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Management effects on the environmental footprint of swine production

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By Aaron M. Jones

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MANAGEMENT EFFECTS ON THE ENVIRONMENTAL FOOTPRINT OF SWINE PRODUCTION

For the degree of Master of Science



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Date

MANAGEMENT EFFECTS ON THE ENVIRONMENTAL FOOTPRINT OF SWINE
PRODUCTION

A Thesis

Submitted to the Faculty

of

Purdue University

by

Aaron M. Jones

In Partial Fulfillment of the

Requirements for the Degree

of

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Dedicated to my family

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NOMENCLATURE

SYMBOL	DESCRIPTION
AA	Amino Acids
ADFI	Average Daily Feed Intake
ADG	Average Daily Gain
AID	Apparent Ileal Digestibility
AmmN	Ammonium Nitrogen
Arg	Arginine
BW	Body Weight
C	Carbon
Ca	Calcium
Cr	Chromium
CH ₄	Methane
CO ₂	Carbon Dioxide
CP	Crude Protein
CTL	Control
CTRL	Control
d	Day

SYMBOL	DESCRIPTION
DDGS	Dried Distillers Grains with Soluble
DE	Digestible Energy
DM	Dry Matter
EPA	Environmental Protection Agency
EU	European Union
FBW	Final Body Weight
FDA	US Food and Drug Administration
GHG	Green House Gasses
GE	Gross Energy
H ₂	Hydrogen Gas
H ₂ O	Water
HCl	Hydrochloric Acid
HNO ₃	Nitric Acid
IBW	Initial Body Weight
ILE	Isoleucine
LCA	Life Cycle Analysis
LLS	Livestock's Long Shadow
LYS	Lysine
ME	Metabolizable Energy
MET	Methionine
SYMBOL	DESCRIPTION
N	Nitrogen

SYMBOL	DESCRIPTION
N ₂	Nitrogen Gas
NH ₃	Ammonia
NH ₄	Ammonium
NO ₂ ⁻	Nitrite
NO ₂	Nitrogen Dioxide
NO ₃ ⁻	Nitrate
NO	Nitric Oxide
N ₂ O	Nitrous Oxide
NRC	National Research Council
O ₂	Oxygen
P	Phosphorous
TN	Total Nitrogen
SBM	Soybean Meal
SID	Standard Ileal Digestibility
THR	Threonine
TID	True Ileal Digestibility
Trp	Tryptophan
Val	Valine
VFA	Volatile Fatty Acids

ABSTRACT

Jones, Aaron M. M.S., Purdue University, May 2015. Management Effects on the Environmental Footprint of Swine Production. Major Professor: Brian T. Richert and John S. Radcliffe.

Livestock production in general is a very small contributor to GHG emissions. However, swine producers will continually be faced with a series of challenges to minimize the environmental impact of swine production. The main objectives of the the studies in this thesis were to evaluate the effects of reducing dietary CP with the supplementation of synthetic AA and the effects of feeding diets with or without antibiotics on manure generation and excretion of N and C. In Exp. 1, thirty-two barrows were used in a metabolism study to evaluate the effect of feeding reduced CP, amino acid (AA) supplemented diets on nutrient excretion. Pigs were assigned to one of four dietary treatments: 1) Control: Corn-SBM-DDGS diets with no synthetic AA, 2) 1X reduction in CP, 3) 2X reduction in CP, and 4) 3X reduction in CP. Diet 4 was balanced on the 7th limiting AA, phenylalanine. Diets 2 and 3 were then formulated to have a stepwise reduction in CP between Diets 1 and 4. Diets 2-4 were supplemented with synthetic amino acids as needed to meet amino acid needs based on NRC 2012 AA minimum ratios for the 7 age phases tested. Low-CP AA supplemented diets significantly reduce N excretion by up to 45%. In addition, VFA concentrations were reduced between 9-17% when dietary CP content was reduced up to 3X levels. Overall fecal C excreted (g/pig/d) was greatest for the lowest CP (3X), largely due to the % C digested being the lowest for

that diet. Both DE and ME, were linearly ($P < 0.0001$) decreased by approximately 6 and 5% respectively with increasing reductions in dietary CP. In Exp. 2, seven hundred twenty-three pigs were placed into eleven identical, environmentally controlled rooms for a wean-to-finish study. Pigs were allotted to one of two dietary treatments: 1) Control: Corn-SBM-DDGS diets with Antibiotics, and 2) Antibiotic Free; treatment 1 less the antibiotics but with alternative supplements. Diets were fed in nine dietary phases. There was a tendency for greater final BW and BW gain per manure pit when pigs were fed the control antibiotic treatment. No significant differences were observed between the two dietary treatments for manure volume (L), manure volume per kg BW gain, DM (g/kg BW gain), N (g/kg BW gain), and AmmN (g/kg BW gain). Manure pH tended to be lower for pigs fed the antibiotic free diet ($P < 0.06$) compared to the control diet. There were no differences observed for manure total C (kg), manure C per kg BW gain, manure C g/pig/d, and manure C g/pig wean-to-finish. In summary, Exp. 1 low CP diets with synthetic AA supplementation result in lower DE and ME values and C digestibility for the lowest CP diets, but significantly reduce N and VFA excretions. In Exp. 2, the antibiotic free diets had similar manure nutrient excretion and generation with lower manure pH which may affect transformation of N_2O during manure land application. The adoption of technologies like these evaluated in this thesis will be of the utmost importance in remaining proactive in finding a way to meet the demands of a growing world population in a manner that is cost effective for the producer, while being environmentally sustainable.

1. LITERATURE REVIEW

1.1. Environmental Footprint of Swine Production

The demand for pork has grown substantially over the past several decades. Most of this growth is the result of changes in consumption patterns as the middle class in developing countries has grown exponentially (FAO, 2014). To meet this ever-growing demand, the US swine industry has increased pork production by 174% since 1977 (USDA-ERS, 2013). This increase in production has happened while the number of swine farms in the US has decreased by 70% (Key and McBride, 2007; USDA-NASS, 2014). This shift in production towards fewer producers is consistent with economies of scale. The driving force behind these consolidations has and will continue to be triggered by technology and the need to make a living wage. As a result, swine production will continue to be concentrated into fewer and larger production systems. This presents an issue within itself from an environmental aspect.

Today we see a greater concentration of animals located on a production site. Given this increase in size, the quantity of manure, odor, and ammonia generated subsequently increases with the size of the facilities. The resulting outcome from this production shift creates growing scrutiny of the swine industry's environmental stewardship. The public's push for tougher environmental regulations from both their state and national government results in more financial hurdles for the producer to deliver

a low-cost product that meets the needs and demands of a growing global population. Unfortunately, this issue can't be answered by a single solution, but rather a series of technological innovations and management alternatives that are financially feasible and protecting the environment while progressing production efficiency. The objective of this thesis is to investigate feeding strategies that minimize nutrient excretion, thereby reducing substrates available for greenhouse gas (GHG) production in manure management for wean-to-finish pigs while maintaining production efficiency.

1.2. Global Environmental Impact of Swine Production

The FAO in 2006 released a detailed life cycle assessment (LCA) outlining livestock's global impact on the environment (FAO, 2006). To better understand this LCA, we must fundamentally understand what a LCA considers and what components are used to determine a predicted outcome. A LCA provides a systematic technique to access the environmental impacts and GHG emissions associated with a product, process, or service across all sectors. Additionally, it estimates quantities of GHG emissions from all sources within the production system (Hermansen and Kristensen, 2011). There are two categories of GHG emissions; indirect and direct. These two forms of GHG arise from livestock through physiological processes (enteric fermentation and respiration), manure storage, land application, fertilizers, animal housing, and treatment of manure slurries (Casey et al., 2006; Monteny et al., 2001). Specifically, direct emissions arise from production of CH₄ and NO₂ via enteric fermentation and nitrification/denitrification of manure and urine (Kaspar and Tiedje, 1981). Indirect emissions result from a wide variety of sources. Those primary sources range from: electricity use, water use, growing

and harvesting of crops, application of fertilizers/manure, manufacturing of items used for production agriculture (i.e. equipment, buildings, etc), deforestation, and transportation, among many others (Pitesky et al., 2009). The Livestock's Long Shadow (LLS) LCA (FAO et al., 2006) also compiled information on manure storage utilized by producers in both developed and developing countries in combination with production systems and agro-ecological zones.

In the FAO's LCA report, anthropogenic GHG emissions were broken down into eight categories. Those major categories are listed in Table 1.1. Results from this report indicated that livestock account for 9% of the CO₂, 35 to 40% of the CH₄, and 65% of the N₂O (FAO, 2006) emitted globally. These numbers have drawn considerable attention from the public based on the FAO's assessment that livestock are responsible for 18% of GHG emissions measured in CO₂ equivalents (CO₂e). However, the validity of these statements have been contested by many researchers regarding the methodology used. In response, the FAO released a follow-up report admitting miscalculations used in the LCA. The EPA (2009) released a report examining the livestock industries contribution to GHG emissions within the United States. In that report, a total of 7,150 Tg CO₂-eq yr⁻¹ of anthropogenic GHG emissions are produced annually in the USA (EPA et al., 2009). Of that 7,150 Tg CO₂-eq yr⁻¹ total, 198 Tg CO₂-eq yr⁻¹ or 2.8% is associated with the livestock sector (EPA, 2009). Of that 2.8% of the total Tg CO₂-eq yr⁻¹ produced, the swine industry contributes only 0.35% of the total U.S. GHG emissions or 3.12% of livestock emissions (Pork, 2011). Based on these estimates it becomes clear that the

livestock sector as a whole is a very small contributor to the GHG emissions within the United States.

However, according to Livestock's Long Shadow (LLS) (FAO et al., 2006) the livestock industry was reported to be a larger contributor to global GHG emissions than that of the transportation sector. Recognizing differences between the FAO (2006) and EPA (2009) GHG estimates, Pitesky et al. (2009) examined the results from both reports, and concluded that LLS (FAO et al., 2006) estimates of GHG emissions for the livestock industry was added up from farm to table (feed, enteric fermentation, processing of meat and milk into foods, etc.). Yet, the analysis for the transportation portion of LLS doesn't use the same process. Instead, it only considered emission from fossil fuels used while driving. In addition, the LLS (FAO et al., 2006) GHG assessment uses a complex LCA for the livestock industry, but doesn't for the transportation sector. It is also important to realize that the LCA GHG estimate for the livestock industry includes land use issues in developing countries, inflating the estimates (Pitesky et al., 2009). LCA models vary considerably in their level of detail, emission factors, functional units applied, allocation techniques, and definition of system boundaries (ISO, 2006). This process presents challenges based on the complexity and variation among characteristics in data sets that are utilized in any LCA. Based on this analysis, LLS's conclusion that livestock production results in more GHG emissions globally than the transportation industry is inaccurate on all accounts. It does, however, highlight sources of GHG emissions produced in the livestock industry and where focus needs to be placed to become more

efficient. This has the potential to help aid in advances for production efficiency from technological advancements and more environmentally conscious management strategies.

1.3. Amino Acids

Amino acids (AA) are organic compounds that serve as building blocks for proteins and as intermediates in metabolic functions. There are ~20 known amino acids that can be classified in many ways based on their chemical structure and biological function. In most cases, AA are categorized into two groups: Essential and Nonessential. The ten AA that are classified as essential must be provided in swine diets for normal health and metabolic processes. Proper utilization and understanding of AA ratios are key in improving production efficiency. Failure to do so can result in diets being inadequate in their AA composition. In recent years, increasing feed cost and environmental concerns about swine have caused many to shift toward feeding diets with lower crude protein (CP). Research has clearly shown that this can be done, provided that the diets are supplemented with synthetic AA (Kerr et al., 1995; Otto et al., 2003). Nonessential AA are those that can be produced by the body rather than having to be directly obtained from the diet. In most instances, the body is able to produce nonessential AA via other metabolites and various sources of nitrogen and glucose (NRC, 2012). A limiting AA can be defined as the AA that is in the shortest supply or lowest quantity in a diet impeding the rate of protein synthesis (NRC, 2012; Mitchell and Block, 1946).

Amino Acids were first discovered in 1810, and have been of great interest to nutritionists since the first discovery of Threonine in 1935 (Vickery and Schmidt, 1931; Kerr, 2006). Since that time, extensive research in the area of AA nutrition has occurred.

The market development of synthetic AA L-Lysine (Lys), D,L-methionine (Met), L-threonine (Thr), and L-tryptophan (Trp) has created a dynamic diet formulation environment in the animal feed sector. D,L- Methionine was the first synthetic AA to be developed and marketed for use in animal feed during the late 1950's and early 1960's (Toride, 2000). Around the same time in the early 1960's, synthetic Lys was also developed for use in livestock feed. These two AA are generally the 1st limiting AA in poultry and swine, respectively. However, as more crystalline AA began to become commercially available, Trp and Thr in the early 1980's, with isoleucine (Ile) and valine (Val) being launched in the late 1990's, swine nutritionists began to better understand the ratios at which other AA should be added to the diet in relation to Lys (Kerr, 2006). These discoveries have helped swine producers lower the cost of the diet and reduce the environmental footprint of swine production through a reduction in N excretion.

1.3.1. Lysine

The dietary significance of Lys for swine has been well understood for more than a half century. Since this time, Lys has been widely accepted as the 1st limiting AA in pig diets. Furthermore, Lys is important on several other fronts. The first being that the concentration of Lys in muscle is approximately 9%; and the second major reason is that many of the feedstuffs that are fed to pigs contain very little Lys. The most common form of synthetic Lys fed in rations is L-Lysine HCl (Toride, 2000; Lewis and Southern, 2001; NRC 2012). Synthetic L-Lys HCl is produced via a fermentation process that uses high performance strains of *Corynebacterium glutamicum* and *Escherichia coli* cultivated in a medium containing glucose or sugar and other ingredients (minerals, vitamins, ammonia

sulphate as a N source, etc.) (Leuchtenberger 1996; Leuchtenberger et al. 2005). Estimated production of feed grade Lys was at 500,000 – 600,000 tons in the early 2000's (Toride, 2000). More recent estimates from 2012 indicated production of Lys had increased to 1,950,000 metric tons (Ajinomoto, 2013). In addition to synthetic L-Lys HCl, Evonik industries produces a Lys source referred to as Biolys®. Biolys® was developed to contain 54.6% L-Lys in the form of a sulfated salt resulting in a granulated, free-flowing product that has been shown to have identical performance responses to that of synthetic L-Lys HCl (Rademacher, 2010). Additionally, Biolys® aids in reducing chloride levels in high lysine starter diets and potentially reducing feed costs by \$0.70 per ton due to additional energy derived from the fermentation process and resulting co-products this Lys source (Rademacher, 2010).

1.3.2. Methionine

Depending on the diet composition and the dietary phase, Met can be the 2nd or 3rd limiting AA for swine. D,L- Methionine is the most commonly found form of Met manufactured and marketed commercially as a feed additive in livestock production. D,L-Metionine is formed from the starting materials of acrolein, hydrocyanic acid, methyl mercaptan, and ammonia (Leuchtenberger et al., 2005). When D,L-Met is ingested by the animal, the AA undergoes a series of enzymatic reactions where it's converted into the nutritive L-form. Oxidase and transaminase are two key enzymes involved in this reaction that allow the pig to directly use the synthetic racemic mixture (Leuchtenberger et al., 2005). What makes D,L-Met unique from other AA is that the L-form is what is biologically available to animals (Leuchtenberger et al., 2005), but is sold

as a D, L mixture. Extensive research has investigated methods of developing a cost effective pure L-form based on knowledge acquired from the fermentation processes of Lys and Thr, but those efforts have not been successfully implemented.

Traditionally the production of the L-form of AA requires the use of production enzymes that can aid in the resolution of N-acetyl D,L-AA via the immobilization of acylase (Chibata, 1978). This process has successfully allowed manufacturers to produce an estimated 500,000 – 600,000 tons of Met annually (Toride, 2000). However, Weckbecker and Hummel (2004) proposed a new enzymatic pathway that converts D,L-Met via enzymes D-amino oxidase and leucine dehydrogenase. This could potentially allow for whole cell catalysts that will aid in the production efficiency and reduction of costs of production for the L-form of Met.

1.3.3. Threonine

Market development of Thr has grown exponentially over the past decades. In the early 2000's it was estimated that Thr was the 3rd most commercially available synthetic AA with production nearing 30,000 tons (Toride, 2000). However, more recent data indicates that the production of Thr has grown to 330,000 metric tons, an increase of 1,000 % from 2000 (Ajinomoto, 2013). The L-form of Thr is the most commercially available form used in the animal feed industry. L-Threonine is produced by fermentation processes using strains of *Corynebacterium glutamicum* and *Escherichia coli* from sugar sources such as glucose, sucrose, and molasses (Debabov, 2003; Lechtenberger et al., 2005). Ikeda (2003) determined the commercial extraction process of using *E. Coli* KY 10935 resulted in an estimated yield anywhere from 40-50 g/100 g of sucrose. Continued

advances in technology will only better the enzymatic reactions, thus aiding in a more commercially available and affordable Thr.

1.3.4. Tryptophan

Synthetic Trp just like Lys, Met, and Thr is commonly used in feeds. Since its 1st production in the 1980's, the availability of L-Trp, like other essential AA, has grown exponentially. The estimated commercial availability of L-Trp was around 1,000 metric tons back in the early 2000's (Toride, 2000). Since this time, production has risen to just over 9,000 metric tons (Ajinomoto, 2013). The process of deriving synthetic L-Trp is the same fermentation process for that of L-Lys and L-Thr described above.

1.3.5. Arginine, Histidine, Isoleucine, Leucine, Phenylalanine, and Valine

As previously mentioned, the primary synthetic Aas used in pig and poultry feed are L-Lys, D,L-Met, L-Thr, and L-Trp. Many of these AA were developed anywhere from 30-50 years ago. Continued improvements in the extraction process and technology have allowed production to increase substantially in the last decade. More recently, L-Ile and L-Val have become available in limited quantities as feed grade AA (Kerr, 2006). However, L-Ile and L-Val are not used widely due to product availability and cost (Kelle, 2009).

1.4. Defining AA Ratios

The relative importance of AA on health and metabolic processes of swine cannot be overstated. The ideal protein concept (Mitchell, 1964), which can be defined as one that provides the exact balance of AA needed for optimum performance and maximum growth was first put into practice with swine in the late 1960's (Cole, 1980; ARC, Agriculture Research Council, 1981; Wang and Fuller, 1989; Wang and Fuller, 1990; Baker, 1997; Miles and Chapman, 2007; NRC, 2012). Extensive research since this time has focused on AA nutrition, specifically looking at the AA requirements of pigs for each dietary phase of growth. The AA requirement of the pig can be expressed in many ways. Generally, AA are expressed as a percentage of the diet, grams per day, grams per unit of energy, or grams per unit of body weight. The requirement of amino acids can also be expressed as a ratio relative to the first limiting amino acid, which in pigs is Lys (NRC, 2012). To increase accuracy of AA availability in diet formulation, amino acid requirements can be expressed as either apparent ileal digestibility (AID), true ileal digestibility (TID), or standardized ileal digestibility (SID) (NRC, 2012). The method in which AA are expressed is dependent upon how the calculation takes into account the ileal AA outflow (Stein et al., 2007). The three forms of ileal digestibility can be calculated and defined by the following equations (Stein et al., 2007):

* The sum of endogenous losses (IAA_{end})

$$(1) \text{ AID}(\%) = [\text{AA intake} - \text{ileal AA outflow}] / \text{AA intake} \times 100$$

$$(2) \text{ TID}(\%) = [\text{AA intake} - (\text{ileal AA outflow} - \text{total } IAA_{end})] / \text{AA intake} \times 100$$

$$(3) \text{ SID}(\%) = [\text{AA intake} - (\text{ileal AA outflow} - \text{basal } IAA_{end})] / \text{AA intake} \times 100$$

1.4.1. Threonine:Lysine

Deficiencies in Thr have the ability to cause small decreases in gain and feed efficiency compared to deficiencies of other AA (Tokach et al., 2012). For younger pigs ranging from 10-20 kg it has been suggested that the optimal Thr:Lys can range from 62 to 66% based on previous research (James et al., 2003; Lenehen et al., 2003 and 2004). Frank et al., (2001) demonstrated that the optimum ratio of Thr:Lys in pigs ranging from 34-65 kg was 65%. This is identical to the results of Buraczewska et al. (2006) and that of the meta-analysis performed on 22 studies by Van Milgen and Le Bellego (2003). In that same meta-analysis performed by Van Milgen and Le Bellego (2003), it was suggested that the Thr:Lys ratio was 58% at 15 kg of body weight. Similarly, Pedersen et al. (2003) found that the optimal Thr:Lys ratio for late finishing stages to be anywhere from 62-64%. Based on these predictions, the minimum requirement for Thr:Lys is approximately 60-62% during the nursery phases, and rises to 64-67% in the late finishing stages. These ratios fall within the ratios reported in the NRC (2012) of 62.5% (20-50 kg), 64.5% (50-80 kg), and 67.2% (80-120 kg).

1.4.2. Met + Cys:Lys (total sulfur AA:Lys)

In addition to the previous AA mentioned, research in the area of total sulfur AA (TSAA):Lys requirements for swine has increased greatly over the years. An extensive review examining the requirement of TSAA ratios in swine was reported by Peek (2005). In that report, Peek examined results for numerous trials spanning a time period of 20 years. Based on the analysis, Peek estimated the TSAA:Lys ratio requirement for pigs on average to be between 56 and 58% depending on the Lys requirement.

The TSAA:Lys estimated for nursery pigs from 8 to 26 kg has been suggested to be 57-61% (Gaines, et al. 2005). Similarly, Yi et al. (2006) found a TSAA:Lys ratio of 58% was ideal for optimal growth of pigs between 12-24 kg. An estimated 60% TSAA:Lys ratio for growing pigs has been reported to be similar to the ratio for nursery pigs (Gaines et al., 2004; Lawrence et al., 2005). For late finishing pigs, Han and Baker (1995) suggested a ratio of 65%. However, when ractopamine HCl is added to swine diets, the TSAA:Lys ratio requirement is estimated at 58% (Frantz et al., 2009). Estimated ratios in the NRC (2012) are: 57, 57.8, and 58.9% for pigs ranging from 20-50, 50-80, and 80-120 kg, respectively.

1.4.3. Tryptophan:Lysine

Ratios of Trp:Lys have been moderately examined over the years with a lot of variability in reported requirements. The low inclusion level of Trp in swine diets presents a challenge in ensuring that the AA has been thoroughly mixed and how much Trp is coming from basal ingredients can dramatically impact the reported ratios (Tokach, 2012). Nevertheless, its importance can't be understated. Research by Guizik et al. (2002) estimated the SID Trp requirements for nursery pigs at 21, 20, and 18% of Lys for 5-7, 6-10, and 10-16 kg pigs, respectively. Similarly, Nemechek et al. (2010) demonstrated that when 8-16 kg pigs were fed at 15% SID Trp:Lys, performance was lower than pigs fed diets with a Trp:Lys of 20%. In a study performed by Quant et al. (2007), the estimated requirement was determined to be at 15.6% of Lys for 25-40 kg pigs. In a follow up study looking at the addition of other AA, it was determined that the estimated Trp:Lys increased to 17% (Quant et al., 2007). Susanbeth (2006) summarized

33 experiments looking at the SID Trp:Lys requirements, and concluded that the SID Trp:Lys requirement was below 17.4% with it being more likely near 16%. However, in that same review, Susanbeth (2006) stated that feeding a 17% SID Trp:Lys would be the safest way to ensure that requirements of pigs were being covered based on biological and ingredient variations. The NRC 2012 estimated the requirements for pigs from 20-50 kg at 17.4% of Lys.

The Trp:Lys requirements for finishing pigs are relatively scarce. Hinson et al. (2010) conducted three experiments examining the Trp:Lys requirements for pigs between 27-45, 67-85, and 96-117 kg. Results from that study estimated the Trp:Lys to be 16% over the entire weight range. More recently, Nitikachana et al. (2013) investigated the SID Trp:Lys of finishing pigs, and reported that Trp:Lys should be no less than 19 to 20%, which is consistent with the findings observed by Slayer et al. (2013). These estimates are slightly higher than the recommendation of SID Trp:Lys at 18.2% for 80-120 kg pigs (NRC, 2012). Interestingly, when Goodband et al. (2014) performed an economic analysis based on the results of Nitikachana and Slayer's experiments, it was reported that it's more economical to be over the Trp estimated requirement than below the requirement estimate.

1.4.4. Isoleucine:Lysine

Research into the requirement of Ile for pigs has been very minimal. Studies that did examine the Ile requirements for growing pigs often utilized spray-dried blood products leading to the overestimate of Ile:Lys (Parr et al., 2003; Tokach et al., 2012) as a result of high levels of Leu found in blood products, leading to a potential imbalance of branched-chain AA (Dean et al., 2005; Fu et al., 2006a,b). Based on these conclusions, research within the last 10 years has examined the optimal Ile:Lys with and without blood products. The SID Ile:Lys requirements has been estimated to be 60% or greater in diets with blood products, and 50% without blood cells (Dean et al., 2005; Fu et al., 2005a,b; Fu et al., 2006a,b,c). Additionally, Dean et al. (2005) concluded that SID Ile:Lys requirements of 50% was adequate for 80 to 120 kg pigs. This is consistent with the 48-52% SID Ile:Lys estimate reported by Lindemann et al., (2010). To further understand the optimal Ile:Lys, a meta-analysis was performed to examine the response to increasing Ile levels in the diets for growing pigs (Van Milgen et al., 2012). Results from the meta-analysis suggested that the SID Ile:Lys requirement be at least 50%. The 2012 NRC estimated requirements for 20-50, 50-80, and 80-120 kg pigs to be 50.8, 51.3 and 52%, respectively.

1.4.5. Valine:Lysine

The ratios of Val:Lys for swine had been scarce until the past decade. Research from the early 50's and late 60's only provided baseline estimates, rather than an optimal estimate (Jackson et al., 1953; Mitchell et al., 1968). More recently, Lewis and Nishimura (1995) reported that the Val requirement for 74 kg pigs was estimated at approximately 50% of Lys. Wiltafsky et al. (2009) reported an SID Val:Lys requirement for 8-25 kg pigs at 65-67%. These estimates are consistent with more recent data reported by Nemechek et al. (2011) for similar size pigs. However, this requirement is below the reported 70% SID Val:Lys requirement suggested by Barea et al. (2009). Current requirement estimates by the NRC (2012) report an SID Val:Lys ratio for 20-50, 50-80, and 80-120 kg pigs to be at 65.8, 66.6, and 67.7% respectively.

1.4.6. Phase Feeding

Feed costs have typically accounted for 65-75% of the total production cost of a pig (Pork Checkoff, 2008). Any inefficiency with the formulation or delivery of diets can subsequently raise the cost of production due to increased feed needs. Several decades ago, pigs were commonly fed two diets during their entire life-cycle prior to market. Today, we now understand that this was inefficient on several accounts. Based on that knowledge, the phase feeding concept was developed. Phase feeding is a term commonly used to refer to the feeding of several diets over short periods of times to more closely meet the nutrient requirements of pigs. Nutrient requirements as a percent of the diet decrease as the pig matures (Hinson, 2005). If market pigs are supplied one or two diets over the course of their life-cycle, pigs will be deficient in nutrients to a certain weight

and excessive for a period of time based on the nutrient profile of the diet (Kornegay and Harper, 1997). Thereafter, nutrients would be provided in excess of the pig's requirement leading to a nutrient imbalance relative to the pigs' requirement Kornegay and Harper, 1997). Henry and Dourmand (1993) provided further evidence to support phase feeding by demonstrating that when pigs were offered a single diet (17% CP) from 25 to 105 kg, N excretion was 31.9 g/d. However, when an additional diet (15% CP) was fed in conjunction with the first diet, N excretion was decreased by 1.9 g/d. If one more phase was implemented (3 total phases), N excretion was further decreased another 2.3 g/d. In total, N excretion was reduced by 16% when fed three dietary phases (17%, 15%, and 13% CP) during the grow-finish phase compared to just one diet throughout the grow-finish period. Based on these results, it's evident that phase feeding has allowed producers to minimize feed costs and reduce nutrient excretion on a whole farm basis by better meeting the pigs' nutritional needs throughout its life cycle.

1.4.7. Impact of Synthetic Amino Acids on Nitrogen Excretion

All animals have six basic nutritional needs for maintenance and growth; water, protein (amino acids), fats (some essential), and carbohydrates (energy), vitamins, and minerals. Even under the most ideal conditions, pigs are not able to utilize 100% of the nutrients that are supplied in their diets. As a result, all undigested nutrients will be excreted in the feces and metabolically unutilized nutrients excreted in the urine. Although, the excretion of undigested nutrients is a natural biological function, it becomes of great concern environmentally. Due to this concern, significant research has focused on technologies and management practices that can aid in the reduction of N

excretion. One of the most practical and cost effective methods to reduce N excretion is by feeding reduced crude protein (CP) diets supplemented with crystalline AA.

Traditionally, the amino acid requirements of swine have been met using corn and soybean meal (SBM). Corn is by far the major cereal grain fed to pigs in the Midwest and throughout the United States and is an excellent source of energy (NRC 2012). However, its protein composition is substantially poorer when compared to other feedstuffs as well as being deficient in certain essential AA. To compensate for the poorer protein and AA composition of corn, SBM has commonly been added to swine diets. Historically, SBM has been one of the more economical feedstuffs added to diets based on their AA content. However, economic conditions have drastically changed within the past decade causing SBM to become more costly to feed. The driving force of higher commodity prices can't be contributed to one thing, but rather a multitude of factors. Today, the global demand for better diets for humans is at an all-time high due to the growing middle class around the world. Globally, we've seen reduced yields contributed by poor weather conditions (ie, droughts) and increased production costs from energy and other sources (Glauber, 2008). In addition to these extrinsic factors, worldwide ethanol production since 2005 has nearly doubled with biodiesel production increasing nearly three-fold (Baier et al., 2009). All of these extrinsic factors in addition to the growing environmental concern over pig production have pushed many producers to examine practical and economically viable feedstuffs that can address the underlying environmental issues.

One method that has grown in use and has been effective in meeting the need for cost effective nutritionally balanced diets is feeding reduced CP diets supplemented with crystalline AA (Gatel and Grosjen, 1992; Dourmad et al., 1993; Lee et al., 1993). Similarly, Kerr and Easter (1995) demonstrated that when CP was reduced from 16 to 12% in a typical corn-soybean meal diet, growth performance was greatly reduced with N excretion being reduced by 10%. However, in that same study Kerr and Easter (1995) were able to show that when Lys, Trp, and Thr were added back into the diet with 12% CP, growth performance was similar to that of the 16% CP diet with N excretion being reduced by 29%. The reduction of N by 29% is consistent with the 28-40% range of N reduction reported by others when CP in diets were lowered by 3 to 4 percentage units and supplemented with AA (Piva et al., 1993; Carter et al., 1996; Sutton et al., 1996). Additionally, Sutton et al. (1997) indicated that N in the slurry could be further reduced when cellulose or a sucrose oligosaccharide was added to the diet. Based on this report, it was suggested that an additive effect of fermentable carbohydrates and reduced protein diets supplemented with AA on N excretion was occurring. However, some research has reported that adding fiber sources to reduced CP diets supplemented with AA can negatively impact growth performance (Kerr et al., 1995; Tuitoek et al., 1997).

Figuroa et al. (2002) fed three standard corn-soybean meal diets and three low-protein diets that were 4% lower in CP compared to each standard diet for gilts starting at 41 kg. The low-protein diets were supplemented with L-Lys, L-Trp, L-Thr, and D,L-Met. From that study, N excretion was decreased by 9 and 13% for the 16 and 14% CP diets, respectively. Furthermore, it was also determined that the reduction of CP by 4

percentage units between the six diets fed (18 to 14, 16 to 12, and 14 to 10) with AA supplementation, N excretion was reduced by 21, 27, and 30%, respectively. Similar research also indicates that N intake can be reduced and accompanied by a decrease in N retention (g/d), but when that data is expressed as a % of N intake, retention is typically increased with low protein-AA supplemented diets (Kerr and Easter, 1995; LeBellego et al., 2001; Figueroa et al., 2002; and Otto et al, 2003).

Nutrients that are excreted in manure are derived from four primary sources: 1) feed wastage, 2) excess nutrients provided in the diet, 3) undigested nutrients provided in the diet, and 4) biological losses from cell turnover (Killpack and Buchholz, 1993). In most cases only 20 to 50% of nitrogen and 20 to 60% of phosphorous is retained (Kornegay and Harper, 1997). As a result, 50 to 80% of the nitrogen supplied in the diet is excreted and 40 to 80% of the phosphorous is excreted (Kornegay and Harper, 1997). This presents an environmental challenge in the management and disposal of manure as its composition may exceed the capacity of land neighboring the production site. As previously mentioned, most manure from a swine facility is utilized as fertilizer for crop production. Nitrogen is typically the most limiting nutrient available for cereal grain production. Swine manure, in general, is a rich source of N that can effectively be utilized for crop production as a substitute for chemical fertilizer N (Sutton, 1982; Chantigny et al., 2008; Deen et al., 2008; Sholly et al., 2010; Seidel et al., 2010).

Typically, swine manure is spread on fields that are in close proximity of the production facilities. These areas are at the greatest risk of accumulating elevated levels of N and minerals, which may be in excess of what the crop production cycle can utilize

(Jongbloed et al., 2009). High transportation costs and time commitments are the major reasons why excessive rates of manure are applied to cropland in close proximity to the manure source (Chang and Janzen 1996). These problems along with the fact that traditionally manure has been applied on fields based on N content presents a problem within itself. Livestock manure contains an incorrect N-to-P ratio compared to the required ratios for plants (Sholly et al., 2010). Typically swine manure consists of a N-to-P ratio of approximately 1:1, which is less than the usual 3:1 ratio required by crops (Swine MMP 1994). The process of applying manure on the basis of N needed by the crop would supply an excess of 2-3 times the amount of P the crop can utilize. The idea of applying manure on the basis of the plants P requirement has some major drawbacks (increased land area, increased transportation cost, increased labor, etc.) to consider (Sholly et al., 2010).

Sholly et al. (2010) investigated changes in manure composition resulting from the feeding of low nutrient excretion (LNE) diets, and the subsequent effects on wheat growth and nutrient uptake when the manure was used as fertilizer. Manure samples were added to soil on a N basis at 325 kg plant-available N ha⁻¹ and on a phosphorous basis at 50 kg P ha⁻¹. Results indicated that soil P increased compared to the negative control diet. This can largely be attributed to the higher P content of stored manures in relation to their N:P ratio. However, there can be considerable variability between manure sources and the nutrient composition based on feedstuffs added to the diet (i.e. synthetic AA or by-product feedstuff use) and the type of manure storage.

1.5. Effects of Swine Manure on Crop Production and The Environment

1.5.1. Terrestrial Carbon Cycle

The carbon cycle involves the movement of C between various reservoirs as a result of numerous chemical, physical, geological, and biological processes (Falkowski et al., 2000). Plants have used the process of photosynthesis for millions of years to effectively utilize atmospheric CO₂ by converting it into C rich sugars and carbohydrates via autotrophs aiding in the development of plants (NYSDEC, 2014). As plants continue to develop and mature, these plants continue to accumulate and sequester more C from the atmosphere (NYSDEC, 2014). This continuous cycle is not only highly efficient in removing CO₂ from the earth's atmosphere, but it's also vital in producing O₂ that's essential for animal life (Falkowski et al., 2000). As plants begin to die and decay, CO₂ is produced and released back into the atmosphere where it will be used again by plants. In the event that atmospheric CO₂ increases, it has been assumed that plants will compensate with rapid consumption of CO₂ in conjunction with growth (Shwartz et al., 2002). However, this might not necessarily be the case after research by Shwartz et al. (2002) looked at this assumption by conducting a three-year experiment examining how the natural ecosystem compensates in minimizing emission rates of CO₂ from the combustion of fossil fuels. From that study, it was concluded that excessive atmospheric CO₂ in reality reduced plant growth therefore reducing CO₂ consumption. This would lead many to believe the original thought that fossil fuel emission could be minimized by transferring large amounts of C in the atmosphere to plants and soils may not actually be the case.

1.5.2. Nitrogen Cycle

On earth, 78% of the atmosphere is comprised of N making it one of the most important elements needed for life in most biological systems (Galloway, 1998). Nitrogen can exist in many forms including, nitrogen gas (N_2), nitric acid (HNO_3), ammonia (NH_3), ammonium (NH_4), nitrate (NO_3^-), nitric oxide (NO), nitrous oxide (N_2O), and many other organic compounds (Brady and Weil, 2000). Nitrogen in the atmosphere or in the soil can go through many complex chemical and biological changes being exchanged into living and non-living material (Brady and Weil, 2000). This exchange of N in the soil and air is commonly referred to as the nitrogen cycle (Figure 1-1). In order for the nitrogen cycle to occur, N must undergo mineralization, fixation, nitrification, and immobilization (Hart et al., 1994).

1.5.3. Fixation and Nitrification

The primary source of N in soil is organic matter. Organic matter found in most fields is composed of a combination of decaying plant materials. However, the majority of the N found in the soil is in the organic form, rendering it useless for plants (Brady and Weil, 2000). In order for the plants to be able to utilize N in the organic form, it must undergo several steps. Nitrification occurs when soil microorganisms, referred to as nitrosomes carry out a redox reaction oxidizing $NH_3 \rightarrow NO_2^-$. Once this occurs, nitrobacter further oxidizes $NO_2^- \rightarrow NO_3^-$ (EPA, 2002). Nitrogen fixation is a process in which Diazotrophs (*cyanobacteria*) and a nitrogenase found in the soil convert atmospheric nitrogen into forms plants are able to utilize (EPA, 2002). The amount of

fixation that occurs is dependent on the moisture content, temperature, oxygen supply, and fertility of the soil (EPA, 2002).

1.5.4. Mineralization and Immobilization

Mineralization is a microbially regulated process in which organic N from manure and crop residues are broken down into NH_4 (Burger and Jackson, 2003). Immobilization is the reverse of mineralization. As previously mentioned, N plays a pivotal role in all biological systems. Both microorganisms in the soil and plants will compete against one another to utilize available N. Immobilization is the process in which NO_2^- , NO_3^- , and NH_4 are taken up by microorganisms in the soil and are subsequently unavailable for crops to utilize (Burger and Jackson, 2003). Temperature, water, oxygen supply (aeration) and moisture in the soil can play a pivotal role in the rate of mineralization (organic nitrogen \rightarrow inorganic nitrogen) and immobilization (inorganic nitrogen \rightarrow organic nitrogen) (EPA, 2002).

Undoubtedly, N is essential in the development and growth of production crops. If N is deficient, root systems and plant growth will be stunted and under-developed leading to severe issues with crop quality (low in crude protein) and potential yield loss (Bates, 1970). On the other hand if there is too much N supplied in the soil, plant maturity could potentially be delayed and cause excessive vegetative growth leading to a loss of grain yield. The management of N as a fertilizer is imperative on many fronts. Any imbalance in the use of N can be an extremely costly mistake economically as well as environmentally. Nitrogen can be lost naturally in four ways during the nitrogen cycle: denitrification, volatilization, runoff, and leaching. This natural occurrence can lead to

excess nutrients in the manure entering the earth's atmosphere and bodies of water causing an environmental hazard to the ecosystems and human health. Denitrification and volatilization account for the majority of N lost in the cycle. Denitrification is a natural cycle that results from a combination of enzymes that use a stepwise reduction of NO_3^- and NO_2^- to the gaseous oxides NO, N_2O , and N_2 (Knowles, 1982). Denitrifying bacteria have the ability to use both oxygen and NO_3^- and NO_2^- as hydrogen acceptors, but in most cases they will only utilize NO_3^- when O_2 isn't readily available (Bremner and Shaw, 1958).

Symbiotic bacteria that are associated with legume root nodules play a key part in this process by taking atmospheric N_2 and reducing it to NH_3 . However, plants are unable to use NH_3 directly from this process. Instead, the NH_3 undergoes a reaction catalyzed by nitrogenase ($\text{N}_2 + 8\text{H}^+ + 8\text{e}^- \rightarrow 2\text{NH}_3 + \text{H}_2$) (Jongsun and Rees, 1994). Once this reaction is carried out, nitrogen compounds (primarily in the form of ammonia) are oxidized into NO_2^- and NO_3^- . The first step in nitrification involves ammonia-oxidizing bacteria (*Nitrosomas*, *Nitrosococcus*, and *Nitrospira*) that act upon ammonia converting it into nitrite: $\text{NH}_3 + \text{O}_2 \rightarrow \text{NO}_2^- + 3\text{H}^+ + 2\text{e}^-$ (EPA, 2002). In the second step *Nitrobacter* oxidizes nitrite: $\text{NO}_2^- + \text{H}_2\text{O} \rightarrow \text{NO}_3^- + 2\text{H}^+ + 2\text{e}^-$ (EPA, 2002) to form nitrate – the most available form of N a plant can utilize.

1.5.5. Volatilization of Nitrogen into Ammonia

N_2 is a relatively stable gas when found in the atmosphere. However, the form of N commonly found in soil has the ability to rapidly change into NH_3 . This transformational loss of N into NH_3 commonly occurs when N is in an organic form known as urea (Espinoza et al., 2005). Urea, in general, is a highly volatile organic compound that when found near the soils surface can be readily converted to NH_3 , subsequently being released into the earth's atmosphere (Killpack and Buchholz, 1993). The occurrence of N volatilization is greater when soil is saturated, extended periods of high soil temperatures, and under alkaline pH conditions (Espinoza et al., 2005). The resulting outcome from ammonia volatilization is a loss of available N from the soil leading to a loss in potential crop production.

1.5.6. Nitrate Leaching

As previously mentioned, N loss in the environment can come from several processes. Leaching can be defined as a process in which water, salts, and soluble organic compounds are carried down through permeable soils below the root zone of vegetation where they eventually reach groundwater (Sutton and Joern, 1992). In most instances, NO_3^- is the primary form of N that is leached into groundwater. High levels of NO_3^- pose a serious health risk for humans, especially infants. If infants have prolonged exposure to high NO_3^- , the resulting outcome could lead to vasodilatory/cardiovascular effects at high levels, and methemoglobinemia at lower levels. If these symptoms are left untreated, infants could become seriously ill and potentially die from excessive exposure. To ensure that levels of NO_3^- do not exceed the standards set forth by the EPA, proper

management practices must be implemented to ensure that livestock manure and chemical forms of nutrients are being efficiently used through nutrient management.

1.5.7. Eutrophication

Eutrophication has been identified as the key culprit in water pollution within the United States, with phosphorus (P) being identified as the nutrient most limiting to eutrophication (US EPA, 1996; Schindler, 1977; Correll, 1999; Sharpley et al., 1987; Sharpley et al., 1994). Eutrophication can be characterized as a process in which excess nutrients from the soil finds their way into surrounding waterways causing an increase algae growth (Correll, 1999). The excessive growth in algae can cause a disruption in the biological equilibrium by reducing oxygen concentrations. This reduction in oxygen due to the greater demand of oxygen needed for bacteria to decompose plant material far outweighs the oxygen that is produced through photosynthesis as very little oxygen diffuses into the water. The resulting outcome from eutrophication is a significant loss in aquatic life.

However, what makes the use of swine manure challenging, is swine manure tends to be inefficiently utilized due to the imbalance of nutrients within the manure relative to crop needs (Lory et al., 2006). This problem leads to the potential for a miscalculation for the rate in which the nutrients should be uniformly applied to the field. In the event that the nutrient availability and nutrient concentration are overestimated, the potential for excess nutrients to be injected in the soil becomes a real possibility. In the event that this does occur, the likelihood of eutrophication to occur in neighboring waterways greatly increases, posing a serious threat for all aquatic life. The resulting

outcome over years of eutrophication can lead to less diversification of fish and plant species, leading to indirect effects on bird and mammal species that are dependent upon those animals to survive (Harper, 1992).

1.6. Air Quality and Ammonia Emissions Effects on Both Human and Pig Health

Air quality within swine facilities is of great importance not only to the performance and health of pigs, but also to the health and well-being of employees who manage and work within those facilities as well as the neighbors in the surrounding environment. Air quality within swine facilities in general is very complex. The air within these facilities typically contains aerial pollutants that arise from organic dust, plant materials, dander, hair, microbial components, and a number of gasses (Cox and Wathes, 1995). In addition to this, the relative humidity, temperature, and ventilation rate also play a pivotal role in air quality. To monitor and control air emissions from swine facilities, the United State Environmental Protection Agency in 2005 launched an air quality assessment to investigate emission rates of ammonia, nitrous oxide, hydrogen sulfide, and volatile organic compounds that arise from livestock production (U.S. EPA, 2005). Based on the outcome from the assessment, standards were then set-forth. Producers who fail to adhere to the regulations set-forth by the EPA are subject to fines and or prosecution depending on the severity and number of offenses that have occurred in the past (U.S. EPA, 2005).

1.6.1. Relationships between Air Quality and Pig Health

Dust and gaseous compounds that arise from swine facilities originate from several sources. The primary source of dust can be associated with feed and bedding, with some coming from the dander produced by the pigs through natural sloughing of dead skin cells. The levels of dust particles in the air is dependent upon the amount of fat supplied in the diets, animal activity, stocking density, size of the pigs, and ventilation rates (Hinson, 2005). Excessive exposure to dust particles and endotoxins in the air have been reported to increase the rate of mortality and reduce weight gain at concentrations greater than 5.2 and 3.7 mg/m³ for nursery and finishing pigs, respectively (Donham, 1991). Wathles et al. (2004) also observed that average daily feed intake (ADFI) and average daily gain (ADG) were negatively impacted for weaned pigs exposed to dust concentrations of 5.1 and 9.9 mg/m³, respectively.

Although concentrations of endotoxins and dust in confinement barns are of primary concern, ammonia levels are of the greatest concern for both human's and swine's respiratory function. Jones et al. (1996) demonstrated that when pigs were exposed to 0, 10, 20, and 40 ppm ammonia, pigs spent significantly less time in ammoniated environments. Research has also shown that at ammonia concentrations at 50 ppm or less, growth performance is not inhibited (Curtis et al., 1975; Malayer et al., 1988), but at moderate concentration levels (19.7 ppm) the onset of puberty is delayed. Stombaugh et al. (1969) also reported that when pigs were exposed to ammonia concentrations ranging from 12 to 145 ppm, ADG and ADFI were subsequently reduced. A maximum concentration of 15 ppm of ammonia has been recommended for optimal

growth of pigs (Urbain et al., 1994). Ammonia levels within a confinement barn can vary depending on several factors (ie. barn, age/size of pig, pit depths, stocking density, season, ventilation, diets, etc.) Ammonia in general is a by-product of anaerobic manure decomposing over time and is hygroscopic in nature, therefore having the tendency to stay in the upper respiratory tract (Donham et al., 1986; Schwartz et al., 1992). The majority of NH_3 in manure originates from urea hydrolysis (Zhu, 2000). The process in which urea is converted to NH_3 via urease occurs within a couple of days (Gay, 2009). Urbain et al. (1994) reported reactive nasal response in pigs after 5 d of exposure to 25 ppm ammonia. This local irritation has been reported to promote local proliferation of bacteria (Drummond et al., 1978). However, other studies examining the relative effects of ammonia concentrations on health and performance of the pig have been inconsistent (Curtis et al. 1975; Urbain et al. 1995; Underdahl et al. 1982). In any case, research would indicate that extended exposure to excessive concentrations of ammonia poses a health concern to pigs. This may be particularly problematic in farrowing and nursery facilities.

1.6.2. Relationship between Air Quality and Human Health

The adverse effects of aerial pollutants such as: endotoxins, dust, and gasses on the health of workers has been well documented (Schiffman et al., 2001; Donham et al., 1985 a,b; Preller et al., 1995). The incidence of symptoms is dependent on exposure time and the level of aerial pollutant concentration in confinement buildings. Donham et al. (1989) reported that when workers were exposed to ammonia concentrations as low as 7 ppm, workers exhibited some form of respiratory distress symptoms (shortness of breath,

bronchitis, etc.). This presents a potential health risk for respiratory tract injury. Largely this can be contributed to the incomplete anaerobic digestion of manure in deep pits resulting in the release and production of organic acids, sulfur-containing compounds and ammonia (Donham et al. 1985b). On average, ammonia concentrations in confinement buildings range from 5-18 ppm (Schiffman et al., 2001). This poses a deep concern to the health and well-being of individuals who are working in these confinement barns for a full workday.

The effects of inhaling dust particles and emissions in confinement buildings have been documented to increase the incidence of coughing, wheezing, chest tightness, pulmonary disease, shortness of breath, nose and eye irritation, asthma, bronchitis, obstructive airways, and many more acute respiratory responses (Donham and Leininger, 1984; Donham et al., 1986; Schwartz et al., 1992; Donham et al., 1994; Reynolds et al., 1996; Preller et al., 1995; Zhang et al., 1998). In recent years, growing public concern has dominated news headlines for not only the workers in these facilities, but also the environmental and community hazards that are posed from swine confinement buildings. Zapletal (1998) stated that higher concentrations of air pollutants were shown to have exhibited a high relation of damaging ecosystems. Specifically, those pollutants play a significant role in acidifying the soil.

In most cases, the number one complaint that many neighbors have about swine confinement barns are the obnoxious odors generated from them (Bundy, 1992). These odors have been reported to cause tension, stress, depression, anger, and fatigue for individuals who live near swine operations (Schiffman et al., 1995). They've also

repeatedly been shown to reduce the volume of air exchanged during breathing (Warren et al., 1992, 1994). In addition, odors have also been shown to induce ocular, nasal, and respiratory mucosae (Cometto-Muniz and Cain, 1992, 1994). These results suggest that people with pre-existing respiratory conditions could presumably be more vulnerable than individuals with no respiratory problems to the emissions from swine facilities. However, there are unfortunately a limited number of studies that have examined the effects of emissions, dust, odors, and other aerial pollutants outside swine facilities. Further research in this area is needed to fully understand the overall implications that airborne emissions have on neighboring communities.

1.7. Role of Antibiotics in Mitigating the Swine Industry's Environmental Footprint

Antibiotics have been widely accepted and used over the last 50 years in the livestock industry to prevent or treat infectious agents, thereby promoting production efficiency. One of the first reported cases that demonstrated the effectiveness in the use of antibiotics for production efficiency of chicks and swine was reported by Moore et al. (1946) and Jukes et al. (1950), respectively. At this time, it's unclear to exactly why feeding subtherapeutic levels of antibiotics to livestock increases production efficiency. However, it's been suggested that antibiotics play a pivotal role in the killing of bacterial that would otherwise reduce the growth of the animal (Visek, 1978; Anderson et al. 1999). In a more recent study, Collier et al. (2003) examined the effects of antibiotics on microflora of younger pigs, and determined that pigs given antibiotic treatments had reduced species diversity and total numbers of bacteria. These results suggest that antibiotics are important in decreasing bacterial colonization within the small intestine.

The performance benefit of using antibiotics in the diets of animals has been well documented. Cromwell (2002) compiled summaries on more than 1,000 growth performance experiments in swine over a 25 year period for both starter pigs and grow-finish pigs. From that analysis, it was reported that starter pigs (7-25 kg) on average had a 16.4% improvement in growth rate and a 6.9% improvement in feed efficiency. Grower pigs (17-49 kg) had a 10.6% improvement in growth rate and a 4.5% improvement in feed efficiency and growing-finishing pigs (24-89 kg) had 4.2 and 2.2% improvements in ADG and feed efficiency, respectively (Cromwell, 2002; Jacela et al., 2009).

1.7.1. Antibiotic Resistance Issues

Growing public concern over the use of antibiotics as growth promoters has risen in the past decade. Those concerns have been based on studies that have suggested that the use of antibiotics has the ability to increase the prevalence of antibiotic-resistant bacteria. One of the first reported incidents of antibiotic resistance in food animals was reported by Starr and Reynolds (1951). In similar studies, it was determined that the use of antimicrobials in animals has the potential to apply selective pressure to the animal's normal and pathogenic microflora (Gaskins et al., 2002). Since that time, other studies have looked at the same issue and the degree of variation between the relationship of the use of antibiotics in feed and antibiotic resistance (Alpharma, 2004; Dawe, 2004, Philips et al., 2004). The demand for answers and the increase in public concern has caused many countries including the European Union to adopt policies and regulations as to the use and the banning of subtherapeutic antibiotic use in the livestock industry. The FDA in

2013 updated its regulation on the use of antibiotics by mandating that the use of antibiotics as growth promotants be phased out.

Many of these reports have found little to no correlation in the development of antibiotic resistance in human pathogens. Cromwell (2001) reported that it was possible for large doses of animal bacteria to colonize in humans, thereby providing a small chance for humans to become exposed to antibiotic resistant organisms. However, although there is a possibility for humans to be exposed to antibiotic resistant bacteria, the likelihood of this occurring is slim (Cromwell, 2001; Holt, 2008). Additionally, the Institute of Medicine (1980, 1989) reported their findings on the risk assessment of using antibiotics in animal agriculture. In those reports, scientists concluded that there was insufficient evidence to support the claims linking subtherapeutic use of antibiotics in livestock to the prevalence of antibiotic resistance in humans. However, in those findings it was suggested that antibiotics fed to animals should be reduced.

Undoubtedly, the implications of these regulations passed by government legislators will change the landscape of the livestock industry. The pressure by the public will only continue to mount, leading government officials to pass tighter regulations. This continual pressure by government regulations domestically and abroad will impose market limitations with import and export restrictions being placed on pork that have been given subtherapeutic antibiotics.

1.7.2. Impact of Antibiotics on Nutrient Excretion

As previously discussed, the movement and consolidation of swine producers has led to a greater concentration of animals in a particular geographical area. This becomes of great concern when the accumulation of excessive nutrients on a farm builds up leading to an imbalance of whole farm nutrients. To combat these problems, swine producers have looked to several methods that aid in the reduction of nutrient excretion. One solution to this issue lies in the improvement of productivity of animals (rate of gain, milk, or egg production). Van Heugten and van Kempen (2000) determined that every 0.1 percent improvement in feed efficiency resulted in a 3.3% reduction in nutrient excretion (assuming similar growth and nutrition). One technology that has been commonly adopted and utilized across all livestock industries is the use of antibiotics. Antibiotics are non-nutritive feed additives that are used in diets for therapeutic potential and to promote growth. Although the mechanism between the relationship of antimicrobial agents and its effect of growth performance hasn't been identified, it's believed to aid in several ways.

Gaskins et al. (2002) proposed that antibiotics improve growth through a series of methods by first inhibiting bacterial infections and reducing microbial metabolism products that have the potential to negatively impact the growth of the pig. The inhibition of bacterial growth will subsequently increase nutrients that are available in the diet allowing nutrients to be more readily absorbed through the intestinal wall. The extent of the response in relation to the enhancement of the overall health status of the pig and growth performance can be variable. Dritz et al. (2002) examined the use of antibiotics in modern production systems and its effect on growth performance. From that study, it was

reported that the effectiveness of feeding antibiotics increased ADG by 5%, but no effect was observed for feed efficiency.

Lindemann et al. (2010) found that the addition of bacitracin and or tylosin concentrations did not improve the digestibility of DM, energy, N, Ca, or P. However, Agudelo et al. (2007) reported that antibiotics were not similar in their effect on P digestibility. Rather, improvements in P digestibility are antibiotic-specific (Agudelo et al., 2007). Specifically, Agudelo et al. (2007) demonstrated that virginiamycin was dependent on the P digestibility of the diet. Additionally, Stewart et al. (2010) examined the effect of virginiamycin on the apparent ileal digestibility of AA in growing pigs. From that study, it was reported that virginiamycin aided in improved apparent ileal digestibility (AID) of most indispensable AA (Stewart et al., 2010). Similarly, Ravindran et al. (1984) found total tract digestibility of CP was improved when virginiamycin was fed to pigs.

1.8. Conclusions

Livestock production in general is a very small contributor to GHG emissions. However, swine producers will continually be faced with a series of challenges to minimize the environmental impact of swine production. Today, swine producers have a growing list of resources that have the ability to address environmental concerns. Continued improvements and advancements in manure handling and application, ventilation systems, and management will be just as important in reducing emissions. The adoption of current and emerging technologies will be of the utmost importance in remaining proactive in finding a way to meet the demands of a growing world population

in a manner that is cost effective for the producer, while being less harmful to the environment. The main objective of the first study in this thesis is to evaluate the effects of reducing dietary CP with the supplementation of synthetic AA has on N and C excretion. The second study in this thesis will examine the effects of feeding standard Corn-SBM-DDGS diets with or without antibiotics on manure generation, N, NH₄, pH, and C. These results will then be utilized in developing a comprehensive C and environmental footprint calculator for the swine industry.

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Table 1.1. Livestock's role in anthropogenic GHG emissions.

Item	Tg CO ₂ -eq yr ⁻¹ . ²		
Enteric Fermentation and Respiration	1,800		
Animal Manure	2,160		
Livestock related land-use changes	2,400		
Desertification linked to livestock	100		
Livestock related release from cultivated soils	230		
Feed Production	240		
On-farm fossil fuel use	90		
Postharvest emissions	10-50		
Total livestock contribution to global anthropogenic GHG emitted. ³			
	CO ₂ (%)	CH ₄ (%)	NO ₂ (%)
	9	35-40	65

¹ Adapted from Pitesky et al. (2009) and FAO (2006).

² Tetragrams of carbon dioxide equivalent.

³ % are expressed over the 1st 7 items. Only CO₂, NH₄, and N₂O emissions are considered the total GHG emissions

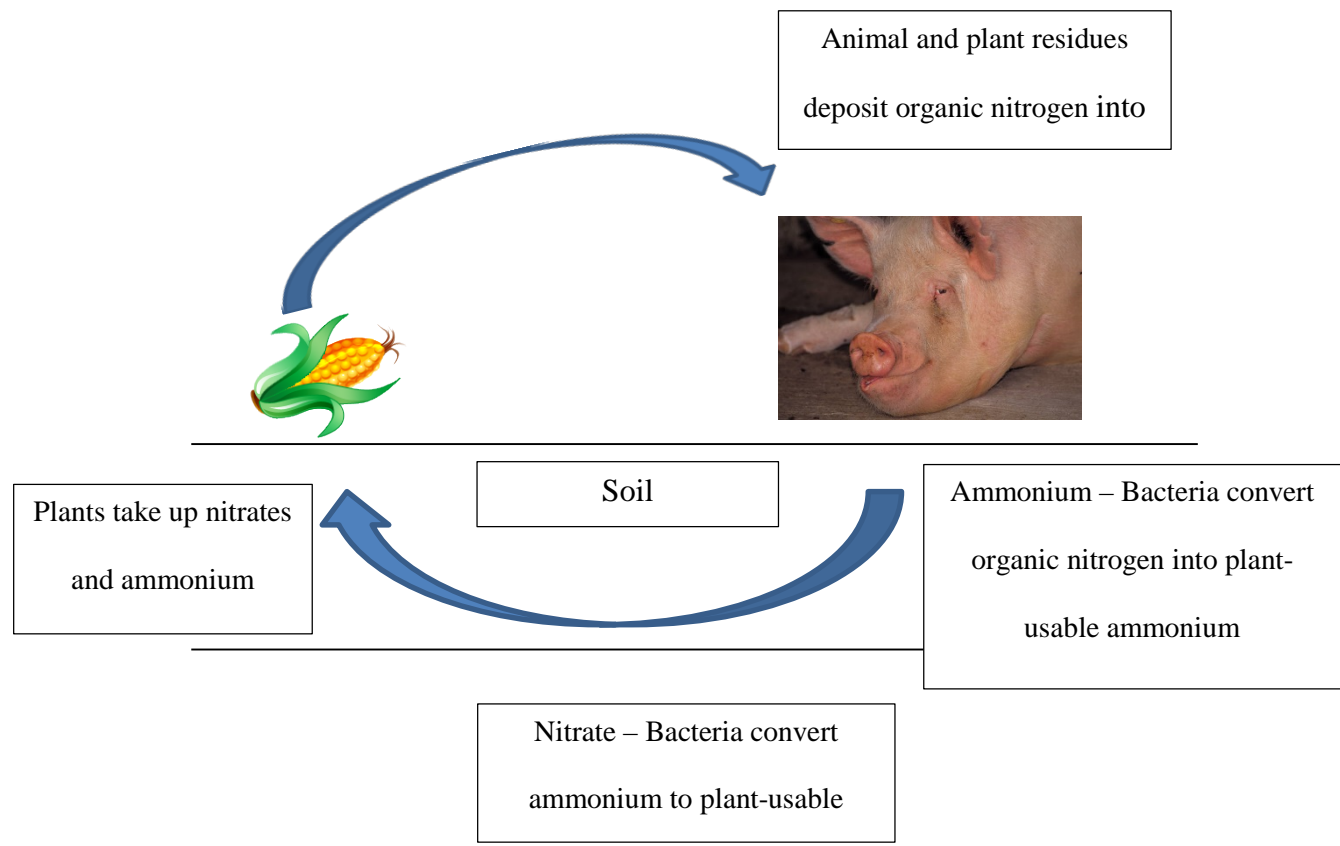


Figure 1-1. Nitrogen Cycle

Adapted from Killpack and Bucholz, 1993

CHAPTER 2. EFFECTS OF FEEDING REDUCED DIETARY CP WITH SUPPLEMENTATION OF SYNTHETIC AA ON N AND C EXCRETION, ENERGY UTILIZATION, AND FECAL VFA CONCENTRATIONS.

2.1. Abstract

Thirty-two barrows (avg initial BW 8.7 ± 0.14 kg) were used to evaluate the effect of feeding reduced CP, amino acid (AA) supplemented diets on nutrient and VFA excretion. Pigs were sorted by BW and genetics, and randomly assigned to one of four dietary treatments: 1) Control: Corn-SBM-DDGS diets with no synthetic AA, 2) 1X reduction in CP, 3) 2X reduction in CP, and 4) 3X reduction in CP. Diet 1 was a standard Corn-SBM-DDGS based diet with no synthetic AA added. Diet 4 was balanced on the 7th limiting AA, phenylalanine. Diets 2 and 3 were then formulated to have a stepwise reduction in CP between Diets 1 and 4. Diets 2-4 were supplemented with synthetic amino acids as needed to meet amino acid needs based on NRC (2012) AA minimum ratios. Feed was supplied twice daily at near ad libitum for each phase. Two nursery phases (d 14-28, d 28-42 post-weaning) and five 21 d grow-finish phases were fed. Pigs were housed in stainless-steel metabolism pens (1.22 m²) equipped with a nipple waterer and stainless-steel feeder. Two pigs were housed per pen during the nursery phase, with one pig being removed on d 42 post-weaning. During nursery phases pigs were allowed an 8 d adjustment period to the diets followed by a 3 d total collection of feces, urine, and

orts. During the grow-finish phases pigs were acclimated to diets for the first 10 d of each phase, and then feces, urine, and orts were collected for 3 d. Results from this study demonstrated that low-CP AA supplemented diets have the ability to significantly reduce N excretion by up to 45% (Lin. $P < 0.0001$). In addition, a reduction of 16, 10, 9, and 17% was detected for acetic, propionic, butyric, and total VFA concentrations, respectively, when dietary CP content was reduced up to 3X levels (Lin. $P < 0.05$). Although, there was no significant difference observed on C intake as dietary CP content was reduced, overall fecal C excreted (g/pig/d) was impacted by dietary CP content with the lowest CP diets with synthetic AA (3X) having the highest fecal C excreted (Quad. $P < 0.05$). This in large part can be contributed to the % C digested being the lowest for that diet at 84.6% (Lin. $P < 0.005$). Both DE and ME, were linearly ($P < 0.0001$) decreased by approximately 6 and 5%, respectively as dietary CP was reduced up to the 3X level. In conclusion, extremely low CP diets with synthetic AA result in lower DE and ME values for the diets, but significantly reduced N and VFA excretions.

1.2. Introduction

As part of a broader effort to reduce the swine industries' environmental impact, significant research has focused on developing management strategies that are both practical and cost effective in mitigating nutrient excretion. Dietary manipulation and management of N in swine diets has been determined to be the most cost-effective way to reduce N excretion (Leneman, et.al, 1993). Research has shown that an 8% reduction in N excretion can be achieved with a one percentage unit decrease in dietary crude protein

when synthetic amino acids (AA) are used to meet AA requirements (Kerr and Easter, 1995). However, if dietary protein is reduced by more than three percentage units and AA included, N retention is subsequently decreased (Carter et al., 1996; Kendall et al., 1999). Today, we know more about the effects dietary protein and AA supplementation on N retention and excretion than we did 15-20 years ago. Yet, when researching the environmental impact that dietary protein and AA supplementation has on C excretion the literature is sparse and unclear as to what extent C excretion can be reduced by dietary manipulation. Velthof et al., (2005) were able to determine that CH₄ emissions were directly related to pH, DM, VFA concentrations, and total C. The main objective of this study was to evaluate the effects of reducing dietary CP with supplementation of synthetic AA on N and C excretion.

2.3. Materials and Methods

This experiment was approved by the Purdue University Animal Care and Use Committee (PACUC# 1303000840).

2.3.1. Experimental Design

Thirty-two crossbred barrows (initial BW 8.7 ± 0.14 kg) were blocked by BW and genetics, and randomly assigned to one of the following dietary treatments: 1) Control: Corn-SBM-DDGS based diets with no synthetic AA, 2) 1X reduction in CP, 3) 2X reduction in CP, and 4) 3X reduction in CP. Diet 1 was a standard Corn-SBM-DDGS

based diet with no synthetic AA added. Diet 4 was balanced on the 7th limiting AA, phenylalanine. Diets 2 and 3 were then formulated to have a stepwise reduction in CP between Diets 1 and 4. Diets 2-4 were supplemented with synthetic AA as needed to meet AA needs based on NRC 2012 AA minimum ratios. Feed was supplied twice daily at near ad libitum levels for each dietary phase to minimize orts. Two nursery phases (d 14-28, d 28-42 post-weaning) and 5 grow-finish phases (Tables 2.1-2.7). Pigs were not collected during phase 1 and 2 of the nursery period due to their short 7d durations. During nursery phases 3 and 4 there were 2 pigs/pen. Each nursery phase had an 8 d acclimation period, followed by 3 d of total collection of feces, urine, and orts. At the end of nursery phase 4, one pig was removed from each metabolism pen for a total of 16 pigs (1 pig/pen) remaining on test during the grow-finish phases. Grow-finish phases had a 10 day acclimation period followed by 3 days of total collections to place the collection period near the middle of each phase. A chromic-oxide premix (0.2%) was included in the metabolism diets as an indigestible marker as a backup for determination of nutrient digestibility.

Pigs were housed in stainless-steel metabolism pens (1.22 m²) equipped with a nipple waterer and stainless steel feeder. Pens had slatted flooring that allowed for total collection of all feces and orts to be collected on a fine-mesh wire screen below the floors. Stainless steel pans were placed under the screens to funnel urine into a plastic bucket. Buckets were acidified with 100 mL of 10% hydrochloric acid to prevent ammonia volatilization. Urine was measured with graduated cylinders, mixed, and a 10% aliquot was frozen along with all feces per pen at -20°C for subsequent analysis. Orts were placed in a forced air drying oven (55 °C) for 72 h after each collection period.

After fecal samples were thawed, the total collection of feces was mixed and homogenized with an equal 1:1 ratio of distilled deionized water to form a slurry. After urine was thawed, samples were thoroughly mixed and a 250 mL subsample was taken and refrozen until subsequent analyses.

2.3.2. Sample Analyses

Fecal slurries were analyzed for dry matter (DM), ash, total Kjeldahl nitrogen (TN), ammonium nitrogen (AmmN), gross energy (GE), carbon (C), volatile fatty acids (VFA's) and chromium (Cr). Urine samples were analyzed for the same with the exception of C and Cr. Diets were analyzed for DM, ash, N, C, Cr, GE. A portion of the feces slurry was placed in a freeze dryer for approximately 7 d. Diets and dried feces were processed through a 1 mm screen in a Wiley mill prior to analysis. Dry matter was determined following 12 h drying period at 100°C. A bomb calorimeter (Parr 1261 Bomb Calorimeter, Parr Instrument Company, Moline IL) was used to measure gross energy (GE) of feed, feces, and urine. For determining urinary energy, 4 mL of urine was pipetted over 1 g of cellulose and dried for 12 h using a VWR® forced air oven (52 °C) (VWR International LLC, Radnor, PA). Based on the GE of feed, feces, and urine, digestible energy (DE) and metabolizable energy (ME) were calculated. Total N (Nelson and Sommers, 1972) and AmmN (Bremmer and Keeney, 1965) were determined by the micro-kjeldahl procedure. Both fecal and feed (Williams et al., 1962) were subjected to a nitric-perchloric digest followed by Cr determination using a Varian SpectraAA 220FS Atomic Absorption Spectrometer (Agilent Technologies, Santa Clara, CA). Fecal VFA's

(acetic, propionic, isoButyric, butyric, isovaleric, and valeric) were analyzed using a Hewlett Packard 5890A Gas Chromatograph (Agilent Technologies, Santa Clara, CA). Carbon was determined on 50 ± 2 mg of feed and freeze dried feces using a Flash EA 1112 Series Nitrogen-Carbon Analyzer (CE Elantech, Inc. Lakewood, NJ). All samples for all assays were conducted in duplicate or triplicate and were within 5 % error of each other and were reanalyzed if they exceeded 5 % error.

2.3.3. Statistical Analysis

Data were analyzed using the Proc GLM procedure of SAS (SAS Inst. Inc., Cary, NC). Metabolism pen was the experimental unit. Linear and quadratic responses were determined for decreasing dietary CP concentration. A P-value ≤ 0.05 was considered significant and $0.05 < P \leq 0.10$ was considered a trend.

2.4. Results

2.4.1. Nursery Phase 3

There were no differences ($P > 0.10$) in initial and final BW of pigs during phase 3 of the nursery (Table 2.8). However, feed intake (g/pig/d) during the collection period displayed a quadratic ($P < 0.04$) response, with feed intake of 508, 706, 667, and 656 g/pig/d for pigs fed CTL, 1X, 2X, and 3X diets, respectively. This increased ADFI resulted in a quadratic increase in fecal (as-is and DM) excretion ($P < 0.04$) with excretion peaking as dietary CP was lowered from CTL to 2X concentrations. The

increased fecal excretion in pigs fed reduced CP diets was accompanied by linear decreases ($P < 0.004$) in DM and energy (DE and ME) digestibilities. Urinary and total N excretion responded quadratically ($P < 0.02$), increasing to 1X and then declining to the 3X CP level. Nitrogen digestibility ($P < 0.08$) tended to respond quadratically to decreasing dietary CP with increasing digestibility as dietary CP was reduced to the 2X concentration, but lowering of N digestibility at the 3X reduction in CP. Nitrogen retention also responded in a quadratic fashion with increasing N retention up to the 2X reduction in dietary CP, but no additional improvement was observed with the 3X reduction. Carbon intake and excretion followed the collection period intakes, peaking at 1X, then plateauing or slightly declining to 3X ($P < 0.03$). Carbon digestibility was linearly ($P < 0.005$) reduced from the CTRL (87.8%) to 3X diet (82.7%) as dietary CP was reduced. Acetic acid ($P = 0.07$) and valeric acid ($P < 0.002$) concentrations in the feces linearly reduced with reduction in dietary CP. However, no differences were observed for total VFA fecal concentrations among dietary treatments.

2.4.2. Nursery Phase 4

The numerically heavier BW at the start of phase 4 for pigs fed reduced CP diets in combination with the tendency ($P = 0.08$) of ADG to linearly increase as CP was reduced resulted in heavier ending BW ($P < 0.03$) for pigs fed reduced CP diets (Table 2.9). Additionally, period and collection ADFI was linearly ($P < 0.02$) increased with reductions in dietary CP, and feed efficiency (G:F) tended ($P < 0.06$) to quadratically decline and then increase with decreasing dietary CP. Digestible and metabolizable

energy (Kcal/kg) responded quadratically ($P < 0.01$) with a reduction in DE at 1X and 2X concentrations, then increasing in the 3X reduction diet with ME decreasing from CTRL to 2X concentrations, then increasing in the 3X reduction diet. Metabolizable energy on a percentage basis also followed this same quadratic pattern ($P < 0.03$). Urinary N excretion also responded quadratically ($P < 0.01$) with reductions in dietary CP with an initial increase in urinary N excretion from CTRL to 1X, but then stepwise decreasing in the 2X and 3X concentrations. This resulted in a linear ($P < 0.015$) reduction in N excretion as dietary CP was reduced with percent N intake retained to linearly ($P < 0.002$) increase. Total C intake (g/pig/d) linearly ($P < 0.01$) increased when pigs were fed reduced CP diets. However, there were no differences ($P > 0.10$) in fecal C excretion or C digestibility. Acetic acid ($P < 0.01$) and total VFA ($P < 0.05$) fecal concentrations linearly decreased as dietary CP was reduced.

2.4.3. Grower Phase 1

Pigs fed reduced CP diets had linearly increased ADFI ($P < 0.01$) and linearly decreased G:F ($P < 0.05$) due to no change in pig ADG during this period (Table 2.10). Heavier initial BW ($P < 0.03$) for pigs fed the low CP AA supplemented diets at the start of this phase were maintained to heavier final BW ($P < 0.02$). The increased fecal energy excretion ($P < 0.11$) in pigs fed reduced CP diets was accompanied by a quadratic response in DE and ME. Both energies (DE and ME) increased at 1X and then decreased, falling below CTRL at 3X. The reduction in urinary N excretion also responded quadratically ($P < 0.0001$) as dietary CP was lowered from CTRL to 3X concentrations.

Total N excreted was linearly ($P < 0.0002$) reduced from 22.1 (CTL) to 13.4 g/pig/d (3X). Nitrogen digestibility ($P < 0.10$) tended to decrease linearly while nitrogen retention ($P < 0.06$) tended to increase linearly as dietary CP was reduced. There were no dietary treatment effects on C response variables during grower phase 1 ($P > 0.27$). Acetic acid and total VFA concentrations tended ($P < 0.11$) to decrease linearly as dietary CP was reduced. Similarly, isoButyric acid concentration in feces were linearly ($P < 0.01$) decreased with a reduction in CP.

2.4.4. Grower Phase 2

There were no differences ($P > 0.10$) in BW, ADG, or ADFI of pigs during grower phase 2 (Table 2.11). However, G:F ($P < 0.06$) tended to linearly decrease in pigs fed reduced CP diets. The linear reduction in urinary energy excretion ($P < 0.01$) resulted from decreased urine output ($P < 0.03$) as dietary CP was reduced. Both DE and ME were linearly ($P < 0.001$) decreased as dietary CP was reduced. Both total N intake and urinary excretion were linearly ($P < 0.0001$) reduced as dietary CP was reduced. This resulted in total N excretion being linearly ($P < 0.0001$) reduced from CTL to 3X concentrations. Carbon intake was linearly decreased ($P < 0.003$) as dietary CP was reduced, but this didn't result in any additional improvement in fecal C excretion ($P > 0.10$). There were no differences observed for fecal VFA concentrations during grower phase 2.

2.4.5. Grower Phase 3

No differences ($P > 0.10$) were observed for growth performance during grower phase 3 (Table 2.12). However, energy intake displayed a quadratic ($P < 0.04$) response, with a slight increase at 1X and 2X and a significant decrease in energy intake in the 3X diet. Additionally, as dietary CP was reduced, DE (Kcal/kg) was linearly ($P < 0.05$) decreased and ME tended ($P < 0.10$) to linearly decrease. The significant decrease in fecal and urine N excretion ($P < 0.006$) as dietary CP was reduced resulted in a linear reduction in total N excretion ($P < 0.0004$) as dietary CP was reduced, primarily a function of reduced N intake ($P < 0.0002$). No differences were observed for N digestibility. Nitrogen retention responded quadratically ($P < 0.009$) with a decrease in retention from CTL to 1X concentrations followed by an increase in N retention in the 2X and 3X reductions. There were no differences ($P > 0.10$) in acetic, propionic, butyric, and total VFA concentrations in feces, while propionic acid tended ($P < 0.07$) to quadratically decrease and then partially increase by the 3X treatment.

2.4.6. Finisher Phase 1

There were no differences ($P > 0.10$) in ADG or initial and final BW during finisher phase 1. However, ADFI displayed a quadratic ($P < 0.05$) response, with ADFI over the entire phase decreasing as dietary CP was reduced to the 1X concentration then increasing in both the 2X and 3X concentrations. Fecal excretion on an as-is and DM basis responded quadratically with a reduction in excretion from CTL to 2X reductions in dietary CP with pigs fed the 3X diet having fecal excretion intermediate between 2X and CTL fed pigs. The DE (Kcal/kg) also responded quadratically ($P < 0.009$), with slightly increasing DE in the 1X and 2X reduced CP diets, but then decreasing in the 3X diet. Nitrogen intake quadratically declined ($P < 0.04$) with declining dietary CP, resulting in linear reduction in urinary N ($P < 0.0007$) and total N ($P < 0.004$) excretion. Nitrogen digestibility ($P < 0.02$) was linearly decreased as dietary CP was reduced. No differences were observed for N retention. Carbon digestibility responded quadratically ($P < 0.05$), with an increase in digestibility at 2X and reduced digestibility in 3X fed pigs relative to CTL fed pigs as a result of the inverse occurring with fecal C excretion ($P < 0.004$) quadratically declining and then increasing by the 3X treatment. Acetic acid ($P < 0.10$) and total VFA ($P < 0.07$) tended to linearly decrease as dietary CP was reduced from CTL to 3X concentrations.

2.4.7. Finisher Phase 2

There were no differences ($P > 0.10$) in growth performance (Table 2.14) during finisher 2. Fecal excretion (DM) tended to quadratically decrease in excretion as dietary CP was lowered from CTRL to 2X concentrations, but increased at 3X. The decrease in fecal excretion in pigs fed reduced CP diets, for the exception of pigs fed the 3X diets, was accompanied by a quadratic ($P < 0.04$) response in DM digestibility, with the largest increase in digestibility with the 2X concentrations which was followed by a decrease at the 3X concentrations. Digestible energy and ME were linearly ($P < 0.05$) decreased as dietary CP was reduced. Nitrogen intake was linearly ($P < 0.0001$) reduced as dietary CP was reduced. This decrease in N intake resulted in urinary N and total N ($P < 0.04$) excreted to be reduced linearly with reductions in dietary CP. Nitrogen digestibility ($P < 0.07$) tended to linearly decrease with reducing dietary CP, but no differences in N retention were detected. Carbon digestibility responded quadratically ($P < 0.02$) with an increase in C digestibility from CTRL to 2X and decrease in the 3X fed pigs as a result of the increased fecal excretion of C by pigs fed the 3X diet (Quad. $P < 0.06$). Although no differences ($P > 0.10$) were observed for the six VFAs tested, total VFA fecal concentrations tended ($P < 0.09$) to linearly decrease with reducing dietary CP.

2.4.8. Overall

Overall, from d 14-147 post-weaning ADFI was linearly increased as dietary CP was reduced, but had no effect on ADG or G:F (Table 2.15). Fecal excretion (DM) tended to respond in a quadratic ($P = 0.08$) fashion with decreasing fecal excretion (DM) up to 2X reduction in CP, but then increasing in 3X fed pigs. Both DE and ME (kcal/kg) were linearly ($P < 0.0001$) reduced as dietary CP was reduced. The linear ($P < 0.0001$) decrease in N intake for pigs fed reduced CP diets was accompanied by linear ($P < 0.0001$) decreases in both urinary and total N excreted. Nitrogen digestibility linearly decreased ($P < 0.0007$) and N retention linearly increased ($P < 0.0001$) with reductions in dietary CP. Overall, there was a linear ($P < 0.03$) reduction in fecal ammonium as dietary CP was reduced. Total C intake and total fecal C excreted tended ($P = 0.06$) to respond quadratically with an increase in both C intake and C excretion up to the 1X reduced CP diets, followed by a decrease in C intake and increasing C excretion to the 3X diet creating a linear ($P < 0.05$) decrease in C digestibility as dietary CP was reduced. Acetic acid, propionic, and valeric acid fecal concentrations were linearly ($P < 0.05$) decreased as dietary CP was reduced. Overall, total VFA fecal concentrations linearly decreased ($P < 0.0005$) up to the 3X CP reduction diet. There were several significant dietary treatment by phase of production interactions indicating that each growth phase does not have the same response to the dietary CP reductions tested in this experiment for energy, N, or C excretion and digestibility.

2.5. Discussion

The results from this study are fairly consistent with published literature related to feeding low CP-AA supplemented diets. Previous research has demonstrated that feeding low CP diets supplemented with synthetic AA reduces N excretion (Kerr et al., 1995; Figueroa et al., 2002; Hinson et al., 2009; Sholly et al., 2009; Gloaguen et al., 2014). In addition to reductions in N excretion, feeding reduced CP, AA supplemented diets improves N retention as a percentage of intake (Kerr and Easter, 1995; Figueroa et al., 2002; Otto et al., 2003). Our results indicate a 45% decrease in total N excretion, which is consistent with the estimated predictions that Kerr and Easter (1995) published. In that paper, it was reported that for every one percentage reduction in CP content with the supplementation of AA, there was a potential to reduce total nitrogen losses by 8% (Kerr and Easter, 1995). Within each phase, there was approximately a 2-2.5% unit reduction in CP between each diet. In our experiment overall dietary CP reductions were between 6-8% and would indicate closer to a 7% decrease in N excretion per each 1% decrease in dietary CP.

Overall, daily fecal N (g/pig/d) across the four dietary treatments was relatively similar. Thus, fecal N didn't have a significant impact on total N excretion. In this study, the largest contributing factor driving the 45% reduction in total N excretion is based on urinary N excretion. Urinary excretion (g/pig/d) was reduced by 12.4 g/pig/d or 60% as CP was reduced and AA were supplemented. This large reduction in urinary N excretion is consistent with previous literature (Kephart and Sherritt, 1990; Kerr and Easter, 1995; Otto et al., 2003). When pigs were fed a higher CP diet, % N digested was 4% greater for

pigs on the CTRL diet compared to our 3X CP reduced diet. These results are consistent with the findings reported by Kephart and Sherritt (1990). It has been suggested that this could be contributed to the higher digestibility of feedstuffs (i.e SBM vs. corn protein) in the high CP diets (Kerr and Easter, 1995). Conversely, % N retained was improved by 24% as CP was reduced with AA supplementation.

Research has demonstrated that when pigs are fed reduced CP AA supplemented diets, carcasses of those pigs tend to contain more fat (Schoenherr, 1992; Tuitoek et al., 1993; Figueroa et al., 2002). Based on those earlier findings, Kerr and Easter (1995) suggested that the N content in low-CP AA supplemented diets affected the metabolism of N and energy. Therefore, the subsequent increase in urinary N excretion in the higher CP diets negatively effects energy utilization (Holmes et al., 1980) due to the metabolic cost of excreting the excess N in the urine. Based on these findings and that the diets that were fed to these pigs contained less SBM and more corn content as CP was reduced would suggest that DE and ME would be higher in our low-CP AA supplemented diets relative to our control. However, this was not the case in our study.

The reasons for DE and ME being reduced are unclear. However, one possibility for the lower DE and ME in our reduced CP AA supplemented diets could've been related the diets being balanced on an ME basis rather than a net energy (NE) basis. Research has shown that balancing diets on an NE basis rather than ME basis will change the energy characteristics (Ajinomoto Inc., 2014). In most cases the protein taken out of diets is usually substituted with either some form of starch or fat (Ajinomoto Inc., 2014). In our case the protein that was removed from the diets and supplemented with crystalline AA and corn. Corn should have a higher NE value than SBM (NRC, 2012). Another

possibility was that when formulating the diets based on nutrient composition book values (NRC, 2012), we over-estimated the energy content of the corn. In this instance, this would subsequently cause reductions in both DE and ME as corn increased in the diet. In addition, there is the possibility that based on the ingredients used to formulate these diets, specifically the substitution of SBM with corn, that the % digestibility being lower in corn than SBM (NRC, 2012) played a significant role in our DE and ME values being lower in the low-CP diets. Further research is needed for a more definitive answer.

It is well known that carbon is a fundamental element on earth and plays an important role in energy containing ingredients (NRC, 2012). However, very little research has focused on looking at the impact that dietary manipulation in general has on total C intake and excretion. More recently, Kerr et al. (2006) examined how dietary protein and cellulose effect on manure composition. In that study, Kerr et al. (2006) found that C content of manure was approximately 0.9% and that approximately 6.5% of the total carbon fed ended up in the manure. Results from our study indicated overall that total C intake was relatively constant at 696.4, 713.5, 709.6, and 693.6 g/pig/d as CP content was decreased (CTRL, 1X, 2X, and 3X, respectively). Although, there was no significant difference observed in C intake as dietary CP content was reduced, overall fecal C excreted (g/pig/d) was impacted by dietary CP content with our lowest CP diets with synthetic AA (3X) having the highest fecal C excreted. This in large part can be contributed to the % C digested being the lowest for that diet at 84.6%. Additionally, the corn that was fed during this trial was from 2012 drought. Research has shown that excessive heat and little rain has the ability to greatly change the complexity of C and N metabolism in plants, thus the likelihood of available nutrients changes as well (Larsson

et al., 1991; Beyrouthy et al., 1994; Foyer et al., 1998). Based on our DE and ME values in the low dietary CP high synthetic AA diets, the relative starch content may have been negatively reduced thus leading to a reduction in carbohydrates and energy digestibilities.

Many concerns or complaints that arise from swine production are directly related to odors. Swine manure in general is comprised of a variety of organic and inorganic compounds that can range in complexity (Sutton et al., 1999). These odors that are associated with swine production arise from the microbial fermentation of volatile organic compounds (VOC) and short chain volatile fatty acids (VFA's) in the gastrointestinal tract (GIT) (Sutton et al., 1999). Research by Mackie et al. (1998) determined that VFA's originated from the deamination of AA via bacteria in both the GIT and feces. In addition, Chung and Baker (1992) found that crystalline AA are absorbed before the hindgut. Thus, a reduction in CP content followed with the addition of synthetic AA should present smaller concentrations of VFA's. In the present study, the reduction of CP in addition to the supplementation of crystalline AA provided a 16, 10, 9, and 17% reduction for acetic, propionic, butyric, and total VFA concentrations, respectively.

2.6. Implications

The reductions in DE and ME as dietary CP is reduced is of concern, in that the pigs on the low CP diets are not efficiently utilizing the energy content within those diets. However, intake (g/pig/d) tended to increase linearly with increasing reductions in CP supporting this potential reduction in energy digestibility being compensated by greater feed intake. Overall, the manipulation of diets to reduce dietary CP is feasible and

practical in reducing fresh N excretion in pigs that will provide a practical method in addressing the growing concerns about environmental pollution arising from production agriculture. The reduced C digestibility and increased C excretion with extremely low CP diets needs further research and how this may impact C:N ratios and potential plant availability of manure nutrients.

2.7 References

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Table 2.1. Ingredient and nutrient composition of nursery phase 3 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	36.944	40.449	44.062	48.096
SBM	16.000	16.000	16.000	16.000
DDGS	15.000	15.000	15.000	15.000
Choice White Grease	2.581	2.637	2.668	2.628
Limestone	1.043	1.048	1.062	1.079
Monocal. Phosphate	-	0.064	0.109	0.160
Vitamin premix ²	0.250	0.250	0.250	0.250
TM premix ³	0.180	0.180	0.180	0.180
Salt	0.350	0.350	0.350	0.350
Soy concentrate	12.732	8.859	4.780	0.145
Fish meal	4.000	4.000	4.000	4.000
Dried whey	10.000	10.000	10.000	10.000
Lysine-HCL	-	0.185	0.382	0.607
DL-Met	-	0.059	0.121	0.191
L-Thr	-	-	0.100	0.213
L-Trp	-	-	0.015	0.053
L-Val	-	-	-	0.081
L-Ile	-	-	-	0.053
Phytase ⁴	0.100	0.100	0.100	0.100
Carbadox ⁵	0.250	0.250	0.250	0.250
Zinc oxide	0.370	0.370	0.370	0.370
Cr premix	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	26.63	24.65	22.65	20.45
DE, kcal/kg	3,661	3,643	3,624	3,600
ME, kcal/kg	3,429	3,429	3,429	3,429
SID Amino acids, %				
Arg	0.91	0.75	0.60	0.44
His	0.42	0.37	0.31	0.26
Ile	0.61	0.52	0.43	0.36
Leu	1.62	1.50	1.38	1.25
Lys	1.35	1.35	1.35	1.35
Met	0.29	0.26	0.24	0.21
Met + Cys	0.54	0.49	0.44	0.39
Phe	0.77	0.68	0.58	0.48
Phe + Tyr	1.35	1.18	1.02	0.84
Trp	0.15	0.12	0.10	0.10
Thr	0.54	0.47	0.42	0.42
Val	0.73	0.64	0.56	0.47

Table 2.1. Cont. Ingredient and nutrient composition of nursery phase 3 diets.

Diet	C	1X	2X	3X
Ca, %	0.53	0.53	0.53	0.53
Avail. P, %	0.21	0.21	0.21	0.21
<u>Analyzed Composition</u>				
Dry matter, %	90.17	89.87	89.99	89.40
CP, %	25.88	23.38	21.31	18.94
Carbon, %	40.66	40.97	41.40	41.29
Gross Energy, kcal/g	4.17	4.17	4.13	4.10

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 6615 IU; vitamin D₃, 662; vitamin E, 44.1 IU; vitamin K, 2.21 mg; vitamin B₁₂, 38.59 µg; riboflavin, 8.82 mg; pantothenic acid, 22.05 mg; niacin, 33.08 mg.

³TM premix supplies the following per kg of diet: iron, 126.1 mg; zinc, 126.1 mg; manganese, 15.63 mg; copper, 11.75 mg; iodine, 0.48 mg; selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Nutrition – Dupont).

⁵Mecadox[®] 10 (Carbadox-Phibro Animal Health, Ridgefield Park, NJ) provided at 55 ppm.

Table 2.2. Ingredient and nutrient composition of nursery phase 4 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	35.840	43.400	51.141	59.198
SBM, 48% CP	38.646	30.758	22.560	13.908
DDGS	20.000	20.000	20.000	20.000
Choice White Grease	2.548	2.474	2.357	2.110
Limestone	0.874	0.919	0.965	1.013
Monocal. Phosphate	0.662	0.707	0.755	0.805
Vitamin premix ²	0.250	0.250	0.250	0.250
TM premix ³	0.180	0.180	0.180	0.180
Salt	0.350	0.350	0.350	0.350
Lycine-HCL	-	0.250	0.515	0.793
DL-Met	-	0.036	0.115	0.199
L-Thr	-	0.024	0.139	0.260
L-Trp	-	-	0.025	0.072
L-Val	-	-	-	0.108
L-Ile	-	-	-	0.104
Phytase ⁴	0.100	0.100	0.100	0.100
Carbadox ⁵	0.250	0.250	0.250	0.250
Cr premix	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	26.82	23.98	21.13	18.28
DE, kcal/kg	3,634	3,618	3,603	3,588
ME, kcal/kg	3,429	3,429	3,429	3,429
SID Amino acids, %				
Arg	1.57	1.34	1.09	0.84
His	0.63	0.55	0.47	0.39
Ile	0.99	0.85	0.71	0.67
Leu	2.14	1.95	1.75	1.54
Lys	1.22	1.22	1.22	1.22
Met	0.39	0.38	0.42	0.47
Met + Cys	0.75	0.71	0.71	0.71
Phe	1.17	1.02	0.88	0.72
Phe + Tyr	2.07	1.81	1.55	1.27
Trp	0.27	0.23	0.21	0.21
Thr	0.85	0.76	0.76	0.76
Val	1.10	0.97	0.83	0.79
Ca, %	0.60	0.60	0.60	0.60
Avail. P, %	0.32	0.32	0.32	0.32

Table 2.2. Cont. Ingredient and nutrient composition of nursery phase 4 diets.

Diet	C	1X	2X	3X
<u>Analyzed Composition</u>				
Dry matter, %	88.94	88.58	88.81	88.30
CP, %	26.69	22.94	19.08	17.75
Carbon, %	46.14	43.90	46.79	45.58
Gross Energy, kcal/g	4.17	4.15	4.11	4.22

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 6615 IU; vitamin D₃, 662; vitamin E, 44.1 IU; vitamin K, 2.21 mg; vitamin B₁₂, 38.59 µg; riboflavin, 8.82 mg; pantothenic acid, 22.05 mg; niacin, 33.08 mg.

³TM premix supplies the following per kg of diet: iron, 126.1 mg; zinc, 126.1 mg; manganese, 15.63 mg; copper, 11.75 mg; iodine, 0.48 mg; selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Health – Dupont)

⁵Mecadox[®] 10 (Carbadox-Phibro Animal Health, Ridgefield Park, NJ) provided at 55 ppm.

Table 2.3. Ingredient and nutrient composition of grower phase 1 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	46.707	53.524	60.688	68.359
SBM, 48% CP	30.126	22.794	15.221	7.192
DDGS	20.000	20.000	20.000	20.000
Choice White Grease	0.850	0.896	0.801	0.500
Limestone	0.960	1.133	1.176	1.118
Monocal. phosphate	0.327	0.369	0.413	0.459
Vitamin premix ²	0.150	0.150	0.150	0.150
TM premix ³	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350
Lysine-HCL	-	0.235	0.478	0.735
DL-Met	-	-	0.056	0.134
L-Thr	-	0.008	0.114	0.226
L-Trp	-	-	0.013	0.056
L-Val	-	-	-	0.080
L-Ile	-	-	-	0.101
Phytase ⁴	0.100	0.100	0.100	0.100
CTC-50 ⁵	0.100	0.100	0.100	0.100
Cr premix	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	23.66	20.98	18.33	15.69
DE, kcal/kg	3,558	3,544	3,530	3,516
ME, kcal/kg	3,374	3,374	3,374	3,374
SID Amino acids, %				
Arg	1.33	1.11	0.89	0.65
His	0.55	0.48	0.41	0.33
Ile	0.85	0.72	0.60	0.56
Leu	1.96	1.78	1.60	1.41
Lys	1.01	1.01	1.01	1.01
Met	0.35	0.32	0.34	0.38
Met + Cys	0.68	0.61	0.59	0.59
Phe	1.02	0.89	0.75	0.61
Phe + Tyr	1.81	1.57	1.33	1.07
Trp	0.23	0.19	0.16	0.16
Thr	0.73	0.64	0.64	0.64
Val	0.97	0.84	0.72	0.66
Ca, %	0.66	0.66	0.66	0.66
Avail. P, %	0.26	0.26	0.26	0.26

Table 2.3. Cont. Ingredient and nutrient composition of grower phase 1 diets.

Diet	C	1X	2X	3X
<u>Analyzed Composition</u>				
Dry matter, %	88.25	88.17	88.30	87.77
CP, %	22.63	20.00	18.14	14.78
Carbon, %	43.23	41.95	42.14	42.58
Gross Energy, kcal/g	4.09	4.10	4.17	3.94

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397; vitamin E, 26.46 IU; vitamin K, 1.32 mg; vitamin B₁₂, 23.15 µg; riboflavin, 5.29 mg; pantothenic acid, 13.23 mg; niacin, 19.85 mg.

³TM premix supplies the following per kg of diet: iron, 87.3 mg; zinc, 87.3 mg; manganese, 10.82 mg; copper, 8.14 g; iodine, 0.33 mg; selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Health – Dupont).

⁵Chlortetracycline (CTC) provided at 110 ppm (Aureomycin 50, Alpharma Inc., Bridgewater, NJ).

Table 2.4. Ingredient and nutrient composition of grower phase 2 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	52.752	59.543	66.479	73.669
SBM, 48% CP	24.146	17.101	9.786	2.085
DDGS	20.000	20.000	20.000	20.000
Choice White Grease	0.786	0.735	0.650	0.443
Limestone	0.963	1.002	1.043	1.087
Monocal. phosphate	0.313	0.354	0.396	0.440
Vitamin premix ²	0.150	0.150	0.150	0.150
TM premix ³	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350
Lysine-HCL	-	0.226	0.460	0.707
DL-Met	-	-	0.026	0.100
L-Thr	-	-	0.098	0.206
L-Trp	-	-	0.022	0.064
L-Val	-	-	-	0.064
L-Ile	-	-	--	0.096
Phytase ⁴	0.100	0.100	0.100	0.100
CTC-50 ⁵	0.100	0.100	0.100	0.100
Cr premix	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	21.33	18.76	16.19	13.63
DE, kcal/kg	3546	3533	3519	3505
ME, kcal/kg	3374	3374	3374	3374
SID Amino acids, %				
Arg	1.15	0.94	0.73	0.50
His	0.50	0.43	0.36	0.28
Ile	0.75	0.63	0.51	0.47
Leu	1.82	1.65	1.48	1.29
Lys	0.86	0.86	0.86	0.86
Met	0.32	0.29	0.28	0.32
Met + Cys	0.62	0.55	0.51	0.51
Phe	0.92	0.79	0.66	0.52
Phe + Tyr	1.62	1.39	1.15	0.91
Trp	0.20	0.16	0.14	0.14
Thr	0.65	0.55	0.55	0.55
Val	0.87	0.75	0.63	0.56
Ca, %	0.53	0.53	0.53	0.53
Avail. P	0.23	0.23	0.23	0.23

Table 2.4. Cont. Ingredient and nutrient composition of grower phase 2 diets.

Diet	C	1X	2X	3X
<u>Analyzed Composition</u>				
Dry matter, %	87.53	87.91	87.51	87.0
CP, %	20.79	17.56	15.73	13.27
Carbon, %	41.07	41.49	39.84	40.21
Gross Energy, kcal/g	4.36	4.02	4.05	3.82

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397; vitamin E, 26.46 IU; vitamin K, 1.32 mg; vitamin B₁₂, 23.15 µg; riboflavin, 5.29 mg; pantothenic acid, 13.23 mg; niacin, 19.85 mg.

³TM premix supplies the following per kg of diet: iron, 87.3 mg; zinc, 87.3 mg; manganese, 10.82 mg; copper, 8.14 mg; iodine, 0.33 mg, selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Health – Dupont)

⁵Chlortetracycline (CTC) provided at 110 ppm (Aureomycin 50, Alpharma Inc., Bridgewater, NJ).

Table 2.5. Ingredient and nutrient composition of grower phase 3 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	57.663	62.970	68.387	74.028
SBM, 48% CP	19.365	13.820	8.085	2.025
DDGS	20.000	20.000	20.000	20.000
Choice White Grease	0.705	0.662	0.578	0.396
Limestone	1.032	1.063	1.095	1.139
Monocal. phosphate	0.245	0.277	0.345	0.345
Vitamin premix ²	0.150	0.150	0.150	0.150
TM premix ³	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350
Lysine-HCL	-	0.178	0.361	0.556
DL-Met	-	-	0.041	0.100
L-Thr	-	0.040	0.178	0.206
L-Trp	-	-	0.031	0.064
L-Val	-	-	-	0.064
L-Ile	-	-	-	0.097
Phytase ⁴	0.100	0.100	0.100	0.100
CTC-50 ⁵	0.050	0.050	0.050	0.050
Cr premix ⁶	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	19.46	17.47	15.48	13.49
DE, kcal/kg	3537	3526	3516	3505
ME, kcal/kg	3374	3374	3374	3374
SID Amino acids, %				
Arg	1.01	0.85	0.68	0.50
His	0.45	0.40	0.34	0.28
Ile	0.67	0.58	0.48	0.47
Leu	1.71	1.57	1.44	1.29
Lys	0.74	0.74	0.74	0.74
Met	0.30	0.28	0.29	0.32
Met + Cys	0.58	0.52	0.51	0.51
Phe	0.83	0.73	0.63	0.52
Phe + Tyr	1.46	1.29	1.10	0.91
Trp	0.17	0.14	0.14	0.14
Thr	0.59	0.55	0.55	0.55
Val	0.79	0.70	0.60	0.56
Ca, %	0.53	0.53	0.53	0.53
Avail. P, %	0.21	0.21	0.21	0.21

Table 2.5. Cont. Ingredient and nutrient composition of grower phase 3 diets.

Diet	C	1X	2X	3X
<u>Analyzed Composition</u>				
Dry matter, %	87.52	87.66	87.72	87.29
Crude protein, %	18.97	15.44	13.69	12.13
Carbon, %	40.88	40.59	41.12	41.48
Gross Energy, kcal/g	4.06	4.02	4.08	3.91

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397; vitamin E, 26.46 IU; vitamin K, 1.32 mg; vitamin B₁₂, 23.15 µg; riboflavin, 5.29 mg; pantothenic acid, 13.23 mg; niacin, 19.85 mg.

³TM premix supplies the following per kg of diet: iron, 87.3 mg; zinc, 87.3 mg; manganese, 10.82 mg; copper, 8.14 mg; iodine, 0.33 mg, selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Health – Dupont)

⁵Chlortetracycline (CTC) provided at 55 ppm (Aureomycin 50, Alpharma Inc., Bridgewater, NJ).

Table 2.6. Ingredient and nutrient composition of finisher phase 1 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	61.200	66.197	71.199	76.302
SBM, 48% CP	15.790	10.605	5.388	0.000
DDGS	20.000	20.000	20.000	20.000
Choice White Grease	0.703	0.665	0.623	0.547
Limestone	1.052	1.081	1.110	1.141
Monocal. phosphate	0.266	0.295	0.325	0.356
Vitamin premix ²	0.150	0.150	0.150	0.150
TM premix ³	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350
Lysine-HCL	-	0.166	0.325	0.506
DL-Met	-	-	-	-
L-Thr	-	-	0.026	0.102
L-Trp	-	-	0.005	0.034
L-Val	-	-	-	0.004
L-Ile	-	-	-	0.018
Phytase ⁴	0.100	0.100	0.100	0.100
CTC-50 ⁵	0.050	0.050	0.050	0.050
Cr premix ⁶	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	18.06	16.17	14.29	12.41
DE, kcal/kg	3,661	3,643	3,624	3,600
ME, kcal/kg	3,429	3,429	3,429	3,429
SID Amino acids, %				
Arg	0.91	0.75	0.60	0.44
His	0.42	0.37	0.31	0.26
Ile	0.61	0.52	0.43	0.36
Leu	1.62	1.50	1.38	1.25
Lys	0.65	0.65	0.65	0.65
Met	0.29	0.26	0.24	0.21
Met + Cys	0.54	0.49	0.44	0.39
Phe	0.77	0.68	0.58	0.48
Phe + Tyr	1.35	1.18	1.02	0.84
Trp	0.15	0.12	0.10	0.10
Thr	0.54	0.47	0.42	0.42
Val	0.73	0.64	0.56	0.47
Ca, %	0.53	0.53	0.53	0.53
Avail. P, %	0.21	0.21	0.21	0.21

Table 2.6. Cont. Ingredient and nutrient composition of finisher phase 1 diets.

Diet	C	1X	2X	3X
<u>Composition analyzed</u>				
Dry matter, %	87.60	87.57	87.41	87.07
CP, %	16.72	14.52	13.48	12.13
Carbon, %	42.44	43.41	41.09	41.11
Gross Energy, kcal/g	3.91	3.90	3.86	3.81

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397; vitamin E, 26.46 IU; vitamin K, 1.32 mg; vitamin B₁₂, 23.15 µg; riboflavin, 5.29 mg; pantothenic acid, 13.23 mg; niacin, 19.85 mg.

³TM premix supplies the following per kg of diet: iron, 87.3 mg; zinc, 87.3 mg; manganese, 10.82 mg; copper, 8.14 mg; iodine, 0.33 mg, selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme, Danisco Animal Health – Dupont)

⁵Chlortetracycline (CTC) provided at 55 ppm (Aureomycin 50, Alpharma Inc., Bridgewater, NJ).

Table 2.7. Ingredient and nutrient composition of finisher phase 2 diets.

	Diet ¹			
	C	1X	2X	3X
<u>Ingredient, %</u>				
Corn	68.723	73.800	78.937	84.278
SBM, 48% CP	27.985	22.656	17.221	11.480
Choice White Grease	0.871	0.819	0.749	0.579
Limestone	0.793	0.823	0.854	0.886
Monocal. phosphate	0.657	0.688	0.719	0.752
Vitamin premix ²	0.150	0.150	0.150	0.150
TM premix ³	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350
Lysine-HCL	-	0.171	0.345	0.529
DL-Met	-	0.023	0.075	0.131
L-Thr	-	0.050	0.126	0.207
L-Trp	-	-	0.004	0.035
L-Val	-	-	-	0.077
L-Ile	-	-	-	0.076
Phytase ⁴	0.100	0.100	0.100	0.100
Ractopamine ⁵	0.030	0.030	0.030	0.030
Cr premix	0.200	0.200	0.200	0.200
<u>Calculated Composition</u>				
Crude protein, %	19.57	17.69	15.82	13.95
DE, kcal/kg	3578	3570	3562	3553
ME, kcal/kg	3417	3419	3421	3422
SID Amino acids, %				
Arg	1.14	0.98	0.82	0.65
His	0.46	0.41	0.36	0.30
Ile	0.71	0.62	0.52	0.50
Leu	1.54	1.41	1.28	1.14
Lys	0.90	0.90	0.90	0.90
Met	0.28	0.27	0.30	0.33
Met + Cys	0.57	0.54	0.54	0.54
Phe	0.84	0.74	0.64	0.54
Phe + Tyr	1.45	1.28	1.10	0.92
Trp	0.20	0.17	0.14	0.14
Thr	0.61	0.59	0.59	0.59
Val	1.56	1.50	1.44	1.45
Ca, %	0.52	0.52	0.52	0.52
Avail. P, %	0.32	0.32	0.32	0.32

Table 2.7. Cont. Ingredient and nutrient composition of finisher phase 2 diets.

Diet	C	1X	2X	3X
<u>Analyzed Composition</u>				
Dry matter, %	86.44	86.70	86.81	86.68
CP, %	19.71	16.02	14.79	13.51
Carbon, %	40.41	40.87	41.95	40.13
Gross Energy, kcal/g	4.08	4.03	3.99	3.98

¹Diet: Control (C), 1X CP reduction (1X), 2X CP reduction (2X), and 3X CP reduction (3X).

²Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397; vitamin E, 26.46 IU; vitamin K, 1.32 mg; vitamin B₁₂, 23.15 µg; riboflavin, 5.29 mg; pantothenic acid, 13.23 mg; niacin, 19.85 mg.

³TM premix supplies the following per kg of diet: iron, 87.3 mg; zinc, 87.3 mg; manganese, 10.82 mg; copper, 8.14 mg; iodine, 0.33 mg, selenium, 0.30 mg.

⁴Phytase activity level 600.1 PU/kg (Phyzyme Danisco, Animal Health – Dupont).

⁵Ractopamine HCl – Paylean-9® (Elanco Animal Health, Greenfield, IN) provided at 6.0 ppm in the diets.

Table 2.8. Effect of reduced CP-AA supplemented diets on nursery phase 3 performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Probability, P ≤		
						Diet	Linear	Quad.
<u>(d 14-28) post-weaning</u>								
Start BW, kg	8.7	8.6	8.6	7.7	0.12	0.7230	0.9648	0.2927
End BW, kg	15.1	15.2	15.4	15.6	0.33	0.4347	0.1231	0.7534
ADG, kg	0.46	0.48	0.50	0.56	0.036	0.1587	0.1146	0.1449
ADFI, kg	0.66	0.65	0.68	0.76	0.037	0.1960	0.1184	0.1741
G:F	0.69	0.75	0.73	0.74	0.021	0.3315	0.2370	0.2865
<u>Collection data</u>								
Intake, g/pig/d as- is	508.3	705.7	666.8	656.1	42.04	0.0398	0.0600	0.0353
Intake, g/pig/d DM	458.4	634.2	600.0	586.6	37.81	0.0423	0.0681	0.0337
Feces, g/pig/d as- is	127.9	223.8	236.9	220.5	7.32	0.0301	0.0199	0.0374
Feces, g/pig/d DM	54.0	93.7	99.9	98.4	7.32	0.0047	0.0021	0.0203
Urine, mL/pig/d	3329.2	3273.8	2805.8	2160.8	871.81	0.7662	0.3348	0.7430
DM, % digest.	88.3	85.2	83.3	83.3	0.93	0.0121	0.0026	0.1309
<u>Energy</u>								
Energy Intake, kcal/pig/d	2377.2	3427.6	3074.0	3043.9	192.78	0.0238	0.0885	0.0206
Fecal Energy excretion, kcal/pig/d	285.4	501.4	517.5	530.8	36.75	0.0032	0.0013	0.0222
Urinary Energy excretion, kcal/pig/d	37.8	46.4	19.7	15.4	6.96	0.0346	0.0144	0.3769

Table. 2.8. Cont. Effect of reduced CP-AA supplemented diets on nursery phase 3 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
DE, kcal/kg	3678	3560	3433	3386	37.7	0.0016	0.0002	0.3687
ME, kcal/kg	3616	3504	3406	3364	37.9	0.0049	0.0007	0.3763
DE, %	88.20	85.37	83.12	82.58	0.91	0.0069	0.0011	0.2401
ME, %	86.72	84.02	82.48	82.06	0.92	0.0224	0.0043	0.2460
<u>Total N</u>								
Intake, g/pig/d	16.4	22.1	22.2	18.6	1.31	0.0327	0.2820	0.0066
Fecal, g/pig/d	3.2	4.0	3.9	4.1	0.39	0.3641	0.1740	0.3629
Urine, g/pig/d	3.7	3.9	2.9	1.6	0.23	0.0005	0.0002	0.0211
Total N excreted, g/pig/d	6.6	7.9	6.8	5.6	0.33	0.0033	0.0199	0.0031
N, % digested	80.9	81.8	82.4	78.3	1.27	0.1803	0.2405	0.0788
N, % retained	54.5	64.3	69.4	69.5	1.23	0.0001	0.0001	0.0021
Fecal AmmN, g/pig/d	0.5	0.8	0.7	0.6	0.06	0.0734	0.6138	0.0175
<u>Total C</u>								
Total C intake, g/pig/d	186.4	259.9	248.4	242.2	15.45	0.0346	0.0503	0.0298
Fecal C excreted, g/pig/d	23.0	39.2	41.8	42.1	3.17	0.0060	0.0023	0.0331
C, % digested	87.8	84.9	83.1	82.9	1.01	0.0241	0.0046	0.2625

Table 2.8. Cont. Effect of reduced CP-AA supplemented diets on nursery phase 3 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
<u>VFA, mmol/mol of feces</u>								
Acetic	75.0	69.3	64.7	61.6	4.70	0.2745	0.0663	0.7840
Propionic	25.8	22.3	21.2	22.4	1.79	0.3400	0.1853	0.2158
isoButyric	2.3	0.4	1.4	0.0	0.77	0.2015	0.1134	0.6924
Butyric	20.8	21.4	20.4	24.5	2.33	0.6085	0.3567	0.4756
isoValeric	0.4	0.4	0.0	0.0	0.24	0.4559	0.1495	0.8578
Valeric	3.2	0.7	0.0	0.0	0.55	0.0074	0.0023	0.0488
Total VFA	127.5	114.4	107.6	108.5	8.97	0.4156	0.1454	0.4521

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean.

Table 2.9. Effect of reduced CP-AA supplemented diets on nursery phase 4 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Probability, P ≤		
						Diet	Linear	Quad.
<u>(d 28 - 42) post-weaning</u>								
Start BW, kg	15.1	15.2	15.4	15.6	0.33	0.4347	0.1231	0.7534
End BW, kg	24.2	25.2	25.7	26.0	0.57	0.1294	0.0276	0.5090
ADG, kg	0.61	0.70	0.70	0.72	0.038	0.2470	0.0828	0.4698
ADFI, kg	0.89	1.08	1.07	1.08	0.041	0.0311	0.0185	0.0671
G:F	0.68	0.64	0.65	0.67	0.013	0.2351	0.7142	0.0556
<u>Collection data</u>								
Intake, g/pig/d as-is	911.0	1074.1	1091.4	1155.9	55.07	0.0597	0.0137	0.3944
Intake, g/pig/d DM	810.2	951.4	969.3	1020.7	48.84	0.0671	0.0156	0.3819
Feces, g/pig/d as-is	229.6	274.8	231.9	251.9	32.02	0.7358	0.8706	0.7035
Feces, g/pig/d DM	103.2	139.9	122.0	131.5	11.91	0.2266	0.2398	0.2837
Urine, mL/pig/d	4289.6	3802.5	4353.3	3645.8	652.63	0.8314	0.6475	0.8696
DM, % digest.	87.5	85.3	87.5	87.1	1.02	0.4062	0.8092	0.3790
<u>Energy</u>								
Energy Intake, kcal/pig/d	4330.2	5085.0	5023.2	5485.5	30.86	0.0757	0.0192	0.5979
Fecal Energy excretion, kcal/pig/d	456.6	623.8	556.7	618.6	55.42	0.1894	0.1254	0.3671

Table 2.9. Cont. Effect of reduced CP-AA supplemented diets on nursery phase 4 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
Urinary Energy excretion, kcal/pig/d	162.8	167.5	247.3	110.9	13.26	0.0004	0.2327	0.0005
DE, kcal/kg	3740	3641	3652	3742	30.9	0.0798	0.9003	0.0136
ME, kcal/kg	3580	3504	3448	3656	32.0	0.0064	0.2622	0.0016
DE, %	89.64	87.75	88.96	88.77	0.74	0.3869	0.6769	0.2760
ME, %	85.81	84.45	83.97	86.72	0.77	0.1086	0.5261	0.0251
<u>Total N</u>								
Intake, g/pig/d	37.1	36.1	33.3	30.5	1.79	0.1071	0.0207	0.6246
Fecal, g/pig/d	5.7	6.3	5.4	5.4	0.60	0.6519	0.4984	0.6099
Urine, g/pig/d	10.6	10.8	7.1	3.9	0.62	0.0001	0.0001	0.0138
Total N excreted, g/pig/d	16.3	17.1	15.7	9.2	1.68	0.0323	0.0146	0.0603
N, % digested	84.8	82.4	84.1	82.5	1.52	0.6233	0.4544	0.7964
N, % retained	56.0	52.5	63.6	69.9	3.06	0.0057	0.0018	0.1072
Fecal AmmN, g/pig/d	1.1	1.2	1.0	0.9	0.12	0.3833	0.2875	0.2345
<u>Total C</u>								
Total C intake, g/pig/d	373.9	417.7	453.5	465.2	22.50	0.0698	0.0132	0.4931
Fecal C excreted, g/pig/d	42.8	60.9	52.0	58.2	4.96	0.1147	0.1265	0.2643
C, % digested	88.75	85.40	88.54	87.50	1.02	0.1500	0.8989	0.2858

Table. 2.9. Cont. Effect of reduced CP-AA supplemented diets on nursery phase 4 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
VFA, mmol/mol of feces								
Acetic	77.4	58.3	64.0	52.7	4.67	0.0240	0.0095	0.4261
Propionic	23.9	21.0	25.0	21.7	1.55	0.2824	0.7272	0.8818
isoButyric	1.8	1.4	1.4	0.9	0.54	0.6958	0.2776	0.8873
Butyric	16.6	14.5	19.2	16.6	1.73	0.3531	0.5738	0.8658
isoValeric	2.4	0.2	0.8	0.2	1.16	0.5232	0.2660	0.5238
Valeric	2.8	2.1	3.3	2.8	1.04	0.8648	0.7978	0.9471
Total VFA	124.9	97.6	113.8	94.7	7.45	0.0579	0.0529	0.5940

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean.

Table 2.10. Effect of reduced CP-AA supplemented diets on grower phase 1 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Probability, P ≤		
						Diet	Linear	Quad.
(d 42 – 63) post-weaning								
Start BW, kg	24.2	25.2	25.7	26.0	0.47	0.1294	0.0276	0.5090
End BW, kg	40.8	41.2	41.6	42.4	0.43	0.0954	0.0194	0.5252
ADG, kg	0.79	0.76	0.76	0.79	0.018	0.4068	0.9752	0.1082
ADFI, kg	1.40	1.36	1.45	1.49	0.027	0.0352	0.0131	0.1963
G:F	0.56	0.56	0.52	0.53	0.013	0.1331	0.0473	0.7126
<u>Collection data</u>								
Intake, g/pig/d as- is	1272.1	1267.3	1328.6	1315.7	41.32	0.6576	0.3259	0.9242
Intake, g/pig/d DM	1122.6	1117.4	1173.1	1154.8	36.46	0.6729	0.3751	0.8614
Feces, g/pig/d as- is	411.1	359.0	444.5	415.6	45.65	0.6266	0.6393	0.8054
Feces, g/pig/d DM	167.2	138.7	178.2	182.5	19.15	0.4133	0.3452	0.4129
Urine, mL/pig/d	3292.5	2475.8	2745.0	3921.7	819.94	0.6135	0.6248	0.2330
DM, % digest.	85.3	87.8	84.6	84.2	1.57	0.4259	0.3952	0.3728
<u>Energy</u>								
Energy Intake, kcal/pig/d	5626.8	5550.8	6024.9	5615.5	175.35	0.2745	0.5879	0.3671
Fecal Energy excretion, kcal/pig/d	796.5	718.1	915.7	924.4	72.29	0.2022	0.1058	0.5618
Urinary Energy excretion, kcal/pig/d	259.4	221.8	222.5	264.3	26.29	0.5417	0.8985	0.1656

Table 2.10. Cont. Effect of reduced CP-AA supplemented diets on grower phase 1 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
DE, kcal/kg	3519	3575	3537	3293	48.1	0.0099	0.0088	0.0125
ME, kcal/kg	3330	3412	3384	3112	42.9	0.0032	0.0063	0.0026
DE, %	86.0	87.1	84.7	83.5	1.20	0.2409	0.1001	0.3623
ME, %	81.4	83.1	81.1	78.9	1.07	0.1204	0.0831	0.1044
<u>Total N</u>								
Intake, g/pig/d	43.4	40.0	38.6	31.1	1.43	0.0012	0.0002	0.1992
Fecal, g/pig/d	7.9	6.0	7.3	7.1	0.95	0.5827	0.8013	0.3918
Urine, g/pig/d	14.2	11.9	9.4	6.3	0.67	0.0001	0.0001	0.0001
Total N excreted, g/pig/d	22.1	17.9	16.7	13.4	0.99	0.0012	0.0002	0.6454
N, % digested	82.1	85.3	80.9	77.3	2.36	0.1840	0.1075	0.1738
N, % retained	48.4	55.3	56.5	57.0	2.64	0.1890	0.0620	0.2954
Fecal AmmN, g/pig/d	1.5	1.2	1.4	1.3	0.17	0.5213	0.5111	0.5956
<u>Total C</u>								
Total C intake, g/pig/d	485.3	468.8	494.3	491.7	15.49	0.6610	0.5341	0.6629
Fecal C excreted, g/pig/d	79.8	66.5	84.3	89.8	9.25	0.3787	0.2762	0.3371
C, % digested	83.8	86.1	82.8	81.8	1.82	0.4288	0.2849	0.3905

Table 2.10. Cont. Effect of reduced CP-AA supplemented diets on grower phase 1 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
<u>VFA, mmol/mol of feces</u>								
Acetic	102.4	87.9	86.6	79.7	8.57	0.3522	0.1034	0.6729
Propionic	25.9	20.0	23.9	20.7	2.89	0.4729	0.3894	0.6599
isoButyric	2.1	2.0	1.6	1.3	0.21	0.0754	0.0143	0.6032
Butyric	17.3	13.8	13.5	13.1	2.12	0.5151	0.2101	0.4861
isoValeric	15.0	8.4	9.4	10.6	2.85	0.4212	0.3582	0.2064
Valeric	3.2	3.1	3.2	2.2	0.61	0.5910	0.3081	0.4528
Total VFA	165.8	135.3	138.2	127.5	13.81	0.2895	0.1031	0.4920

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean.

Table 2.11. Effect of reduced CP-AA supplemented diets on grower phase 2 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹						<u>Probability, P ≤</u>		
	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
<u>(d 63 - 83) post-weaning</u>								
Start BW, kg	40.6	41.5	40.4	40.6	0.64	0.6437	0.6797	0.5524
End BW, kg	54.5	54.8	53.9	53.0	0.88	0.3879	0.1268	0.4466
ADG, kg	0.73	0.67	0.71	0.65	0.028	0.2492	0.1430	0.9656
ADFI, kg	1.80	1.78	1.81	1.80	0.042	0.9530	0.8930	1.000
G:F	0.41	0.38	0.39	0.37	0.011	0.1438	0.0611	0.8399
<u>Collection data</u>								
Intake, g/pig/d as-is	1704.8	1699.2	1705.6	1702.5	17.93	0.9924	0.9929	0.9381
Intake, g/pig/d DM	1429.2	1493.7	1492.6	1481.2	15.68	0.8978	0.5761	0.6514
Feces, g/pig/d as-is	635.4	698.1	628.1	664.1	81.85	0.9090	0.9599	0.8571
Feces, g/pig/d DM	252.7	262.0	227.0	264.1	22.45	0.5298	0.9907	0.4992
Urine, mL/pig/d	4216.7	3262.5	3054.2	2431.7	684.76	0.1242	0.0312	0.7580
DM, % digest.	83.1	82.5	84.8	82.2	1.47	0.4724	0.9426	0.4476
<u>Energy</u>								
Energy Intake, kcal/pig/d	7877.4	7252.8	7396.2	6989.2	63.21	0.0001	0.0001	0.0853
Fecal Energy excretion, kcal/pig/d	1177.6	1243.0	1087.4	1253.6	109.79	0.5976	0.8649	0.6147
Urinary Energy excretion, kcal/pig/d	350.4	257.1	243.0	167.3	53.17	0.0430	0.0099	0.8326

Table 2.11. Cont. Effect of reduced CP-AA supplemented diets on grower phase 2 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
DE, kcal/kg	3708	3332	3456	3134	57.9	0.0002	0.0001	0.6058
ME, kcal/kg	3514	3108	3324	3043	66.3	0.0014	0.0014	0.3111
DE, %	85.1	82.9	85.3	82.1	1.87	0.2310	0.2542	0.6700
ME, %	80.6	77.3	82.0	79.7	1.63	0.2354	0.7589	0.7584
<u>Total N</u>								
Intake, g/pig/d	49.0	46.0	42.9	36.2	0.47	0.0001	0.0001	0.0016
Fecal, g/pig/d	10.8	10.8	8.8	9.1	1.23	0.4322	0.1696	0.8672
Urine, g/pig/d	22.8	16.3	12.4	6.3	1.05	0.0001	0.0001	0.8557
Total N excreted, g/pig/d	33.6	27.1	21.1	15.4	1.19	0.0001	0.0001	0.7413
N, % digested	78.0	76.5	79.7	74.7	2.78	0.5112	0.5396	0.4911
N, % retained	31.5	41.2	50.8	57.4	2.18	0.0001	0.0001	0.4467
Fecal AmmN, g/pig/d	1.9	1.9	1.5	1.5	0.16	0.0654	0.0157	0.9654
<u>Total C</u>								
Total C intake, g/pig/d	1058.3	1057.6	981.9	933.9	27.74	0.0156	0.0026	0.3595
Fecal C excreted, g/pig/d	115.9	118.5	105.3	123.2	10.21	0.5373	0.8247	0.4205
C, % digested	89.1	88.8	89.3	86.8	0.95	0.1823	0.1173	0.2219

Table 2.11. Cont. Effect of reduced CP-AA supplemented diets on grower phase 2 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
VFA, mmol/mol of feces								
Acetic	70.9	59.7	61.8	59.5	3.58	0.4898	0.2416	0.4712
Propionic	29.7	26.4	27.5	29.9	2.98	0.8313	0.9071	0.3811
isoButyric	2.3	1.9	1.7	1.7	0.35	0.4000	0.1442	0.4650
Butyric	21.5	19.4	17.9	16.9	4.89	0.8700	0.4360	0.8973
isoValeric	5.5	3.7	2.9	4.5	1.64	0.6133	0.5686	0.2751
Valeric	5.0	3.6	3.5	4.3	1.54	0.8262	0.6989	0.4274
Total VFA	134.9	114.6	115.1	116.8	16.37	0.6995	0.4099	0.4660

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean.

Table 2.12. Effect of reduced CP-AA supplemented diets on grower phase 3 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Probability, P ≤		
						Diet	Linear	Quad.
<u>(d 83 – 104) post-weaning</u>								
Start BW, kg	59.7	59.7	59.8	60.4	0.95	0.9583	0.6705	0.7679
End BW, kg	81.1	78.7	80.8	80.6	1.51	0.6584	0.9386	0.4448
ADG, kg	1.07	0.95	1.05	1.01	0.042	0.2695	0.7389	0.3270
ADFI, kg	2.21	2.10	2.14	2.11	0.038	0.3520	0.3027	0.3196
G:F	0.49	0.46	0.49	0.48	0.021	0.7935	0.9831	0.8208
<u>Collection data</u>								
Intake, g/pig/d as-is	2341.3	2328.3	2417.1	2300.9	86.38	0.6796	0.9287	0.4690
Intake, g/pig/d DM	2079.9	1889.5	2137.4	1977.8	89.84	0.2830	0.9000	0.8566
Feces, g/pig/d as-is	839.0	781.0	742.2	643.5	53.22	0.1839	0.0448	0.6871
Feces, g/pig/d DM	297.0	289.5	290.2	263.2	19.76	0.6707	0.3380	0.6046
Urine, mL/pig/d	2281.2	2760.3	3572.2	2995.8	820.51	0.6368	0.4551	0.4306
DM, % digest.	85.8	84.6	86.5	86.6	0.81	0.4012	0.3191	0.4010
<u>Energy</u>								
Energy Intake, kcal/pig/d	9954.4	10066.8	10024.4	9743.7	99.02	0.0785	0.1329	0.0355
Fecal Energy excretion, kcal/pig/d	1419.0	1323.3	1424.3	1285.7	86.66	0.5946	0.5117	0.7936

Table 2.12. Effect of reduced CP-AA supplemented diets on grower phase 3 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Diet	Linear	Quad.
<u>Urinary</u>								
Energy excretion, kcal/pig/d	362.2	417.9	453.3	383.9	63.99	0.7356	0.7625	0.3146
DE, kcal/kg	3477	3438	3473	3394	20.2	0.0573	0.0527	0.2890
ME, kcal/kg	3343	3262	3321	3226	31.6	0.0934	0.1017	0.8270
DE, %	86.0	85.6	85.9	86.6	0.69	0.8240	0.5915	0.4613
ME, %	82.5	81.1	81.5	82.6	0.78	0.4221	0.8497	0.1183
<u>Total N</u>								
Intake, g/pig/d	62.7	48.3	48.3	40.9	2.18	0.0006	0.0002	0.1144
Fecal, g/pig/d	11.1	9.8	8.9	7.2	0.68	0.0326	0.0057	0.7715
Urine, g/pig/d	28.7	24.9	18.8	11.7	2.18	0.0041	0.0006	0.4392
Total N excreted, g/pig/d	39.7	34.6	27.7	18.9	2.51	0.0028	0.0004	0.4523
N, % digested	82.3	79.9	81.6	82.3	0.98	0.2953	0.7437	0.1198
N, % retained	35.6	29.2	41.6	53.1	2.93	0.0031	0.0022	0.0093
Fecal AmmN, g/pig/d	2.1	2.0	1.6	1.3	0.16	0.0379	0.0061	0.5862
<u>Total C</u>								
Total C intake, g/pig/d	837.5	828.1	871.9	833.1	31.22	0.6612	0.8165	0.5654
Fecal C excreted, g/pig/d	135.2	129.1	134.1	121.5	8.86	0.6922	0.4415	0.6929
C, % digested	84.2	83.1	84.8	85.1	0.87	0.4834	0.3320	0.4063

Table 2.12. Cont. Effect of reduced CP-AA supplemented diets on grower 3 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Diet	Linear	Quad.
VFA, mmol/mol of feces								
Acetic	78.2	74.5	73.1	75.0	8.10	0.9739	0.7962	0.7141
Propionic	20.4	16.1	16.7	18.1	1.48	0.2045	0.4202	0.0658
isoButyric	0.5	0.3	4.6	3.4	1.31	0.1800	0.0844	0.6829
Butyric	15.2	13.6	14.9	12.7	1.79	0.7347	0.5056	0.8428
isoValeric	3.8	1.3	2.0	3.4	1.52	0.5757	0.9342	0.1954
Valeric	0.9	1.2	0.6	0.5	0.62	0.9021	0.6297	0.7273
Total VFA	119.0	107.0	111.9	113.1	10.86	0.8728	0.8197	0.5268

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Pooled SEM = Standard error of the mean.

Table 2.13. Effect of reduced CP-AA supplemented diets on finisher phase 1 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Probability, P ≤		
						Diet	Linear	Quadratic
<u>(d 104 - 125) post-weaning</u>								
Start BW, kg	81.1	78.7	80.8	80.6	1.51	0.6584	0.9386	0.4448
End BW, kg	100.2	96.4	100.0	100.0	2.28	0.6037	0.7880	0.3955
ADG, kg	0.90	0.84	0.91	0.92	0.049	0.6269	0.5813	0.4037
ADFI, kg	2.71	2.63	2.72	2.72	0.017	0.1897	0.4870	0.0456
G:F	0.34	0.32	0.34	0.34	0.022	0.7843	0.7120	0.4690
<u>Collection data</u>								
Intake, g/pig/d as-is	2549.0	2340.3	2565.5	2573.7	58.42	0.0624	0.3364	0.0739
Intake, g/pig/d DM	2203.3	2029.0	2227.1	2230.9	50.61	0.0657	0.2998	0.0877
Feces, g/pig/d as-is	792.3	630.7	597.8	737.2	50.52	0.0549	0.4560	0.0101
Feces, g/pig/d DM	305.0	283.8	256.7	332.3	19.39	0.0799	0.5891	0.0238
Urine, mL/pig/d	3037.6	3732.0	5088.8	2904.9	576.60	0.0708	0.7493	0.0237
DM, % digestibility	86.2	86.1	88.5	85.1	0.80	0.0606	0.8257	0.0500
<u>Energy</u>								
Energy Intake, kcal/pig/d	10342. 6	9700.3	10247.3	10124.8	142.8	0.0429	0.8859	0.0789
Fecal Energy excretion, kcal/pig/d	1429.2	1314.5	1238.2	1593.8	86.1	0.0557	0.3614	0.0156
Urinary Energy excretion, kcal/pig/d	418.5	398.4	326.9	177.8	74.7	0.2137	0.0643	0.3713
DE, kcal/kg	3367	3374	3394	3209	31.8	0.0079	0.0191	0.0091
ME, kcal/kg	3209	3215	3270	3142	55.9	0.4440	0.6132	0.2248
DE, %	86.2	86.5	87.9	84.2	0.8	0.0496	0.3028	0.0270
ME, %	82.2	82.4	84.7	82.5	1.5	0.5970	0.6692	0.3758

Table 2.13. Effect of reduced CP-AA supplemented diets on finisher phase 1 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quad.
<u>Total N</u>								
Intake, g/pig/d	68.4	56.7	55.3	50.1	1.43	0.0001	0.0001	0.0370
Fecal, g/pig/d	10.4	9.8	9.3	11.5	1.07	0.4738	0.5988	0.1921
Urine, g/pig/d	33.5	23.2	21.2	14.0	2.40	0.0035	0.0007	0.4904
Total N excreted, g/pig/d	43.9	33.0	30.4	25.5	3.01	0.0177	0.0042	0.3110
N, % digested	85.1	83.0	83.1	77.2	1.62	0.0430	0.0180	0.2296
N, % retained	36.4	42.3	44.8	49.3	4.67	0.3964	0.1150	0.8628
Fecal AmmN, g/pig/d	1.7	1.7	1.6	2.1	0.23	0.4430	0.4192	0.2196
<u>Total C</u>								
Total C intake, g/pig/d	948.9	890.8	921.7	921.5	2.85	0.3227	0.6506	0.1790
Fecal C excreted, g/pig/d	141.8	136.8	124.3	160.9	9.20	0.0838	0.3626	0.0362
C, % digested	85.1	84.8	86.5	82.5	0.87	0.0463	0.2067	0.0479
<u>VFA, mmol/mol of feces</u>								
Acetic	63.9	59.5	46.1	46.9	6.96	0.3456	0.0992	0.7004
Propionic	20.9	23.8	15.3	18.8	1.69	0.0554	0.1170	0.8587
isoButyric	0.5	1.0	0.3	0.6	0.42	0.6659	0.8344	0.8413
Butyric	14.8	14.8	12.2	12.6	1.30	0.4849	0.1885	0.8808
isoValeric	0.1	0.7	0.0	0.9	0.46	0.4195	0.4138	0.7821
Valeric	0.0	0.0	0.2	0.5	0.21	0.2416	0.0649	0.4594
Total VFA	100.0	99.8	74.1	80.3	8.29	0.1914	0.0732	0.6843

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean.

Table 2.14. Effect of reduced CP-AA supplemented diets on finisher phase 2 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Probability, P ≤		
						Diet	Linear	Quad.
(d 125 – 144) post-weaning								
Start BW, kg	100.2	96.4	100.0	100.0	2.28	0.6037	0.7880	0.3955
End BW, kg	119.3	116.7	120.0	121.1	4.32	0.9144	0.7043	0.6533
ADG, kg	0.96	1.02	1.00	1.05	0.117	0.9494	0.6488	0.9908
ADFI, kg	2.99	2.85	2.93	2.97	0.058	0.3000	0.9941	0.1107
GF	0.35	0.36	0.34	0.36	0.037	0.9758	0.9346	0.9276
<u>Collection data</u>								
Intake, g/pig/d as-is	2727.6	2943.3	2802.0	2874.3	94.07	0.4206	0.5446	0.4279
Intake, g/pig/d DM	2389.4	2577.6	2449.2	2502.5	82.24	0.4313	0.6235	0.3954
Feces, g/pig/d as-is	537.6	497.0	417.4	601.1	75.48	0.3683	0.7775	0.1400
Feces, g/pig/d DM	240.9	235.8	218.3	318.2	28.23	0.1148	0.1664	0.0733
Urine, mL/pig/d	3671.1	4092.4	4345.2	2863.9	860.02	0.4181	0.5844	0.1760
DM, % digestibility	90.0	90.8	91.1	87.3	1.09	0.1042	0.1923	0.0441
<u>Energy</u>								
Energy Intake, kcal/pig/d	11727.5	12245.2	11666.0	1185.0	362.44	0.6856	0.9674	0.6747
Fecal Energy excretion, kcal/pig/d	933.6	951.9	876.9	1216.2	103.64	0.1546	0.1735	0.1260
Urinary Energy excretion, kcal/pig/d	458.1	431.6	511.4	333.0	51.95	0.1534	0.2887	0.1461
DE, kcal/kg	3759	3706	3692	3573	31.1	0.0189	0.0054	0.2766
ME, kcal/kg	3601	3565	3516	3461	41.5	0.2328	0.0518	0.8129
DE, %	92.1	92.2	92.5	89.8	0.77	0.1105	0.1305	0.0761
ME, %	88.2	88.7	88.1	87.0	1.02	0.7169	0.4350	0.4208

Table 2.14. Effect of reduced CP-AA supplemented diets on finisher 2 pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Diet	Linear	Quadratic
<u>Total N</u>								
Intake, g/pig/d	86.0	75.5	66.3	62.0	2.55	0.0010	0.0001	0.2179
Fecal, g/pig/d	9.9	8.5	9.6	10.4	1.45	0.8195	0.7221	0.4393
Urine, g/pig/d	31.4	24.2	25.9	13.1	3.15	0.0198	0.0085	0.3596
Total N excreted, g/pig/d	41.3	32.7	35.6	23.5	4.16	0.0886	0.0394	0.6642
N, % digested	88.4	88.6	85.6	83.3	1.77	0.2458	0.0669	0.4684
N, % retained	52.3	56.7	46.6	61.6	4.85	0.2067	0.4857	0.2673
Fecal AmmN, g/pig/d	1.5	1.5	1.6	1.6	0.28	0.9907	0.8179	0.9886
<u>Total C</u>								
Total C intake, g/pig/d	952.5	1042.5	1019.7	999.4	33.19	0.4202	0.5441	0.4274
Fecal C excreted, g/pig/d	109.5	106.0	100.4	146.7	12.69	0.0944	0.1317	0.0612
C, % digested	88.6	89.8	90.2	85.3	1.20	0.0565	0.1613	0.0220
<u>VFA, mmol/mol of feces</u>								
Acetic	58.6	53.6	53.1	51.0	5.69	0.8081	0.3945	0.7557
Propionic	19.8	19.9	15.8	17.5	1.54	0.3384	0.1955	0.6071
isoButyric	1.0	0.0	0.8	0.6	0.42	0.3805	0.8909	0.3227
Butyric	11.4	11.2	12.9	11.5	1.51	0.8468	0.8098	0.6704
isoValeric	1.3	0.0	0.3	0.2	0.69	0.5496	0.5470	0.4405
Valeric	0.6	0.0	0.3	0.2	0.46	0.8248	0.7674	0.5218
Total VFA	103.8	85.7	82.6	79.4	8.04	0.2440	0.0932	0.3406

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Diets: C = Control, 1X = 1X reduction in CP, 2X = 2X reduction in CP, 3X = 3X reduction in CP.

² Pooled SEM = Standard error of the mean

Table 2.15. Effect of reduced CP-AA supplemented diets on overall pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets ¹	C	1X	2X	3X	SEM ²	Phase	Diet	Probability, P ≤		Phase*Diet
								Linear	Quad.	
Overall (d 14 -145) wean-to-finish										
ADG, kg	0.73	0.74	0.77	0.77	0.021	0.0001	0.4214	0.1271	0.8619	0.2871
ADFI, kg	1.64	1.67	1.70	1.70	0.016	0.0001	0.1205	0.0340	0.3041	0.1297
GF	0.45	0.44	0.45	0.46	0.013	0.0001	0.8648	0.5812	0.6344	0.3459
<u>Collection data</u>										
Intake, g/pig/d as-is	1726.4	1776.1	1786.8	1787.0	23.36	0.0001	0.1258	0.0624	0.2688	0.0338
Intake, g/pig/d DM	1514.1	1495.9	1568.2	1558.2	30.50	0.0001	0.3011	0.1424	0.8845	0.2415
Feces, g/pig/d as-is	512.0	494.2	469.4	503.3	19.28	0.0001	0.4184	0.5569	0.1709	0.0261
Feces, g/pig/d DM	204.7	206.5	198.3	225.4	7.48	0.0001	0.0614	0.1100	0.0868	0.0118
Urine, mL/pig/d	3536.4	3378.6	3711.0	2876.4	270.65	0.0826	0.1297	0.1662	0.1925	0.6599
DM, % digestibility	86.5	85.7	86.6	85.2	0.47	0.0001	0.0902	0.1410	0.4931	0.0244

Table 2.15. Cont. Effect of reduced CP-AA supplemented diets on overall pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Phase	Diet	Linear	Quad.	Phase*Diet
<u>Energy</u>										
Energy Intake, kcal/pig/d	7486.1	7642.6	7606.3	7532.9	88.19	0.0001	0.5576	0.7812	0.1751	0.0012
Fecal Energy excretion, kcal/pig/d	939.0	962.3	936.4	1049.8	31.23	0.0001	0.0341	0.0309	0.1414	0.0161
Urinary Energy excretion, kcal/pig/d	294.2	274.0	289.5	206.0	18.87	0.0001	0.0021	0.0028	0.0780	0.5613
DE, kcal/kg	3603	3515	3522	3394	15.0	0.0001	0.0001	0.0001	0.1678	0.0001
ME, kcal/kg	3452	3362	3385	3293	17.3	0.0001	0.0001	0.0001	0.9024	0.0001
DE, %	87.5	86.7	87.0	85.5	0.38	0.0001	0.0021	0.0012	0.3029	0.0110
ME, %	83.8	82.9	83.5	82.9	0.43	0.0001	0.3164	0.2677	0.7200	0.0098

Table 2.15. Cont. Effect of reduced CP-AA supplemented diets on overall pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Phase	Diet	Linear	Quad.	Phase*Diet
<u>Total N</u>										
Intake, g/pig/d	52.0	46.7	43.6	38.3	0.68	0.0001	0.0001	0.0001	0.9393	0.0001
Fecal, g/pig/d	8.5	7.9	7.6	7.8	0.36	0.0001	0.3099	0.1277	0.2697	0.3620
Urine, g/pig/d	20.6	16.6	13.8	8.2	0.63	0.0001	0.0001	0.0001	0.2129	0.0001
<u>Total N excreted, g/pig/d</u>										
	29.0	24.5	21.8	16.0	0.82	0.0001	0.0001	0.0001	0.4210	0.0015
<u>N, % digested</u>										
	83.0	82.6	82.5	79.4	0.69	0.0001	0.0011	0.0007	0.0541	0.4844
<u>N, % retained</u>										
	45.3	48.8	53.4	59.6	1.09	0.0001	0.0001	0.0001	0.2356	0.0018
<u>Fecal AmmN, g/pig/d</u>										
	1.5	1.4	1.3	1.3	0.07	0.0001	0.189	0.0319	0.9685	0.1816
<u>Total C</u>										
<u>Total C intake, g/pig/d</u>										
	696.4	713.5	709.6	693.6	9.22	0.0001	0.3088	0.7644	0.0632	0.0008
<u>Fecal C excreted, g/pig/d</u>										
	93.3	94.2	91.3	105.3	3.49	0.0001	0.0222	0.0378	0.0571	0.0201
<u>C, % digested</u>										
	86.7	86.1	86.5	84.6	0.47	0.0001	0.0065	0.0054	0.1501	0.0278

Table 2.15. Cont. Effect of reduced CP-AA supplemented diets on overall pig performance, fecal and urinary excretion, composition, and nutrient digestibility.

Diets	C	1X	2X	3X	SEM ¹	Phase	Diet	Linear	Quad.	Phase*Diet
<u>VFA, mmol/mol</u>										
<u>of feces</u>										
Acetic	73.1	65.9	64.6	61.6	2.47	0.0001	0.0117	0.0017	0.3766	0.6357
Propionic	23.7	21.5	20.8	21.4	0.78	0.0001	0.0489	0.0299	0.0745	0.3461
isoButyric	1.6	1.1	1.6	1.2	0.30	0.0174	0.4412	0.6118	0.9637	0.3258
Butyric	16.9	15.7	15.8	15.3	0.81	0.0001	0.5750	0.2232	0.6482	0.924
isoValeric	4.1	2.2	2.1	2.8	0.55	0.0001	0.0349	0.0938	0.0169	0.8207
Valeric	2.3	1.5	1.6	1.4	0.28	0.0001	0.1041	0.0415	0.2449	0.6353
Total VFA	124.5	108.2	106.4	103.5	3.99	0.0001	0.0013	0.0005	0.0851	0.7610

BW = body weight, DM = Dry matter, DE = digestible energy, ME = metabolizable energy, N = nitrogen, AmmN = ammonium-nitrogen, C = carbon, VFA = volatile fatty acids.

¹ Pooled SEM = Standard error of the mean.

CHAPTER 3. EFFECTS OF DIETARY ANTIBIOTICS AND ANTIBIOTIC-FREE DIETS WITH ALTERNATIVE ADDITIVES ON MANURE GENERATION AND CHARACTERISTICS.

3.1. Abstract

Seven hundred twenty-three (395 barrows and 329 gilts) (York x Landrace; Temple Genetics, Inc.) maternal cross pigs (avg. initial BW = 6.70 kg) were placed into eleven identical, environmentally controlled rooms for a wean-to-finish study. Pigs were allotted by sex and weight to one of two dietary treatments: 1) Control: Corn-SBM-DDGS diets with Antibiotics, and 2) Antibiotic Free; treatment 1 less the antibiotics with alternatives. Diets were formulated to meet or exceed the nutrient requirements for swine (NRC, 2012) during each phase. Each room contained 6 pens with 10 or 11 pigs/pen at a stocking density of 0.84 to 0.76 m²/pig. Diets were fed in nine dietary phases, four nursery phases and five 21-d grow-finish phases. Each room contained a separate ventilation system and two manure pits. Manure storage pits were initially charged with fresh water until the pit floor was completely submerged (average initial pit depth = 5.10 cm). There was a tendency for greater total final BW and total BW gain per manure pit when pigs were fed the control antibiotic treatment. No significant differences were observed between the two dietary treatments for manure volume (L), manure volume per

kg BW gain, or DM (g/kg BW gain), N (g/kg BW gain), and AmmN (g/kg BW gain) excretion. Manure pH tended to be lower for pigs fed the antibiotic free diet ($P < 0.06$) compared to the control diet. There were no differences observed for manure total C (kg), manure C per kg BW gain, manure C g/pig/d, and manure C g/pig wean-to-finish. Carbon intake was significantly higher ($P < 0.03$) for pigs fed the antibiotic free diets during nursery phase 1 and lower ($P < 0.01$) during nursery phase 4. The C intake for the remaining nursery phases (2 & 3) along with all three grower phases and two finisher phases and the overall wean-to-finish period was not significantly affected by dietary treatments. From the tested diets utilized in this trial, the antibiotic free diets had similar manure nutrient excretion and generation with lower manure pH which may affect transformation of N_2O during manure land application.

3.2. Introduction

Traditionally, antimicrobial agents have been added to feed and used extensively by livestock producers since their introduction in 1951 (Radostits, 1994) to help control the spread of infectious diseases and to enhance production efficiency. Much of the research evaluating the effects of antibiotics has indicated significant economic benefits from their use (Jukes et al., 1950; Brorsen et al., 2001; Cromwell et al., 2002). Due to their economic benefits, Cromwell (1991) estimated antibiotics were being used at sub-therapeutic levels in about 90% of nursery diets, 70% of grower diets, and 50% of finisher diets. Antimicrobials are also used at higher therapeutic levels to prevent disease in exposed animals and to treat infected animals (Cromwell, 1991). More current

estimates reported by the USDA (2006) estimated subtherapeutic-levels for nursery pigs at 85.3% and 81.2% for grow-finish diets. Additionally, research has shown that antibiotics have the potential to positively impact nutrient digestibility by reducing the concentration of bacteria (dependent upon antibiotic) in the gastrointestinal tract, reducing the production ammonia, lactic acid, and amines (Vervacke et al., 1979; Cromwell, 2001; Stewart et al., 2010). This reduction in bacteria concentration reduces competition with the host for nutrients, thus, it may increase the digestibility of nutrients by reducing the rate of passage (Gaskins et al., 2002; Kim et al., 2007; Stewart et al., 2010). This increase in nutrient digestibility can lead to a reduction in nutrient excretion.

However, growing concern about the perceived over-use of antimicrobial agents both in humans and animals and its contribution to the development of antimicrobial resistance has caused many governments around the world to consider it a major threat to public health. The European Union in 2001 launched an EU strategy to combat the threat of antimicrobial resistance by phasing out the use of antibiotics for non-therapeutic use in livestock and poultry by January 1, 2006 (EU, 2005). More recently, the FDA launched an initiative to phase out the subtherapeutic use of medically important antibiotics for livestock and poultry plus requiring the oversight of all remaining therapeutic antibiotic uses under written veterinarian direction (FDA, 2013). The concern over sub-therapeutic use of antibiotics isn't something new. Since its first use in feeds at sub-therapeutic levels, countless reports have attempted to link incidents of antibiotic resistance to food animals (Starr and Reynolds, 1951). Yet, today there has been no clear-cut evidence to definitively link the use of sub-therapeutic levels of antibiotics in feed with antimicrobial resistance in humans (Institute of Medicine, 1989; Regassa et al., 2008; NPPC, 2014).

This chapter will examine the effects of feeding standard Corn-SBM-DDGS diets with or without antibiotics, on manure generation, nitrogen (N), ammonium (NH₄), pH, and carbon (C) excretion.

3.3. Materials and Methods

The use of pigs for this experiment was approved by the Purdue University Animal Care and Use Committee (PACUC# 1112000447).

3.3.1. Experimental Design

Seven hundred twenty-three (York x Landrace; Temple Genetics, Inc.) maternal cross pigs were placed into eleven identical environmentally controlled rooms for a wean-to-finish study. Pigs were received in two delivery groups (group) two weeks apart from the same farm. Delivery 1 filled rooms 1-6 (3 rooms/treatment) and delivery 2 filled rooms 7-11 (2 control, 3 antibiotic free). Room 12 was used as an off-test room to house pigs that had to be removed from the study, primarily from the antibiotic free rooms where pigs were not treated with antibiotics. Pigs were allotted by sex and weight to one of two dietary treatments by room: 1) Control: Corn-SBM-DDGS diets with Antibiotics, and 2) Antibiotic Free; treatment 1 less the antibiotics, but with antibiotic alternatives. The antibiotics used and the alternatives used in the antibiotic free diets are detailed in Table 3.1. Diets were formulated to meet or exceed the nutrient requirements for swine (NRC, 2012) during each phase (Tables 3.2 – 3.5). Each room contained 6 pens with 10

or 11 pigs/pen at a stocking density of 0.76 to 0.84 m²/pig. Each pen contained two nipple waterers and two single-hole self-feeders. Diets were fed in nine dietary phases, four nursery phases (phase 1, 2, and 3 on feed budgets and phase 4 until d 42) and five 21-d grow-finish phases. Each room was independently controlled for ventilation and within each room there were 2 manure pits. Manure storage pits were initially charged with fresh water until the pit floor was completely submerged (average manure pit depth = 5.10 cm). Pigs were weighed individually along with feeders at diet changes to calculate performance and manure excretion per kg of BW gain. Pigs were marketed when the mean pig weight in a room was at a minimum of 125 kg.

3.3.2. Sample Collection and Analysis

A subsample of corn, SBM, and DDGS used in the diets that were fed was pooled and processed through a 1 mm screen in a Wiley mill prior to analysis. Carbon content of the following feed ingredients: Corn, Soybean Meal, and DDGS were determined by weighing out 50 ± 2 mg of sample into a tin capsule and assayed using a Flash EA 1112 Series Nitrogen-Carbon Soil Analyzer (CE Elantech, Inc. Lakewood, NJ). For other feed ingredients carbon content was calculated based on the equation in the swine NRC (2012).

Manure pit depths were taken at three separate locations per pit in the middle of the isle for each room at the center of each pen using a steel ruler with a metal extension arm. Pit depths were measured at d 0, 7, 14, 28, 42, 63, 84, 105, 126, 147, and marketing of pigs (d 147 or 153). Four vacuum manure column samples were taken in accordance to

Figure 1 for each pen (total of 12 samples from each pit). After the 12 vacuum samples (Figure 3.1 and Figure 3.2) were collected from manure each pit, samples were pooled, mixed thoroughly, and two, 250 mL subsamples were collected and placed in -20°C freezer for later analysis. After manure samples were thawed, samples were stirred and then sampled for analysis of DM, ash, TN, ammonium nitrogen (AmmN), carbon and pH. Manure DM was analyzed by drying overnight (12 + hrs) at 100°C using a forced air drying oven and then ashed (6 + hrs) at 600°C using a muffle furnace (Thermo Fisher Scientific, Inc. Waltham, MA). Manure TN and AmmN were determined by the Kjeldahl procedure (Nelson and Sommers, 1972). Manure pH was determined using an Orion 310 basic PerpHecT® LogR pH meter (Thermo Fisher Scientific Inc. Waltham, MA). Manure samples analyzed for carbon were oven dried for 72 h at 55°C and then processed with a mortar and pestle. Manure carbon was determined using a Flash EA 1112 Series Nitrogen-Carbon Soil Analyzer (CE Elantech, Inc. Lakewood, NJ).

3.3.3. Statistical Analysis

Data were analyzed for dietary effects using the GLM procedure of SAS (SAS Inst. Inc., Cary NC) which included group and body weight block as fixed effects in the model with room serving as the experimental unit. A P-value of ≤ 0.05 was considered significant and $0.05 < P \leq 0.10$ was considered a trend.

3.4. Results

3.4.1. Body Weights and Gain

No differences were observed for initial body weight (IBW) over pits and ADG (Table 3.6). However, there was a tendency ($P < 0.09$) for pigs fed the control diet to have a higher final body weight and total live weight gain over a manure pit. This tendency for pigs fed the control diet to have a higher final body weight can be contributed to the control pigs have a tendency ($P < 0.08$) to consume more feed and the fact that there were fewer pigs over each pit in the antibiotic free rooms by the end of the study. Overall, feed efficiency was significantly better when pigs were fed the antibiotic free diets ($P \leq 0.03$).

3.4.2. Manure Generation and Composition

The composition of stored manure is shown in Table 3.6. Overall, total manure volume (L), manure volume per kg BW gain and output of DM (g/kg BW gain) were not affected by the dietary treatments. However, a group effect and diet x group interaction was observed for all these variables. Pigs fed antibiotic free diets had lower manure volume (total and per kg BW gain) and DM per kg of gain in group 1 (1st set of pigs), but higher manure volume and DM generation in group 2 (Table 3.8). There were no significant dietary differences observed for manure N and AmmN (g/kg BW gain). Manure pH had a tendency ($P < 0.06$) to be more acidic when pigs were fed the antibiotic

free treatment. Pigs in group 2 had a significantly ($P < 0.04$) higher manure N (g/kg BW gain) and pH (Table 3.7).

3.4.3. Carbon Data

Dietary treatments had no effect on the percent carbon analyzed from manure pit samples. However, pigs in group 1 had a higher ($P < 0.04$) analyzed percent manure C than pigs in group 2 (Table 3.7). Interactions between diet and group ($P < 0.03$) were observed for total manure C (kg), manure C per kg BW gain (g/kg), manure C (g/pig/d), and total wean-to-finish manure C (g/pig). These interactions were the result of the control treatment having greater manure C values during group 1, but lower manure C excretion values in group 2 relative to the antibiotic free fed pigs. (Table 3.6).

The amount of C consumed during nursery phase 1 was significantly higher ($P < 0.03$) for pigs fed the antibiotic free diet while control pigs had greater ($P < 0.01$) C intake during the nursery 4 period and control fed pigs tended ($P < 0.11$) to have greater C intake during grower 2 and finisher 2 phases as well (Table 3.6). A significant replicate effect ($P < 0.04$) was observed with pigs in group 1 having higher C consumption during nursery periods 1, 2, 4 and a tendency to have greater ($P < 0.07$) overall during the nursery phase C consumption (Table 3.7). The amount of C intake between groups was significantly greater ($P < 0.04$) for group 1 in grower phase 1, with group 2 having the greater C intake ($P < 0.05$) for grower phases 2 and 3, finisher 2, and a tendency ($P < 0.06$) for greater C intake during the whole grow-finish period. No interactions were observed between diets and groups for grower 2 and 3, but grower phase 1 had a

tendency ($P < 0.08$) for group 1 pigs fed both diets having a higher C intake with greater reduction in C intake by group 2 control fed pigs than pigs fed the antibiotic free diet. Total carbon intake for the entire wean-to-finish period was not affected by dietary treatment or an interaction between diets and groups.

3.5. Discussion

Antibiotics have been widely accepted and used over the last 50 years in the livestock industry to prevent or treat infectious diseases. However, growing public concern over antibiotic resistance or “super bugs” has brought about questions regarding the safety of antibiotics being used as growth promotants in livestock and poultry. Many studies have examined the risk of developing cross-resistance of pathogens to antibiotics used in human medicine, but none have been able to find definitive results that would indicate a public health concern (Cromwell, 2001). Still, many countries have taken extensive action that has resulted in the restriction of antibiotic use (EU, 2005; Carlet et al., 2012). As a result, the livestock industry has actively pursued alternatives to antibiotics that can serve the same purpose as subtherapeutic antibiotics with similar performance.

Previous research has extensively examined the effects that antibiotics have on the growth performance of pigs at different stages of their life cycle. Results from most of those studies reported a significant improvement in growth performance when antibiotics were added to diets (Jukes, 1955; Clawson and Alsmeyer, 1973; Beames, 1969; Cromwell, 2001; Cromwell, 2002). It is believed that antibiotics are able to reduce the

microbial use of nutrients and microbial production of ammonia which is toxic to the animal (Coates, 1980; Anderson et al., 1999). In addition, antibiotics role in reducing N excretion has been reported at 5% or less (Han et al., 2001). However, Gaskins (2002) reported that when pigs were fed 10-50 ppm tylosin, apparent N digestibility and retention was significantly increased with N excretion being subsequently decreased by approximately 10%. Additionally, the supplementation of carbadox to swine diets has been demonstrated to improve apparent ileal digestibility of amino acids and nitrogen retention (Stahly et al., 1994; Partanen et al., 2001; Wang et al., 2005). Both of these antibiotics were used in our study.

A comparison of the initial body weight (BW), final BW, and BW gain demonstrated no significant differences in our experiment. The lack of growth response observed in this study is contrary to previous research with feeding antibiotics. One possibility of why the growth response for this trial wasn't as significant as previous studies could be contributed to the source of pigs used for this trial being from a relatively high-health herd. Research, has shown that high-health pigs that are placed into facilities with strict biosecurity don't demonstrate a growth response as well to antibiotics compared to pigs of lower-health status (Van Lumen, 2003). Additionally, several alternatives to antibiotics were used in the antibiotic free pigs that have been previously reported to have some potential as replacements giving similar or only slight reductions in growth performance (Walsh et al., 2007; Walsh et al., 2012).

When pigs were fed the antibiotic free diet, manure pH was reduced by 0.10 unit (control-pH 6.95; antibiotic free-pH 6.85). This decrease in manure pH has the potential to reduce N loss, therefore the volatilization loss of ammonia could potentially be

significantly reduced in antibiotic free rooms (Sutton et al., 1999; Rotz, 2004; Velthof et al., 2005). The reduction in manure pH could be correlated to the fact that the alternatives used in the antibiotic-free diets were more influential on the intestinal microflora thus reducing competition between the host lowering the pH. However, we did not observe any differences in manure N or AmmN, which would be the primary sources of aerial ammonia. The manure pH reduction may simply be related to the antibiotic free treatment use of water acidification during the first 2 nursery phases.

Emissions of N₂O from manure application are directly correlated to the amount of N and C applied (Velthof et al., 2003). Carbon, just like nitrogen, plays an important role in biological systems. The amount of carbon in swine diets will vary depending on the type of feedstuffs added. Research has demonstrated that the application of manure containing easily degradable C increases the risk of denitrification in the soil that is then transformed into N₂O (Paul and Beuchamp, 1989). Traditionally, the greatest proportions of ingredients added to swine diets are corn, soybean meal, and dried distillers grains with solubles (DDGS). For this study, samples of all three ingredients were analyzed for carbon content and used to determine the C composition of the diets fed, the total C consumed, and the C content of manure. Carbon consumption was approximately 22% higher for pigs fed the antibiotic free diet during nursery phase 1. Nursery phase 2 and 3 had no differences in C consumption. However, C intake for nursery phase 4 was reduced by nearly 12% in the antibiotic free fed pigs. Overall, there were no differences for total C intake for the nursery phases. These differences in nursery intakes are likely related to the water acidification early in the nursery period for the antibiotic free pigs and the

carbadox improving weight and intake in the later nursery phase. Carbon intakes during the grow-finish phases were fairly consistent across both treatments with total C intake from wean-to-finish being slightly higher (~4%) for pigs fed the control diet. The total C found in manure (pig) for wean-to-finish pigs was reduced by nearly 5% for pigs fed the antibiotic free diet. However, when manure C is expressed on a per kg of BW gain basis there was no difference between treatments. Although this reduction was not significantly different, the potential decrease in manure C has the potential to reduce CO₂ and CH₄ emissions due to the decrease in manure C that has the potential to be transformed.

3.6. Implications

The removal of antibiotics in wean-to-finish diets and using alternatives as potential replacements in these high health pigs has been demonstrated to be viable in having no negative impact on manure C, N, pH or total volume. From the tested diets utilized for this trial, the antibiotic free diets had the greatest potential in reducing N loss during land application because of the lower manure pH.

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Table 3.1. Antibiotic and Antibiotic Alternative use programs

Phase	Duration/Amount	Antibiotic program	Antibiotic free program
Nursery 1	1.36 kg/hd	Carbadox ¹ (55 ppm) + 3,000 ppm Zn from ZnO	Water Acidification ² + 3,000 ppm Zn from ZnO
Nursery 2	2.49 kg/hd	Carbadox ¹ (55 ppm) + 2,500 ppm Zn from ZnO	Water Acidification ² – (13 days total) + 2,500 ppm Zn from ZnO
Nursery 3	8.16 kg/hd	Carbadox ¹ (55 ppm) + 2,000 ppm Zn from ZnO	DFM ³ + 2,000 ppm Zn from ZnO
Nursery 4	Ad lib to day 42	Carbadox ¹ (55 ppm) + 189 ppm Cu from CuSO	DFM ³ + 250 ppm Cu from CuSO
Grower 1	21 days	CTC ⁴ – 110 ppm	DFM ³ + 126 ppm Cu from CuSO
Grower 2	21 days	Linco ⁵ – 110 ppm	DFM ³
Grower 3	21 days	Linco ⁵ – 44 ppm	DFM ³
Finisher 1	21 days	Tylan ⁶ – 22 ppm	Oregano ⁷
Finisher 2	21 days	Tylan ⁶ – 11 ppm	Oregano ⁷

¹Carbadox (Zoetis, Florham Park, NJ).

²Water Acid – Kemin Industires, Inc. KemSan Produced.

³DFM – Dupont, Danisco Animal Nutrition 75,000 CFU Bacillus/gram of feed.

⁴Chlortetracycline (CTC) – Aureomycin (Zoetis, Florham Park, NJ).

⁵Lincomix (Zoetis, Florham Park, NJ).

⁶Tylan (Elanco Animal Health, Greenfield, IN).

⁷Oregano-Regaon E (Ralco Animal Health, Marshall, MN).

Table 3.2. Ingredient and nutrient composition of nursery phase 1 and 2 dietary treatments.

Phase Dietary Treatments	Nursery 1		Nursery 2	
	Control	Antibiotic Free	Control	Antibiotic Free
<u>Ingredient, %</u>				
Corn, yellow dent	32.200	32.590	35.735	36.125
SBM, 47.5% CP	13.200	13.200	18.550	18.550
Soybean Oil	5.000	4.860	4.000	3.860
Limestone	0.730	0.730	0.570	0.570
Monocal. Phosphate	0.530	0.530	0.410	0.410
Vitamin premix ¹	0.250	0.250	0.250	0.250
TM premix ²	0.175	0.175	0.175	0.175
Phytase ³	0.100	0.100	0.100	0.100
Salt	0.250	0.250	0.250	0.250
Plasma protein	6.500	6.500	2.50	2.500
Spray dried blood meal	1.500	1.500	1.25	1.250
Soy concentrate	4.500	4.500	2.50	2.500
Fish meal	4.000	4.000	4.00	4.000
Dried whey	25.000	25.000	28.50	28.500
Lactose	5.000	5.000	-	-
Lysine-HCL	0.090	0.090	0.220	0.220
DL-Methionine	0.220	0.220	0.230	0.230
L-Threonine	0.040	0.040	0.100	0.100
L-Tryptophan	-	-	0.010	0.010
Carbadox-10 ⁴	0.250	-	0.250	-
Zinc oxide	0.415	0.415	0.350	0.350
Hemicell-HT 1.5 ⁵	0.025	0.025	0.025	0.025
Rabon ⁶	0.025	0.025	0.025	0.025
<u>Calculated Composition</u>				
ME, Kcal/kg	3,593.0	3,594.6	3,526.6	3,528.2
NE, Kcal/kg	2,746.2	2,746.0	2,674.3	2,674.1
Carbon content, %	39.33	39.38	40.75	40.80
Crude protein, %	24.17	24.21	22.98	23.01
Lysine, %	1.72	1.72	1.65	1.65
SID Lysine, %	1.55	1.55	1.50	1.50
SID Met, %	0.52	0.52	0.54	0.54
SID Met. + Cys., %	0.91	0.91	0.87	0.87
SID Thr, %	0.97	0.97	0.93	0.94
SID Trp, %	0.27	0.27	0.26	0.26
SID Ile, %	0.85	0.85	0.83	0.83
SID Val, %	1.10	1.10	1.00	1.00

Table 3.2. Cont. Ingredient and nutrient composition of nursery phase 1 and 2 dietary treatments.

Lactose, %	22.40	22.40	19.95	19.95
Calcium, %	0.85	0.85	0.80	0.80
Avail. Phosphorus, %	0.60	0.60	0.55	0.55
Zinc, ppm	2,991.0	2,991.0	2,522.0	2,522.0

¹Vitamin premix provided per kilogram of the diet: vitamin A, 6,615 IU; vitamin D₃, 661.5 IU; vitamin E 44.1 IU; vitamin B₁₂ 38.6 µg; riboflavin 8.8 mg; d-Pantothenic Acid 22.05 mg; niacin 33.1 mg.

²Available trace mineral per kilogram of the diet: Iron, 121.25 mg; Zinc, 121.25 mg; Manganese, 15.03 mg; Copper, 11.3 mg; Iodine, 0.46 mg; Selenium, 0.30 mg.

³Phytase activity level at 600 PU/kg (Phyzyme, Danisco Animal Health – Dupont)

⁴Carbadox (Zoetis, Florham Park, NJ) provided at 55 ppm.

⁵Hemicell HT (Elanco Animal Health, Greenfield, IN) – 1.5 enzyme provided at 0.08 mannase of the diet.

⁶Rabon larvacide (Bayer HealthCare LLC, Animal Health Division, Shawnee Mission, KS) provided at 19.3 ppm tetrachlorvinphos.

Table 3.3. Ingredient and nutrient composition of nursery phase 3 and 4 dietary treatments.

Phase Dietary Treatments	<u>Nursery 3</u>		<u>Nursery 4</u>	
	Control	Antibiotic Free	Control	Antibiotic Free
<u>Ingredient, %</u>				
Corn, yellow dent	45.653	45.928	49.04	49.27
SBM, 47.5% CP	25.132	25.108	30.066	30.00
DDGS, 7% fat	5.000	5.000	15.000	15.00
Choice white grease	-	-	2.000	2.00
Soybean oil	3.000	3.000	-	-
Limestone	0.847	0.847	1.355	1.355
Monocal. Phosphate	0.364	0.363	0.636	0.636
Vitamin premix ¹	0.250	0.250	0.250	0.250
TM premix ²	0.175	0.175	0.175	0.175
Phytase ³	0.100	0.100	0.100	0.100
Salt	0.300	0.300	0.350	0.350
Fish meal	4.000	4.000	-	-
Dried whey	14.000	14.000	-	-
Lysine-HCL	0.276	0.276	0.329	0.329
DL-Methionine	0.167	0.167	0.200	0.200
L-Threonine	0.087	0.087	0.060	0.060
Cu sulfate	-	-	0.075	0.100
Zinc oxide	0.375	0.375	-	-
Carbadox ⁴	0.250	-	0.250	-
Hemicell-HT 1.5 ⁵	-	0.025	0.025	0.025
Banmith dewormer ⁶ , 48	-	-	0.100	0.100
DFM ⁷	-	0.025	-	0.025
Rabon ⁸	0.025	0.025	0.025	0.025
<u>Calculated Composition</u>				
ME, Kcal/kg	3,442.2	3,450.8	3,385.8	3,392.7
NE, Kcal/kg	2,699.4	2,706.2	2,819.7	2,825.4
Carbon content, %	40.40	40.50	40.36	40.38
Crude protein, %	21.75	21.76	23.07	23.06
Lysine, %	1.46	1.46	1.43	1.43
SID Lysine, %	1.31	1.31	1.25	1.25
SID Met, %	0.47	0.47	0.46	0.46
SID Met. + Cys., %	0.76	0.76	0.73	0.73
SID Thr, %	0.81	0.81	0.77	0.77
SID Trp, %	0.22	0.22	0.22	0.22

Table 3.3. Cont. Ingredient and nutrient composition of nursery phase 3 and 4 dietary treatments.

Phase Dietary Treatments	Nursery 3		Nursery 4	
	Control	Antibiotic Free	Control	Antibiotic Free
SID Ile, %	0.80	0.80	0.81	0.81
SID Val, %	0.88	0.88	0.92	0.92
Calcium, %	0.80	0.80	0.75	0.75
Avail. Phosphorus, %	0.38	0.37	0.29	0.29
Zinc, ppm	2702	2702	-	-
Copper, ppm	-	-	189	252

¹Vitamin premix provided per kilogram of the diet: vitamin A, 6,615 IU; vitamin D₃, 661.5 IU; vitamin E 44.1 IU; menadione, 2.2 mg; vitamin B₁₂ 38.5 µg; riboflavin 8.82 mg; d-Pantothenic Acid 22.05 mg; niacin 33.1 mg.

²Available trace mineral per kilogram of the diet: Iron, 121.25 mg; Zinc, 121.25 mg; Manganese, 15.03 mg; Copper, 11.3 mg; Iodine, 0.46 mg; Selenium, 0.30 mg.

³Phytase activity level at 600 PU/kg (Phyzyme, Danisco Animal Health – Dupont).

⁴Carbadox (Zoetis, Florham Park, NJ) provided at 55 ppm.

⁵Hemicell – HT (Elanco Animal Health, Greenfield, IN) 1.5 enzyme provided at 0.08 MU/kg mannase of the diet.

⁶ Banimith dewormer (Phibro Animal Health Corp., Teaneck, NJ) provided at 480 ppm.

⁷DFM activity level at 75,000 CFU/g.

⁸Rabon larvacide (Bayer HealthCare LLC, Animal Health Division, Shawnee Mission, KS) provided at 19.3 ppm tetrachlorvinphos.

Table 3.4. Ingredient and nutrient composition of grower phase 1, 2, and 3 dietary treatments.

Phase Dietary Treatments	Grower 1		Grower 2		Grower 3	
	Control	AB ¹ Free	Control	AB ¹ Free	Control	AB ¹ Free
<u>Ingredient, %</u>						
Corn, yellow dent	55.308	55.222	56.148	56.234	59.505	59.523
SBM, 47.5% CP	15.259	15.270	9.622	9.611	6.596	6.593
DDGS, 7% fat	25.000	25.000	30.000	30.000	30.000	30.000
Choice White Grease	1.000	1.000	1.000	1.000	1.000	1.000
Limestone	1.527	1.527	1.519	1.519	1.450	1.450
Monocal. phosphate	0.265	0.265	0.024	0.024	-	-
Vitamin premix ²	0.125 ^a	0.125 ^a	0.150 ^b	0.150 ^b	0.150 ^b	0.150 ^b
TM premix ³	0.140	0.140	0.140	0.140	0.140	0.140
Salt	0.350	0.350	0.350	0.350	0.300	0.300
Lysine-HCL	0.570	0.570	0.579	0.579	0.548	0.548
DL-Methionine	0.081	0.081	0.040	0.040	0.028	0.028
L-Threonine	0.142	0.142	0.116	0.116	0.066	0.066
L-Tryptophan	0.032	0.032	0.048	0.048	0.034	0.034
Phytase ⁴	0.100	0.100	0.100	0.100	0.080	0.080
CTC ⁵	0.050	0.050	-	-	-	-
Lincomix ⁶	-	-	0.100	-	0.040	-
Cu sulfate	-	0.050	-	-	-	-
Zymanase ⁷			0.025	0.025	0.025	0.025
Hemicell-HT Enzyme ⁸	0.025	0.025	-	-	-	-
DFM ⁹	-	0.025	-	0.025	-	0.025
Rabon ¹⁰	0.025	0.025	0.038	0.038	0.038	0.038
<u>Calculated Composition</u>						
ME, Kcal/kg	3,363.0	3,360.5	3,364.8	3,367.4	3,372.6	3,373.2
NE, Kcal/kg	3,128.3	3,126.2	3,286.8	3,288.9	3,311.6	3,312.0
Carbon content, %	40.57	40.53	40.88	40.90	41.01	41.01
Crude protein, %	19.64	19.64	18.42	18.43	17.18	17.18

Table 3.4. Ingredient and nutrient composition of grower phase 1, 2, and 3 dietary treatments.

Phase Dietary Treatments	Grower 1		Grower 2		Grower 3	
	Control	AB ¹ Free	Control	AB ¹ Free	Control	AB ¹ Free
<u>Calculated Composition</u>						
Lysine, %	1.28	1.28	1.17	1.17	1.06	1.06
SID Lysine, %	1.10	1.10	0.98	0.98	0.88	0.88
SID Met, %	0.36	0.36	0.31	0.31	0.29	0.29
SID Met. + Cys., %	0.64	0.64	0.57	0.57	0.53	0.53
SID Thr., %	0.69	0.69	0.61	0.61	0.52	0.52
SID Trp., %	0.18	0.18	0.17	0.17	0.14	0.14
SID Ile., %	0.61	0.61	0.54	0.54	0.49	0.49
SID Val., %	0.74	0.74	0.68	0.68	0.63	0.63
Calcium, %	0.71	0.71	0.65	0.65	0.61	0.61
Avail. Phosphorus, %	0.25	0.25	0.22	0.22	0.21	0.21
Copper, ppm	-	126	-	-	-	-

¹AB = Antibiotic Free

^{2a} Grower 1 Vitamin premix provided per kilogram of the diet: vitamin A, 3307 IU; vitamin D₃, 331 IU; vitamin E 22.05 IU; menadione, 1.1 mg; vitamin B₁₂ 19.3 µg; riboflavin 4.41 mg; d-Pantothenic Acid 11.03 mg; niacin 16.5 mg

^{2b} Grower 2 & 3 Vitamin premix provided per kilogram of the diet: vitamin A, 3969 IU; vitamin D₃, 397 IU; vitamin E 26.5 IU; menadione, 1.3 mg; vitamin B₁₂ 23.2 µg; riboflavin 5.29 mg; d-Pantothenic Acid 13.23 mg; niacin 19.9 mg

³Available trace mineral per kilogram of the diet: Iron, 87.3 mg; Zinc 87.3 mg; Manganese 10.82 mg; Copper 8.14 mg; Iodine 0.33 mg; Selenium, 0.30 mg.

⁴Phytase activity level at 600 PU/kg (Grower 1 & 2); Phytase activity level at 480 FTU/kg (Grower 3) (Phyzyme, Danisco Animal Health – Dupont).

⁵Chlortetracycline (CTC) – Aureomycin (Zoetis, Florham Park, NJ) provided at 55 ppm.

⁶Lincomix (Zoetis, Florham Park, NJ) provided lincomycin at 110 ppm of diet during grower phase 2 and 44 ppm during grower phase 3.

⁷Zymannase (Elanco Animal Health, Greenfield, IN) included at a rate of 0.08 MU/kg β-glucanase & 0.10 MU/kg β-mannanase, respectively.

⁸Hemicell-HT 1.5X (Elanco Animal Health, Greenfield, IN) 1.5 enzyme provided at 0.08 MU/kg mannanase of diet.

⁹DFM activity level at 41.34 CFU/g of diet (Danisco Animal Health – Dupont).

¹⁰Rabon larvacide (Bayer HealthCare LLC, Animal Health Division, Shawnee Mission, KS) provided at 19.29 ppm tetrachlorvinphos for grower phase 1 and provided at 29.3 ppm tetrachlorvinphos for grower phases 2 and 3.

Table 3.5. Ingredient and nutrient composition of finisher 1 and 2 dietary treatments.

Phase Dietary Treatments	<u>Finisher 1</u>		<u>Finisher 2</u>	
	Control	Antibiotic Free	Control	Antibiotic Free
<u>Ingredient, %</u>				
Corn, yellow dent	63.172	63.172	77.146	77.146
SBM, 47.5% CP	3.068	3.068	4.315	4.315
DDGS, 7% fat	30.000	30.000	15.000	15.000
Choice white grease	1.000	1.000	1.000	1.000
Limestone	1.373	1.373	1.219	1.219
Monocal. phosphate	-	-	0.114	0.114
Vitamin premix ¹	0.125	0.125	0.125	0.125
TM premix ²	0.100	0.100	0.100	0.100
Salt	0.300	0.300	0.209	0.209
Lysine-HCL	0.533	0.533	0.434	0.434
DL-Methionine	-	-	0.016	0.016
L-Threonine	0.105	0.105	0.105	0.105
L-Tryptophan	0.043	0.043	0.038	0.038
Phytase ³	0.080	0.080	0.080	0.080
Zymanase ⁴	0.025	0.025	0.025	0.025
Tylan 40 ⁵	0.025	-	0.025	-
Oregano ⁶	-	0.025	-	0.025
Rabon ⁷	0.050	0.050	0.050	0.050
<u>Calculated composition</u>				
ME, Kcal/kg	3,378.8	3,378.8	3,396.7	3,396.7
NE, Kcal/kg	3,338.3	3,338.3	2,977.8	2,977.8
Carbon content, %	41.06	41.04	40.46	40.46
Crude protein, %	15.81	15.81	13.23	13.23
Lys, %	0.95	0.95	0.81	0.81
SID Lys, %	0.78	0.78	0.69	0.69
SID Met, %	0.24	0.24	0.22	0.22
SID Meth + Cyst., %	0.47	0.47	0.43	0.43
SID Thr, %	0.51	0.51	0.46	0.46
SID Trp, %	0.13	0.13	0.12	0.12
SID Ile, %	0.43	0.43	0.38	0.38
SID Val, %	0.57	0.57	0.50	0.50
Calcium, %	0.57	0.57	0.53	0.53
Avail. Phosphorus, %	0.21	0.21	0.15	0.15

¹Vitamin premix provided per kilogram of the diet: vitamin A, 3307 IU; vitamin D₃, 331 IU; vitamin E 22.05 IU; menadione, 1.1 mg; vitamin B₁₂ 19.3 µg; riboflavin 4.41 mg; d-Pantothenic Acid 11.03 mg; niacin 16.5 mg.

²Available trace mineral per kilogram of the diet: Iron, 48.5 mg; Zinc, 48.5 mg; Manganese, 6.01 mg; Copper, 4.52 mg; Iodine, 0.18 mg; Selenium, 0.30 g.

³Phytase activity level at 480 PU/kg (Phyzyme, Danisco Animal Health – Dupont).

⁴Zymannase (Elanco Animal Health, Greenfield, IN) included at a rate of 0.08 MU/kg β -glucanase & 0.10 MU/kg β -mannanase, respectively.

⁵Tylan 40 (Elanco Animal Health, Greenfield, IN) tylosin phosphate provided at 22.05 ppm of the diet.

⁶Oregano (Ralco Nutrition, Inc., Marshall, MN) provided at 250 ppm of the antibiotic-free diet.

⁷Rabon larvacide (Bayer HealthCare LLC, Animal Health Division, Shawnee Mission, KS) provided at 38.59 ppm tetrachlorvinphos of the diet.

Table 3.6. Effect of dietary antibiotics and antibiotic-free diets with alternative additives on manure generation and characteristics.

Dietary Treatment	Control	Antibiotic Free	SE	Diet	Grp	Diet X Grp
Initial BW over pit, kg	217.33	217.51	7.50	0.9856	0.6822	0.8920
Final BW over pit, kg	3964.72	3777.63	78.10	0.0890	0.1648	0.3453
Gain over pit, kg	3748.06	3560.76	73.46	0.0719	0.1312	0.3101
ADG, kg	0.787	0.785	0.032	0.7583	-	-
ADFI, kg	2.082	2.050	0.092	0.0823	-	-
G:F	0.377	0.383	0.016	0.0274	-	-
% Removals	3.0	8.3	-	-	-	-
<u>Manure</u>						
Volume, L	22675.24	21239.34	986.45	0.2875	0.0011	0.0314
Volume, L/kg BW gain	6.06	5.95	0.22	0.7219	0.0012	0.0029
DM, g/kg BW gain	503.72	518.78	20.60	0.5880	0.0012	0.0105
N, g/kg BW gain	45.62	45.15	0.90	0.6960	0.0411	0.2851
AmmN, g/kg BW gain	33.63	32.58	1.27	0.5424	0.7986	0.0866
pH	6.95	6.85	0.04	0.0582	0.0376	0.3016
<u>Carbon Data</u>						
Manure C, (%)	44.43	44.36	0.19	0.7676	0.0365	0.1567
Manure Total C, kg	909.83	864.61	42.19	0.4297	0.0678	0.0334
Manure Total C, g/kg BW gain	243.10	242.74	9.70	0.9785	0.0061	0.0032
Manure Total C, g/pig/d	188.00	182.90	7.02	0.5909	0.0015	0.0040
Manure Total C, g/pig (wean-to-finish)	27548.70	26234.86	1041.27	0.3544	0.0013	0.0066
C Intake, g/pig/d						
Nursery 1	7.53	9.60	0.67	0.0318	0.0339	0.3535
Nursery 2	30.43	34.10	2.19	0.2237	0.0001	0.2543
Nursery 3	128.46	127.78	4.89	0.9180	0.1824	0.6881
Nursery 4	216.20	191.40	6.49	0.0107	0.0388	0.4803
Grower 1	447.93	472.66	15.24	0.2378	0.0198	0.0799
Grower 2	727.31	682.38	19.54	0.1015	0.0446	0.2118
Grower 3	824.58	789.67	18.87	0.1811	0.0011	0.4322
Finisher 1	844.85	802.51	22.65	0.1768	0.1284	0.7805
Finisher 2	939.72	884.13	24.74	0.1088	0.0419	0.5612
Total C Intake Nursery, g/pig/d	382.62	362.89	11.96	0.2304	0.0720	0.6275
Total C Intake Grow-Finish, g/pig/d	3784.40	3631.35	84.94	0.1920	0.0638	0.9642
Total C Intake Wean-to-Finish, g/pig/d	4167.02	3994.24	94.86	0.1875	0.1431	0.9191

Table 3.7. Effect of group on manure generation and characteristics.

Group	Group 1	Group 2	SE	P ≤
Initial BW over pit, kg	219.49	215.35	7.50	0.6822
Final BW over pit, kg	3795.96	3946.39	78.10	0.1648
Gain, kg	3577.12	3731.15	73.46	0.1312
<u>Manure</u>				
Volume, L	19365.44	24549.15	986.45	0.0011
Volume, L/kg BW gain	5.42	6.59	0.22	0.0012
DM, g/kg BW gain	564.96	457.54	20.60	0.0012
N, g/kg BW gain	44.06	46.71	0.90	0.0411
AmmN, g/kg BW gain	33.32	32.88	1.27	0.7986
pH	6.84	6.96	0.04	0.0376
<u>Carbon Data</u>				
Manure C, (%)	44.68	44.11	0.19	0.0365
Manure Total C, kg	941.89	832.54	42.19	0.0678
Manure Total C, g/kg BW gain	263.19	222.65	9.70	0.0061
Manure Total C, g/pig/d	203.13	167.77	7.02	0.0015
Manure Total C, g/pig/d (wean-to-finish)	29577.75	24205.82	1041.27	0.0013
<u>C Intake, g/pig/d</u>				
Nursery 1	9.59	7.54	0.67	0.0339
Nursery 2	41.34	23.19	2.19	0.0001
Nursery 3	123.61	132.63	4.90	0.1824
Nursery 4	213.47	194.14	6.49	0.0388
Grower 1	486.39	434.20	15.24	0.0198
Grower 2	676.68	733.01	19.54	0.0446
Grower 3	757.55	856.70	18.87	0.0011
Finisher 1	799.66	847.70	22.65	0.1284
Finisher 2	875.73	948.13	24.74	0.0419
Total C Intake Nursery, g/pig/d	388.00	357.51	11.96	0.0720
Total C Intake Grow- Finish, g/pig/d	3596.00	3819.75	84.94	0.0638
Total C Intake Wean-to- Finish, g/pig/d	3984.00	4177.25	94.86	0.1431

Table 3.8. Effect of dietary antibiotics and antibiotic-free diets with alternative additives and group on manure generation and characteristics.

	<u>Control</u>		<u>Antibiotic Free</u>		SE	P ≤
	Group 1	Group 2	Group 1	Group 2		
IBW over pit, kg	220.08	214.58	218.90	216.13	11.85	0.8920
FBW over pit, kg	3839.27	4090.16	3752.65	3802.61	123.49	0.3453
Gain, kg	3619.84	3876.28	3534.39	3587.13	116.14	0.3101
<u>Manure</u>						
Volume, L	21622.12	23728.37	17108.76	25369.92	1559.71	0.0314
Volume, L/kg BW gain	6.00	6.13	4.85	7.06	0.36	0.0029
DM, g/kg BW gain	596.88	410.55	533.03	504.54	32.57	0.0105
N, g/kg BW gain	45.87	45.37	42.24	48.05	1.43	0.2851
AmmN, g/kg BW gain	35.38	31.87	31.26	33.90	2.01	0.0866
pH	6.87	7.04	6.82	6.88	0.06	0.3016
<u>Carbon Data</u>						
Manure C, (analyzed)	44.90	43.96	44.46	44.26	0.30	0.1567
Manure Total C, kg	1029.46	790.19	854.32	874.89	66.71	0.0334
Manure Total C, g/kg BW gain	285.62	200.58	240.77	244.72	15.34	0.0032
Manure Total C, g/pig/day	221.24	154.76	185.03	180.78	11.09	0.0040
Manure Total C, g/pig (wean-to-finish)	32381.77	22715.64	26773.73	25696.00	1646.39	0.0066
C Intake, g/pig/d						
Nursery 1	8.13	6.92	11.04	8.16	1.05	0.3535
Nursery 2	37.79	23.07	44.89	23.32	3.47	0.2543
Nursery 3	125.27	131.69	121.95	133.62	7.74	0.6881
Nursery 4	222.77	209.64	204.17	178.63	10.26	0.4803
Grower 1	492.88	402.99	479.91	465.41	24.09	0.0799
Grower 2	682.33	772.29	671.03	693.74	30.90	0.2118
Grower 3	764.95	884.22	750.15	829.19	29.84	0.4322
Finisher 1	816.59	873.12	782.73	822.28	35.82	0.7805

Table 3.8. Cont. Effect of dietary antibiotics and antibiotic-free diets with alternative additives and group on manure generation and characteristics.

	<u>Control</u>		<u>Antibiotic-Free</u>		SE	P ≤
	Group 1	Group 2	Group 1	Group 2		
C Intake, g/pig/d						
Finisher 2	913.23	966.21	838.22	930.05	39.12	0.5612
Total C Intake						
Nursery,	393.95	371.29	382.05	343.73	18.92	0.6275
g/pig/d						
Total C Intake						
Grow-Finish,	3669.97	3898.83	3522.04	3740.66	134.30	0.9642
g/pig/d						
Total C Intake						
Wean-to-	4063.92	4270.12	3904.09	4084.39	149.99	0.9191
Finish, g/pig/d						

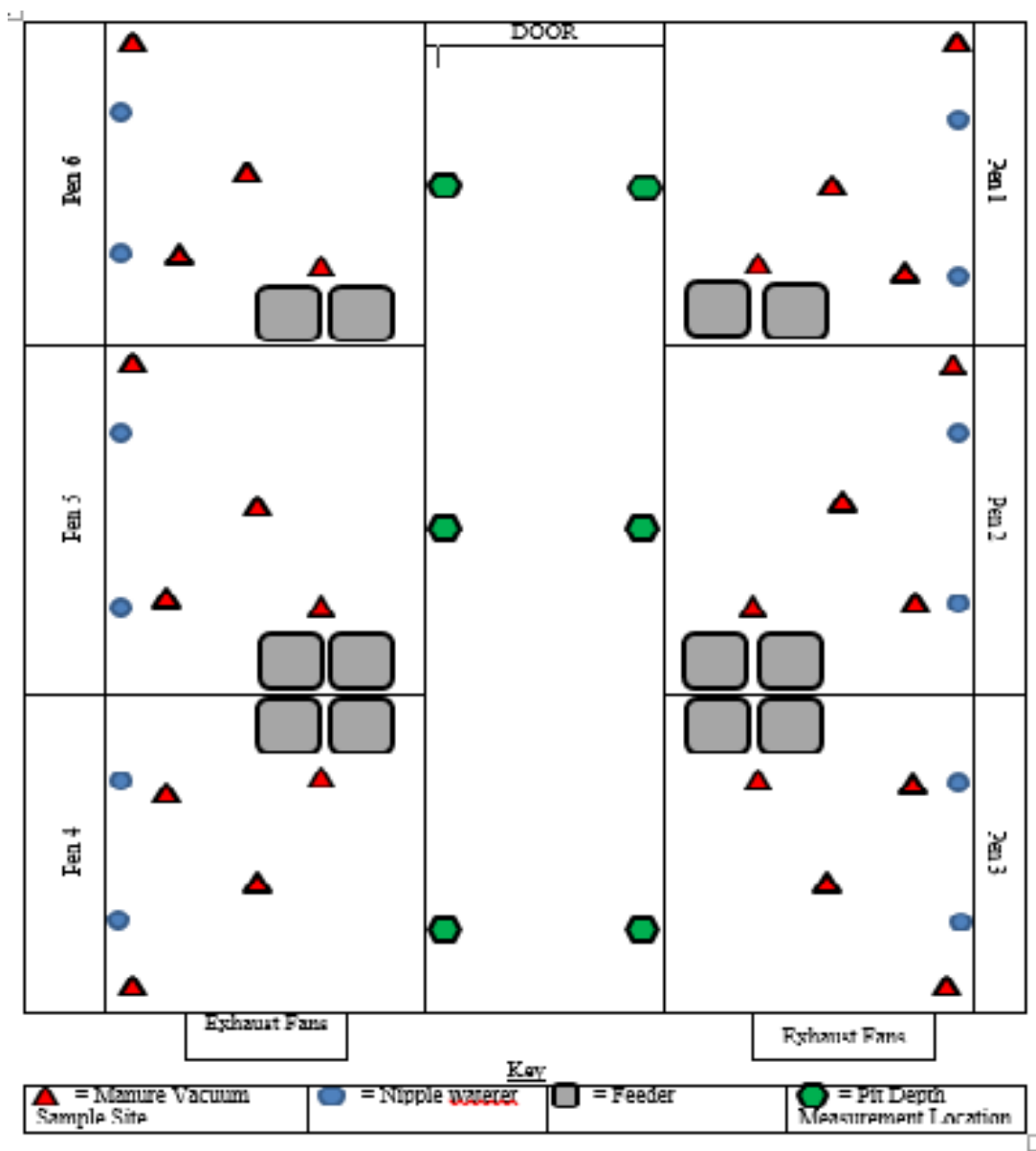


Figure 3.1. Manure Vacuum and Pit Sampling Layout.



Figure 3.2. Manure Sampling.

CHAPTER 4. SUMMARY

Dietary manipulation along with other management practices can serve as an effective way in reducing nutrient excretion. The reduction in dietary protein and the supplementation of synthetic AA has clearly been shown to significantly reduce N excretion (Kerr and Easter, 1995; Otto et al., 2003; Hinson, 2005). In addition to the reduction in N excretion, economic analysis on the dietary manipulation and management of N has been shown to be one of the more cost effective methods (Leneman et al., 1993). However, if dietary protein is lowered by more than three percentage units and AA supplemented, N retention has been reported to subsequently decrease (Carter et al., 1996; Kendall et al., 1999). What is unclear from the literature is the role in which reducing dietary protein affects C excretion.

The objective of the first study was to evaluate the effects of reducing dietary CP with supplementation of synthetic AA on N and C excretion, energy utilization, and fecal VFA concentrations. It was hypothesized that the reduction of dietary protein with the supplementation of synthetic AA to the seventh limiting AA should greatly reduce N and VFA excretions without effecting energy utilization. Our results suggest that formulating diets with lower dietary protein and supplementing with synthetic AA decreases N and

VFA excretions relative to pigs fed the control diet. However, E utilization and C excretion were negatively impacted.

Results from this study suggest that the manipulation of diets to reduce dietary CP is feasible and practical in reducing fresh N excretion in pigs and will aid in developing a model that will help predict losses between excretion and storage. However, the reduced C digestibility and increased C excretion with extremely low CP diets needs further research and how this may impact C:N ratios and potential plant availability of manure nutrients.

A second study was conducted to determine the impact that a feeding program with or without antibiotics in addition to alternatives has on manure generation, N, NH₄, pH, and C excretion. In this study, it was concluded that the removal of antibiotics in wean-to-finish diets with the use of alternatives as the potential replacements in high health pigs has been demonstrated to be viable in having no negative impact on manure C, N, pH, or total volume. The manure pH of antibiotic-free diets with alternatives was reduced leading to the potential to reduce N loss during land application, thus reducing the potential for volatilization loss of ammonia. Additionally, total manure C (g/kg BW gain) for wean-to-finish was reduced by nearly 5% when pigs were fed the antibiotic-free diet.

Overall, feeding reduced CP diets significantly reduced N excretion up to 45%. However, there were negative effects on DE, ME, and C digestibility at the highest inclusion level of synthetic AA. In addition, the removal of antibiotics in wean-to-finish diets and using alternatives as potential replacements in high health pigs has been demonstrated to be viable in having minimal impact on manure C, N, or total volume.

4.1. References

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