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AMINO ACID DIGESTIBILITY AND NITROGEN UTILIZATION OF CORN HYBRIDS FED TO GROWING PIGS

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AMINO ACID DIGESTIBILITY AND NITROGEN UTILIZATION OF CORN HYBRIDS FED TO GROWING PIGS

Ву

Janet Lynn Snow

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ABSTRACT

AMINO ACID DIGESTIBILITY AND NITROGEN UTILIZATION OF CORN HYBRIDS FED TO GROWING PIGS

By

Janet Lynn Snow

Protein quality of seven corns (high oil (HO), isogenic high oil (IHO), waxy (WX), isogenic waxy (IWX), high lysine (HL) and two yellow dent corns (STD 1, STD 2)) were investigated using growing barrows. Apparent ileal digestibilities (AID) for amino acids (AA) between the corn hybrids were investigated. Numerically higher AID resulted for HO and WX compared to IHO, IWX and STD 2 (P > .1). The AID were lower for HL compared to STD 1 ($P \le .1$) and similar to STD 2 (P > .1). In the second experiment, the barrows were fed a casein and a protein free diet to determine true ileal amino acid digestibilities (TID). Secretions of AA ($P \le .1$) were greater with casein compared to protein free for all essential AA (P > .1) and both treatments had TID close to 100 % for the majority of AA, therefore TID could not be determined. The objective of experiment three was to determine the corns' nitrogen utilization. Nitrogen (N) retention was similar between HO vs IHO, WX vs IWX and HL vs STD 1, but all hybrids were superior to STD 2 (P > .1). Overall, protein quality of HO and WX were superior compared to isogenics and STD 2 (P > .1). Significantly lower AID (P < .05) but similar N retention (P > .1) was reported for HL compared to STD 1.

I would like to dedicate this work to the people I love most: my husband and my two wonderful families. My husband helped me through the collections and the rigors of this degree. He was my loving supporter through it all and for that he deserves this dedication and my thanks. In addition, I dedicate this to my families who gave me continuous support and words of encouragement after long days and a lot of time spent apart.

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INTRODUCTION

Corn is the number one cereal grain produced in the U. S. and the major energy source in diets fed to livestock. Even though corn provides sufficient energy, a diet containing corn alone does not provide an adequate source of nutrients, especially for swine. The protein requirements in swine diets are commonly balanced by the addition of soybean meal and synthetic amino acids to corn. Specialty corns have the potential to be used in the swine industry as an alternative nutrient dense feedstuff, but the protein quality has not been evaluated. This question is addressed in the following chapters. Protein quality is characterized by the following components: the concentration and profile of amino acids, the digestibility, and utilization. Chapters one and two will specifically address the first two components of protein quality in high oil, high lysine and waxy corn. Protein utilization will be addressed in the third chapter. While some information exists on the nutrient profile of specialty corns, protein digestibility and utilization have not been studied extensively. As the swine industry moves towards formulation of diets on a digestible basis and the use of models, as was evident in the recently published National Research Council nutrient requirements for swine (1998), data regarding the protein digestibility and utilization in corn hybrids, with potential for swine, are essential.

LITERATURE REVIEW

Corn is the number one cereal grain used in livestock diets in the United States, but corn alone is not a adequate source of nutrients. Many researchers have developed corn hybrids with increased nutrient concentrations that offer advantages over standard yellow dent corn. Some examples of "value added" or "specialty corns" that currently exist are high oil, high lysine (opaque-2) and waxy corn. In the past, the production of these corns had not been feasible because of low yields and poor agronomic traits, but research and field tests have lead to corn with a more nutrient dense kernel and yields similar to conventional corn. This provides a potential nutritional tool for swine producers, but before the use of these hybrids can be advocated the protein digestibility and utilization must be researched.

Nutrient composition of the standard yellow dent corn

The corn kernel consists of four major parts: germ, endosperm, pericarp and tip cap. A standard kernel contains approximately 70 % starch, 8.5 % protein and 4.4 % oil (Boyer, 1994). Constituents of the kernel can be genetically manipulated to change the corn nutrient profile (Figure 1).

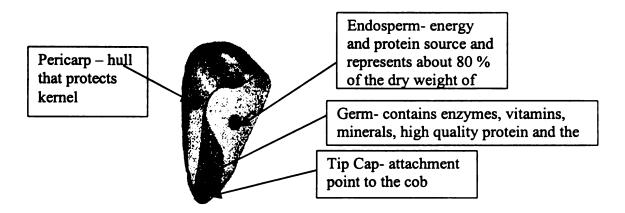


Figure 1: Diagram of a corn kernel

Adapted from the Ohio Corn Marketing Program web site (1997)

Cornstarch

Eighty to 90 percent of the total starch in standard corn is found in the endosperm (Boyer, 1994). Cornstarch exists in two major forms, amylose and amylopectin. Amylose, a linear starch, has primarily α -1, 4 linkages of D glucose units; amylopectin, a globular starch, contains small chains of α -1, 4 linkages with an α -1, 6 linkage every 20 to 25 glucose residues (Rooney and Pflugfelder, 1986). The α -1, 6 linkages of amylopectin create a randomly branched structure endosperm (Boyer, 1994). An intermediate starch polymer is also present in minute amounts, but is hard to identify because it has characteristics of both amylose and amylopectin. Amylose, amylopectin and the intermediate starch are packed together in tiny, insoluble granules in the kernel. The exact arrangement of the starch granule is unknown. Once mixed with water and heated, the breakdown of hydrogen bonds causes starch to become soluble. Gelatinization, the breakdown of the starch molecule by water, swells the molecule and increases its flexibility (White, 1994). Fats and protein can be bound to the starch

molecule and can hinder gelatinization (Rooney and Pflugfelder, 1986; White, 1994). The swelling of the starch allows the majority of amylose to leach out of the granule and exposes the starch to catabolic enzymes.

Corn proteins

The majority of the protein in standard corn is found in the endosperm and germ portions of the kernel. Standard kernel protein is composed of 7 % albumins, 5 % globulins, 52 % prolamines (zeins) and 25 % glutelins on a percent nitrogen basis (Boyer, 1994). Albumins, glutelins and globulins are the most soluble proteins in the kernel and provide a better balance of essential amino acids (AA) for animals than prolamines (Vasal, 1994). Prolamines contain disproportionately large amounts of glutamine, proline and alanine, which are non-essential AA in swine. The endosperm, the largest component of the kernel, may contain up to 80 % of the total kernel protein. The endosperm accounts for 60 % of the kernel prolamines and 34 % of the kernel glutelins. The germ, only 8-10 % of the dry matter of the kernel, consists of primarily albumins (60 %) and a small portion of prolamines (5-10 %). High quantities of prolamines in the whole kernel protein do not provide sufficient essential AA such as lysine and tryptophan for swine (Boyer, 1994).

Corn lipids

The standard corn kernel contains approximately 4.4 % oil, the majority of which is found in the germ (83-85 %) (Boyer, 1994). Endosperm, pericarp and tip caps, together, contain 10 to 13 % of kernel oil. Triacylglycerides, a mixture of unsaturated and saturated fatty acids, are the major component of corn oil. Corn oil is comprised of

50 % linoleic (18:2), 40 % oleic (18:1), 12 % palmitic (16:1), 2 % steric (18:0) and 1 % linolenic (18:3) acid (Boyer, 1994). Several fatty acids (FA) are essential for different species, but swine require linoleic and arachidonic acid. Arachidonic acid can be synthesized *in vivo* from linoleic acid, therefore only linoleic needs to be supplemented in swine diets. Corn oil is a high quality fat source because of the large percentage of linoleic acid and low linolenic acid (Boyer, 1994). The digestibility of fats is affected by the FA chain length and the degree of unsaturation. Short chain FA are easily digested, although, as the chain length increases to 20 carbons or more, the degree of unsaturation also increases, which augments the digestibility of the longer chain FA.

Specialty corns

Waxy Corn

Waxy corn, a genetic mutant discovered in China in the early 1900s, was differentiated because of its dull and waxy-like kernel appearance. This is caused by a single gene recessive mutation that occurs on chromosome 9 (Fergason, 1994). The homozygous recessive plants only produce the granular starch amylopectin. Other variations of the waxy mutant (Argentine waxy) produce small amounts of amylose (< 5 %). The waxy gene is epistatic to all known endosperm mutants, thus causing the absence of amylose (White, 1994). Today, the backcross method is the most popular breeding method for the waxy trait (Fergason, 1994).

Agronomic considerations

Waxy corn is not commercially produced by most seed corn companies for livestock feeds, although it is relatively competitive in yields when compared to standard

corn. Yield comparisons indicate that waxy corn differs by less than 5 % compared to normal yellow dent. Evidence exists that attests to waxy grain having a higher test weight than normal corn, which should offset the minimal yield differences (Fergason, 1994).

Practical applications of waxy corn

Currently, most production of starch from waxy corn is used in paper, textile, corrugating and adhesive industries. Waxy corn is not used in commercial animal feeds, and very little information exists on the nutritional value of waxy corn. Some research has shown a potential for waxy grain feeds compared to normal dent as demonstrated by increases in average daily weight gains in lambs (McDonald, 1973). In addition, McDonald also showed that waxy based diets had improved feed efficiency in finishing beef cattle. Carcasses of beef fed waxy corn had lower back fat and higher carcass grades (Fergason, 1994). Waxy corn has been tested in nursery pigs compared to yellow dent corn and found to have no effect on average daily gain, average daily feed intake, or feed conversion (Johnston, 1991). A grow-finish study using waxy and normal corn reported similar results as the nursery study, where no differences in performance between the waxy fed and non-waxy fed pigs were found (Hanson, 1946). Utilization of waxy corn has been minimal because its use has not been advocated and publicized. Therefore, there is little to no demand for production of this potentially valuable corn from the animal industry.

High lysine or opaque corn

Lysine and tryptophan are severely deficient in corn and breeding efforts have been targeted towards improving lysine and tryptophan content of corn to improve overall protein quality. Prolamine, the most abundant protein fraction in corn, is low in lysine and tryptophan. High lysine or opaque breeding programs have decreased the prolamine content and increased content of albumins, globulins and glutelins (Wilson, 1992). Developments in altering the prolamine fraction led to high lysine varieties with decreased prolamine content by approximately 50 % which in term amplified the remaining high quality protein types and increasing lysine content by 62 % (Boyer, 1994). Increases in other AA (percent increase in parentheses) such as tryptophan (30 %), histidine (17 %), arginine (33 %), aspartic acid (49 %) and glycine (20 %) has been reported to be associated with the high lysine corn (Pond and Maner, 1984; Vasal, 1994).

Inheritance of the opaque characteristics are achieved by a backcross system and the opaque kernels are then inter-mated further until a hybrid of desirable quantities of the high lysine and tryptophan is created. Discovery of this mutant spurred the search for new mutants with different alterations of the germ-endosperm ratio. Another effort has begun to attempt to combine mutants to create a more ideal hybrid (Ahmadi et al., 1995).

Agronomic considerations

Many different chromosomes with a completely recessive inheritance modulate opaque characteristics. The mutations affect the zein synthesis when the homozygous recessive genotype is present. All opaque mutants have three things in common, including low prolamine portion, a soft chalky endosperm (that is why it was named opaque), and a deficiency in the amount of dry matter produced (Boyer, 1994). In

addition, several poor agronomic effects are associated with the opaque gene, which include lower yields compared to standard corns due to lower endosperm size and reduced kernel weight. The soft chalky appearance of high lysine kernel is not desirable to producers and it causes lower kernel density. Finally, other problems such as increased moisture content, higher incidence of ear rot, greater damage by stored grain insect pests, poor germination, increased kernel breakage, thicker pericarp, larger germ size, and reduced cob weight have discouraged crop producers from planting this corn.

Practical applications of high lysine corn

Nutritionally, the opaque and high lysine varieties, in rats, have been shown to have a protein efficiency ratio (PER) similar to casein and a higher niacin availability, respectively. In pigs, high lysine corn is adequate as the only source of protein for the finishing, pre-gestation and gestation periods (Hawton et al., 1996). Burgoon et al. (1992) tested high lysine corn in starter and finisher pigs and found that it was not adequate for starter pig but was sufficient for finishing pigs. Cromwell (1984) investigated opaque-2 hybrids modified with single mutant genes (i.e. sugary-2, floury-2, waxy) and found that when these corn replaced normal corn, it tended to increase the rate and efficiency of gain in 14-kg pigs. Baker (1970) studied opaque-2, normal corn and corn-soybean meal based diets fed to first parity gilts during pregnancy and lactation and found that gilts receiving the opaque corn lost less weight during lactation and supported good postparturient performance of offspring when compared to those receiving the other treatments. Even with the agronomic disadvantages of the high lysine corn, the protein quality offsets the lower agronomic performance (West and Kincer, 1985). Grain and

livestock producers may find that growing and feeding high lysine corn might be less expensive than supplementing diets with soybean meal and synthetic AA.

High-oil corn

High-oil corn is characterized by oil contents greater than 6 % while normal corns usually range from 3.5 to 5 % (Lambert, 1994). The manipulation of the germ affects the rate of oil accumulation in the kernel from day 15 to 45 after pollination (Brunson et al., 1948). In addition to the increasing oil content, the composition also changes; the oil in corn was reported to contain higher concentration of linoleic acid (Alrefai et al., 1995). Alteration of the germ not only increases oil in high-oil corn but also the protein content. This is believed to occur because the genetics of high-oil corn also affects the germ to endosperm ratio.

The University of Illinois started one of the earliest breeding programs for highoil corn. Initially, a backcross system was initiated to augment the oil content of corn.
This system produced an oil content 20 % higher than the normal corn average (Sprague and Brimhall, 1948). Another breeding system recently developed by DuPont (Optimum, a trademark of DuPont), consists of a "topcross" system where a 90-92 % male sterile hybrid seed corn is mixed with 8-10 % high-oil pollinator. The top cross system usually equates to corn with oil contents of 6-8 %, similar to what is obtained with the backcross method.

Agronomic considerations

The major problem with the backcross genetics system is that yields are sacrificed at oil levels greater than 8 % (Lambert, 1994). Undesirable agronomic traits of the

backcross high oil corn have prevented planting and harvesting at the same rate as normal yellow dent. For example, high-oil corn has been reported to produce reduced ear length, smaller ear diameter, lighter kernel weight, reduced plant and ear height and early flowering. On the other hand, on a energy per acre or a lysine per acre basis, the added value of high oil corn may offset the disadvantages. In contrast, the top cross system has an advantage over backcross system because it has yields, ears, oil content and weights similar to normal corn (Hawton et al., 1996). In addition, the top cross system can easily incorporate other value added traits into the corn produced by altering the genetics of sterile corn seed.

Practical applications of high oil corn

Nutritional benefits of the high oil-corn are numerous. The energy per unit of feed being greater than normal yellow dent varieties could be useful in diets where fat is usually added to increase the energy density and palatability. Lactation and nursery diets may be an area where high-oil corn could have an impact (Trottier and Snow, 1997). As stated earlier, the increase in oil content is associated with an increase in protein resulting from an increase in the germ to endosperm ratio, therefore a higher protein quality (Lambert, 1994).

In swine, high oil corn has been tested in grower and finisher diets to evaluate the nutritional potential. Feed efficiency was increased with decreased feed intake in grower and finisher pigs (Orban et al., 1994). In a study by Nordstrom et al. (1972) the high oil corn reduced by 6 % the feed necessary to reach market. Gestating sows fed high-oil corn diets during the last 30 d of gestation gained more weight and had higher fat levels in colostrum, but no effects on litter size, birth weight or weaning weights were found

(Boyer, 1994). Average daily gain was shown to increased by 9 % in pigs fed diets containing high oil corn as compared to pigs fed normal corn diets (Pettigrew and Yang, 1997).

These studies indicate that kernel oil is as readily used as the normal fats added to diets and yet use of high-oil corn eliminates many problems associated with the addition of animal fats to diets. Diets using high oil corn have also seen benefits besides the nutritional applications. Diets including high oil corn have had increased storage time and feed flow from feed storage bins and feeders (Adeola and Bajjalieh, 1997). Although, pigs utilize extracted fats more efficiently than in-seed fats, Adams and Jensen (1984) noted that it was much easier to handle high oil corn. Also, types of oils fed to weanling pigs to improve dry matter digestibility were investigated. It was determined that corn oil possessed the highest digestibility compared to lard and tallow (Cera et al., 1988). The only detrimental effect to feeding high-oil corn was shown when finishing pigs were fed diets with large inclusions rates of high oil corn, fat quality was "soft" and of poor appearance (Monahan et al., 1992). On the other hand, a 15 % inclusion of high-oil corn in the diet did not produce soft fat in a study published by Nordstrom (1972).

Added value corns such as waxy, high lysine and high oil have potentially great advantages over the normal yellow dent variety in swine diets. Waxy corn maybe beneficial with its increased digestible starch. Opaque or high lysine corn provides a better balance of AA and could decrease the inclusion rate of synthetic AA and soybean meal. The higher oil and protein content in the high-oil corn could also decrease the inclusion rate of synthetic AA and eliminate the need to add fat to diets.

Protein digestibility and utilization in swine

Minimal data has been published on the protein digestibilities of specialty com hybrids. Carr (1992) looked at the apparent protein digestibilities of high-oil com compared to normal yellow dent. The essential AA digestibilities are summarized in Table 0.1. High-oil and waxy corn were evaluated for protein digestibilities and found that the digestibility coefficients were similar between corn hybrids (Soltwedel, 1994). Results of essential AA digestibilities from Soltwedel (1994) are summarized in Table 0.2. Only two studies have looked at the protein utilization of high oil corn, but waxy corn has not been evaluated for nitrogen utilization (Adeola and Bajjalieh, 1997; Carr, 1994). Limited data are available on the protein utilization and digestibility of high lysine corn. Burgoon (1992)looked at high lysine corn storage and moisture effects and reported a few values for nitrogen utilization and digestibility, but no other research has been done. It is evident that more research is necessary before the true value of these potentially valuable corn hybrids in swine diets is known.

Digestibility of AA can be measured by two different methods. One is the direct and the other is the indirect difference method. The direct method usually involves cereal grains that pigs will willingly consume when formulated into single feed ingredient diets. The single ingredient supplies the only source of dietary protein therefore the digestibility can be directly determined by the difference between the amount ingested and the amount at the terminal ileum. The indirect difference method is usually used with feedstuffs that are unpalatable or cannot be investigated with the direct method. Digestibility is measured indirectly by the addition of a cereal grain to make a complete diet along with using a diet that is primarily comprised of the cereal grain. Then, by difference, the

digestibility of the test feedstuff can be determined. Also, digestibility can be measured by obtaining two types of samples, feces and digesta from the terminal ileum. It has been stated that the use of ileal digesta is the preferred and most accurate site to determine digestibility (Moughan and Smith, 1985; Sauer et al., 1989). Reasons for the preference of measuring ileal digestibility values instead of fecal is because of the microbial activity in the colon alter AA resulting in higher fecal digestibilities than the ileal method (Moughan and Smith, 1985; Sauer et al., 1977). In addition, the ileal method was shown to be more sensitive for measurement of AA in grains (Taverner and Farrell, 1981). Using either the direct or indirect difference method leads to three types of digestibility coefficients that can be calculated which are discussed in the next section (Sauer et al., 1989).

Factors affecting amino acid digestibility

Digestibility can be affected by many factors that can potentially increase or decrease the AA digestibilities. Some of the factors include age, fiber content, particle size of the diet, protein intake, antibiotics (den Hartog et al., 1989; Donkoh and Moughan, 1994; Laplace et al., 1989; Sauer et al., 1989; Stein, 1998). Stein found that different age pigs particularly, growing pigs and sows in different physiological states, had different digestibility coefficients. Fiber was shown to decrease AA digestibilities Laplace (1989), Grala (1998) and Schulze (1994). Wondra et al.(1995) demonstrated that decreasing particle size increased protein digestibility. Protein intake is also been reported to increase apparent AA digestibility by Donkoh and Moughan (1994) and Sauer et al (1989). Some antibiotics have also been shown to increase apparent AA digestibility by decreasing microbial activity (den Hartog et al., 1989).

Mearsurements of digestibility: apparent digestibility

Apparent digestibility measures the difference between the AA ingested minus the AA in the ileal digesta. The main disadvantage of measuring apparent digestibility underestimates the actual digestibility of a feedstuff because it does not take into account endogenous nitrogen secretions. However, many researchers report apparent digestibility only due to the labor and complications associated with trying to estimate endogenous nitrogen (Leterme et al., 1991; Southern, 1991)

True digestibility

True digestibility modifies the apparent digestibility measurement by taking into account the endogenous nitrogen (Stein, 1998). Therefore, true digestibility would be the most accurate estimation of digestion of the AA by taking samples at the ileum. But, as mentioned earlier, the lack of accurate methods results in many complications with estimating endogenous losses.

Methods for estimating endogenous losses

Endogenous secretions are proteins that originate from saliva, gastric secretions, pancreatic juice, bile acids, and intestinal secretions (Grala et al., 1998). Estimation of endogenous losses in the gut can be measured by several methods. The regression method utilizes the feeding of incremental levels of crude protein (CP) in several diets to determine a linear relationship between CP intake and the percent nitrogen in the digesta. Once the linear relationship is established, the y-intercept, which is equal to endogenous losses, is extrapolated (Fan and Sauer, 1994). This method assumes that endogenous losses are constant with varying CP levels (de Lange et al., 1989). Another method is

feeding a protein free diet (de Lange et al., 1989). This diet is fed and the ileal samples are collected and analyzed for endogenous losses (Donkoh and Moughan, 1994). The problem with this method is that the absence of protein *per se* may not be reflective of the endogenous secretions produced by a protein-containing diet. Therefore, it assumes that there is no relationship between CP intake and endogenous losses (Southern, 1991). For this reason, the protein free diet is now more commonly used to evaluate the non-diet specific endogenous losses that are discussed below. A casein diet is another method for estimating endogenous losses where the animal is fed a diet that contains a readily available protein source: casein (Chung and Baker, 1992). The procedures are the same as for a protein free diet. In this case, casein is assumed to be absorbed completely and the animal maintains a close to normal plane of nutrition (Stein, 1998).

Factors that affect endogenous secretion

Increased dry matter intake, specifically increased protein and fiber levels, was shown to augment endogenous nitrogen secretion (Sauer et al., 1977). Moreover, increased fiber intake increased the total dry matter flow at the terminal ileum, reduced nitrogen utilization, and resulted in a increase endogenous and undigested dietary nitrogen (Schulze et al., 1995). A study by den Hartog (1989) showed that increased fiber in protein free diet increased the endogenous nitrogen at the terminal ileum and was thought to increase sloughing off of mucosal cells and increase mucus production. In addition, dietary protein source and intake was shown to be positively related to endogenous nitrogen secretions (den Hartog et al., 1989). It has also been shown, in the same review, that increased poorly digested proteins in diets resulted in increased enzyme secretion.

Standardized ileal digestibility

In the past decade, new research from France has addressed the problems associated with measurement of true digestibilities and estimation of endogenous losses. They have proposed the idea of standardized ileal digestibilities (SID). Standardized ileal digestibilities separate endogenous losses into two categories: specific endogenous losses and non-specific endogenous losses (Jondreville et al., 1995). Specific endogenous losses are related to the nutrient quality of a feedstuff. For example, if a feedstuff is high in fiber the specific losses would be related to the amount of fiber in the feedstuff. Non-specific endogenous losses are influenced only by the amount of dry matter intake. At the Technical Institute for Cereals and Forages (ITCF, Paris, France), non-specific endogenous losses are typically estimated by feeding a protein free diet. Consequently, the feedstuff specific endogenous losses are estimated with a model approach. With these two endogenous estimations they can modify the apparent digestibilities to standardized ileal digestibilities.

Ileal cannulation

Cannulation of the terminal ileum is a common procedure for obtaining ileal digesta samples. Re-entrant cannulas, one of the first methods for obtaining ileal digesta, (Sauer et al., 1989) need no dietary markers to estimate the percentage of sample obtained because of a total diversion of the digesta (Figure 2). This total diversion of the gastrointestinal tract can influence the normal functioning of the small intestine(Easter and Tanksley, 1973). In addition, the surgery to place the cannula is intricate and disrupts the electrochemical pulses in the intestine (Sauer et al., 1989). However, this

older method is not used as commonly today as the simple T cannula and the ileo-cecal shunt.

The simple T cannula method requires a dietary marker because only a portion of the digesta is sampled (Figure 3; (Gargallo and Zimmerman, 1980; Hamilton et al., 1985). The surgery is less stressful on the gastrointestinal tract compared to the reentrant method (Sauer and Ozimek, 1986; Stein et al., 1998). The functioning of the small intestine is not impaired. It has become the most common method for measuring feedstuff ileal digestibility in pigs and dogs (Hill et al., 1996) (Muir et al., 1996). The simple T method seems to be the best for obtaining samples from the ileum.

Within the last ten years, a method called the ileo-cecal shunt has been developed. This procedure separates the ileum from the cecum and attaches it to the rectum. This way digesta samples can be collected directly from the anus. This method is commonly used in European countries where digestibility trials are frequently performed.

Conclusion

From the above literature review, it is evident that there are many nutrient advantages for the specialty corn hybrids. Although, the amount of swine research available on the digestibility and utilization of these hybrids is scarce, the potential these corns offer to swine nutrition warrant further investigation. Protein digestibility and utilization of the corns must be researched more extensively before they can be advocated for use in swine diets. Several methods have been identified in this review to enable the investigation of protein metabolism. Therefore, it was the objective of this thesis to examine the protein digestibility and utilization of high oil, waxy and high lysine corn

Small Intestine

Figure 2: Re-entrant cannula

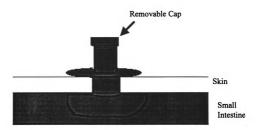


Figure 3: Simple T cannula

Table 1: Apparent amino acid digestibilities for normal and high-oil corn based on swine ileal digesta collections (Adapted from Carr, 1994)

Essential Amino Acid	Normal corn	High-oil TC-2	Difference	P-value	SEM
Arg (%)	65.26	72.62	7.36	.005	1.53
His (%)	69.07	74.14	5.07	.021	1.52
Ile (%)	73.95	77.41	3.46	.045	1.30
Leu (%)	70.31	76.25	5.94	.015	1.62
Lys (%)	83.99	83.84	-0.15	.897	1.10
Phe (%)	74.93	80.95	6.03	.004	1.20
Thr (%)	72.82	73.31	0.49	.759	1.50
Val (%)	67.23	72.48	5.26	.018	1.51

Table 2: Apparent digestibilities of amino acids (Adapted from Soltwedel, 1996)

Essential Amino Acid	Normal (2560)	High-oil / Waxy (X571-20)	High-oil (X577-3)	High - oil (X577-14)	Waxy	High-oil (7003.08)	SEM
Arg (%)	88.72	72.99	76.20	80.05	71.56	72.10	2.85
His (%)	70.69	72.56	72.87	73.11	66.72	67.77	1.83
Ile (%)	75.95	76.44	75.79	76.97	72.33	72.76	1.45
Leu (%)	73.80	76.96	75.91	77.31	69.00	71.55	1.89
Lys (%)	82.43	83.20	84.21	85.08	82.97	83.54	0.69
Met (%)	86.29	81.98	86.66	76.84	81.51	81.35	1.06
Phe (%)	80.12	73.66	81.75	75.23	73.28	70.22	1.68
Thr (%)	70.17	68.36	76.91	72.44	66.60	65.43	1.41
Trp (%)	78.42	59.79	77.43	77.47	74.47	78.52	1.23
Val (%)	73.32	72.09	72.63	72.98	66.87	68.47	1.75

CHAPTER 1: APPARENT AND TRUE ILEAL AMINO ACID DIGESTIBILITIES OF CORN HYBRIDS FED TO GROWING PIGS

Abstract

Four barrows (52.8 \pm 1.7 kg) were used consecutively in two 4 x 4 Latin squares to compare ileal amino acid digestibilities between different corn hybrids, (high lysine (HL) vs standard yellow dent 1 (STD 1), high oil (HO) vs isogenic high oil (IHO), waxy (WX) vs isogenic waxy (IWX), and all specialty hybrids vs standard yellow dent two (STD 2)). Each diet consisted of a test corn hybrid (96.62 %), dicalcium phosphate (1.25 %), limestone (0.88 %), vitamin premix (0.50 %), trace-mineralized premix (0.25 %), salt (0.25 %), chromic oxide (0.25 %) and an amino acid top dress to meet or exceed NRC (1988) amino acid requirements for growing pigs. Diets were offered in three equal daily meals for a period of seven days and amino acid top dresses were discontinued on d 5. A casein diet was fed at the end of the second square to estimate endogenous losses. Apparent and true amino acid digestibilities were determined for all seven hybrids. All amino acid digestibility coefficients were higher for HO and WX compared to their isogenics but not statistically different (P > .1). The amino acid digestibility coefficients were lower for the HL corn compared to STD 1 ($P \le .1$). All protein and amino acid digestibilities were superior in the HO and WX compared to STD 2 (P > .1). High lysine corn had superior digestibilities to STD 2 in Arg, His, Lys, Met, Gly, and Proline (P > .1). Overall, STD 1 and HO corns had the highest amino acids digestibility coefficients of all the hybrids studied.

Introduction

Swine nutritionists are moving toward diet formulation on a digestibility basis in an attempt to better meet the pig's nutrient requirements despite the lack of information regarding digestibility coefficients for common feedstuffs (1998). Currently, digestibility coefficients for amino acids (AA) exist for the yellow dent corn, but there is no published data available on AA digestibilities of novel feedstuffs such as "value added" or "specialty" corns. Several genetically engineered corn hybrids such as high lysine (opaque-2), waxy and high oil corns have been suggested to offer nutritional advantages in swine diets over standard corns (Pond and Maner, 1984) but no research has been performed on their nutrient availability and utilization. The goal of this experiment was to measure the apparent and true ileal amino acid digestibility of high lysine, waxy and high oil corns in growing pigs.

Materials and Methods

Six corn hybrids high oil (HO; Cargill 5990 TC), isogenic high oil (IHO; Cargill 5990), waxy (WX; Pioneer 3528E), isogenic waxy (IWX; Pioneer 3527), high lysine (HL; Crows), and yellow dent 1 referred as standard one (STD 1; Crows) were obtained from different seed companies to investigate their amino acid digestibility in growing pigs. Three seed companies provided a specialty hybrid and its respective isogenics. The HL corn did not have an isogenic so the company provided a standard corn with genetics similar to the HL corn. These corns were planted at Michigan State University (MSU) farms in 1200 ft² adjacent plots. The plots were monitored throughout growth for dry matter, ear density and plot density. The centers of the corn plots were harvested

individually, at approximately 25 % moisture, using buffer rows to maintain identity of the hybrids. All hybrids were commercially dried and shipped back to MSU for grinding in a Raymond Mill (No. 82; Chicago, IL) and mixed into experimental diets. A second standard yellow dent corn (STD 2) was purchased to represent a normal industry corn which had no known genetic similarities to the six test hybrids.

The Michigan State University All University Committee for the Use and Care of Animals approved all animal protocols. Initially seven Yorkshire pigs, with and initial body weight of 40 kg, were fitted with simple T cannulas for a 7 x 7 Latin square design. Cannulas were surgically inserted and maintained according to Stein et. al. (1998). Barrows were moved to individual pens (1.5 m x 2.75 m) and allowed fourteen days for recuperation after surgery. Daily feed intake, body temperature, and integrity of the cannula were monitored. Ampicillin was administered for the first three days post surgery. All pigs were provided free access to water. After three pigs lost their cannulas during the initial sampling period the pens were modified by adding smooth siding to further ensure the integrity of the cannulas. As a result the four remaining littermate barrows (52.8 \pm 1.7 kg) were rearranged into two consecutive 4 x 4 Latin squares. The barrows were fed each of the seven diets with the STD 2 diet repeated in both squares to allow pre-planned comparisons of the corns to be made across the two squares. The pigs were allowed five days to adjust to the diets. On days six and seven of each period, digesta from each pig was collected for 12 hours. Pigs were weighed weekly to monitor growth. Environmental temperature of the room ranged from 16 - 29°C with a mean temperature of 21°C.

Diets

Experimental diets consisted of one of the respective test corns included at 96.62 % as the sole source of protein and energy. Vitamins, minerals, dicalcium phosphate, limestone and salt were included at 3.13 % of the diet. All vitamins and minerals met or exceeded NRC (1988) requirements. Chromic oxide was included in the diet as an indigestible marker at 0.25 %. Barrows were provided ad libitum access to feed during the post-surgical recovery period and then restricted to 90 % of ad libitum intake during the experiment. Diets were offered in three equal daily meals. Amino acid top dresses were added to the daily feed allotments during the adjustment period to meet the amino acid requirements for the five deficient amino acids: isoleucine, lysine, tryptophan, methionine and threonine (NRC, 1988). Amino acid top dresses were removed one day prior to sampling of ileal digesta. At the end of the second Latin square, a casein diet was fed to all four pigs at the same time to estimate endogenous nitrogen secretion at the terminal ileum. The casein diet was a semi-purified diet consisting of 71.43 % cornstarch, 11.75 % casein, 5.0 % Solka Floc, 4.0 % corn oil, 4.0 % sugar, 2.25 % dicalcium phosphate, 0.32 % limestone, 0.50 % vitamin premix, 0.25 % trace mineral mix, salt and chromic oxide (Table 6).

Sample Preparation and analysis

Digesta for each pig was homogenized and pooled over the two-day collection and stored at -20 °C until samples were prepared for analysis. Digesta samples were thawed, freeze-dried and finely ground using a coffee grinder for dry matter, crude protein, amino acid, and chromium analysis. Dry matters of the corns, digesta and diets were determined using a vacuum oven at 60 °C for twelve hours. A Hach-kjeldahl was

used to determine crude protein (Hach et al., 1987). Particle size was determined using weight distribution over standard U. S. sieves according to Ensor et. al.(1970). Gross energy of the corns and diets were determined via adiabatic bomb calorimetry (Parr Instrument Co., Moline, IL).

Amino acid determination was performed using the Pico•Tag ® method (Waters Co., Milford, MA) which included acid hydrolyzation in an autoclave for 24 hours at 110 °C. Samples were then brought up to volume and filtered. The amino acid filtrate was then subsampled and dried using vacuum centrifugation. The AA hydrolysate was reconstituted, dried again, derivatized with PITC® and separated using a Waters high pressure liquid chromatographer (Waters, Co., Milford, MA) fitted with a 15 cm hydrolysate column.

Chromium (Cr) was determined at two different laboratories. At Michigan State University, a wet ash procedure was employed to oxidize the Cr (Bolin, 1952). One gram of sample was digested using concentrated nitric acid and heat. After digestion perchloric acid was added to oxidize the Cr ³⁺ to Cr ⁶⁺. Samples were then weighed, diluted and shipped to a commercial laboratory to be analyzed by atomic absorption spectrophotometry (University of Illinois, Urbana, IL; see appendix 2 for the AA digestibility values using these results). Due to complications with the previous mentioned procedure, a commercial laboratory also performed Cr analysis and these values were used to calculate the digestibilities reported in this chapter (University of Missouri Agriculture Experiment Station, Columbia, MO; (AOAC, 1990)).

Apparent and true amino acid digestibilities and endogenous amino acid losses were calculated according to Stein (1998) and these calculations are summarized and included in appendix 1.

Statistical analysis

Data was analyzed using both GLM and MIXED procedures of SAS (1990) in a Latin square design. Data from the two squares were pooled and STD 2 corn was used as the reference corn. A linear model with main effects of square, collection nested within square, diet and pig as a random variable were used in the statistical model. LSMEANS with stderr and pdiff options in GLM and pdiff option in MIXED were used to determine difference between the following preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1, HO vs STD 2, WX vs STD 2 and HL vs STD 2. Both analysis were evaluated and the MIXED procedure was used for statistical difference because of the multiple sources of variance in the model and MIXED ability to handle negative variance.

Results

The pigs remained healthy during this experiment and had an average daily gain of 0.45 kg/d and an average daily feed intake of 2.70 kg/d. Table 3 represents the nutrient compositions of the seven corn hybrids. Dry matter, gross energy and ash were relatively similar across all corn hybrids. The major differences in nutrient concentrations were seen in crude protein and ether extracts. Crude protein (CP) ranged from 8.36 to 10.81 % in the hybrids with the STD 1 and the HO having the first and second greatest protein content, respectively. The HO also had the greatest ether extract at 7.93 % while all of the corns ranged from 3.97 to 5.44. Amino acid contents of the

corn hybrids were also determined. The highest lysine contents were found in the HL and STD1 corns at 0.36 and 0.29 %, respectively. Arginine, methionine, threonine and valine concentrations ranged from 0.45 to 0.61, 0.19 to 0.24, 0.26 to 0.36, and 0.43 to 0.54 %, respectively. These amino acids are reported as a percentage of the total CP in table 4. Glutamic acid and leucine are the most abundant amino acids in the corns providing approximately 17 % and 10 % of the total protein, respectively. High lysine corn has a higher percentage of lysine (4.14) than the rest of the corn hybrids (3.23-2.57). Arginine content in the HL corn is 1.26 % greater than the next highest concentration that is in the STD 2 corn. Also, HL corn has less leucine as a percentage of total nitrogen at 7.24 % and the other hybrids have 10 - 11 %. The other essential amino acids as a percentage of CP are similar across the hybrids. The mean particle size for each of the corn hybrids are reported in Table 5. The particle sizes were similar across all corn and ranged from 0.81 to 0.95 mm. Dry matter was comparable across all diets (Table 7). Inclusion of the corn hybrids in the diets resulted in CP contents ranging from 8.27 to 10.93 %. Crude protein content of the casein diet was 12.06 %. Gross energy was highest in the HO diet at 4.51 Mcal/kg whereas all the other diets contained approximately 4.36 Mcal/kg. Average daily feed intakes (ADFI) ranged from 2.37 - 2.82 kg/d, but feed wasted by the pigs was not measured. Amino acid top dresses were used to better meet the barrows requirements for the amino acids that were deficient in the corn-based diets. Inclusion rates of the five amino acids are reported in table 1.6 in g/kg of diet.

Apparent ileal amino acid (AID) digestibilities are reported in table 9. Although notable numerically higher, there were no differences (P > .1) in AID between HO vs

IHO and WX vs STD 2 corns. When comparing WX to IWX, Arg, His, Phe, Val and Tyr were higher ($P \le .1$) and all other amino acids were similar. Histidine, Ile, Leu, Lys, Met, Thr, Asp, Glu, Pro were lower ($P \le .1$) in the HL compared to STD 1. In addition, Phe, Val, Ala, Ser and Tyr were lower (P < .05) in HL compared to STD 1. When comparing HO to STD 2, differences were seen in His, Ile, Leu, Lys, Met, Ala ($P \le .1$) and Pro (P < .05). When HL vs STD 2 AID were compared, Arg, Leu, Lys, Ala ($P \le .1$) and Pro (P < .05) were lower. Overall, STD 1 had higher absolute AID than all other hybrids. Consequently, the STD 1 corn had the greatest mean protein digestibility followed by the HO and the WX corn. Total AID were not different between HO vs IHO, WX vs IWX, WX vs STD 2, and HL vs. STD 2. Significant ($P \le .1$) total AID were shown in the HL vs STD 1 and HO vs STD 2.

True ileal amino acid digestibilities (TID) were calculated using the endogenous nitrogen losses reported in table 11. The five greatest endogenous amino acid losses from highest to lowest were Pro, Glu, Ser, Leu and Val. These losses were crucial in calculating the TID in table 10. True ileal digestibilities for the amino acids ranged from 70.12 to 130.93 %. There were no differences in TID when comparing between WX and IWX. Differences between HO and IHO were found for Met and Gly ($P \le .1$). Differences in Lys, Met, Ala ($P \le .1$), Phe, Thr , Asp, and Tyr (P < .05) were found between the HL and STD 1. Comparisons of the STD 2 corn to HO digestibilities showed differences for Glu and Pro at P < .05. Glu was the only amino acid that was different in the WX vs STD 2 contrast. When comparing HL and STD 2 differences were found for Phe, Tyr ($P \le .1$), Thr, Ala, Glu and Pro (P < .05). Overall, total protein TID did not differ between any of the six preplanned comparisons.

Apparent ileal amino acid digestibilities of the casein diet are reported in table 12. Digestibilities range from 58.81 ± 3.97 to 94.26 ± 0.55 with a mean amino acid digestibility of 84.73 ± 1.09 %.

Discussion

Amino acid concentrations of the corn hybrids were similar to reported values for other varieties of high oil, waxy, high lysine and standard yellow dent corns (Adeola, 1996; Adeola and Bajjalieh, 1997; Burgoon et al., 1992; Carr, 1994; Dupont, 1997; Soltwedel, 1996). Some variations were seen with leucine across literature and hybrids but all differences in concentrations were less than 0.20 %. In addition, the reported ratios of amino acids as a percent of protein were similar to those reported by Pond and Maner (1984) for HL and standard corns at 4.2 and 2.6 %, respectively. The high lysine amino acid data (AA/16 g N) also demonstrate increases in arg, asp and gly, but decreases in ala and leucine equivalent to the findings of Pond and Maner (1984).

Apparent amino acid digestibilities for high oil, waxy and standard yellow dent corns were similar to other essential amino acid digestibilities reported for these corns (Carr, 1994; Soltwedel, 1996; Southern, 1991). Differences in AID between hybrids and standard 2 were seen for the non-essential amino acids, particularly for asp, glu, gly, pro, and tyr (P < .05). The low AID for standard 2 corn reported by this study are difficult to explain, but the low digestibilities may be a result of constant addition of amino acids by the endogenous losses. Endogenous losses have been shown to include high proportions of glu, gly and pro (Sauer et al., 1977). Apparent digestibility values for high lysine corn were lower in this study than reported by Burgoon (1992). One possible reason for this

difference is that the pigs used in Burgoon's study varied between 97 and 73 kg whereas, this study used smaller pigs (40 kg).

True digestibility values for these specialty corns have not been reported. In this experiment, TID values from normal yellow dent corn were greater than reported by Stein (1998) who also used a casein diet to estimate endogenous secretions or by Southern (1991). True digestibilities in this study are higher because the estimations of endogenous secretions from the casein diet were higher than others published for similar size pigs (Stein, 1998). It is believed the overestimation of endogenous secretions were the primary reason this study reported several TID greater than 100 %. This observation raises the question of whether a protein free diet would give a more accurate estimation of endogenous losses. This question is addressed in the next chapter.

When comparing the apparent ileal digestibilities of the casein diet to other published values, it was found that the AID in this study were slightly lower than those published by Stein (1998) and Chung (1992).

In conclusion, a definite difference in nutrient concentrations between corn hybrids was found. The apparent amino acid digestibilities of waxy corn were similar and high oil and high lysine corns were superior to the commercially purchased yellow dent corn.

Implications

This study provides apparent amino acid digestibility coefficients for increasingly popular corn hybrids, which will be valuable for the formulation of swine diets. In addition, this study shows that the amino acids in specialty corns are adequately and in some cases better digested when compared to standard yellow dent two corn.

Table 3: Nutrient composition of corn hybrids on a dry matter basis

				Corn hyb	rids		
Items	НО	IHO	wx	IWX	HL	STD 1	STD 2
Dry matter, %	88.21	88.34	87.72	87.97	88.35	88.13	85.60
Crude protein, %	10.10	8.82	8.70	8.80	8.70	10.81	8.36
Ether extract, %	7.93	5.44	4.49	4.49	3.97	4.96	4.23
Gross energy, Mcal/kg	4.65	4.50	4.51	4.40	4.45	4.55	4.59
Ash, %	1.90	2.06	1.64	1.90	1.65	2.08	1.63
Amino acid, %							
Essential							
Arg	0.48	0.45	0.47	0.46	0.61	0.56	0.48
His	0.33	0.30	0.32	0.32	0.34	0.37	0.30
Ile	0.37	0.33	0.30	0.32	0.30	0.40	0.34
Leu	1.15	0.96	0.88	0.94	0.63	1.27	0.85
Lys	0.26	0.24	0.26	0.25	0.36	0.29	0.27
Met	0.24	0.21	0.21	0.22	0.20	0.24	0.19
Phe	0.49	0.42	0.40	0.42	0.33	0.54	0.40
Thr	0.33	0.30	0.30	0.30	0.32	0.36	0.26
Val	0.49	0.43	0.43	0.44	0.45	0.54	0.44
Non-essential							
Ala	0.74	0.61	0.58	0.62	0.50	0.78	0.67
Asp	0.65	0.57	0.66	0.49	0.82	0.73	0.30
Glu	1.81	1.51	1.49	1.49	1.27	1.99	1.42
Gly	0.37	0.33	0.35	0.34	0.40	0.40	0.35
Pro	0.86	0.73	0.75	0.77	0.68	0.95	0.66
Ser	0.48	0.43	0.43	0.43	0.40	0.53	0.43
Tyr	0.34	0.31	0.31	0.29	0.30	0.37	0.30
ΣAmino Acids	9.39	8.13	8.14	8.10	7.91	10.32	7.66

Table 4: Amino acid concentration as a percent of total nitrogen

Amino acid/ 16 g N, %	НО	IHO	WX	IWX	HL	STD 1	STD 2
Essential							
Arg	4.75	5.10	5.40	5.23	7.01	5.18	5.74
His	3.27	3.40	3.68	3.64	3.91	3.42	3.59
Ile	3.66	3.74	3.45	3.64	3.45	3.70	4.07
Leu	11.39	10.88	10.11	10.68	7.24	11.75	10.17
Lys	2.57	2.72	2.99	2.84	4.14	2.68	3.23
Met	2.38	2.38	2.41	2.50	2.30	2.22	2.27
Phe	4.85	4.76	4.60	4.77	3.79	5.00	4.78
Thr	3.27	3.40	3.45	3.41	3.68	3.33	3.11
Val	4.85	4.88	4.94	5.00	5.17	5.00	5.26
Non-essential							
Ala	7.33	6.92	6.67	7.05	5.75	7.22	8.01
Asp	6.44	6.46	7.59	5.57	9.43	6.75	3.59
Glu	17.92	17.12	17.13	16.93	14.60	18.41	16.99
Gly	3.66	3.74	4.02	3.86	4.60	3.70	4.19
Pro	8.51	8.28	8.62	8.75	7.82	8.79	7.89
Ser	4.75	4.88	4.94	4.89	4.60	4.90	5.14
Тут	3.37	3.51	3.56	3.30	3.45	3.42	3.59

Table 5: Particle size of the corn used in experimental diets (mm)

Hybrids	Geometric mean diameter a	Standard deviation by weight
НО	0.92	2.86
IHO	0.93	2.84
WX	0.95	2.90
IWX	0.92	2.86
HL	0.81	3.02
STD 1	0.93	2.83
STD 2	0.85	2.93

Coefficient of variation = 5.8 %

Table 6: Composition of experimental diets on as-fed basis (%)

Ingredients	Corn-based	Casein
Com ^a	96.62	
Cornstarch ^b		71.43
Casein ^c		11.75
Solka Floc ^c		5.00
Com oil ^d		4.00
Sugar		4.00
Dicalcium phosphate	1.25	2.25
Limestone	0.88	0.32
Vitamin mix ^e	0.50	0.50
Trace mineral mix ^f	0.25	0.25
Salt	0.25	0.25
Chromic oxide ^g	0.25	0.25

^a Corn varieties used were STD 1, STD 2, HO, IHO, WX, IWX and HL. ^b Argo Foods, CPC International, Inc., Englewood Cliffs, NJ 07632-9976.

^c Harland Teklad, Madison, WI 53744-4220.

^d Kraft Foodservice, Inc., Glenview, IL 60025.

^e Provided the following per kilogram of diet: 4,583 IU vitamin A, 458 IU vitamin D₃, 55 IU vitamin E, 11 mg vitamin K, 3.66 mg menadione, 0.0275 mg vitamin B₁₂, 3.66 mg riboflavin, 14.67 mg d-pantothenic acid, 22 mg niacin, 0.913 mg thiamine, 0.825 mg pyridoxine.

^f Provided the following per kilogram of diet: 335 g Ca, 5 g Fe, 5 g Zn, 5 mg Cu, 5 mg Mn, 150 µg Se, and 75 µg I.

⁸ Fisher, Itasca, IL 60143.

Table 7: Nutrient composition of the diets on a dry matter basis

Items	НО	IHO	WX_	IWX	HL	STD 1	STD 2	Casein
Composition, calculated								
Crude protein, %	10.06	8.54	8.56	8.56	7.93	10.36	7.75	11.16
Gross energy, Mcal/kg	4.49	4.35	4.36	4.25	4.30	4.40	4.43	4.67ª
Calcium, %	0.70	0.69	0.70	0.70	0.69	0.69	0.70	0.66
Phosphorus, %	0.57	0.57	0.57	0.57	0.57	0.57	0.57	0.55
ADFI, kg/d	2.37	2.51	2.63	2.82	2.35	2.51	2.55	2.76
Composition, analyzed								
Dry matter, %	87.61	87.95	87.22	87.63	88.09	88.15	87.18	91.46
Crude protein, %	9.64	9.51	8.43	8.55	8.65	10.93	8.27	12.06
Gross energy, Mcal/kg	4.51	4.36	4.39	4.36	4.33	4.38	4.30	4.38
Amino acid, %								
Essential								
Arg	0.47	0.42	0.42	0.39	0.64	0.50	0.40	0.58
His	0.36	0.31	0.30	0.29	0.38	0.38	0.27	0.48
Ile	0.38	0.32	0.29	0.31	0.28	0.39	0.26	0.73
Leu	1.09	0.98	0.86	0.87	0.62	1.30	0.74	1.14
Lys	0.25	0.22	0.25	0.23	0.34	0.26	0.23	0.96
Met	0.23	0.21	0.20	0.21	0.21	0.25	0.16	0.43
Phe	0.46	0.42	0.38	0.38	0.34	0.54	0.34	0.67
Thr	0.32	0.29	0.30	0.30	0.32	0.35	0.26	0.50
Val	0.47	0.43	0.40	0.40	0.45	0.51	0.36	0.85
Non-essential								
Ala	0.70	0.63	0.58	0.57	0.51	0.80	0.51	0.43
Asp	0.62	0.56	0.58	0.58	0.80	0.69	0.49	0.81
Glu	1.69	1.46	1.35	1.44	1.26	1.89	1.16	2.67
Gly	0.34	0.31	0.32	0.31	0.39	0.38	0.30	0.26
Pro	0.80	0.76	0.73	0.72	0.68	0.95	0.60	1.26
Ser	0.48	0.42	0.42	0.40	0.41	0.53	0.37	0.73
Tyr	0.33	0.30	0.29	0.27	0.31	0.36	0.26	0.68
ΣAmino Acids	8.99	8.04	7.67	7.67	7.94	10.08	6.71	13.18

Calculated as DE/.90, Mcal/kg

Table 8: Amino acid top dress composition

Amino acid, g/kg of diet	НО	IHO	wx	IWX	HL	STD 1	STD 2
Ile	0.53	0.93	1.23	0.92	0.77	1.23	1.27
Lys	3.96	4.07	4.50	4.24	4.35	3.38	4.97
Met	0.60	1.22	0.98	0.65	0.69	0.74	1.02
Thr	1.79	2.15	2.29	2.12	2.16	2.21	2.40
Trp	0.15	0.15	0.15	0.15	0.15	0.15	0.15

Table 9: Apparent amino acid digestibilities (%) of the corn hybrids

	НО	IHO	wx	IWX	HL	STD 1	Pooled SEM	STD 2
Essential								
Arg	73.19 ^{ad}	66.04ª	67.49 ^{ad}	56.76 ^b	77.16 ^{ae}	81.12ª	5.52	65.27 ± 3.33^{d}
His	76.32 ^{ac}	69.73ª	71.07 ^{ad}	58.76 ^b	70.89 ^{ad}	83.01 ^b	5.10	64.57 ± 2.95^{d}
Ile	76.17 ^{ae}	68.60 ^a	68.83 ^{ad}	58.95ª	51.90 ^{ad}	80.05°	6.31	61.68 ± 3.64^{d}
Leu	82.89 ^{ac}	78.17 ^a	78.15 ^{ad}	69.19ª	60.79 ^{af}	87.99 ^c	4.73	73.39 ± 2.73^{d}
Lys	64.67 ^{ac}	52.27ª	54.90 ^{ad}	43.05ª	58.12 ^{ad}	73.76 ^b	6.83	50.33 ± 3.94^{d}
Met	78.32 ^{af}	69.57ª	69.93 ^{ad}	60.29ª	64.60 ^{ad}	83.49 ^c	5.60	62.77 ± 3.23^{d}
Phe	79.32 ^{ad}	75.52ª	75.82 ^{ad}	65.30 ^b	60.57 ^{ad}	86.40 ^c	5.06	70.22 ± 2.92^{d}
Thr	56.89 ^{ad}	52.68ª	53.94 ^{ad}	39.07ª	35.45 ^{ad}	72.82°	8.72	43.08 ± 5.03^{d}
Val	69.68 ^{ad}	63.46ª	64.23 ^{ad}	50.89 ^b	54.19 ^{ad}	78.98 ^c	6.41	57.10 ± 3.70^{d}
Non-								
essential	70 20 86	71 51 8	con ce ad	C2 01 8	50 27 8 0	01 62 ^c	4.02	(1.7(+ 2.05d
Ala	72.30 ae	71.51 2	67.65 ad	62.01	50.27 ac	81.52°	4.93	61.76 ± 2.85^{d}
Asp	78.17 ad	68.53 a	74.46 ad	63.41	73.58 ad	89.45 b	6.68	76.32 ± 3.92^{d}
Glu	78.59 ad	76.07 °	75.60 ad	69.63 ª	67.37 ad	88.80°	4.93	76.65 ± 2.85^{d}
Gly	39.16 ad	17.05 ª	35.36 ad	16.79 ª	31.40 ad	59.64 a	14.81	16.85 ± 8.75^{d}
Pro	57.35 af	24.72 a	16.22 ad	11.82 a	33.16 af	63.36 b	17.51	-9.30 ± 13.26^{d}
Ser	73.79 ad	66.87*	67.45 ad	55.33 a	58.59 ad	82.56 °	6.18	64.17 ± 3.57^{d}
Tyr	63.25 ad	64.55ª	63.02 ad	48.96 ^b	52.45 ad	74.80 ^c	6.56	56.75 ± 3.78^{d}
Mean AA Digestibility	70.00 ac	61.56ª	62.76 ad	51.89°	56.28 ad	79.23 °	6.38	55.73 ± 3.69 ^d

a,b Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 ($P \le .10$)

a.c Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 (P < .05)

de Means in rows with different superscripts are different among these preplanned comparisons: HO vs STD 2, WX vs STD 2, HL vs. STD 2 ($P \le .10$)

df Means in rows with different superscripts are different among these preplanned comparisons: HO

vs STD 2, WX vs STD 2, HL vs. STD 2 (P < .05)

Table 10: True amino acid digestibilities (%) of the corn varieties

							Pooled	
	НО	IHO	WX	IWX	HL	STD 1	SEM	STD 2
Essential								
Arg	96.72 ad	91.76ª	92.83 ^{ad}	93.05 ª	96.48 ^{ad}	98.94 ª	3.45	94.86 ± 2.73^{d}
His	95.24 ad	92.44 ª	93.88 ad	89.66 a	92.37 ad	97.34 a	3.48	92.44 ± 2.73^{d}
Ile	118.13 ad	120.49 ª	123.72 ad	129.09 a	120.87 ad	112.57ª	6.87	129.16 ± 4.85^{d}
Leu	100.74 ad	98.68 a	101.01 ad	100.07 a	100.85 ad	99.73 a	3.48	101.42 ± 3.16^{d}
Lys	114.61 ad	105.01 a	103.37 ad	111.22 a	98.88 ad	113.18°	6.86	107.74 ± 5.32^{d}
Met	102.74 ad	97.42 ^b	98.17 ad	97.58ª	97.65 ad	101.81 ^b	3.19	100.50 ± 2.82^{d}
Phe	99.37 ad	97.90ª	99.79 ^{ad}	97.91 ª	94.60 ac	99.91 °	3.35	99.68 ± 2.88^{d}
Thr	101.39 ad	102.47ª	101.19 ad	103.05 a	86.88 af	105.44 ^c	4.91	101.33 ± 3.71^{d}
Val	111.79 ^{ad}	108.99 a	112.01 ad	116.07ª	106.80 ad	109.79 a	6.95	114.40 ± 5.93^{d}
Non-esser	ntial							
Ala	94.86 ad	98.40ª	95.67 ^{ad}	98.25 ª	88.15 af	96.88 ^b	3.71	97.43 ± 2.28^{d}
Asp	89.42 ad	81.83 a	87.27 ^{ad}	80.72 a	83.42 ad	97.57°	4.87	92.39 ± 2.81 ^d
Glu	96.30 af	98.58ª	98.79 af	98.49 ª	97.29 af	100.98 a	4.50	106.13 ± 4.06^{d}
Gly	93.32 ad	70.12 ^b	77.94 ^{ad}	81.99 a	80.44 ad	93.60 a	9.35	78.21 ± 6.83^{d}
Pro	111.96 af	89.45 a	79.92 ad	92.55ª	110.67 af	97.88ª	13.88	78.44 ± 9.78 d
Ser	120.60 ad	121.25 ª	121.45 ad	130.93 a	124.66 ad	116.56ª	9.50	129.86 ± 8.20^{d}
Tyr	93.13 ^{ad}	96.77ª	95.82 ad	95.36ª	90.68 ae	96.77°	3.26	96.67 ± 2.58 ^d
Mean AID	102.52 ad	98.23 ª	98.93 ^{ad}	101.00 a	98.17 ad	102.43 a	3.91	101.29 ± 3.30 d

<sup>a.b Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 (P ≤ .10)
a.c Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 (P < .05)
d.e Means in rows with different superscripts are different among these preplanned comparisons: HO vs STD 2, WX vs STD 2, HL vs. STD 2 (P ≤ .10)
d.f Means in rows with different superscripts are different among these preplanned comparisons: HO vs STD 2 WX vs STD 2 HI vs. STD 2 (P ≤ .05)</sup>

vs STD 2, WX vs STD 2, HL vs. STD 2 (P < .05)

Table 11: Endogenous amino acid losses (mg/kg dry matter intake) at the terminal ileum determined by feeding a casein diet

	Pig A	Pig B	Pig C	Pig D	Mean ± SEM
Amino Acid					
Essential					
Arg	628.47	652.59	791.69	754.07	706.71 ± 39.28
His	364.93	478.24	468.68	487.41	449.82 ± 28.55
Ile	904.63	978.39	1137.25	1185.19	1051.37 ± 65.92
Leu	1032.67	1184.84	1619.43	1255.24	1273.05 ± 124.45
Lys	664.58	818.87	863.26	787.45	783.54 ± 42.59
Met	323.39	348.32	434.86	374.93	370.38 ± 23.93
Phe	519.24	571.39	727.46	583.80	600.47 ± 44.58
Thr	734.14	835.46	999.71	1145.03	928.59 ± 90.55
Val	1033.96	1206.63	1524.89	1296.15	1265.41 ± 102.18
Non-essential					
Ala	957.28	1148.06	872.63	1325.23	1075.8 ± 101.14
Asp	342.99	469.66	557.04	493.18	465.72 ± 44.88
Glu	971.06	2498.90	1997.78	2760.28	2057.01 ± 395.04
Gly	850.29	1303.77	953.34	1175.41	1070.7 ± 103.13
Pro	2345.33	3642.31	1944.49	4277.69	3052.46 ± 545.95
Ser	937.20	1508.41	1757.81	1664.81	1467.06 ± 183.96
Tyr	568.51	621.98	681.14	661.10	633.18 ± 24.81

Table 12: Apparent digestibility of casein diet (%)

	Casein	SEM
Amino Acid	- Custin	02
Essential		
Arg	87.73	0.68
His	90.67	0.59
Ile	85.65	0.90
Leu	88.83	1.09
Lys	86.78	1.29
Met	91.43	0.55
Phe	91.06	0.66
Thr	81.49	1.81
Val	85.10	1.20
Non-essential		
Ala	75.24	2.33
Asp	94.26	0.55
Glu	92.28	1.48
Gly	58.81	3.97
Pro	75.80	4.33
Ser	79.80	2.53
Тут	90.72	0.36
Mean AA Digestibility	84.73	1.09

CHAPTER 2: COMPARISON OF ENDOGENOUS AMINO ACID SECRETIONS FROM GROWING PIGS FED CASEIN AND PROTEIN FREE DIETS

Abstract

Four barrows (98.5 ± 5.6 kg body weight) fitted with simple-T cannulas at the terminal ileum were used to determine differences in endogenous amino acid secretions between casein and protein free diets. A switch back design was used where pigs were randomly allotted to treatments in pairs. The case in diet consisted of 71.43 % cornstarch and 11.75 % casein while the protein free diet consisted of 81.98 % cornstarch. The remainder of the ingredients in both diets were Solka Floc (5 %), corn oil (4 %), and sugar (4 % in the casein and 5 % in the protein free diet). Vitamins, minerals, dicalcium phosphate, limestone and salt were included in both diets to meet or exceed NRC requirements (1988). Chromic oxide was included in the diets as an indigestible marker at 0.25 %. Pigs were fed the diets for a period of seven days that included a five day diet adjustment period followed by two days of ileal digesta collections (12 hours each day). Digesta was pooled during the two day collection period and then analyzed for amino acid content. In the casein and protein free diets, proline, glutamic acid, serine and glycine were the amino acids secreted in the greatest amounts. Significant differences (P ≤ .1) in endogenous amino acid secretions were seen in all amino acids with the exception of arginine, aspartic acid, proline and serine (P > .10). The casein diet resulted in higher amino acid secretions than the protein free diet for all amino acids.

Introduction

True ileal digestibilities (TID) are a more accurate estimation of the digestibility of a feedstuff than apparent digestibility because the calculation takes into account endogenous secretions. Unfortunately, researchers have struggled with methods to estimate endogenous losses at the terminal ileum for several years. Endogenous nitrogen secretions originate from saliva, gastric juice, pancreatic juice, bile, intestinal juices, mucosa and sloughed mucosal cells (Stein, 1998). Secretions of nitrogen are influenced by several factors such as age, body weight, dry matter intake, quality of protein, and fiber (Grala et al., 1998; Sauer et al., 1977; Sauer et al., 1977; Taverner et al., 1981). Many methods exist for the determination of endogenous secretions in the digestive system of the pig, but none of them are without criticism. Several studies have investigated the protein free diet (de Lange et al., 1989; de Lange et al., 1989; Sauer et al., 1977; Taverner et al., 1981) and the casein diet (Chung and Baker, 1992; Chung and Baker, 1991; Stein, 1998). The protein free diet is believed to underestimate the endogenous losses but it is advantageous because is the least expensive and easiest (Stein, 1998). Increased protein intake has been shown to directly increase endogenous amino acid secretions in the gastrointestinal tract (Sauer, 1986 #36). The major disadvantage to the protein free diet is that it does not estimate diet specific endogenous losses or account for protein level specific endogenous secretions (Lewis and Bayley, 1995). The other method that has been investigated is the casein diet. In this method casein is assumed to be 100 % digestible and therefore any amino acids at the terminal ileum are of endogenous origin. This method accounts for both the diet specific and the protein level

specific endogenous secretions. Baker and Chung (1992) tested the digestibility of casein and found that is readily digested in pigs with true digestibility coefficients ranging from 94.8 - 99.9 % for all essential amino acids. Therefore, it could be used to estimate endogenous losses and assumed to be 100 % digestible. Stein (1998) used this method but believed that the endogenous secretions obtained from this method were overestimated. It was the goal of this experiment to investigate the endogenous secretions at the terminal ileum in pigs fed both a casein and a protein free diet. In addition, it was an objective of this experiment to how these diets would affect true ileal digestibilities.

Materials and Methods

Animals, experimental design and diets were approved by Michigan State University All University Committee for Animal Use and Care. Four, littermate barrows $(98.5 \pm 5.6 \text{ kg}, \text{Yorkshire})$, used in the previous experiment (chapter one), were randomly assigned to diets and arranged in a switchback design (Gill, 1978; Neter et al., 1996) to investigate the differences in endogenous secretion at the terminal ileum of pigs fed a casein and a protein free diet. The two diets were fed to the four pigs such that six individual observations were obtained for each diet. Environmental temperature in this experiment ranged from 21-29°C with a mean temperature of 24°C. Housing, feeding and digesta collection procedures were performed as described in chapter one.

Diets

The experimental diets are reported in table 13. The casein diet provided protein and energy to the barrows whereas the protein free diet only provided energy. Vitamins,

minerals, dicalcium phosphate, limestone and salt were included to meet or exceed NRC requirements (1988). Chromic oxide was included in the diets as an indigestible marker at 0.25 %.

Chemical analysis

Digesta, diets and corns analyses were performed as described in chapter one.

Statistical analysis

Data were analyzed using MIXED procedures of SAS (1990). A linear model with no interactions was used and included pig and diet as main effects. Sequence and carryover effects were evaluated and removed from the model because they were not significant at the lowest P-values of .29 and .34, respectively. Least square means and the pdiff option were used to determine differences between means.

Results

All pigs remained healthy during this study. Pigs on the casein diet had an ADFI of 2.58 kg/d and an ADG 0.48 kg/d. The pigs on the protein free diet had an ADFI and ADG of 0.054 kg/d and 2.03 kg/d, respectively. One pig in the last collection period refused to eat the protein free diet so normal finishing diet was given and no digesta sample was taken. Nutrient compositions of the diets are reported in table 14. The casein diet provided 12.06 % crude protein to the barrows whose requirement was 13 % (NRC, 1988). In addition, the casein diet met or exceeded the barrows requirements for essential amino acids and energy. On the other hand, the protein-free diet did not provide protein but did provide adequate amount of energy. Table 15 reports the endogenous losses in pigs fed the casein and protein-free diets. Differences in endogenous amino acid

secretions were found for His, Lys, Phe, Gly and Tyr ($P \le .1$). In addition, Ile, Leu, Met, Thr, Val, Ala and Glu were different at P < .05. Overall, endogenous losses at the terminal ileum were greater for the casein diet compared to the protein-free diet. Tables 16 and 17 are calculations of TID using both the protein free and the casein diets, respectively. The data are from the corns fed in chapter one. These two tables illustrate the differences that diets for endogenous estimation can have on true ileal digestibilities (TID). The range of TID values are 75.24 to 118.62 and 72.73 to 130.06 % for the protein free and the casein diets, respectively. Overall, the TID using the casein are greater than those for the protein free diet with the exception of three amino acids: Arg, Asp and Pro.

Discussion

Taverner's (1981) experiment where protein free diets were fed reported that the most abundant amino acids in the ileal endogenous digesta were proline, glycine, glutamic acid, aspartic acid, serine and threonine in order of most to least. Similar results were seen in this study for endogenous losses in both the casein and protein free diet. The most prevalent amino acids in the protein free diet (in order of highest to lowest) were proline, glutamic acid, glycine, serine, alanine, leucine and valine. In the casein diet, proline, glutamic acid, serine, glycine, valine, alanine and leucine had the greatest amount protein in the endogenous digesta from highest to lowest.

The values obtained for endogenous losses from the casein diet were at least two times the amount seen in the protein free diet. It is logical to attribute the differences in endogenous secretions to the differences in crude protein between the two diets. Data presented by Jondreville (1995) state that endogenous nitrogen has two fractions: the

protein that is secreted but it is not specific to the feedstuff (only affected by the dry matter intake) and protein that depends on the composition of the feedstuff (affected by the quantity and type of feed ingested). The researchers at the Technical Institute for Cereals and Forages use a protein free diet to estimate the non-specific endogenous losses. Therefore, in this study, the protein free diet only estimates the non-specific endogenous losses and the casein diet estimates the diet specific endogenous losses and the non-specific endogenous secretions. Since casein was used and not a corn based diet the diet specific endogenous losses were thought to be overestimated.

Similar endogenous amino acid losses from the protein free diet were found when compared to published values (Chung and Baker, 1992; de Lange et al., 1989; de Lange et al., 1989; Sauer et al., 1977; Stein, 1998; Taverner et al., 1981). Endogenous losses from the casein diet were definitely higher than values published by Stein (1998) and Chung and Baker (1992).

Implications

This study implies that neither the casein nor the protein free diet was adequate to estimate endogenous losses for the corn diets.

Table 13: Experimental diets on as-fed basis (%)

Ingredients	Casein	Protein free
Cornstarch ^a	71.43	81.98
Casein ^b	11.75	
Solka Floc ^b	5.00	5.00
Com oil ^c	4.00	4.00
Sugar	4.00	5.00
Dicalcium phosphate	2.25	2.70
Limestone	0.32	0.07
Vitamin mix ^d	0.50	0.50
Trace mineral mix ^e	0.25	0.25
Salt	0.25	0.25
Chromic oxide ^f	0.25	0.25

^a Argo Foods, CPC International, Inc., Englewood Cliffs, NJ 07632-9976. ^b Harland Teklad, Madison, WI 53744-4220.

^c Kraft Foodservice, Inc., Glenview, IL 60025.

^d Provided the following per kilogram of diet: 4,583 IU vitamin A, 458 IU vitamin D₃, 55 IU vitamin E, 11 mg vitamin K, 3.66 mg menadione, 0.0275 mg vitamin B₁₂ 3.66 mg riboflavin, 14.67 mg d-pantothenic acid, 22 mg niacin, 0.913 mg thiamine, 0.825 mg pyridoxine.

Provided the following per kilogram of diet: 335 g Ca, 5 g Fe, 5 g Zn, 5 mg Cu, 5 mg Mn, 150 μg Se, and 75 μg I.

Fisher, Itasca, IL 60143.

Table 14: Nutrient composition of the diets on a dry matter basis

	Exper	imental Diets
Items	Casein	Protein-free
Composition, calculated		
Crude protein, %	11.16	0.00
Gross energy, Mcal/kg ^a	4.67	4.64
Calcium, %	0.66	0.56
Phosphorus, %	0.55	0.46
Composition, analyzed		
Dry matter, %	91.27	91.54
Crude protein, %	12.06	1.32
Gross energy, Mcal/kg ^a	4.38	3.97
Amino acid, %		
Essential		
Arg	0.58	-
His	0.48	-
Ile	0.73	-
Leu	1.14	-
Lys	0.96	-
Met	0.43	
Phe	0.67	-
Thr	0.50	-
Val	0.85	-
Non-essential		
Ala	0.43	
Asp	0.81	-
Glu	2.67	
Gly	0.26	-
Pro	1.26	-
Ser	0.73	-
Tyr	0.68	-
ΣAmino Acids	13.18	-

Table 15: Endogenous losses in casein and protein free diets (mg/kg dry matter intake)

	Casein ^a ± SEM	Casein ^b ± SEM	Protein-free b ± SEM
Amino Acid			
Essential			
Arg	706.71 ± 39.28	522.96 ± 61.99°	362.45 ± 69.31 °
His	449.82 ± 28.55	538.37 ± 69.19^{c}	255.10 ± 77.35 d
Ile	1051.37 ± 65.92	887.99 ± 79.08^{c}	288.45 ± 86.43 °
Leu	1273.05 ± 124.45	1090.69 ± 147.10^{c}	473.09 ± 163.89 °
Lys	783.54 ± 42.59	914.68 ± 143.04°	373.82 ± 159.92 d
Met	370.38 ± 23.93	369.51 ± 41.31 °	173.82 ± 46.16 °
Phe	600.47 ± 44.58	645.54 ± 101.07°	311.21 ± 112.99 d
Thr	928.59 ± 90.55	907.15 ± 80.42 °	407.87 ± 89.92 °
Val	1265.41 ± 102.18	1194.32 ± 143.06°	447.20 ± 158.52 °
Non-essential			
Ala	1075.80 ± 101.14	1109.51 ± 128.85°	465.55 ± 144.05 °
Asp	465.72 ± 44.88	627.42 ± 133.08°	386.54 ± 148.79 °
Glu	2057.01 ± 395.04	2107.95 ± 224.52°	717.17 ± 251.02 °
Gly	1070.70 ± 103.13	1295.63 ± 190.36°	647.71 ± 212.83^{d}
Pro	3052.46 ± 545.95	2419.47 ± 580.81 °	$2047.65 \pm 606.96^{\circ}$
Ser	1467.06 ± 183.96	1302.57 ± 116.76°	487.71 ± 129.89°
Tyr	633.18 ± 24.81	605.28 ± 85.81°	307.23 ± 95.94 ^d

The casein amino acid endogenous losses obtained from experiment 1 are shown here again for comparison to the casein endogenous losses in this experiment. Data were not used for statistical comparison.

b The casein and protein free diets were used in a switch back design.

c,d Means within the same row with different superscripts are significantly different at P ≤ .10

c,e Means within the same row with different superscripts are significantly different at P < .05

Table 16: True amino acid digestibilities (%) of the corn varieties using protein free diet for endogenous losses

	НО	IHO	wx	IWX	HL	STD 1	Pooled SEM	STD 2 ± SEM
Amino Acid			-					
Essential								
Arg	91.66	90.83	91.81	90.16	92.62	94.39	2.29	91.75 ± 2.45
His	93.23	95.79	97.14	92.77	90.16	95.20	1.65	93.60 ± 2.25
Ile	96.49	96.84	98.87	96.54	87.35	95.41	2.43	93.60 ± 2.23
Leu	92.17	94.61	96.14	91.70	85.90	93.09	1.90	92.99 ± 1.75
Lys	103.35	101.98	100.93	103.82	89.93	103.11	4.89	101.55 ± 3.48
Met	96.06	95.70	96.37	93.47	90.07	96.04	1.47	95.28 ± 1.93
Phe	95.97	98.31	100.06	96.38	91.38	96.83	2.18	98.70 ± 2.09
Thr	89.06	96.10	95.97	93.88	75.24	95.28	4.26	90.64 ± 3.56
Val	93.64	96.13	98.42	94.94	86.10	95.46	3.02	95.38 ± 2.70
Non-								
essential Ala	88.18	95.66	93.01	94.37	79.09	91.58	2.19	90.45 ± 1.42
Asp	91.24	91.68	96.99	90.29	85.50	97.70	5.34	102.49 ± 4.56
Glu	88.60	92.79	93.14	89.84	86.20	95.12	2.66	97.35 ± 2.49
Gly	93.89	83.68	89.75	97.81	77.57	92.01	6.29	85.10 ± 7.25
Pro	118.62	105.30	92.96	109.65	114.35	101.22	13.70	93.11 ± 22.08
Ser	100.64	100.62	101.14	100.57	97.33	101.70	3.07	104.68 ± 1.91
Туг	87.37	94.00	93.15	90.12	85.03	91.95	2.59	91.88 ± 2.85
Mean AA Digestibility	95.63	95.01	95.39	95.99	88.37	96.01	1.34	95.13 ± 1.26

Table 17: True amino acid digestibilities (%) of the corn varieties using casein^b diet for endogenous losses

	НО	IHO	wx	IWX	HL	STD 1	Pooled SEM	STD 2 ± SEM
Amino Acid							-	
Essential								
Arg	89.47	89.16	90.39	88.79	89.78	92.92	2.68	88.63 ± 3.06
His	100.31	101.42	103.64	103.69	98.42	101.00	5.31	100.76 ± 4.35
Ile	118.51	115.92	119.82	126.36	120.21	113.55	8.23	124.66 ± 3.69
Leu	97.58	99.07	101.50	100.35	95.60	97.11	3.81	99.08 ± 3.00
Lys	129.22	122.43	121.18	139.85	110.25	125.06	15.24	122.42 ± 6.99
Met	103.48	101.73	102.82	103.76	99.00	102.08	4.27	103.54 ± 3.39
Phe	101.08	102.47	105.02	105.37	98.10	100.77	5.24	103.27 ± 3.53
Thr	101.10	106.38	105.76	109.09	86.70	104.89	6.08	102.97 ± 4.54
Val	113.50	111.87	115.72	121.97	108.67	111.12	8.81	116.41 ± 4.51
Non-essential								
Ala	97.37	102.93	101.36	108.03	93.30	98.54	5.03	100.33 ± 3.00
Asp	90.18	91.13	96.65	91.49	83.19	97.18	3.49	99.76 ± 4.54
Glu	98.67	101.49	102.66	103.80	100.51	102.88	2.90	109.36 ± 2.77
Gly	109.38	93.70	99.25	118.33	93.85	102.96	18.06	95.71 ± 11.52
Pro	104.12	89.69	81.55	94.33	108.04	90.74	14.73	72.73 ± 17.68
Ser	122.43	119.02	120.01	130.06	125.22	118.76	8.86	129.05 ± 3.59
Тут	93.27	98.82	98.52	100.78	90.71	96.85	6.29	96.45 ± 4.69
Mean AA Digestibility	102.92	104.35	109.13	104.12	100.10	103.52	2.55	104.07 ± 3.53

^b Endogenous values from this experiment

CHAPTER 3: EFFECTS OF CORN HYBRIDS FED TO GROWING PIGS ON NITROGEN METABOLISM

Abstract

Eight barrows (19.11 \pm 0.49 kg, Yorkshire) were arranged in two different 4 x 4 Latin squares to investigate the effects of feeding different corn hybrids on nitrogen metabolism. The corn hybrids were: high oil (HO), isogenic high oil (IHO), waxy (WX), isogenic waxy (IWX), high lysine (HL), standard yellow dent one (STD 1) and standard yellow dent two (STD 2). Standard two corn was used in both Latin squares to serve as the reference corn so that cross square comparisons could be preformed. Pigs within each square were littermates. Pigs were fed corn-based diets consisting of 96.87 % test corn. Vitamins, minerals, dicalcium phosphate, limestone and salt were included at levels to meet or exceed NRC requirements (, 1988 #231). Pigs were allowed an diet adjustment period (5 days) followed by five days of total urine and fecal collection. Feces and urine were pooled across collections and analyzed for nitrogen content using a Hach-kjeldahl procedure. Nitrogen retained was not different between HO vs IHO, WX vs IWX and HL vs STD 1 (P > .1). Nitrogen retention as a percent of N intake and N absorbed was similar between HO vs STD 2 and WX vs STD 2 (P > .1), but was different when HL was compared to STD 2 ($P \le .1$). Nitrogen digestibility was highest with the HO, STD 1 and HL corns at 83.31, 83.30 and 81.06 % respectively. Overall, pigs fed the STD 1 HL and HO diet retained numerically more nitrogen consumed and absorbed than all other corns.

Introduction

In the previous chapters the amino acid digestibility of the corn hybrids (high oil (HO), isogenic high oil (IHO), waxy (WX), isogenic waxy (IWX), high lysine (HL), standard one (STD 1) and standard two (STD 2) were investigated, but to fully investigate the protein quality of these corns it is crucial to measure the nitrogen retention and utilization. The objective of this experiment was to determine the nitrogen digestibility, absorption and utilization in the growing pig fed different corn varieties. Currently, there is not a plethora of data regarding the nitrogen retention of pigs fed these corn hybrids. Therefore, it is important that nitrogen balance experiments be performed on these hybrids to determine differences in utilization before they can be advocated for use in swine diets.

Materials and Methods

Animals, diets and experimental design were approved by Michigan State University All University Committee for Animal Use and Care. Eight barrows (Yorkshire) with an initial average body weight of 19.1 ± 0.49 kg were used to investigate nitrogen utilization of seven corn hybrids: high oil (HO), isogenic high oil (IHO), waxy (WX), isogenic waxy (IWX), high lysine (HL), standard one (STD 1) and standard two (STD 2). Pigs were randomly assigned to diets containing 96.87 % of the respective corn hybrid and arranged in two 4 x 4 Latin squares. The diet assignments within squares were the same as described in chapter one. Standard corn two was present in both squares, as the reference corn and the pig within each square were littermates. The test corns provided the only source of protein. Vitamins, minerals, dicalcium phosphate, limestone and salt were included at levels to meet or exceed NRC

requirements (1988). Environmental temperature ranged from 16 - 27°C with a mean temperature of 21°C. The metabolism room was mechanically ventilated to allow adequate air exchange. Pigs were individually housed in stainless steel, metabolism cages (1.2 m x 0.75 m) containing low-pressure nipple waterers that provided free access to water. The metabolism cages allowed for separate collection of urine and feces.

Feed intake was adjusted to 90 % of ad libitum intake and maintained at that level during the trial. Pigs were fed equal meals four times daily during the entire experiment. Pigs were allowed five days to adjust to the diets followed by a five-day separate urinary and fecal collection. Five grams of Ferric Oxide was mixed with 100 g of diet to be used as an indigestible marker to mark the beginning and end of the fecal collection. Feces and urine were collected once daily, with volumes and weights recorded. Urine was preserved by the addition of 100 ml of 20 % sulfuric acid to each daily collection. Daily fecal samples were pooled and frozen at -20°C, and 20 % of the daily total urinary volume were pooled and frozen at -20°C until analyses were performed.

Chemical analysis

Fecal samples were thawed and homogenized in a Hobart mixer for 10 minutes and sub-sampled. The sub-samples were freeze dried and ground through a 1 mm screen using a cyclone mill (Cyclotec Sample Mill 1093, Sweden). A 2 g sample of fecal material was dried further in a vacuum oven for 12 hours at 60°C for determination of total dry matter. Corn, diets, freeze dried feces and liquid urine were analyzed for nitrogen using a Hach-kjeldahl procedure (Hach et al., 1987).

Statistical analysis

Data was analyzed using MIXED procedures of SAS (1992) in a Latin square design. Data from the two squares were pooled and STD 2 corn was used as the reference corn. The linear model included main fixed classification effects of square, collection nested within square and diets; pig nested within square was included as a random effect. Least square means with pdiff option were used to determine difference between the following preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1, HO vs STD 2, WX vs STD 2 and HL vs STD 2.

Results

During this experiment the pigs remained healthy with average daily feed intake of 1.5 kg/d and average daily gain of 229 g/d. Diet compositions are reported in table 18; the main difference between these diets and chapter one's diets is the exclusion of chromic oxide, which accounts for the 0.25 % increase in test corn. Nutrient composition of the diets is the same as those reported in chapter one (Table 20). Nitrogen utilization and dry matter digestibility of the seven corn hybrids are reported in table 21. Initial and final weights of the barrows were similar across the corn diets. Nitrogen intake did not differ between HO vs IHO, WX vs IWX, and HL vs STD 1 (P > .10). However, nitrogen intakes for the hybrids were higher when comparing WX to STD 2 (P < .10), HO to STD 2 (P < .05) and HL to STD 2 (P < .05). Fecal nitrogen was greater in WX compared to IWX (P < .10). Urine nitrogen was similar for all contrasts investigated (P > .10). All hybrids (HO, WX, and HL) had higher nitrogen absorbed when compared to the standard yellow dent corn (P < .10). Nitrogen absorbed was greater for the hybrids HO vs IHO (P < .10) and lower for HL vs STD 1 (P < .10). Nitrogen retained was greater in HO and HL

compared to STD 2 (P < .05). Digestibility of nitrogen was greater for HO and HL vs STD 2 (P < .05). Consequently, IWX had higher nitrogen digestibility than WX (P \leq .1). Nitrogen retention as a percent of nitrogen intake and as a percentage of nitrogen absorbed was lower between of HL and STD 2 (P < .05). Significantly higher differences in dry matter (DM) intake were found comparing HL to STD 1 (P < .05), WX to STD 2 (P \leq .10), HL to STD 2 (P < .05). Dry matter digestibility was significantly higher when comparing HO to IHO (P < .05). Isogenic waxy and STD 1 had greater dry matter digestibility than WX and HL, respectively (P < .05). Finally, HO corn was the only hybrid to have a significantly higher DM digestibility compared to the STD 2 corn (P < .05).

Discussion

Nitrogen metabolism in pigs fed specialty coms is an important topic to fully understand the possible applications of these value-added corns. Published information on this subject is clearly lacking. Carr (1994), using 63 kg pigs, investigated nitrogen metabolism of HO corn varieties. Carr recorded ranges of nitrogen digestibilities for HO corn from 61 to 77 %, nitrogen retention as a percent of intake from 59 to 81 % and nitrogen retentions as a percent of absorbed from 75 to 76 %. Adeola (1997), using 25 kg pigs, also tested varieties of HO corns and published nitrogen digestibilities of 76 %, nitrogen retention as a percent of intake from 47 to 55 % and nitrogen retention as a percent of absorbed from 62 to 72 %. The values obtained from this study, 83.31 ± 2.13 % for nitrogen digestibility, 49.74 ± 5.35 % retention as a percent of intake and 59.73 ± 6.52 % retention as a percent of absorbed, are comparable to those published by Adeola (1997).

Ashce (1986) reported nitrogen utilization of high lysine corn, using 35 kg growing pigs. Nitrogen digestibilities varied from 79 to 80 % and nitrogen retentions from 47 to 52 %. Values for nitrogen digestibility and retention in this study were comparable with 81.06 ± 2.13 % and 57.22 ± 5.35 %, respectively. The retention values for standard yellow dent corn were also similar to other published values (Lin et al., 1987) Adeola(1997) Asche(1986) (Carr, 1994). Ranges of nitrogen digestibilities for standard yellow dent corns were 76 to 85 %. Nitrogen retentions as a percent of intake ranged from 38 to 61 % and as a percent of absorbed were from 44 to 79 %. These values are similar to data from this study: 74.12 ± 1.74 % for nitrogen digestibility, 38.48 ± 3.44 % for nitrogen retention as a percent of intake and 52.34 ± 4.07 % for nitrogen retention as a percent of absorbed. No published values for waxy corn were available to compare to this study.

Nitrogen digestibilities of the corn hybrids were greatest for the high oil, standard 1, and high lysine corn. Interestingly, nitrogen utilization expressed as nitrogen retention were greater for the standard 1 and high lysine compared to the high oil corn. This implies that the 20 kg pig was able to better utilize the protein from the standard 1 and the high lysine corns. Nitrogen digestibilities and retentions for the waxy corn were similar to standard 2 corn. However, nitrogen digestibilities and retentions for the high oil and high lysine corns were superior to those of the standard 2 corn that was the commercially purchased corn. As a whole, STD 1, HL and HO corns were superior to the other hybrids for nitrogen digestibility, nitrogen retention as a percent of intake and nitrogen retention as a percent of nitrogen absorbed.

Implications

This study implies that feeding the specialty corns will have result in similar and in some cases superior nitrogen utilization by the growing pig compared to feeding standard yellow dent two. Particularly, the high oil and high lysine hybrids should be considered as the corn hybrids of choice for feeding swine.

Table 18: Experimental diets on as-fed basis (%)

Ingredients	Experimental
Com ^a	96.87
Dicalcium phosphate	1.25
Limestone	0.88
Vitamin mix ^b	0.50
Trace mineral mix ^c	0.25
Salt	0.25

^a Corn varieties used were STD 1, STD 2, HO, IHO, WX, IWX and HL.

^b Provided the following per kilogram of diet: 4,583 IU vitamin A, 458 IU vitamin D₃, 55 IU vitamin E, 11 mg vitamin K, 3.66 mg menadione, 0.0275 mg vitamin B₁₂, 3.66 mg riboflavin, 14.67 mg d-pantothenic acid, 22 mg niacin, 0.913 mg thiamine, 0.825 mg pyridoxine.

^c Provided the following per kilogram of diet: 335 g Ca, 5 g Fe, 5 g Zn, 5 mg Cu, 5 mg Mn, 150 µg Se, and 75 µg I.

Table 19: Amino acid top dress composition

	НО	IHO	WX	IWX	HL	STD 1	STD 2
Amino acid, g/kg of diet							
Ile	0.53	0.93	1.23	0.92	0.77	1.23	1.27
Lys	3.96	4.07	4.50	4.24	4.35	3.38	4.97
Met	0.60	1.22	0.98	0.65	0.69	0.74	1.02
Thr	1.79	2.15	2.29	2.12	2.16	2.21	2.40
Trp	0.15	0.15	0.15	0.15	0.15	0.15	0.15

Table 20: Nutrient composition of the diets on a dry matter basis

Items	НО	IHO	wx	IWX	HL	STD1	STD 2
Composition, calculated							
Crude protein, %	10.09	8.56	8.59	8.58	7.96	10.40	7.77
Gross energy, Mcal/kg	4.49	4.35	4.36	4.25	4.30	4.40	4.43
Calcium, %	0.70	0.69	0.70	0.70	0.69	0.69	0.70
Phosphorus, %	0.57	0.57	0.57	0.57	0.57	0.57	0.57
Composition, analyzed							
Dry matter, %	87.61	87.95	87.22	87.63	88.09	88.15	87.18
Crude protein, %	9.64	9.51	8.43	8.55	8.65	10.93	8.27
Gross energy, Mcal/kg	4.51	4.36	4.39	4.36	4.33	4.38	4.30
Amino acid, %							
Essential							
Arg	0.47	0.42	0.42	0.39	0.64	0.50	0.40
His	0.36	0.31	0.30	0.29	0.38	0.38	0.27
Ile	0.38	0.32	0.29	0.31	0.28	0.39	0.26
Leu	1.09	0.98	0.86	0.87	0.62	1.30	0.74
Lys	0.25	0.22	0.25	0.23	0.34	0.26	0.23
Met	0.23	0.21	0.20	0.21	0.21	0.25	0.16
Phe	0.46	0.42	0.38	0.38	0.34	0.54	0.34
Thr	0.32	0.29	0.30	0.30	0.32	0.35	0.26
Val	0.47	0.43	0.40	0.40	0.45	0.51	0.36
Non-essential							
Ala	0.70	0.63	0.58	0.57	0.51	0.80	0.51
Asp	0.62	0.56	0.58	0.58	0.80	0.69	0.49
Glu	1.69	1.46	1.35	1.44	1.26	1.89	1.16
Gly	0.34	0.31	0.32	0.31	0.39	0.38	0.30
Pro	0.80	0.76	0.73	0.72	0.68	0.95	0.60
Ser	0.48	0.42	0.42	0.40	0.41	0.53	0.37
Tyr	0.33	0.30	0.29	0.27	0.31	0.36	0.26
ΣAmino Acids	8.99	8.04	7.67	7.67	7.94	10.08	6.71

Table 21: Nitrogen utilization and dry matter digestibility of the corn-based diets

Į,	CH.	OUI	A /II	7/11/	111	eTD1	Pooled	Acro Corro
Item	On	Our	<	۲ <u>»</u> ۲	пГ	SIDI	SEM	SID 2 ± SEM
u	4	4	4	4	4	4		7
Initial weight, kg	19.15	20.39	20.39	20.39	19.15	19.15		19.77
Final weight, kg	33.25	33.44	33.44	33.44	33.25	33.25		33.35
N intake, g/d	16.32ªf	15.09	14.81 20	14.33	17.70 af	18.24	0.85	13.33 ± 0.60^{d}
N in feces, g/d	2.84ad	3.09	3.77 ad	3.11 ^b	3.48 ad	3.17	0.41	3.39 ± 0.32^{d}
N in urine, g/d	5.90°d	4.89	4.09 ad	4.93	4.51 ad	4.06 ª	0.72	4.61 ± 0.49 ^d
N absorbed, g/d	13.83 ^{af}	11.65 ^b	10.70ac	10.88	14.56 af	15.42 ^b	0.67	9.60 ± 0.55^{d}
N retained, g/d	7.92ªf	6.78	6.62 ad	5.97	10.05 af	11.34	0.95	4.99 ± 0.59 ^d
N digestibility, %	83.31 ^{af}	78.90	74.04 ad	77.80 ^b	81.06 af	83.30	2.13	74.12 ± 1.74^{d}
N retention, % of intake	49.74ªd	45.58	44.55 ^{ad}	42.03	57.22 af	62.36	5.35	38.48 ± 3.44 ^d
N retention, % of absorbed	59.73ªd	57.73	59.14 *d	53.39 2	70.75 ^{af}	75.06	6.52	52.34 ± 4.07 ^d
DM intake, g/d	1066.5 ad	1052.0	1118.3 **	1068.9	1275.1 ^{af}	1076.0 °	54.85	1002.32 ± 39.60^{d}
Fecal DM, g/d	110.08 af	125.95	162.11 ac	134.42	179.58 af	122.15°	13.58	136.03 ± 10.84^{d}
DM digestibility, %	90.16 ^{af}	87.65°	85.19 ad	87.11°	86.36 **	89.07°	0.91	86.14 ± 0.76^{d}

 4e Means in rows with different superscripts are different among these preplanned comparisons: HO vs STD 2, WX vs STD 2, HL vs. STD 2 (P \leq 10) 4f Means in rows with different superscripts are different among these preplanned comparisons: HO vs STD 2, WX vs STD 2, HL vs. STD 2 (P \leq .05) ^{4,c} Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 (P < .05) ^{2,b} Means in rows with different superscripts are different among these preplanned comparisons: HO vs IHO, WX vs IWX, HL vs STD 1 (P ≤ .10)

SUMMARY AND GENERAL CONCLUSION

The goal of this thesis was to evaluate the protein quality of high oil, isogenic high oil, waxy, isogenic waxy, high lysine and two yellow dent corns in growing pigs.

Apparent and true amino acid digestibilities were determined for each of the corn hybrids. It was found that the high oil and the waxy corns had digestibilities superior to the commercial yellow dent corn. In addition, high lysine corn had digestibilities similar to commercial yellow dent corn two. When true ileal digestibilities were calculated, the values were greater than 100 % and it was concluded that the casein diet overestimated the endogenous losses. Therefore, we designed a second experiment to compare the casein diet to a second method for determining endogenous secretions.

In the second experiment, a protein free diet was compared to a casein diet in an attempt to better estimate endogenous losses and obtain accurate true amino acid digestibilities. The results showed that the endogenous secretions from the casein diet were two times greater than endogenous losses from the protein free diet. The study proved that accurate true ileal digestibilities could not be obtained. Therefore, it would be recommended that a protein free diet should be fed within the Latin square design during the first experiment which may have given acceptable true ileal digestibility values.

Nitrogen utilization of the corn hybrids were determined in the third experiment.

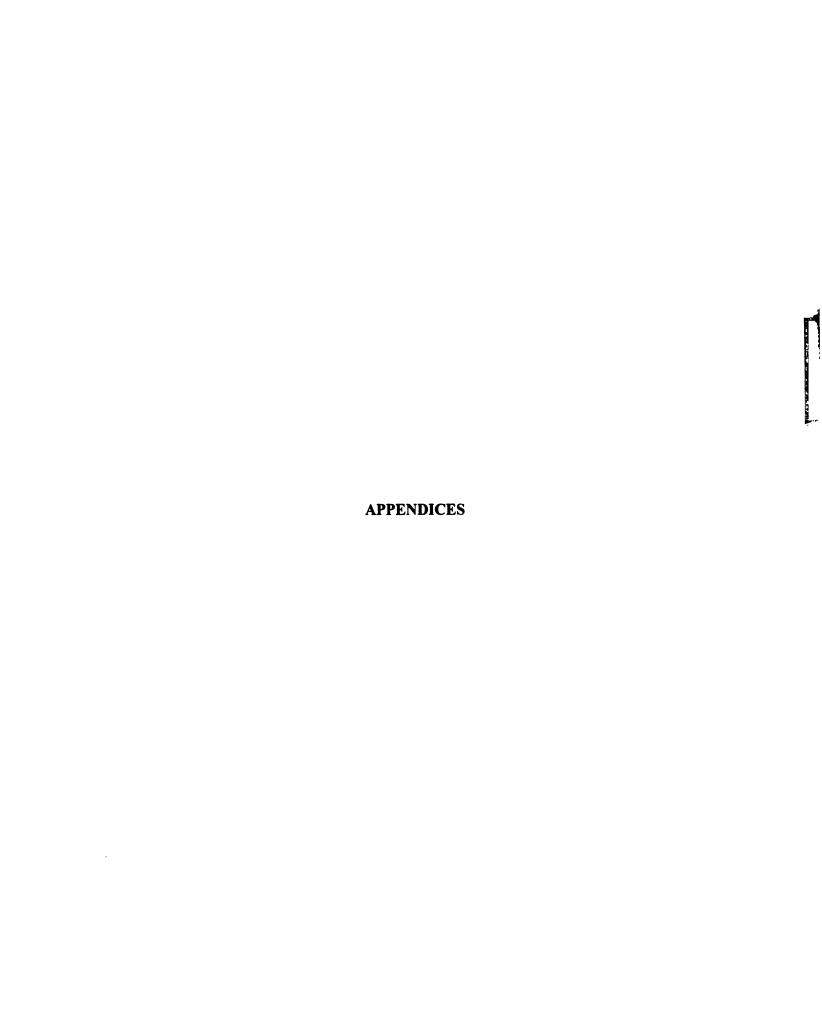
The results showed that the nitrogen utilization of the high lysine corn was definitely

superior to the commercial yellow dent corn and the other corn hybrids. Waxy and high oil corn were found to have nitrogen utilization similar to the commercial yellow dent.

In conclusion, this work suggests that the specialty corns, particularly high oil and high lysine, are superior to the yellow dent hybrids. In addition to their superior nutrient density and (or) profile, these specialty corns have higher protein and (or) nitrogen digestibility and nitrogen utilization.

VITA

Janet L. Snow is originally from Columbia, Missouri where she was raised on a commercial cattle farm. She was interested in animals since she was a child and was involved in the husbandry of cattle, swine, sheep and poultry. She was an eleven year member of 4-H and was involved in FFA in high school. She was active in showing livestock and she participated in county fairs until her Sophomore year in college. In 1995, she married her high school sweetheart, Mr. Scott S. Snow. She received her Bachelors degree in Animal Science from the University of Missouri-Columbia in May of 1996 and then moved to Michigan in June to pursue her Masters degree in swine nutrition at Michigan State University under the supervision of Dr. Nathalie L. Trottier.



Appendix A

Calculations for digestibility:

Adapted from Stein, 1998

Apparent ileal digestibility (AID):

 $AID = (100 - [(AAd/AAf) \times (Crf/Crd)]) \times 100\%$

Where:

AAd = amino acid concentration in the digesta

AAf = amino acid concentration in the feed

Crf = Chromium in the feed

Crd = Chromium in the digesta

Endogenous amino acid loss (EAL):

 $EAL = (AAd \times (Crf/Crd))$

Where:

EAL = Endogenous flow of each amino acid, mg lost/kg DMI

AAd = Amino acid concentration in the digesta

Crf = Chromium in the feed

Crd = Chromium in the digesta

True ileal digestibility (TID):

 $TID = [AID + (EAL/AAf)] \times 100 \%$

Where:

AID = Apparent ileal digestibility

EAL = Endogenous flow of each amino acid, mg lost/kg DMI

AAf = amino acid concentration in the feed

Apparent ileal digestibility obtained from the chromium results from samples analyzed at University of Illinois

Appendix B

	НО	IHO	wx	IWX	HL	STD 1	STD 2
Essential							
Arg	70.06	70.67	72.24	68.28	72.66	86.64	71.40
His	71.65	74.99	76.078	70.76	66.00	87.22	71.53
Ile	72.50	73.50	73.94	70.72	47.07	85.75	68.55
Leu	79.15	82.43	82.61	78.59	57.07	90.64	78.19
Lys	56.68	61.77	64.67	61.35	48.96	79.70	59.46
Met	73.26	75.27	75.78	72.46	58.86	87.24	69.55
Phe	75.45	80.12	80.66	75.57	56.10	89.54	75.60
Thr	50.22	61.33	62.81	57.46	27.94	79.62	53.11
Val	63.86	70.28	71.35	65.67	47.53	84.07	64.79
Non-essential							
Ala	67.77	76.57	73.20	73.70	46.15	86.31	68.51
Asp	73.56	75.27	81.67	74.99	67.23	90.16	81.43
Glu	75.58	80.03	79.62	78.56	63.45	91.23	81.80
Gly	28.73	31.98	48.88	42.91	16.97	69.49	34.37
Pro	43.99	41.45	29.38	40.21	19.38	71.20	14.05
Ser	69.58	72.36	72.94	67.94	53.38	87.01	70.76
Tyr	59.52	69.48	68.08	62.55	47.37	82.73	64.30
Total Protein	64.47	68.59	69.66	66.36	49.76	84.28	64.21

Appendix C



High Oil Cargill 5990 TC



Isogenic High Oil Cargill 5990



High Lysine Crows



Crows Standard Yellow Dent 1



Waxy Pioneer 3528 E



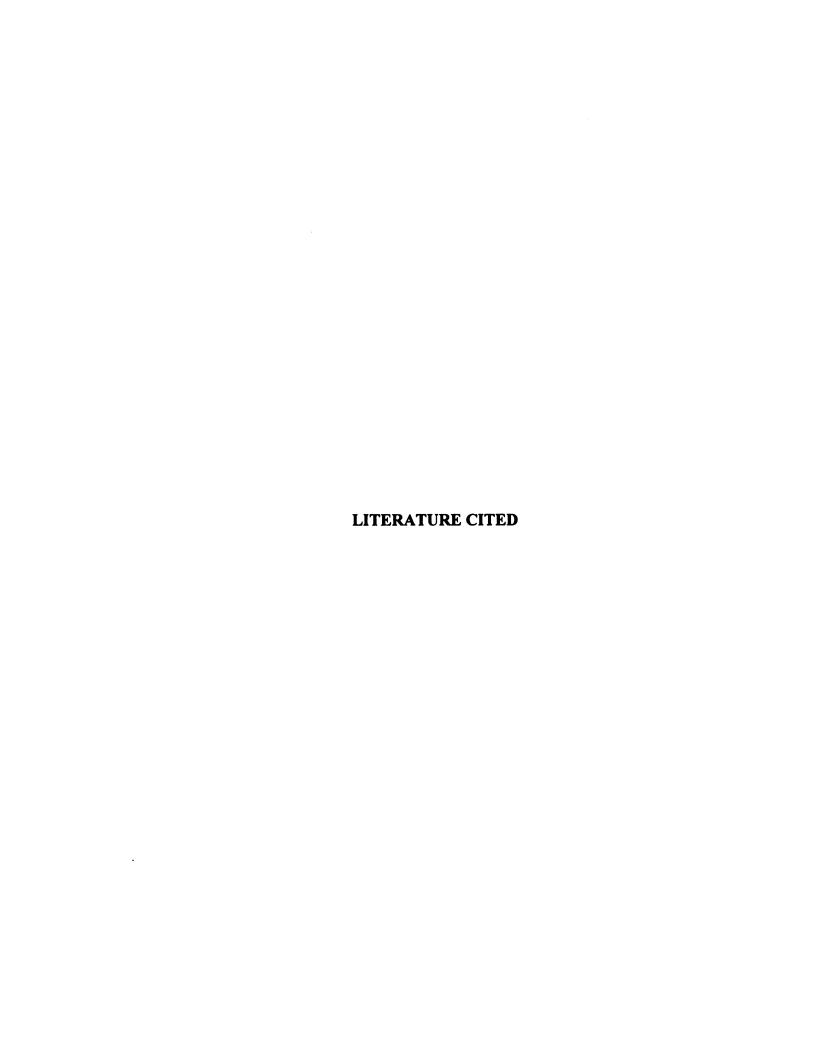
Isogenic Waxy Pioneer 3527



Barrow fitted with a simple T cannula



Digesta collected from a cannulated pig



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