 and 1985 and Tetrick et al., 1995) was performed by first preparing a standard curve to ensure proper function of the enzyme and the assay. The standard curve was obtained by using known amounts of β-hydroxybutyric acid in place of plasma samples

(Appendix Figure 1). Plasma samples and reagents were kept on ice during the assay. Thawed plasma samples were neutralized prior to performing the assay by adding equal amounts of plasma and 10% perchloric acid to a tube. The solution was then centrifuged (Sorvall RC-5B Refrigerated Superspeed Centrifuge, DuPont Instruments, Wilmington, DE) for 10 min at 5000 RPM/min (2516 x g). The supernatant was decanted and adjusted to a pH of 7 by using potassium hydrogen carbonate. Once a pH of 7 was achieved, the samples were centrifuged again, and the supernatant was decanted into a clean tube. Samples were chilled on ice for 15 min and then transferred to a 56°C water bath for 15 min. One milliliter micro-cuvettes with a 1 mm light path were used and the spectrophotometer (DU-640, Beckman Instruments, Fullerton, CA) was set to read at 546 nm for a total of 40 min at 5 minute intervals. The assay was performed by adding 40 µl of Reagent 1 (0.025 M NAD in phosphate buffer saline solution, PBS pH 8.5), 40 µl of Reagent 2 (0.0084M FeCl<sub>3</sub> in distilled water then 0.022 M BPS added to FeCl<sub>3</sub> solution), 300 µl of plasma (or 300 µl of neutralized perchloric acid for the blank) and 20 µl of Reagent 3 (0.0098 M PMS in distilled water then 10 µl were diluted with 1.55 ml of distilled water) into the cuvette. The solution in the cuvette was stirred and the spectrophotometer was started. After 10 min a constant drift was achieved and 15 µl of β-hydroxybutyrate dehydrogenase was added to the cuvette and mixed. The enzyme was derived from *Pseudomonas lemoignei* in lyophilized powder form (Sigma Chemical Co., St. Louis MO) and reconstituted with 1.5 ml of water. Thirty min after adding the enzyme the reaction was complete and a constant drift was achieved. Change in absorbance was calculated by:  $(A_2 - A_1)_{\text{sample}} - (A_2 - A_1)_{\text{blank}}$ .  $A_2$  is the absorbance at 40 minutes and  $A_1$  is the absorbance at 10 min.

Ileal and jejunal digesta samples were collected into 50 ml conical tubes and stored at -20°C until analysis. Ileal digesta samples were freeze dried (Heto Power Dry LL300,

Appropriate Technical Resources, LLC, Laurel, MD) to a fine powder in order to extract the lipid from the samples. Samples were subsequently assayed for total lipid content and for fatty acid profiles. Total lipid was determined using the Folch fat extraction method (Folch et al., 1957) and was modified as follows. Dry Ileal digesta samples were mixed and then 0.85 g of sample was weighed out into a 50 ml glass test tube. Next, 0.15  $\mu$ l of 12 N hydrochloric acid was added to prevent soap formation and then 20 ml of 2:1 chloroform:methanol was added to the test tube and homogenized for 1 minute. The homogenized solution was then poured through a funnel using Whatman Paper 4 into another 50 ml glass test tube. The original 50 ml test tube was washed with an additional 5 ml of the Chloroform:Methanol mixture, vortexed and poured through the funnel as well. Five milliliters of 0.9% saline was added to the filtrate which was then capped and vortexed until the filtrate became a homogeneous solution. Next, the samples were either held at room temperature for 20 min or were centrifuged for 10 min at 830 x g (4°C) to allow for separation of the lipid (organic) phase. The lower phase was transferred into a 25 ml glass test tube that had been baked (at 104°C for 2 hours then placed in a desiccator to cool) and weighed. The sample was dried under nitrogen gas and then baked at 104°C for 2 hours. After 2 hours the samples were placed into a desiccator to cool and then the test tubes were weighed again. Total fat was calculated by subtracting the weight of the clean test tube from that same test tube containing the extracted fat. The dried test tubes containing the lipid were filled with nitrogen gas, capped and stored at 4°C until further analysis.

Fatty acid methylation was performed in part by the method described by Gatlin et al. (2002). The method to analyze C36 by gas chromatography (GC) was developed by the Odle Laboratory for this experiment. Prior to methylation, 10  $\mu$ l of internal standard (100 mg C17:0 dissolved in 20 ml of ethanol) was added to each sample. The GC column (Phenomenex,

Torrance, CA; Zebron # ZB-5HT) measured 30 m in length with a 0.25 mm I.D., 0.1  $\mu$ m film thickness and maximum temperature rating of 430°C. Analysis was performed by using an HP-5890 gas chromatograph (Agilent Technologies, Santa Clara, CA) with a flame ionization detector (FID). The oven temperature was programmed to progress from 50°C for 1 min to 210°C at 15°C per min. After 3 min the temperature was increased further to 360°C at 15°C per min. All of the samples and standards were chromatographed using a 50:1 split ratio. The injection temperature was 260°C and the FID temperature was set to 400°C.

Cobalt analysis of diet and digesta samples, for apparent ileal fatty acid digestibility calculations, was performed by the Analytical Services Laboratory on the NCSU campus. A dry ashing procedure was used to determine the amount of cobalt present in the samples (AOAC, 1990). Apparent ileal fatty acid digestibilities were calculated based on the marker-ratio method previously described by Jagger et al. (1992) and Gatlin et al. (2005).

For ileal and jejunal morphology measurements, a 1-inch section of fresh tissue was fixed in formalin. After 24 h, the formalin was replaced with 70% ethanol for storage of the sections. For histology, each sample was sliced by hand in 2 mm wide sections with a razor blade and placed into a histology cassette and transported to the NCSU Veterinary School Histology laboratory for Hemotoxin/Eosin (H/E) staining. Two sections per sample were placed onto one slide. Slides were read using an Olympus Vanox-S Microscope (Olympus Corporation, Lake Success, NY). Readings were taken at three random locations for villus height, villus width (at half of height) and crypt depth. From the measurements, villus area and the villus height to crypt depth ratio were calculated (Corl et al., 2007 and Argenzio et al., 1990).

### *Statistical Analysis*

Data were analyzed using the GLM procedure of SAS according to a 2x2x2 factorial randomized complete block design with pen as the experimental unit. For intestinal morphology measurements, days post weaning (7 versus 14) was modeled as an additional independent variable. Differences were considered significant when  $P < 0.05$ .

## **RESULTS**

### ***Animal Performance***

Initial piglet body weights were similar among treatments (Figure 2, Appendix table 1). Inclusion of the dietary emulsifier did not affect growth performance ( $P > 0.1$ ; Appendix table 1), so data in figures 2-5 were averaged across this effect. Liquid-fed pigs did not lose weight after weaning; whereas, piglets fed the dry diet lost weight immediately postweaning (Figure 2). By the fourth day of the trial, the body weight of the pigs fed the dry diet had rebounded and their weight had surpassed their day 0 body weight. Beginning on day 2 and continuing to the end of the trial, the liquid-fed pigs were heavier than the dry-fed pigs ( $P < 0.05$ ). Overall, on day 14, the dry-fed pigs (11.45 kg) weight approximately 2 kg less than the liquid-fed pigs (13.61 kg), regardless of triglyceride chain length. During the second week of the trial, LCT-fed pigs were heavier than the MCT-fed pigs ( $P < 0.05$ ) such that on day 14, pigs fed LCT had a final body weight of 13.1kg while those fed MCT weighed 11.96 kg (Figure 2). Accordingly, pigs fed the liquid LCT (14.37 kg) diet were heaviest at the end of the trial. Furthermore, pigs fed the liquid MCT diet (12.85 kg) were heavier than those fed dry LCT diet (11.84kg), and pigs fed dry MCT diet (11.06kg) were lightest.

Average daily gain (ADG; Figure 3; Appendix Table 1) was affected ( $P < 0.05$ ) by diet physical form for the entire length of the trial, with liquid-fed pigs gaining 44% faster than dry-

fed pigs (0.49 kg/d vs 0.34 kg/d, respectively). Pigs fed LCT also gained faster than pigs fed MCT ( $P < 0.05$ ) during the first week and overall, but not during the second week. Gains were 23% higher for the LCT-fed pigs compared to the MCT fed pigs when the data were averaged across all 14 days of the trial (0.46 kg/d vs 0.37 kg/d, respectively). Total body weight gains were highest for the piglets fed the liquid LCT diet followed by those fed the liquid MCT diet, dry LCT diet and was lowest for piglets fed the dry MCT diet.

Average daily feed intake (ADFI) was calculated on a dry matter (DM) basis. ADFI was significantly ( $P < 0.05$ ) affected by chain length for the entire trial with LCT fed pigs eating more than MCT fed pigs (Figure 4; Appendix Table 1). During the first week of the trial, piglets ate only 0.29 kg/d of the LCT diet and 0.25 kg/d of the MCT diet. The greatest difference in feed intake was observed during the second week of the trial when LCT-fed pigs consumed 42% more feed than the MCT-fed pigs (0.64 kg/d vs 0.45 kg/d, respectively). Overall, ADFI was increased by 21% when pigs were fed the LCT diet versus the MCT diet (0.40 kg/d vs 0.33 kg/d, respectively). Diet physical form affected ( $P < 0.05$ , liquid  $>$  dry) ADFI during the second week of the trial and when averaged over the entire 14 day trial. Piglets consumed roughly the same amount of feed during the first week of the trial. During the second week of the trial, piglets fed the liquid diets consumed 17% more feed than those fed the dry diets (0.62 kg/d vs 0.53 kg/d, respectively). For the ADFI over the entire 14 day trial, pigs fed the liquid diets consumed 0.39 kg of diet per day while those on the dry diet consumed 0.34 kg/d (15% more liquid vs dry).

Feed efficiency (G:F, Figure 5; Appendix table 1) during the first week postweaning was greater for pigs fed the liquid diets (diet form effect,  $P < 0.05$ ) and was greater for pigs fed LCT than MCT diets (chain length effect,  $P < 0.05$ ). Efficiency was greatest for pigs fed the liquid diet containing LCT with a value of 1.36. During the second week and for the overall trial, the

liquid-fed pigs continued to display superior efficiency ( $P < 0.05$ ). Overall, gain:feed was improved by 29% ( $P < 0.05$ ) in the liquid fed pigs compared to the dry fed pigs (1.26 v 0.98, respectively).

### ***Digestibility Marker***

Two inert digestibility markers were compared; one being a lipophilic marker (an alkane, C-36) and the other a mineral-based marker (Co-EDTA). The use of Co-EDTA resulted in a statistically ( $P < 0.05$ ) greater digestibility for all fatty acids examined, except for C-8 (Figure 6). Based on these findings, C-36 was used to compute fat digestibilities for this experiment.

### ***Fat Digestibility***

Ileal fat digestibility coefficients were calculated from samples obtained on day 7 and 14 using the C-36 alkane as a fat digestibility marker. Diet physical form had no detectable effect on fat digestibility at both 7 and 14 days post-weaning ( $P > 0.21$ ; Figure 7, Figure 8; Appendix Table 2). On days 7 and 14 post-weaning, the digestibility for total fat was highest for the MCT diet compared to the LCT diet ( $P < 0.001$ , 98.42% vs 93.39%, respectively). An interaction between chain length and emulsification was detected ( $P < 0.03$ ) for the fatty acids constituting the largest portion of the diet (except C 8:0) and for total fat digestibility. Specifically, addition of the Tween-80 emulsifier increased the digestibility of the long chain saturated fatty acids, but had no effect or reduced the digestibilities of the medium chain fatty acids. The greatest impact of added emulsification was observed at 14 days post-weaning in stearic acid (C 18:0) when the addition of Tween-80 significantly increased FA digestibility over non-emulsified LCT diet ( $P < 0.001$ , 82% vs 69%, respectively).

### ***Small Intestine Morphology***

Table 3 shows the effect of diet physical form, fatty acid chain length and emulsification on jejunal morphology. The villus height, width, crypt depth and the area of the jejunum increased as the pigs aged ( $P < 0.05$ ). Diet physical form tended to affect crypt depth ( $P < 0.10$ , liquid > dry). A chain length by age interaction was observed in jejunum villus width ( $P < 0.05$ ) and a trend was observed in the jejunum area ( $P < 0.10$ ).

Table 4 shows the effect of diet physical form, fatty acid chain length and emulsification on morphology of the ileal mucosa. Ileal height, villus to crypt ratio and the area of the villus all increased as the piglets aged ( $P < 0.05$ ). Villus width in the ileum also tended to be larger for the pigs at 14 days of age compared to 7 days of age ( $P < 0.10$ ).

### ***Plasma Ketone Body Concentration***

Pigs fed diets containing MCT had significantly greater circulating levels of  $\beta$ -hydroxybutyrate in their plasma compared to the pigs fed the LCT diets ( $P < 0.0001$ ) (Figure 8). Pigs fed the dry MCT diet had an elevated  $\beta$ -hydroxybutyrate concentration than liquid MCT fed pigs ( $P < 0.05$ ). However, this effect was reversed in the LCT fed pigs wherein liquid LCT fed pigs had significantly greater concentrations of plasma ketone body levels than dry LCT fed pigs ( $P < 0.05$ ).

## **DISCUSSION**

### ***Growth Performance***

As expected, diet physical form had a significant impact on weanling piglet growth performance with liquid outperforming dry fed pigs (Zijlstra et al., 1996). A decrease in body weight is commonly observed at weaning; however, in the liquid fed pigs there was no reduction in piglet body weights. The addition of an emulsifier to the diet did not significantly impact



growth performance. Piglets fed the LCT diet showed significant improvements in growth performance. Dietary fat source (medium vs long) is thought to be a major factor effecting post weaning growth performance. Fat in the current study was included at 12% of the diet. Eusebio et al. (1960) and Peo et al. (1957) conducted several weanling pig trials with fat levels ranging from 0 to 38% of the diet and pigs were weaned at an average age of 17.3 days and 7.9 days, respectively. In one of the experiments conducted by Eusebio et al. (1960) piglets weaned onto the diet containing soybean gained significantly less than pigs weaned onto the control diet (no fat) or the coconut oil diet. Piglets on the control diet with no added fat outperformed piglets weaned onto a diet containing dietary fat. In another experiment by Eusebio et al. (1957) piglets that were weaned onto diets containing lard had significantly lower body weights than piglets on a tallow, soybean or control (no fat) diet. Peo et al. (1957) and Eusebio et al. (1960) reported a decrease in body weight gain when fat was included above 5% in the diet. On the other hand, Allee et al (1971) and Sewell and Miller (1965) reported that dietary fat levels as high as 13% did not significantly affect body weights in piglets weaned at 14 days and 22.7 days, respectively. In the experiment conducted by Allee et al. (1971) the dietary fat source was corn oil and average daily gain was not significantly affected by the level of fat inclusion. Sewell and Miller (1965) used diets containing corn oil, lard and beef tallow. Piglets fed the corn oil diet outperformed those on the control (no fat), tallow or lard diet when fat was included at 8% of the diet. Oliver et al. (2005) reported that including fat (mixture of tallow and lard) at either 2% or 25% of the diet did not significantly affect body weight gains in pigs weaned at 10 days of age. In the current experiment ADG and ADFI were significantly increased in the pigs fed the liquid LCT diet. Additionally, G/F was increased in liquid LCT fed pigs over all other treatment groups.

### *Digestibility Marker*

In a comparison between C36 and Co-EDTA, the fat digestibility was consistently higher in the measurements made using Co-EDTA. To our knowledge, this is the first experiment using C36 as a fat digestibility marker in weanling pigs. Past research has shown that C36 is 100% recovered in the feces when sprayed onto leaves and stems that were fed to goats (Giraldez et al., 2006) and close to 100% recovery in pigs (Choct & van Barneveld 1991). Co-EDTA is a mineral based water soluble marker that does not go through the same digestive processes as fat. Therefore, it has been suggested that the use of a marker with fat like qualities would be more accurate for performing fat digestibility studies (Ohajuruka and Palmquist, 1991).

### *Digestibility*

The digestibility of fat can be affected by many factors, such as fatty acid chain length, the use of an emulsifier or the degree of saturation of the fat. Previous research has shown that MCT are a more readily available source of energy and can be directly absorbed into the bloodstream unlike LCT (Odle 1997 and Greenberger et al., 1966). Therefore, as observed in the current experiment and in past research MCT have a much higher digestibility than LCT (Cera et al., 1988 and 1989). Typical weaning diets include fat at low levels and use a LCT source of fat. In the current experiment the digestibility of the emulsified LCT fat was improved over the non-emulsified LCT. Past research on including added emulsifiers in the diet has conflicting results. Tween-80 was shown to increase the digestibility of pure MCT in pigs compared to soy-lecithin or gum-arabic (Wieland et al., 1993). However, in other studies the digestibility of coconut oil was unaffected by lecithin or lysolecithin while lecithin improved the digestibility of tallow (Jones et al., 1992). Jones et al. (1992) observed a decrease in digestibility when lecithin or lysolecithin were included above 5% of the fat. The ratio of unsaturated (u) to saturated (s) fatty

acids can also affect the digestibility of fat. A U/S ratio under 1.3 has been shown to dramatically reduce the digestibility of the fat (Stahly 1984). During the current experiment the LCT fat source had a U/S ratio of 0.86. Digestibility of saturated LCFA were consistently greater in pigs fed the emulsified LCT diet. Therefore, emulsifying LCT fats appears to be beneficial in improving fat digestibility.

#### *Small Intestine Morphology*

Major dietary changes have been shown to negatively affect the enzymes of the digestive system in pigs (Lindermann et al., 1986 and Corring et al., 1978). Along with affecting enzyme production, changes in small intestine morphology have been observed (Kelly et al., 1991 and Zijlstra et al., 1996). A decrease in jejunal and ileal villus heights has been reported in newly weaned pigs (Kelly et al., 1991) and this effect is evident to at least 1 week post weaning (Zijlstra et al., 1996). Placing piglets on a liquid diet not only eliminates a depression in growth post weaning it also results in an increase in villi height compared to dry fed or sow reared piglets (Kelly et al., 1991 and Zijlstra et al., 1996). However, during our experiment diet physical form did not impact ( $P > 0.10$ ) jejunal or ileal villus height. Diet physical form only affected jejunal crypt depths with liquid fed pigs being greater than dry fed pigs ( $P < 0.10$ ). One possible explanation for the lack of effect could be that our complex diets containing spray dried fats was suffice in enhancing intestinal villus heights post weaning. Encapsulating the fat could emulsify the fat enough to improve post-weaning piglet performance with out the need for adding additional emulsifiers to the diet.

#### *Plasma Ketone Body Concentration*

Piglets have very low levels of circulating plasma ketone bodies. Ketone bodies are an available energy source when nutrient intake is non-existent although they are not normally a

source of energy in piglets. When needed, piglets can receive up to 40% of their energy requirements from ketone bodies (Tetrick et al., 1995). Circulating plasma ketone body concentrations were higher in MCT fed pigs than LCT fed pigs during the current experiment. During fat digestion, MCT are absorbed and transported to the liver more quickly than LCT resulting in a greater rate of MCFA being oxidized to ketone bodies than LCFA. The majority of MCFA are oxidized to ketone bodies while LCFA can either be oxidized to CO<sub>2</sub> or ketone bodies or LCFA can be stored in adipose. The rate of LCFA oxidation into CO<sub>2</sub> or ketone bodies is dependent on the animals nutritional state. If the animal is in a state of starvation more LCFA will be converted to ketone bodies, however, if the animal is in a fed state then the majority of LCFA will be stored or oxidized to CO<sub>2</sub> (Odle 1997 and Greenberger 1966).

#### *Spray Dried Fat*

All fats in this experiment were spray dried (encapsulated) which forms an emulsified fat that has better handling qualities. Encapsulating fat has been shown to increase ADG and G/F in nursery pigs (Xing et al., 2004). This new processing technique may emulsify the fat enough to improve growth performance of newly weaned pigs while they adapt to a traditional dry weaning diet. However, the current study did not directly study the effect of a spray dried fat versus a non spray dried fat on animal performance. All diets in the current study contained spray dried fats. Granted all the diets contained a spray dried fat, an improvement was observed in the digestibility of long chain saturated fatty acids when Tween-80 was included in the diet. Leading to the conclusion that the use of spray dried fats may improve performance without having to include an emulsifier in a diet containing a spray dried fat.

## **IMPLICATIONS**

Liquid feeding newly weaned pigs a diet containing LCT can increase daily gains by nearly 30%. Utilization of C36 as a digestibility marker may give a more accurate representation of fat digestibility due to its fat like qualities compared to a mineral based marker. Addition of tween-80 as an emulsifying agent further increased digestibility of LCT containing diets over non-emulsified LCT but not over the emulsified MCT diet. Emulsifying a highly saturated LCT diet can improve fat digestibility. However, spray drying the fats may provide enough emulsification to enhance piglet growth and fatty acid digestibility without the use of added emulsifiers. Further research into the use of spray dried fats is needed to resolve this issue.

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**Table 1.** Diet formulation and calculated composition.

Ingredients	% (as fed)
Experimental Fat Source (1-4) Choice white grease vs. MCT +/- emulsifier	11.86
Whey Protein (with lactose)	69.83
Modified Wheat Proteins (80%)	8.13
Plasma Protein (78%)	5.00
L-Lysine HCl	0.31
DL-Methionine	0.11
Limestone	0.75
Dicalcium Phosphate	0.55
Mineral Premix <sup>a</sup>	0.50
Vitamin Premix <sup>b</sup>	0.13
Antibiotics <sup>c</sup>	0.14
Other <sup>d</sup>	2.69
<b>Composition (calculated)</b>	
ME, Mcal/kg	3.70
Lactose, %	43.20
Crude protein, %	24.90
Crude fat, %	13.00
Crude fiber, %	0.10
Crude ash, %	8.34
Lysine, %	1.98
Calcium, %	0.91
Phosphorous, %	0.78

- a. Provided the following (g/kg premix): Choline, 60; Zn, 23.5; Fe, 20.0; K, 20.0; S, 16.3; Ca, 10.0; P, 5.5; Mn 5.0; Na, 2.8; Cu, 1.8; Mg, 1.0; I, 0.4; Cl, 0.4; Co, 0.2; Se, 0.06.
- b. Provided the following per kg premix: vitamin A, 33,000,000 IU; vitamin D3, 6,600,000 IU; vitamin E, 55,000 IU; vitamin K, 5.12g; vitamin C 117g; niacin, 33.07g; pantothenate, 29.98g; riboflavin, 8.38g; vitamin B6, 4.00g; folic acid, 2.76g; thiamin, 2.04g; biotin, 66mg; vitamin B12, 44mg.
- c. Provided the following per kg diet: oxytetracycline, 154 mg (d 0 to 14) and 165 mg (d 15 to 35); neomycin base, 154 mg (d 0 to 14) and 115.5 mg (d 15 to 35).
- d. Including citric acid, potassium sorbate, vitamin E, flavor additive, Fe, Zn, Mn, Chromic Oxide, flow agent (liquid diet only), bentonite (pellet diet only).

**Table 2.** Analyzed fatty acid profiles of MCT and LCT diets (% w/w of total fatty acids).

<b>Fatty Acid</b>	<b>MCT</b>	<b>LCT</b>
<b>8:0</b>	38.67	1.52
<b>10:0</b>	38.59	1.79
<b>12:0</b>	1.25	1.05
<b>14:0</b>	2.15	4.72
<b>14:1</b>	0.16	0.23
<b>16:0</b>	7.02	33.26
<b>16:1</b>	0.58	4.52
<b>18:0</b>	1.92	10.83
<b>18:1</b>	5.36	27.98
<b>18:2</b>	4.10	13.22
<b>Other</b>	0.18	0.88
<b>Total</b>	99.98	100.00
<b>Saturated</b>	89.60	53.17
<b>Unsaturated</b>	10.20	45.95
<b>Medium-chain</b>	80.84	9.30
<b>Long-chain</b>	18.99	89.82

**Table 3.** Effect of diet physical form, fatty acid chain length and emulsification on jejunal mucosal morphology.

	Day	Dry				Liquid				SEM
		MCT		LCT		MCT		LCT		
		+ Emul	- Emul	+ Emul	- Emul	+ Emul	- Emul	+ Emul	- Emul	
Height (um) <sup>a</sup>	7	523.13	510.67	412.68	608.29	436.35	370.84	509.71	551.61	74
	14	651.48	586.85	659.4	599.66	672.76	609.69	618.81	550.44	74
Width (um) <sup>a, d</sup>	7	140.9	128.82	138.48	149.73	137.37	144.09	162.89	155.11	12
	14	155.09	179.47	167.53	152.87	181.03	171.09	152.32	160.59	12
Crpyt Depth (um) <sup>a, c</sup>	7	239.26	260.67	268.29	260.41	292.65	256.76	260.49	295.22	26
	14	320.54	275.95	279.23	266.01	292.65	345.06	330.05	308.7	26
Villus:Crypt Ratio <sup>b</sup>	7	2.79	1.93	1.55	2.34	1.52	1.52	2.00	1.86	0.10
	14	2.03	2.16	2.59	2.35	2.38	1.82	1.93	1.77	0.10
Area (mm <sup>2</sup> ) <sup>a, e</sup>	7	0.47	0.50	0.43	0.62	0.41	0.39	0.59	0.6	0.30
	14	0.81	0.61	0.74	0.60	0.82	0.69	0.64	0.67	0.30

a, Day 14 > Day 7, P < 0.05

b, Day 14 > Day 7, P < 0.10

c, Form, Liquid > Dry, P < 0.10

d, Chain\*Day interaction, P < 0.05

e, Chain\*Day interaction, P < 0.10

**Table 4.** Effect of diet physical form, fatty acid chain length and emulsification on ileal mucosal morphology.

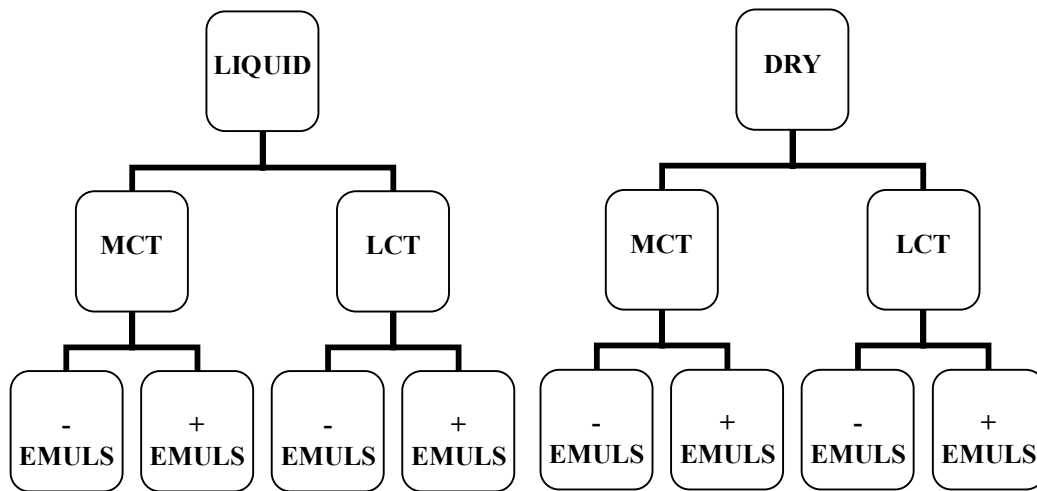
	Day	Dry				Liquid				SEM
		MCT		LCT		MCT		LCT		
		+ Emul	- Emul	+ Emul	- Emul	+ Emul	- Emul	+ Emul	- Emul	
Height <sup>a, c</sup>	7	361.17	260.61	379.54	310.61	369.3	312.60	329.39	330.62	36
(um)	14	390.21	354.33	401.48	397.46	392.28	403.76	429.40	391.16	36
Width <sup>b, d</sup>	7	113.96	109.45	112.23	145.84	117.74	128.85	136.6	128.5	16
(um)	14	142.96	124.58	130.5	170.86	117.19	150.06	124.94	143.1	16
Crpyt Depth	7	205.44	212.69	173.23	204.42	208.80	203.33	204.88	194.17	20
(um)	14	187.71	219.33	182.81	184.69	180.74	186.29	187.96	201.56	20
Villus:Crypt	7	1.79	1.28	2.23	1.58	1.77	1.57	1.67	2.08	0.29
Ratio <sup>a</sup>	14	2.11	1.88	2.29	2.24	2.38	2.25	2.31	2.08	0.29
Area <sup>a</sup>	7	0.2809	0.1923	0.2832	0.3213	0.3067	0.2775	0.3123	0.3015	0.06
(mm <sup>2</sup> )	14	0.3818	0.3288	0.3615	0.4802	0.3120	0.4178	0.3743	0.3838	0.06

a, Day 14 > Day 7, P < 0.05

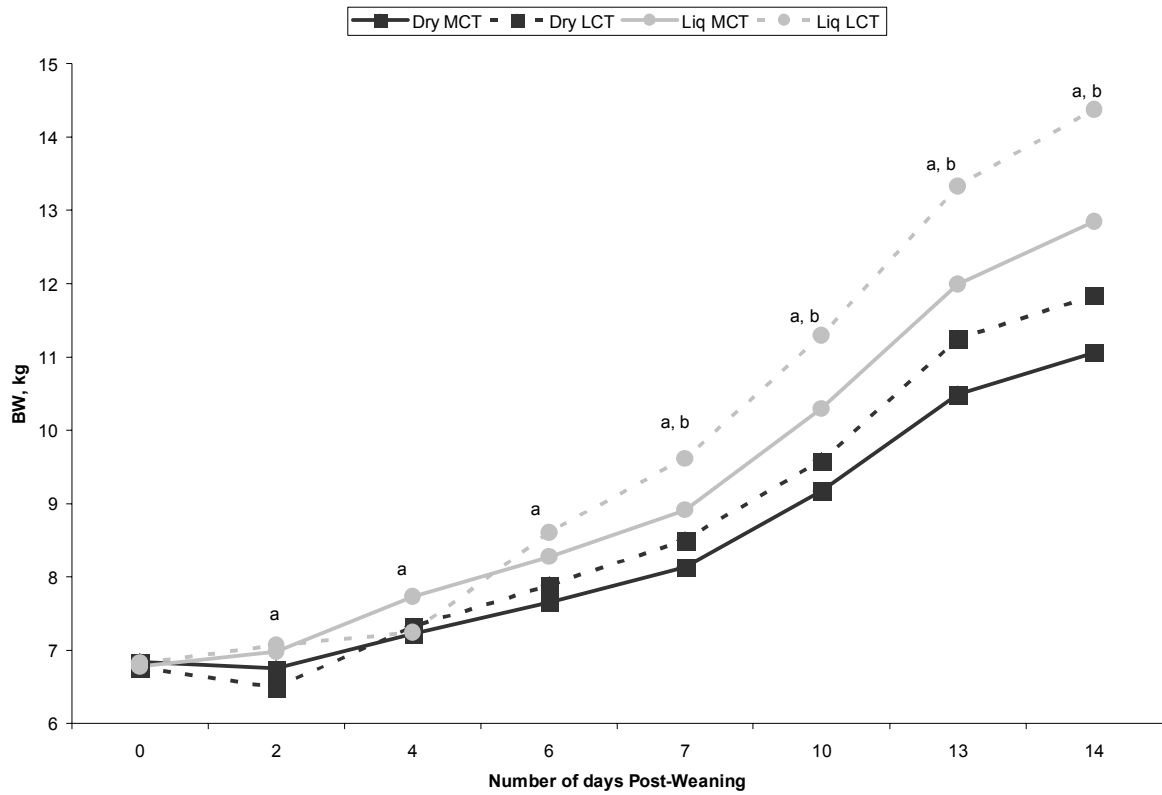
b, Day 14 > Day 7, P < 0.10

c, Emulsification main effect, P < 0.05

d, Emulsification main effect, P < 0.10

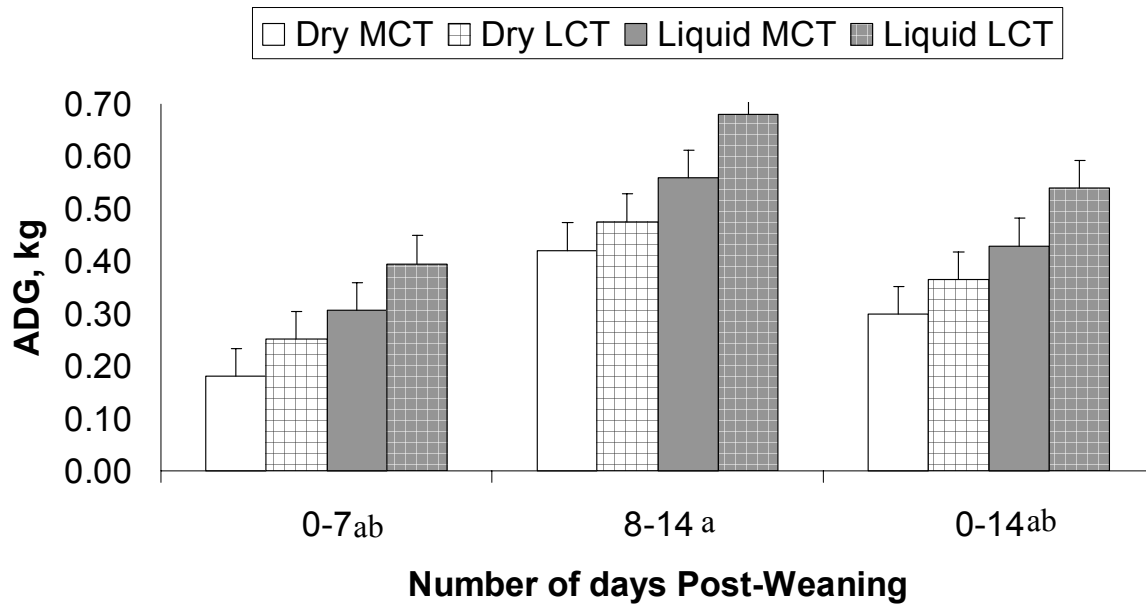


**Figure 1.** Schematic of dietary treatments in a 2x2x2 factorial design. Diet physical form (liquid vs dry), fatty acid chain length (medium chain triglyceride, MCT, vs long chain triglyceride, LCT) and with or without emulsification (Tween-80).



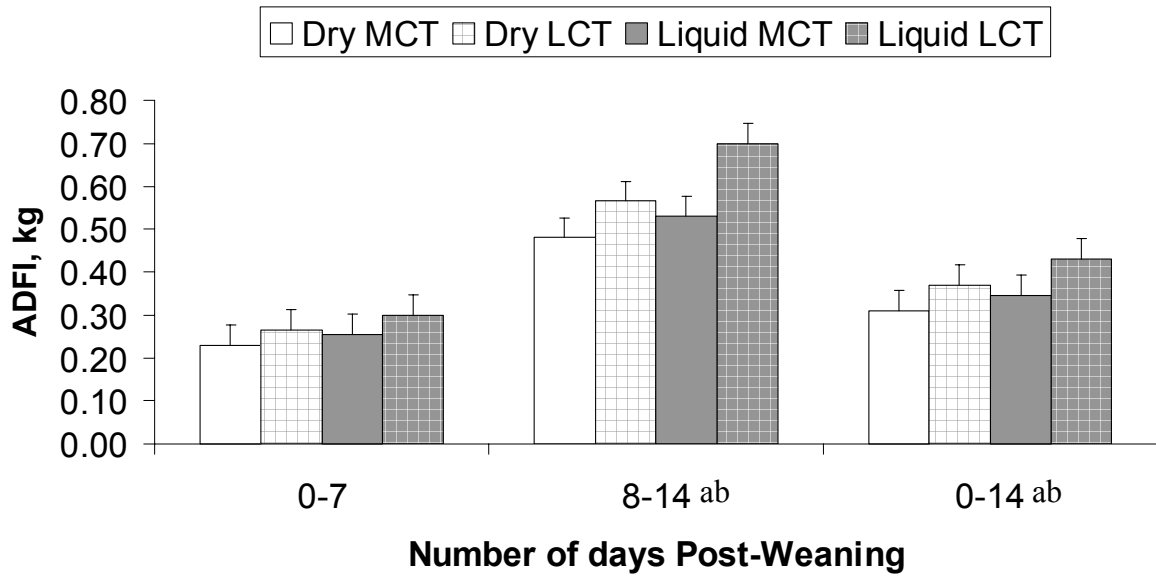
a, Form, Liquid > Dry,  $P < 0.05$       b, Chain, LCT > MCT,  $P < 0.05$

**Figure 2.** Effect of diet physical form and fatty acid chain length on growth performance of weanling pigs. Emulsification had no detectable effect.



a, Form, Liquid > Dry, P < 0.05      b, Chain, LCT > MCT, P < 0.05

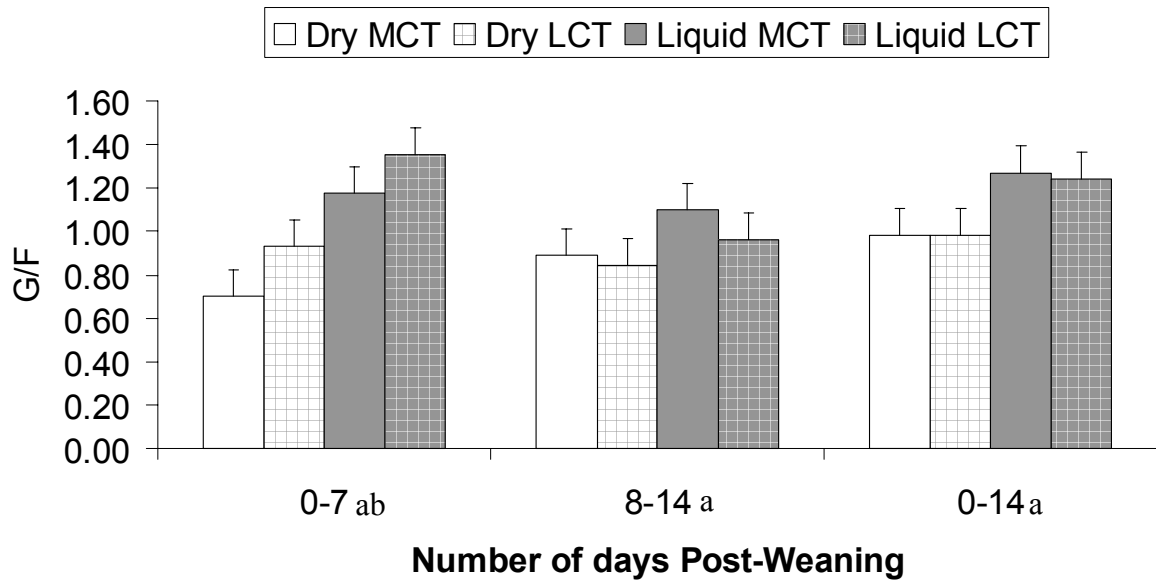
**Figure 3.** Effect of diet physical form and fatty acid chain length on ADG of weaned pigs. Emulsification had no detectable effect.



a, Form, Liquid > Dry,  $P < 0.05$       b, Chain, LCT > MCT,  $P < 0.05$

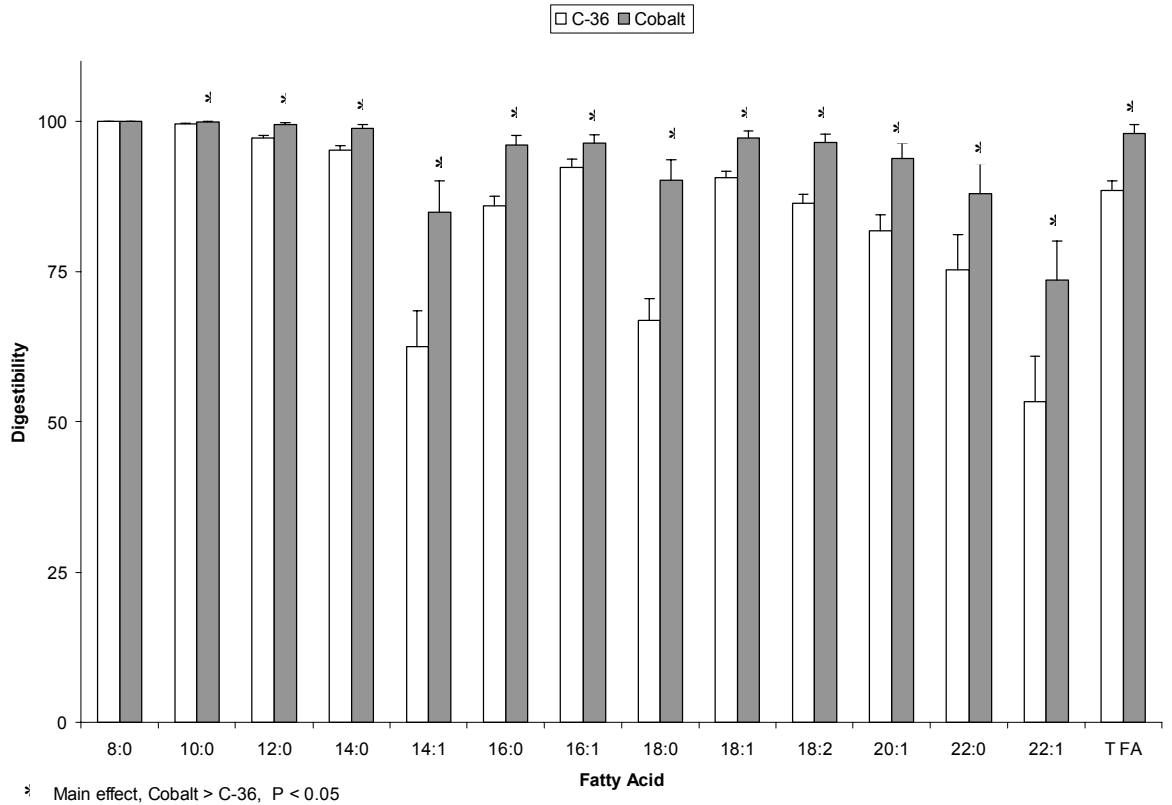
**Figure 4.** Effect of diet physical form and fatty acid chain length on ADFI of weaned pigs. Emulsification had no detectable effect.



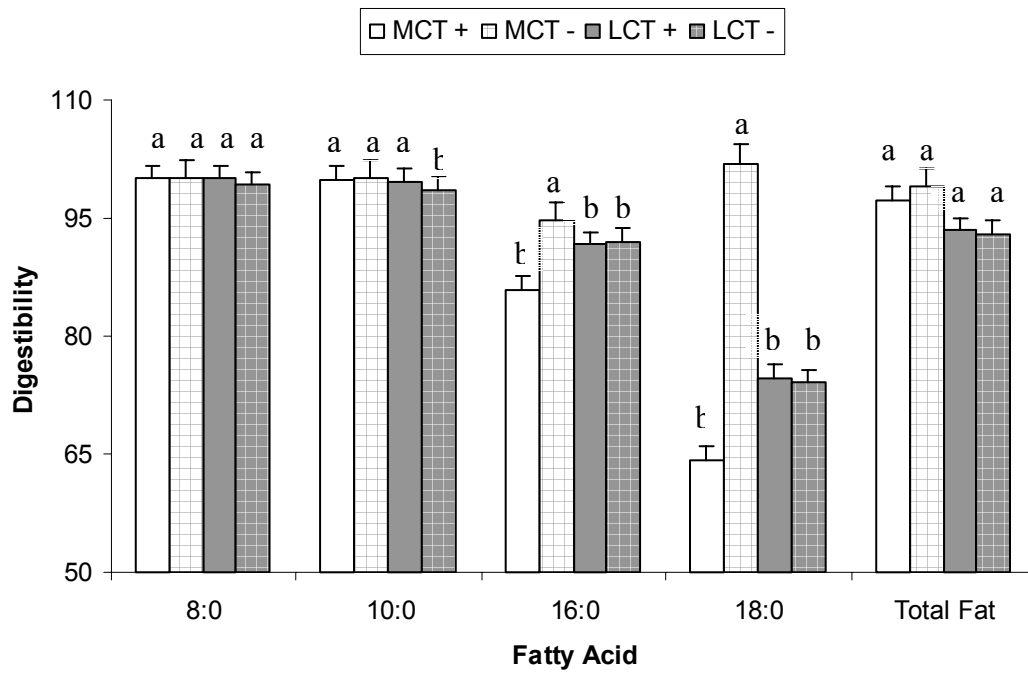


a, Form, Liquid > Dry,  $P < 0.05$       b, Chain, LCT > MCT,  $P < 0.05$

**Figure 5.** Effects of diet physical form and fatty acid chain length on G/F of weaned pigs. Emulsification had no detectable effect.

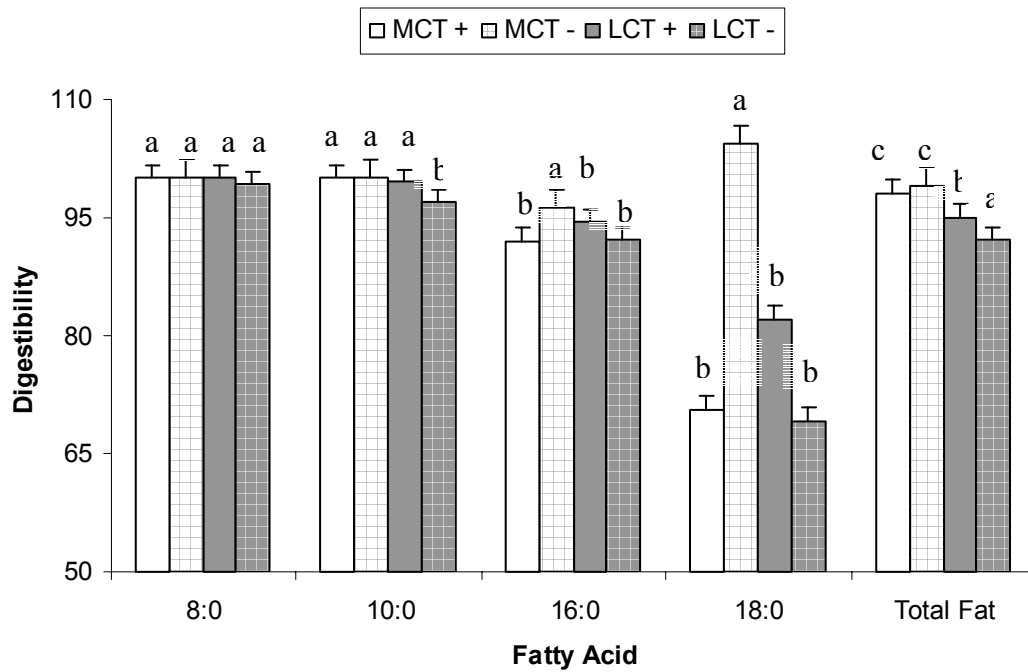


**Figure 6.** Effect of dietary marker on fatty acid digestibility. Comparison of Co-EDTA and C36.



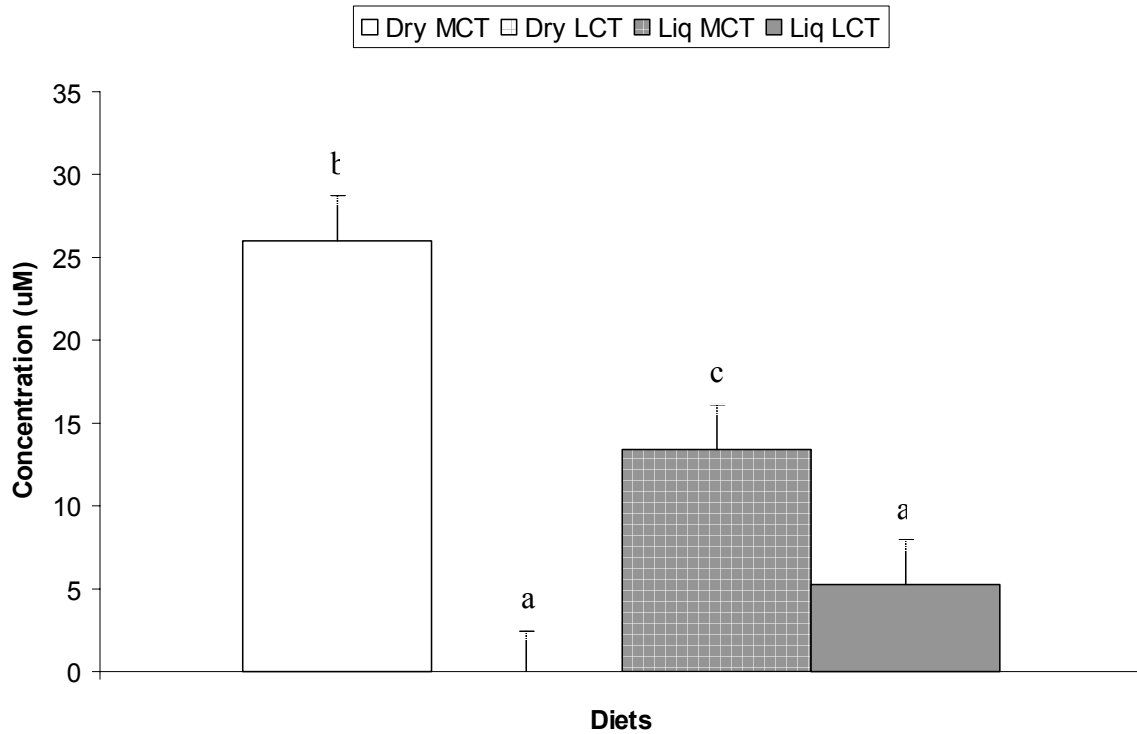
Chain length and Emulsification interaction,  $P < 0.05$   
 a,b,c, Bars lacking a common superscript differ,  $P < 0.05$

**Figure 7.** Effect of fatty acid chain length and emulsification on apparent ileal fatty acid digestibility in piglets at 7 days post weaning. Diet physical form had no detectable effect.



Chain length and Emulsification interaction,  $P < 0.05$   
 a,b,c,d Bars lacking a common superscript differ,  $P < 0.05$

**Figure 8.** Effect of fatty acid chain length and emulsification on apparent ileal fatty acid digestibility in piglets at 14 days post weaning. Diet physical form had no detectable effect.



Diet physical form and Chain length interaction,  $P < 0.05$   
 a,b c Bars lacking a common superscript differ,  $P < 0.05$

**Figure 9.** Effect of diet physical form and fatty acid chain length on circulating plasma  $\beta$ -hydroxybutyrate concentrations. Emulsification had no detectable effect.

## APPENDIX

**Table 1.** Effects of diet physical form, fatty acid chain length and emulsification on growth performance in weaned pigs.

	Day	Dry				Liquid				SEM	P - values		
		MCT		LCT		MCT		LCT			Form	Chain	Emuls
		+	-	+	-	+	-	+	-				
Emuls	Emuls	Emuls	Emuls	Emuls	Emuls	Emuls	Emuls	Emuls					
BW, kg	0	6.93	6.77	6.73	6.79	6.76	6.80	6.86	6.80	0.09	0.9746	0.7798	0.6074
ADG kg	0-7	0.18	0.19	0.25	0.25	0.26	0.35	0.41	0.38	0.05	0.0002	0.0203	0.5128
	7-14	0.39	0.45	0.44	0.51	0.54	0.59	0.64	0.72	0.07	0.0018	0.0926	0.2134
	0-14	0.28	0.32	0.35	0.38	0.40	0.47	0.53	0.55	0.04	<.0001	0.0097	0.1675
ADFI kg	0-7	0.21	0.25	0.26	0.27	0.23	0.28	0.32	0.28	0.04	0.2326	0.0995	0.5915
	7-14	0.43	0.54	0.57	0.56	0.48	0.58	0.69	0.72	0.06	0.0321	0.0033	0.1731
	0-14	0.28	0.34	0.37	0.37	0.31	0.38	0.44	0.42	0.04	0.0431	0.0056	0.2543
G/F	0-7	0.77	0.63	0.94	0.92	1.11	1.24	1.27	1.44	0.22	<.0001	0.0043	0.5750
	7-14	0.94	0.84	0.76	0.93	1.14	1.06	0.92	1.00	0.08	0.0075	0.1197	0.7227
	0-14	1.03	0.94	0.93	1.04	1.27	1.27	1.18	1.31	0.07	<.0001	0.7746	0.4379

**Table 2.** Effects of diet physical form, fatty acid chain length and emulsification on apparent ileal fatty acid digestibility by piglets at one and two weeks postweaning.

	Day	Dry				Liquid				SEM
		MCT		LCT		MCT		LCT		
		+ Emuls	- Emuls	+ Emuls	- Emuls	+ Emuls	- Emuls	+ Emuls	- Emuls	
8:0	7	99.98	100.00	100.00	100.00	100.00	100.00	100.00	98.69	0.44
	14	99.98	100.00	100.00	99.54	100.00	100.00	99.91	99.15	0.44
10:0 <sup>a,b,f</sup>	7	99.93	99.98	99.74	98.74	99.91	99.98	99.53	98.55	0.41
	14	99.97	99.96	99.58	97.55	99.94	99.97	99.38	96.57	0.41
12:0 <sup>a,f,g</sup>	7	96.92	99.36	98.37	97.86	98.67	99.45	98.00	98.14	0.88
	14	99.35	98.78	98.58	95.29	98.59	99.06	98.41	95.61	0.88
14:0 <sup>f</sup>	7	97.73	99.14	97.49	97.11	96.42	98.75	96.68	96.72	0.91
	14	95.58	98.41	97.74	97.58	97.45	99.31	98.56	96.09	0.91
14:1	7	58.65	76.41	71.18	80.57	71.23	89.52	81.48	81.03	11.00
	14	64.13	72.02	79.44	92.88	84.34	68.00	67.44	91.97	11.94
16:0 <sup>b,f</sup>	7	86.93	95.16	92.80	93.67	84.89	94.36	90.44	90.36	2.34
	14	92.19	95.78	92.74	94.11	91.75	96.84	96.03	90.06	2.34
16:1 <sup>a,b,d,f,h</sup>	7	86.05	96.96	98.81	98.56	84.23	95.53	97.03	97.13	2.27
	14	95.01	95.34	98.29	98.49	89.28	97.16	99.46	97.65	2.27
18:0 <sup>a,b,f</sup>	7	61.29	104.45	79.02	79.79	67.19	99.52	70.49	68.34	8.02
	14	73.34	104.73	76.04	79.21	69.75	103.12	88.17	59.17	7.94
18:1 <sup>a,b,f</sup>	7	91.02	94.30	75.44	94.71	93.04	93.85	68.53	98.33	6.55
	14	95.22	94.52	80.73	93.23	94.39	96.56	73.86	94.59	6.37
18:2 <sup>a,d,f</sup>	7	89.51	95.68	98.87	97.71	88.21	91.95	98.61	91.45	2.41
	14	94.34	94.40	98.83	98.17	92.93	98.01	99.09	95.09	2.41
Total Fat <sup>a,f</sup>	7	97.65	99.12	94.28	94.48	97.15	98.97	92.52	91.59	1.41
	14	98.18	99.01	93.58	94.13	97.99	99.32	96.52	90.02	1.41

Form had no detectable effect,  $P > 0.10$

a, Chain length main effect,  $P < 0.05$

b, Emulsification main effect,  $P < 0.05$

c, Day main effect,  $P < 0.05$

d, Day main effect,  $P < 0.10$

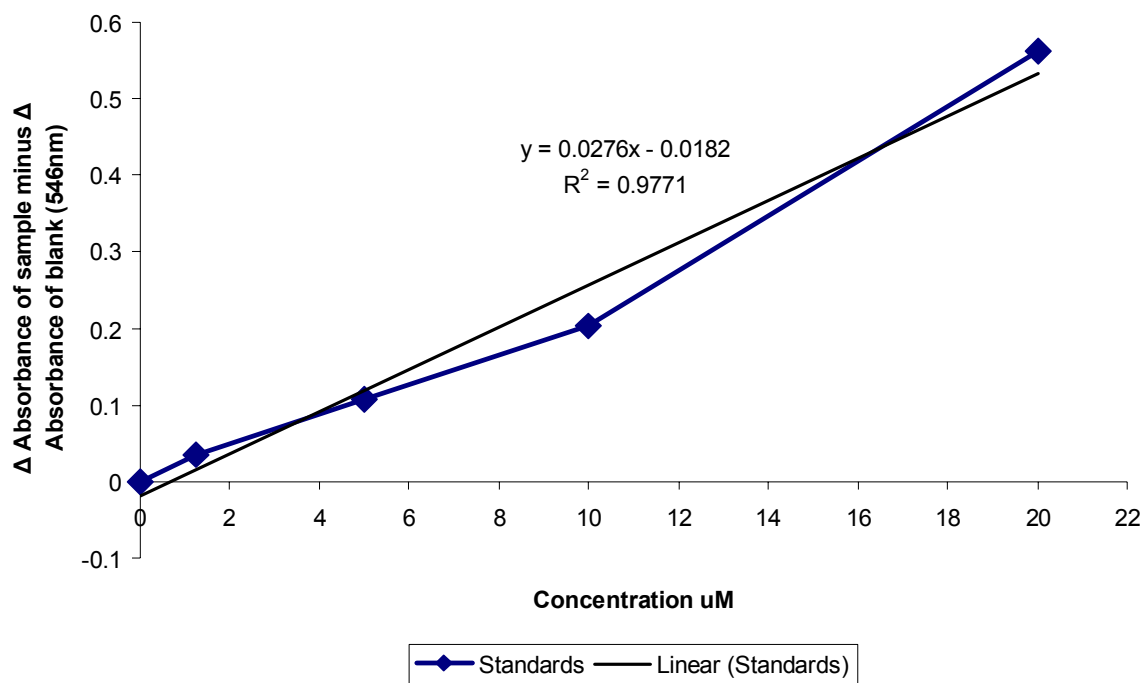
e, Chain X Day interaction,  $P < 0.05$

f, Chain X Emulsification interaction,  $P < 0.05$

g, Emulsification X Day interaction,  $P < 0.05$

h, Emulsification X Day interaction,  $P < 0.10$





**Figure 1.** Standard curve for plasma  $\beta$ -hydroxybutyrate colorimetric assay.