ABSTRACT

PHILLIPS, CHRISTINA ELLEN. Effects of Enzyme Supplementation in Pigs Fed Corn-Soybean Diets. (Under the direction of Dr. Eric van Heugten).

Three experiments were designed to measure the effects of enzyme supplementation on growth performance, ileal digestibility and fecal digestibility of dry matter, protein, gross energy, and amino acids as well as excretion of odor compounds. In experiment 1, an enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase was incorporated in corn or soybean meal for growing pigs. The results showed that addition of the enzyme did not improve fecal digestibility, ileal digestibility, or amino acid digestibility when either chromic oxide or acid insoluble ash were used as markers.

In experiment 2, the same enzyme cocktail was incorporated in corn-soybean meal diets with or without 30% soy hulls for growing pigs. Results showed that addition of the enzyme to corn-soybean meal based diets alone or with 30% added soy hulls did not improve the apparent ileal or fecal digestibilities of nutrients in growing pigs.

In experiment 3, the same enzyme cocktail was incorporated into a corn-soybean meal based diet with 20% soy hulls and compared to typical corn-soybean diet as well as a corn-soybean meal based diet with 20% soy hulls without enzyme. There were no significant effects of the enzyme added to the corn/soybean meal/soy hull diet of pigs on growth performance, carcass characteristics, ammonia emission or odor. However, the addition of enzyme improved gross energy digestibility and also improved the digestibilities of Ca, P, Mg, K, Fe, Zn, Cu, and Mn.

Overall, our data indicate that addition of enzymes to higher fiber diets, not traditional corn soybean diets, show promising results in increasing the digestibility of these feedstuffs.

Effects of Enzyme Supplementation in Pigs Fed Corn-Soybean Meal Diets

by
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BIOGRAPHY

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Chapter 1

Literature Review
Enzyme Supplementation in Swine Diets

Components of Swine Feed

Introduction

Environmental conditions during grain production, genetic differences, and interactions between environment and genetics can cause nutrient composition and bioavailability to differ widely among cereal grains (Sauber, 2001). Differences in soil fertility, growing conditions, maturity, and yield may cause grains of the same genetic strain, grown in different regions to have different nutrient compositions (Sauber, 2001). Grains of different genetic strains grown in the same region may also react differently to identical growing conditions (Sauber, 2001). Increased development of genetically modified grains is likely to increase this variability. The drying, grinding, and flaking processes of grains can modify nutrient availability (Sauber, 2001). The differences in gender and genetic line of pigs consuming the feed may also alter energy and nutrient digestibility (Sauber, 2001). These are all factors to consider when formulating a swine diet, along with an understanding of the nutrient requirements and knowledge of available feed ingredients.

Sources of Energy

Because grains are widely available, energy dense, easily transported, readily consumed by swine, and usually are the most economical source of metabolizable energy (ME) they are the primary ingredient of most swine feed in the Western Hemisphere (Ewan, 1996, Sauber, 2001).

Grains contain energy from three main categories: carbohydrates, protein, and fat. Carbohydrates consist of sugars, starch, and non-starch polysaccharides. Starch is the main carbohydrate and energy source in most diets fed to pigs (NRC, 1998).

Fiber is composed of cellulose, hemi-cellulose, and lignin. Cellulose represents 50 to 80 percent of the dietary fiber in typical feedstuffs, hemi-cellulose represents 20 percent and lignin represents 10 to 50 percent (Van Soest and McQueen, 1973). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) are methods that have been developed to quantify fiber based on solubility. NDF is an estimate of the total plant cell wall, which consists primarily of cellulose, hemi-cellulose, and lignin. ADF is an estimate of just cellulose and lignin, excluding hemi-cellulose (Goering and Van Soest, 1970).

Adding fiber to swine diets decreases the DE concentration of the diet (King and Taverner, 1975; DeGoey and Ewan, 1975; Kennelly et al., 1978; Kennelly and Adherne, 1980b). Pigs attempt to maintain DE intake by increasing feed intake (Baird et al., 1975; Agricultural Research Council, 1981; Low, 1985). However, pigs may lower feed intake when consuming feedstuffs of excessive bulk or reduced palatability, or when crude fiber approaches 10 to 15 percent of the diet (Braude, 1967). During low temperatures, equal growth rates have been maintained by both high-fiber and lower–fiber diets (Coffey et al., 1982; Stahly, 1984). However during higher temperatures, high-fiber diets usually depress the growth rate compared to lower-fiber diets (Coffey et al., 1982; Stahly, 1984).

There are differing hypotheses on the effect of fiber on protein digestibility. One hypothesis is that protein digestibility is not significantly affected by the level of fiber when the source of the fiber does not contain significant amounts of protein (Friend, 1970; Kennelly and Aherne, 1980a). The other hypothesis is that protein digestibility is decreased when there is an increase in the dietary level of fiber (Pond et al., 1962, Cole et al., 1967; Kass et al., 1980; Just et al., 1983; Frank et al., 1983; Noblet and Perez, 1993).

Sources of Protein and Amino Acids

Improving the efficiency of feed utilization is important because feed accounts for the largest cost in pig production. Protein sources are the most costly ingredients in swine feed, so this holds especially true with them in particular (Chiba, 2001). Many different cereal grains, such as corn, barley, sorghum, oats, rye, triticale, and wheat, are included in swine diets to supply energy, essential amino acids, and other nutrients (Gabert et al., 2001, Sauber, 2001). Cereal grains usually provide 30 to 60 percent of the amino acid requirement (NRC, 1998). Soybean meal, cottonseed meal, peanut meal, sunflower meal, and canola meal are sources of vegetable protein that are used in swine diets to ensure adequate amounts of and proper balance of amino acids (NRC, 1988, Gabert et al., 2001). Animal by-products, including fish meal, meat and bone meal, meat meal, blood meal, and spray-dried animal plasma, are also sources of protein used in swine diets (Gabert et al., 2001). To increase levels of amino acids not supplied in adequate amounts from feedstuffs, crystalline amino acids can be added to a diet (NRC, 1998).

The feedstuffs used in a diet determine the levels of additional protein supplementation to that diet that are necessary to provide adequate levels of essential amino acids. Using feedstuffs that contain high quality proteins will meet the essential amino acid requirement at lower dietary protein levels than using feedstuff that contain lower quality proteins (NRC, 1998). Meeting the essential amino acid requirement by feeding lower dietary protein levels or using supplements of crystalline amino acids is important to minimize nitrogen excretion (NRC, 1998).

A fraction of each amino acid in most swine diets cannot be fully broken down, digested and absorbed by the animal (NRC, 1998). Also, not all amino acids that are absorbed are metabolically available (NRC, 1998). Amino acids are more available in some diets than others. For example, the amino acids in some proteins such as milk products, used in weanling pig diets are almost fully available (Southern, 1991; Lewis and Bayley, 1995). Amino acids in other plant proteins are not as available as expected due to antinutritional factors (Southern, 1991; Lewis and Bayley, 1995).

Antinutritional Factors

Swine diets contain antinutritional factors that interfere with digestion of proteins and carbohydrates (Xavier, 2005). The most important antinutritional factors in swine diets are protease inhibitors, lectins, and tannins because they are found in cereal grains and at high levels in soybeans (Huisman, 1989). Other antinutritional factors include non-starch polysaccharides (NSP), phytate, alkaloids, estrogens and flatulence factors. These compounds have negative effects on digestive and metabolic processes of pigs (van Heugten, 2001). It is difficult to predict the effect of antinutritional factors on animal growth performance and metabolism because it is dependent on a variety of factors including animal age, stage of production, type of feedstuff, level of inclusion, and diet composition. Also, because antinutritional factors are present in different combinations in swine diets (van Heugten, 2001).

Trypsin and chymotrypsin inhibitors, the most well known protease inhibitors, are found in large quantities in soybeans, peas and beans (Gueguen et al., 1993; van Heugten, 2001). Growth performance in pigs is reduced by trypsin inhibitors which interfere with the

digestion and absorption of proteins. Lectins are glycoproteins that bind to molecules containing carbohydrates (Grant and van Driessche, 1993; Liener, 1994). *Phaseolus* beans and soybeans contain high levels of lectins, whereas peas and field beans contain lower levels (Gatel, 1994). They are resistant to proteolytic degradation in the digestive tract. Tannins are polyphenolic compounds that also bind to carbohydrates but also have the ability to bind to proteins preventing their absorption (van Heugten, 2001). Sorghum grains contain high concentrations of tannins, faba beans contain moderate concentrations, and barley, millet, peas, *Phaseolus* beans, chickpeas, cowpeas and lentils contain low concentrations (Jansman, 1993; Jansman and Longstaff, 1993).

Non-starch polysaccharides consist of many different plant polymers, including β-glucans, pentosans, cellulose, hemicellulose, pectic substances and α-galactosides (Xavier, 2005). Non-starch polysaccharides are present in legume seeds in the form of oligosaccharides, hemicellulose and pectin, and in grasses as β-linked glucans and pentosans (Xavier, 2005). NSP increase the excretion of nitrogen in feces by reducing the absorption of protein therefore lowering plasma urea (Xavier, 2005).

Supplemental Enzymes used to Improve Pig Performance

Introduction

Enzymes are feed additives along with animal drugs, growth-promoting minerals, organic acids, and probiotics. Enzymes are proteins consisting of long chains of amino acids (Sheppy, 2001). They are catalysts and are involved in all anabolic and catabolic pathways of digestion and metabolism (Liu, 1997). The primary objective of adding enzymes to animal feeds is to improve the utilization of nutrients in feedstuffs

(Sheppy, 2001). In doing so, pig performance is increased and waste is minimized, thus having beneficial effects on the environment. This is achieved by one or more of the following mechanisms: 1) degradation of specific bonds in ingredients not usually degraded by endogenous digestive enzymes, 2) degradation of anti-nutritive factors that lower the availability of nutrients, 3) increased accessibility of nutrients to endogenous digestive enzymes, and 4) supplementation of enzyme capacity of young animals (Sheppy, 2001).

Proteins, fat, and carbohydrates must be broken down in order to be absorbed (Liu, 1997). They are absorbed in the form of free amino acids, dipeptides, short-chain peptides, fatty acids, and monosaccharides, or oligopeptides–saccharides (Liu, 1997). Specific enzymatic reactions must occur in order for these digestive processes to take place. The endogenous enzymes produced by animals may be insufficient to break down and absorb all of the nutrients available in feedstuffs (Liu, 1997). Supplementation of diets with exogenous enzymes will improve digestive capacity. Enzymes will further break down the cell-wall structure, leading to increased nutrient availability and changes
in the physical properties of non-starch polysaccharides (Liu, 1997). Non-starch polysaccharides will be more efficiently digested causing changes in the composition and content of bacteria in the small and large intestine (Liu, 1997). This will result in reduced maintenance requirements and therefore reduce waste and environmental pollution (Liu, 1997). Enzyme addition can also improve the accuracy of feed formulations by reducing the variability in nutritive value between feedstuffs of the same category (Sheppy, 2001).

Feed enzymes are becoming increasing popular due to changing social views and emphasis on efficient animal production with minimal environmental damage. Consumers

are demanding an all-natural feed supply. Feed enzymes are produced by microbial fermentation, and can be manufactured (Liu, 1997). Feed enzymes are acceptable alternatives to growth promoting antibiotics because they are common in daily life.

The poultry industry is the largest user of feed enzymes (Marquardt, 1997). They are accepted as a feed enhancer to improve nutrient digestibility and efficiency of nutrient utilization in poultry (Liu, 1997). Progress with exogenous enzymes used in pig diets has been slower due to variable response and difficulty in measuring their effects commercially (Sheppy, 2001; Liu, 1997). In European markets where viscous cereals are the predominant energy source, fiber-degrading enzymes are prevalent (Sheppy, 2001). However, enzyme manufacturers find it challenging to market their products in North American markets, where corn-soybean are predominant components in swine feeds (Sheppy, 2001). Corn-soybean meal diets are historically regarded as the 'gold standard' in terms of consistency of animal response (Sheppy, 2001). However, this diet is not digested 100%, and many factors cause variability, therefore it still has room for improvement. Phytase has already gained both global acceptance and global application in the animal feed industry (Sheppy, 2001). Now the emphasis has turned to the apparent potential to improve the availability of amino acids and trace minerals in the diet, along with improving the digestibility of fiber and starch components of corn-soybean meal diets (Sheppy, 2001).

Two diet formulation methods exist to apply enzyme treatments in practice: (1) formulate diets to a regular nutrient content and supplement with enzyme with the goal of an improvement in feed efficiency, or (2) formulate diets to a reduced nutrient content and count

on an increased availability of nutrients by the enzyme, while reducing feed costs. There are four types of enzymes that can be used in swine diets: enzymes that break down fiber, protein, starch and phytic acid. Tables 1, 2 and 3 summarize the results of several studies on the growth performance, ileal digestibility, and fecal digestibility responses of pigs fed diets with enzyme supplements.

Fiber-Degrading Enzymes

Fiber, a type of non-starch polysaccharide, is defined as the plant cell wall components that are left over after enzymes break down starch, fat and protein. Pigs do not produce enzymes to digest fiber. There is a direct, negative correlation between dietary fiber content and metabolizable energy value (Grieshop et al., 2001). As fiber content increases, the ability of the pig to utilize the energy of the feedstuff decreases. Fiber also produces negative effects on amino acid and fat digestibility (Grieshop et al., 2001). Corn and soybean meal have a relatively low content of fiber compared to other ingredients (NRC, 1998). Supplemental enzymes have been explored to improve the digestibility of fiber to improve growth performance and reduce the incidence of digestive disorders linked with non-digestible fiber in the diet.

Much of the research with fiber-degrading enzymes has been conducted with beta-glucanase (Graham, 1986; Baas and Thacker, 1996; Li et al., 1996a; Li et al., 1996b; Baidoo et al., 1998; Thacker et al., 1988; Graham et al., 1989; Thacker et al., 1989; Bedford et al., 1992; Thacker et al., 1992b; Gdala et al., 1997; Jensen et al., 1998), which is commonly used in diets that consist of barley and/or wheat because of their high fiber content. Some research has been conducted with beta-glucanase mixed with other fiber degrading enzymes such as

xylanase, galactosidase, mannanase, mannosidase, or pentosanse (Graham et al., 1988; Thacker et al., 1992a; Hogberg et al., 2004).

Other fiber-degrading enzymes that have been researched include galactosidase (Gdala et al., 1997a; Gdala et al., 1997; Baucells et al., 2000), xylanase (Gdala et al., 1997; Mavromichalis et al., 2000; Choct et al., 2004; Barrera et al., 2004; Diebold et al., 2004), pentosanase (Thacker et al., 1991; Bedford et al., 1992), beta-mannanase (Pettey et al., 2002), carbohydrase mixtures (Kim et al., 2003; Ji and Kim, 2004, Kim et al., 2006), and a fiber degrading enzyme cocktail (Moeser and van Kempen, 2002).

β-glucanase

In a study by Graham et al. (1986) β-glucanase supplementation of a barley-based diet fed to 3-4 month old pigs did not significantly affect the apparent digestibilities of any components at the terminal ileum or faeces. These results are supported by an experiment by Baas and Thacker (1996) who supplemented barley-based growing-finishing pig rations with any of the five enzyme preparations, which failed to improve average daily gain, feed intake or feed efficiency. The digestibility coefficients for dry matter, crude protein and energy for pigs fed a diet supplemented with one of the five enzyme preparations were significantly higher compared with all other diets. The remaining four enzyme-supplemented diets did not differ from the control diet. These authors concluded, there would appear to be little justification for the routine supplementation of barley-based grower-finisher diets with β-glucanase.

Graham (1989) found there was no effect of β-glucanase supplementation in the barley-based diets on the ileal or fecal apparent digestibilities of dry matter, energy or crude

protein. Gdala et al. (1997) showed the supplementation of soybean meal with β-glucanase had no effect on the apparent ileal or fecal digestibilities of dry matter, crude protein, and fat. In a study by Jensen et al. (1998) live weight gain, feed intake and feed/gain ratio were not influenced by the β-glucanase enzyme treatment supplemented to barley-based diets.

Li et al. (1996a) revealed β-glucanase supplementation to a hulless barley-soybean meal diet increased the ileal digestibilities of GE, CP, and the majority of the amino acids and the fecal digestibilities of GE, CP, and all amino acids measured. There was no effect of β-glucanase supplementation to a wheat-soybean meal diet on the ileal digestibilities of any criteria measured. However, supplementation of β-glucanase to the wheat-soybean meal diet increased the fecal digestibility of energy.

Li et al. (1996b) found β-glucanase supplementation to a barley-soybean meal diet improved the digestibilities of dry matter, energy and crude protein in young pigs. There was no effect of β-glucanase supplementation to a wheat-soybean meal, corn-soybean-meal or rye-soybean meal diet.

Baidoo (1998) performed digestion trials with two hulless barley (Condor and CDC Buck) based diets and demonstrated that enzyme supplementation significantly improved the digestibilities of DM, GE and amino acids in hulless barley/canola meal diets for young pigs. This improvement was confirmed in a performance study that showed a significant increase in average daily gain.

Thacker (1988) found that β-glucanase supplementation to a barley-soybean meal diet improved the digestibilities of dry matter, crude protein and energy. The higher digestibility coefficients for dry matter, crude protein and energy were not reflected in a significant

improvement in pig performance. Beta-glucan is not a major factor detracting from the nutritional value of hulless barley as a feedstuff for use in swine production. As a consequence, there would appear to be little justification for the routine inclusion of beta-glucanase in hulless barley-based diets fed to swine (Thacker, 1988).

In another study by Thacker (1989), there was no significant improvement in average daily gain, feed intake or feed efficiency as a result of supplementation with β-glucanase either when fed alone or in combination with sodium bentonite. The digestibility of dry matter and crude protein was not improved as a result of supplementation with β-glucanase or the combination of β-glucanase and sodium bentonite. However the digestibility coefficient for energy was significantly higher for pigs fed the diet supplemented with both sodium bentonite and β-glucanase in comparison with those fed β-glucanase alone. The overall results of these experiments

support a previous study (Thacker, 1988), which cast doubt on the efficacy of beta-glucanase supplementation as a means of improving the performance of pigs fed barley-based diets.

In experiment 1 (Thacker et al., 1992b), supplementation with β-glucanase or propionic acid alone, significantly increased dry matter digestibility. However, when the additives were fed in combination, the digestibility coefficient for dry matter was not significantly different from the control. Digestibility coefficients for crude protein and energy were not affected by any treatment. Growth rate, feed intake, feed efficiency and carcass characteristics were also not improved by supplementation with enzyme or acid treatment. In experiment 2 (Thacker et al., 1992b), digestibility coefficients for dry matter, crude protein and energy were not affected by enzyme or acid treatment. In addition, there

were no differences in growth, feed intake or feed efficiency as a result of any treatment. Supplementation with organic acids would therefore appear to be ineffective in potentiating the response to dietary β-glucanase in pigs fed barley-based diets. The results of these experiments support previous reports from the same laboratory showing little benefit from supplementation of swine diets with beta-glucanase or pentosanase (Thacker et al., 1992a, Thacker et al., 1992b).

A study by Bedford (1992) which showed a significant improvement in weight gain was brought about by beta-gluclanase supplementation of a hulless barley-based diet.

The results concerning the effect of β-glucanase on nutrient digestibility in the GI tract are contradictory, presumably because the degradation of β-glucan is influenced by several factors, including age of animals, feedstuffs included in the diet and source of the enzyme (Gdala et al., 1988).

β-glucanase and other enzymes

In a study by Graham et al. (1988), the inclusion of a β-glucanase and xylanase enzyme preparation increased the ileal digestibility of crude protein, however enzyme supplementation had no effect on apparent fecal digestibility.

Thacker et al. (1992a) observed that supplementation of a barley-based diet with an enzyme preparation of β-glucanase and pentosanase or salinomycin, alone or in combination, had no significant effect on weight gain, feed intake, feed efficiency or nutrient digestibility. Pigs fed the combination of additives had a digestibility coefficient for crude protein significantly higher than those fed the unsupplemented diet or pigs fed either additive individually. Supplementation with the enzyme preparation, alone or in combination, had no

significant effect on weight gain, feed intake or feed efficiency of pigs fed rye-based diets (Thacker et al. 1992a). Supplementation with the enzyme preparation of β-glucanase and pentosanase or salinomycin, alone or in combination, had no significant effect on the digestibility of dry matter in comparison with the unsupplemented rye-based control diet. However, digestibility coefficients for crude protein and gross energy were significantly higher for pigs fed the rye-based diets supplemented with salinomycin or the combination of additives in comparison with the unsupplemented rye-based control (Thacker et al. 1992a).

Hogberg et al. (2004) found no improvement in average daily gain or ileal or total tract digestibilities when a β-glucananse and xylanase enzyme mixture was supplemented in pigs fed a triticale, wheat and barley mixture.

Other fiber-degrading enzymes

The supplementation of α-galactosidase to pig diets revealed conflicting results. Gdala et al. (1997a) found that the addition of α-galactosidase to a lupin seed meal diet did not significantly affect crude protein digestibility, but increased the ileal digestibility of methionine and threonine. Gdala et al. (1997) observed that supplementation of soybean meal with α-galactosidase or xylanase had no effect on the apparent ileal or fecal digestibilities of dry matter, protein and fat. Baucells et al. (2000) found the supplementation of galactosidase to soybean meal based diets of grower pigs caused a significant increase in gain/feed, however, there was no change in weight gain. Supplementation of galactosidase to soybean meal based diets of finisher pigs however, resulted in a significant increase in gain/feed and weight gain. Enzyme supplementation improved dry matter and crude protein digestibility of the finishing diets (Baucells et al., 2000).

There are mixed results on the addition of xylanase to pig diets. Mavromichalis et al. (2000) found that enzyme supplementation of xylanase to a wheat-based diet had no effect on growth performance or fecal digestibilities in pigs. However, Choct et al. (2004), Barrera et al. (2004), and Diebold et al. (2004) found at least some improvements in digestibility and pig performance with the addition of xylanase.

Choct et al. (2004) revealed that both steeping and xylanase addition increased feed intake and growth rate, suggesting that both techniques influence the non-starch polysaccharide composition of a wheat-based diet. Barrera et al. (2004) found an increase in the apparent ileal digestibilities of crude protein, lysine, methionine, and threonine as a result of xylanase supplementation to a wheat-based diet, however, it did not signifanlty improve growth performance or gain:feed ratio. Diebold et al. (2004) showed that there was a positive effect on ileal digestibility of crude protein and methionine with xylanase and phospholipase supplementation. Supplementation of both enzymes provided the largest improvement in the ileal digestibility value of crude protein and methionine. There was no effect of xylanase or the combined enzyme supplementation on the digestibilities of crude protein or gross energy. However, supplementation with phospholipase alone decreased the total-tract digestibilities of crude protein and gross energy (Diebold et al., 2004).

Neither Thacker et al. (1991) nor Bedford et al. (1992) found positive results with the addition of pentosanase to pig diets. Thacker et al. (1991) found that enzyme supplementation with pentosanase did not have a significant effect on fecal digestibility of rye or barley-based diets. Enzyme supplementation did not affect average daily feed intake or average daily gain in either experiment; however, pigs fed the enzyme-supplemented diet

were more efficient in one experiment, but not in the second. Bedford et al. (1992) observed that pentosanase supplementation of rye-based diets for weanling pigs did not provide any benefit in terms of animal performance or nutrient digestion.

Pettey et al. (2002) found that the addition of β-mannanase to pig diets improved growth performance but not ileal digestibility. In experiments 1 and 2, pigs fed corn-soybean meal diets with β-mannanase had greater gain:feed ratio than pigs fed diets with no added enzyme. In experiment 3, the addition of β-mannanase increased average daily gain and the gain to feed ratio. There were no differences in the apparent digestibility of energy, crude protein, or dry matter in pigs supplemented with enzyme (Pettey et al., 2002).

The addition of carbohydrases had mixed results when added to diets for finishing pigs (Kim et al., 2003), but positive results were observed when carbohydrases are supplemented to sow diets (Ji and Kim, 2004). Kim et al. (2003) reported that gain:feed ratio was higher for pigs consuming a corn-soybean meal based diet supplemented with carbohydrases than for pigs fed the control diet; however, there were no differences in average daily gain or average daily feed intake. Apparent ileal digestibility of lysine, threonine, and tryptophan but not methionine was greater in the carbohydrase diet than the control diet. In the study performed by Ji and Kim (2004) there was no difference in sow body weights at farrowing between the corn-soybean meal based control group and the carbohydrase group. However the carbohydrase group had lower body weight loss of sows after lactation than the control group.

A fiber degrading enzyme was added to a corn and soybean meal diet in which soy hulls were added to increase fiber (Moeser and van Kempen, 2002). Inclusion of the fiber-

degrading enzyme improved dry matter digestibility by 2% and energy digestibility by 3% while decreasing fecal output by 10%. A trend of increased nitrogen digestibility was also observed with the addition of the enzyme. Moeser and van Kempen (2002) concluded that this particular enzyme was able to effectively break down fiber, allowing for more nutrient utilization.

Protein-Degrading Enzymes

Proteases are of particular interest because protein is the second most expensive item in animal diets next to energy. Protease enzyme supplementation is thought to be beneficial particularly in young pigs, 4-6 weeks of age, due to the fact that the proteolytic and amylolytic digestive system is not fully developed. Supplemental enzymes may be used during the weaning period to lessen the nutritional stress of abruptly switching from a liquid diet to a solid diet. Proteases can also be used to increase the availability of protein origin from plant sources such as soybean meal by helping to breakdown proteinaceous ANF. The response of pigs to supplementation of diets with proteolytic enzymes alone had been studied in earlier experiments. The results were inconclusive (Caine et al., 1997; Gdala et al., 1997).

Caine et al. (1997) found that neither spraying nor incubating soybean meal with protease improved the growth performance or apparent digestibilities of crude protein and amino acids in newly weaned pigs. Gdala et al. (1997) reported that the supplementation of soybean meal with protease had no effect on the apparent ileal or fecal digestibilities of dry matter, protein and fat.

Starch-Degrading Enzymes and Multi-enzyme Preparations

The digestibility of feed ingredients is usually measured relative to corn because of it's consistently high digestibility (Sheppy, 2001). However, corn is not digested 100% and therefore could benefit from the addition of an exogenous starch-degrading enzyme. Adding amylase to a swine diet can help increase the break down of starch, increasing nutrient absorption and improving growth rates (Sheppy, 2001). Some of the research with starch-degrading enzymes has been conducted with α-amylase (Gdala et al., 1997). However, most of the work conducted with starch-degrading enzymes has been done in combination with fiber and/or protein-degrading enzymes (Inborr and Ogle, 1988; Tangendjaja et al., 1988; Inborr et al., 1993; Officer, 1995; Gill et al., 2000; Medel et al., 2002; Omogbenigun et al., 2004; Nyachoti et al., 2006). Gdala et al. (1997) found that the supplementation of soybean meal with α-amylase had no effect on the apparent ileal or fecal digestibilities of dry matter, protein and fat.

Inborr and Ogle (1988) studie the effects of treating cooked and toasted barley with carbohydrate degrading enzymes (α-amylase, β-glucanase and glucoamylase) and then adding a mixture of amylolytic, cellulolytic and proteolytic enzymes to the complete diets, on the performance and feed intake of piglets. There were no significant treatment effects on daily liveweight gain, growth performance or feed conversion ratios over the entire experimental period. Although not significant, improved feed conversion ratios noted for all the enzyme treated diets suggest a positive effect on diet digestibility.

In the study by Tangendjaja et al. (1988), amylase and cellulase were evaluated as a means of improving the feeding value of rice bran as a feed ingredient for pigs. Rice bran

contains more than 30% starch and about 29% of neutral detergent fiver. This high fiber content is a limiting factor in the utilization of rice bran for monogastric animals. There were no significant differences in the daily gain of pigs fed rice bran treated with enzymes. Enzyme treatment resulted in higher feed intake, causing feed per gain ratios to be higher than that of untreated rice bran or cooked rice bran. The results of the digestion trial showed no differences among treatments in digestible dry matter, crude protein or gross energy (Tangendjaja et al., 1988).

Inborr et al. (1993) found no significant difference in growth performance when comparing a barley-wheat based control diet with a diet supplemented with a multi-enzyme preparation (β-glucanase, xylanase, and α-amylase). Crude protein digestibility was numerically higher in pigs fed the enzyme-treated diets compared with the pigs fed the control diet. Results agree with Officer (1995) and Medel et al. (2002). Officer (1995) indicated that multi-enzyme supplementation of unpelleted wheat-based diets had either no effect, or was detrimental to piglet performance. Responses seen in pelletted feeds are often more prevelant than responses seen in unpelleted feeds. This is because the pelleting of diets is an effective way to improve feed efficiency for all phases of swine production (Cline and Richert, 2001). Medel et al. (2002) found no effect of the multi-enzyme preparation (α-amylase, xylanase, and β-glucanase) supplementation of barley-based diet on growth performance or fecal digestibility of piglets.

In the study by Gill et al. (2000) there was no significant difference in average daily gain or feed intake when comparing barley-based or sugar beet pulp-based control diets *versus* enzyme supplemented diets. However, piglets given enzyme-supplemented diets

showed a significant improvement in the conversion of gain to feed as compared with those given unsupplemented diets. The results of the digestibility trial showed that there were no significant effects of enzyme supplementation on dry matter, crude protein or gross energy digestibility (Gill et al., 2000).

Enzyme supplementation of a multi-enzyme blend to pigs fed a barley-based diet with raw peas improved the apparent ileal digestibilities of lysine and threonine, but not crude protein (Nyachoti et al., 2006). Enzyme supplementation of a multi-enzyme blend to pigs fed a barley-based diet with micronized peas had no effect on the apparent digestibilities of crude protein or amino acids. Two experiments with young pigs were conducted to investigate the effect of multienzyme preparations on nutrient digestibility and growth performance in pigs fed corn/barley/wheat based-diets (Omogbenigun et al., 2004). The multienzyme preparations used were: 1) cellulase, galactanase, and mannanase, 2) cellulase and pectinase, and 3) cellulase, galactanase, mannanase, and pectinase. Pigs fed enzyme-supplemented diets had a higher average daily gain and gain:feed ratio than those fed the control diet. However, there were no differences in average daily gain and gain:feed ratio among the enzyme-supplemented diets. The results of this study demonstrate that, with an appropriate enzyme combination, young pigs can effectively utilize diets containing ingredients that are normally poorly utilized (Omogbenigun et al., 2004). This will increase the flexibility in feed formulation allowing for the use of lower-quality ingredients, with the potential to reduce feed costs. Results agree with Baidoo (1997). Apparent ileal digestibility of crude protein was similar among enzyme-supplemented diets and all were higher than that observed for the control diet. Compared with the control, enzyme supplementation increased

total-tract digestibilities of dry matter, crude protein and gross energy. This is similar to the findings of Li et al. (1996) who found an increase of gross energy and crude protein digestibilities, when a hulless barley/soybean meal diet supplemented with 0.2% β-glucanase was fed to young pigs. Conversely, Thacker et al. (1992) in studies with 7- to 8- week old pigs, reported no effect of β-glucanase supplementation on the fecal digestibilities of crude protein and gross energy.

Phytic acid-Degrading Enzymes

Phosphorus is supplemented in swine diets because most of the phosphorus found in feedstuffs is tied up as phytic acid and cannot be broken down by endogenous enzymes (Sheppy, 2001). Phytase can be added to the diet to break down phytic acid in feedstuffs, and allow the pig to utilize the phytate phosphorus. This helps the environment because the pig produces less phosphorus in feces, and it can also reduce the cost of feed because supplemental phosphorus can be reduced or eliminated from the diet (Sheppy, 2001).

Conclusions

There are inconsistent responses in growth performance and digestibility from the supplementation of feed enzymes to swine diets. These inconsistent responses may be due to a combination of factors: 1) a complex interaction between the endogenous and exogenous enzymes and substrate, 2) age and sex of the pigs, 3) different feedstuffs and 4) the experimental procedure (Liu, 1997).

As consumer resistance to traditional growth promoters increases, the replacement by more consumer-friendly substances, such as natural enzymes, will become more common. The addition of enzymes to feed will not only lead to an improved nutritional

value of the feed but may have a major beneficial impact on the environment in terms of composition and quantity of excreta.

There are still many obstacles to overcome before enzymes can be globally accepted as an animal feed enhancer. An enhanced understanding of the interaction between the enzyme, its substrate, and the environment of the animal's gut is needed (Sheppy, 2001). It is also important to understand how and which feedstuffs will respond to different enzymes at the amount that will have the most economic and cost-effective response. (Sheppy, 2001). A major concern for enzyme manufacturers is the enzyme's ability to withstand heat processing in order to be manufactured in pelleted feeds (Sheppy, 2001). Another concern is the ability to monitor enzyme activity. Each enzyme manufacturer has its own method, and measurements of enzyme activity in feeds are often inaccurate and subject to high variation (Liu, 1997). Therefore, it is important to develop a consistent and reliable method for both quality control and rating of the enzyme products sold. After all of these concerns are addressed, the use of enzymes in swine feed manufacturing will be cost effective and beneficial to the environment.

The possible impact of enzymes on the environment, odor, and fiber

Introduction

The agriculture industry is not the only industry that produces waste materials. However, in order to prevent environmental pollution from these wastes they must be well managed. In recent years, more concerns have been raised regarding the environmental pollution resulting from animal husbandry. Nitrogen and phosphorus in animal excreta are the biggest threats to our environment. Air can be polluted by ammonia, which is caused by

excessive nitrogen (Liu, 1997). Soil and water can be polluted by nitrate, a compound that is converted from nitrogen by bacteria (Liu, 1997). If fiber digestibility can be increased due to the addition of feed enzymes to swine diets, then it is possible that there would be a decrease in fecal output. This would also increase animal performance while decreasing environmental pollution and odor.

The impact of enzymes in the diet on the environment

Many factors determine how well a non-ruminant animal can digest a feed grain. These factors include the age and species of the animal as well as the type, state of maturity, and location the grain was grown (Bedford, 1995). An important concept required to effectively use enzyme supplementation to increase digestibility is to understand what is causing the decrease in digestion in the intestinal lumen. This will vary by feedstuff and age of the animal, for example a lack of proteolytic and amylolytic enzyme activity in the young pig. Understanding these important concepts will result in the more efficient use of grains through enzyme supplementation, and a reduction in manure output. With less animal waste there will be obvious environmental benefits (Bedford, 1995).

Monogastric animals lack endogenous phytase required to free phosphorus that is bound to phytate. This undigested phosphorus contributes to phosphorus pollution. Inorganic phosphorus may be added to diets to meet the requirements for phosphorus in swine. The addition of inorganic phosphorus to diets takes up space that could otherwise be used by cheaper ingredients. There have been studies in which phytase addition to diets deficient in available phosphorus enhanced the utilization of phosphorus, as well as studies that have shown improved growth performance with low levels of available

phosphorus (Liu, 1997). Enzymes which increase feed digestibility, will decrease the excretion of manure, ultimately minimizing environmental pollution. As environmental damage becomes an important issue with the consumer, the interest in the use of enzymes in swine diets will increase.

The impact of enzymes in the diet on odor

Manure from pigs is mostly a mixture of urine and feces. It contains undigested dietary components, endogenous end products, and indigenous bacteria from the lower gastrointestinal tract (Sutton et al., 1999). After feces is excreted many compounds are emitted from the microbial fermentation that took place in the gastrointestinal tract and its continued fermentation. These compounds include odorous volatile organic compounds, short-chain VFA, and other volatile carbon-, nitrogen-, and sulfur-containing compounds (Sutton et al., 1999). The conversion of urea to ammonia occurs shortly after excretion. Poorly digested proteins lower the efficiency of nitrogen utilization therefore contributing to odor problems and ground water pollution (Moughan, 2000). Perhaps with the addition of protease enzymes, proteins can be better digested. When an ingredient or diet is utilized more efficiently, there is a reduction in manure output and environmental pollution is lessened (Moughan, 2000).

Feed additives are being considered in controlling odor from swine production as the issue is gaining more public, regulatory, and legal attention (Williams, 1995). Feed additives have been used to try and control odors, including those that bind ammonia, change digesta pH, affect specific enzyme activity, and mask odors (Sutton et al., 1999).
These attempts have been either costly or not consistently successful. Sutton et al. (1999)

focused on manipulating the diet when conducting odor research. It is thought that increasing the nutrient utilization of the diet will reduce the products excreted, subsequently minimizing odors. Manipulating the diet to enhance the microbial metabolism in the lower digestive tract will reduce the release of odor-causing compounds. If it is possible to manipulate the diet in order to change the physical characteristics of urine and feces, this will reduce odor emissions. Excess degradable proteins and lack of specific fermentable carbohydrates during microbial fermentation cause the primary odor-causing compounds. Excessive protein intake will increase both ammonia emission and odor emission (Le et al., 2005). Therefore we can hypothesize that the supplementation of protein degrading enzymes to swine diets will decrease both ammonia emission and odor emission.

The impact of enzymes on fiber

Non-ruminants are unable to utilize cell walls as efficiently as ruminants except through hindgut fermentation (Dierick et al., 1984). Therefore nutrients from the break down of fiber must be made available for digestion within the small intestine prior to entering the ileum (Dierick et al., 1994). This is a primary objective of the use of feed additives. This approach may increase the number of feed ingredients used in pig diets, for example lower quality feedstuffs may be used, as well as improve the utilization of currently used feedstuffs (Dierick et al., 1994). Non-starch polysaccharides are complex, and for that reason an enzyme must have many different specifications. Protein and storage polysaccharides may be released from the partial degradation of endosperm walls,
causing an increase in the amount of diet digested in the small intestine (Dierick et al., 1994). As fiber content in a diet increases, the ability of the pig to utilize the energy of the feedstuff

decreases. With an increase in fiber digestion from the addition of enzymes, an increase in animal performance is to be expected.

Conclusion

In order to obtain maximum benefit from inclusion of enzymes in feed, it is necessary to ensure that the enzyme or enzyme cocktail added is chosen on the basis of feed composition as well as the age and species of the animal. A choice of enzyme cocktail rather than addition of a single enzyme may be more effective and the proportion of each enzyme present in a cocktail should reflect the proportion of its substrate present in the feed. If used in this manner, enzymes have the capability to reduce environmental pollution and odor, and increase animal performance.

Literature Cited

Agriculture Research Council. 1981. The Nutrient Requirements of Pigs: Technical Review. Rev. ed. Slough, England. Commonwealth Agricultural Bureaux. xxii, 307 pp.

Baidoo, S. K., Y. G. Liu, and D. Yungblut. 1998. Effect of microbial enzyme supplementation on energy, amino acid digestibility and performance of pigs fed hulless barley based diets. Can. J. Anim. Sci. 78:625-631.

Baird, D. M., H. C. McCampbell, and J. R. Allison. 1975. Effect of level of crude fiber, protein and bulk in diets for finishing hogs. J. Anim. Sci. 41:1039-1047.

Barrera, M., M. Cervantes, W. C. Sauer, A. B. Araiza, N. Torrentera, and M. Cervantes. 2004. Ileal amino acid digestibility and performance of growing pigs fed wheat-based diets supplemented with xylanase. J. Anim. Sci. 82:1997-2003.

Bass, T. C., and P. A. Thacker. 1996. Impact of gastric pH on dietary enzyme activity and survivability in swine fed β-glucanase supplemented diets. Can. J. Anim. Sci. 76:245-252.

Baucells, F., J. F. Perez, J. Morales, and J. Gasa. 2000. Effect of α-galactosidase supplementation of cereal-soya-bean-pea diets on the productive performances, digestibility and lower gut fermentation in growing and finishing pigs. J. Anim. Sci. 71:157-164.

Bedford, M.R., 1995. Mechanism of action and potential environmental benefits from the use of feed enzymes. Anim. Feed Sci. Technol. 53:145-155.

Bedford, M.R., J. F. Patience, H. L. Classen, and J. Inborr. 1992. The effect of dietary enzyme supplementation of rye- and barley-based diets on digestion and subsequent performance in weanling pigs. Can. J. Anim. Sci. 72: 97-105.

Braude, R. 1967. The effect of changes in feeding patterns on the performance of pigs. Proc. Nutr. Soc. 26:163-181.

Caine, W. R., W. C. Sauer, S. Tamminga, M.W.A. Verstegen, and H. Schulze. 1997. Apparent ileal digestibilites of amino acids in newly weaned pigs fed diets with protease-treated soybean meal. J. Anim. Sci. 75:2962-2969.

Chiba, L. I. 2001. Protein Supplements. In: Swine Nutrition, Second Ed. Austin J. Lewis and L. Lee Southern, Eds., U.S.A. Pp. 803-837.

Choct, M., E. A. D. Selby, D. J. Cadogan, and R. G. Campbell. 2004. Effect of liquid to feed ratio, steeping time, and enzyme supplementation on performance of weaner pigs. Australian Journal of Agricultural Research. 55:247-252.

Cline, T. R. and B. T. Richert. 2001. Feeding Growing-Finishing Pigs. In: Swine Nutrition, Second Ed. Austin J. Lewis and L. Lee Southern, Eds., U.S.A. Pp. 717-723.

Coffey, M. T., R. W. Seerley, D. W. Funderburke, and H. C. McCampbell. 1982. Effect of heat increment and level of dietary energy and environmental temperature on the performance of growing-finishing swine. J. Anim. Sci. 54:95-105.

Cole, D. J. A., J. E. Duckworth, and W. Holmes. 1967. Factors affecting involuntary feed intake in pigs. 1. The effect of digestible energy content of the diet on the intake of castrated male pigs housed in holding pens and in metabolism crates. Anim. Prod. 9:141-148.

DeGoey, L. W., and R. C. Ewan. 1975. Effect of level of intake and diet dilution on energy metabolism in the young pig. J. Anim. Sci. 42:1045-1051.

Diebold, G., R. Mosenthin, H.-P. Piepho, and W. C. Sauer. 2004. Effect of supplementation of xylanase and phospholipase to a wheat-based diet for weanling pigs on nutrient digestibility and concentrations of microbial metabolites in ileal digesta and feces. J. Anim. Sci. 82:2647-2656.

Dierick, N. A. and J. A. Decuypere. 1994. Enzymes and growth in pigs. Pages: 169-195. In: Principles of Pig Science. D. J. A. Cole, J. Wiseman, and M. A. Varley, Eds., U.K.

Ewan, R. C. 1996. Energy Values of Feed Ingredients. 5th Revised Edition. Ames: Iowa State University. 101 pp.

Frank, G. R., F. X. Aherne, and A. H. Jensen. 1983. A study of the relationship between performance and dietary component digestibilities by swine fed different levels of dietary fiber. J. Anim. Sci. 57:645-654.

Friend, D. W. 1970. Comparison of some milling products of barley and rye when fed in diets to rats. Can. J. Anim. Sci. 50:345-348.

Gabert, V. M., H. Jorgensen, and C. M. Nyachoti. Bioavailability of Amino Acids in Feedstuffs for Swine. In: Swine Nutrition, Second Ed. Austin J. Lewis and L. Lee Southern, Eds., U.S.A. Pp. 151-186.

Gatel, F. 1994. Protein quality of legume seeds for non-ruminant animals: a literature review, Anim. Feed Sci. Technol., 45:317-348.

Gdala, J., H. N. Johansen, K. E. Bach Knudsen, I. H. Knap, P. Wagner, and O. B. Jorgensen. 1997. The digestibility of carbohydrates, protein and fat in small and large intestine of piglets fed non-supplemented and enzyme supplemented diets. Anim. Feed Sci. Technol. 65:15-33.

Gdala, J., A. J. M. Jansman, L. Buraczewska, J. Huisman, and P. van Leeuwen. 1997a. The influence of α-galactosidase supplementation on the ileal digestibility of lupin seed carbohydrates and dietary protein in young pigs. Anim. Feed Sci. Technol. 67:115-125.

Gill, B. P., J. Mellange, and J. A. Rooke. 2000. Growth performance and apparent nutrient digestibility in weaned piglets offered wheat-, barley- or sugar-beet pulp-based diets supplemented with food enzymes. Anim. Sci. 70:107-118.

Goering, H. K., and P. J. Van Soest. 1970. Forage fiber analysis: Apparatus, reagents, procedures and some applications. Agricultural Handbook 379. Washington, D.C.: U.S. Department of Agriculture. iv, 20 pp.

Graham, H., J. G. Fadel, C. W. Newman, and R. K. Newman. 1989. Effect of pelleting and β-glucanase supplementation on the ileal and fecal digestibility of a barley-based diet in the pig. J. Anim. Sci. 67:1293-1298.

Graham, H., K. Hesselman, and P. Aman. 1986. Influences of β-glucanase supplementation of a barley-based diet in the pig gastrointestinal tract. Nutr. Rep. Int. 34:1089-1096.

Graham, H., W. Lowgren, D. Pettersson, and P. Aman. 1988. Effect of enzyme supplementation on digestion of a barley/pollard-based pig diet. Nutr. Rep. Int. 38:1073-1079.

Grant, G., and E. van Driessche. 1993. Legume lectins: Physicochemical and nutritional properties. In Recent Advances of Research in Antinutritional Factors in Legume Seeds, van der Poel, A. F. B., J. Huisman, and H. S. Saini, Eds., Wageningen Pers, The Netherlands, 219.

Grieshop, C. M., D. E. Reese, and G. C. Fahey. 2001. Nonstarch Polysaccharides and Oligosaccharides in Swine Nutrition. In: Swine Nutrition, Second Ed. Austin J. Lewis and L. Lee Southern, Eds., U.S.A. Pp. 563-383.

Guenguen, J., M. G. van Oort, L. Quillien, and M. van Hessing. 1993. The composition, biochemical characteristics and analysis of proteinaceous antinutritional factors in legume seeds. A review. In Recent Advances of Research in Antinutritional Factors in Legume Seeds, van der Poel, A. F. B., J. Huisman, and H. S. Saini, Eds., Wageningen Pers, The Netherlands, 9-30.

Hogberg, A., and J.E. Lindberg, 2004. Influence of cereal non-starch polysaccharides and enzyme supplementation on digestion site and gut environment in weaned piglets. Anim. Feed Sci. Technol. 116:113-128.

Huisman, J. 1989. Antinutritional factors (ANFs) in the nutrition of monogastric farm animals. In Nutrition and Digestive Physiology in Monogastric Farm Animals, van Weerden, E. J., and J. Huisman, Eds., Pudoc, Wageningen, The Netherlands. 17-35.

Inborr, J., M. Schmitz, and F. Ahrene. 1993. Effect of adding fiber and starch degrading enzymes to a barley/wheat based diet on performance and nutrient digestibility in different segments of the small intestine of early weaned pigs. Anim. Feed Science Technol. 44:113-127.

Inborr, J. and R. B Ogle. 1988. Effect of enzyme treatment of piglet feeds on performance and post-weaning diarrhoea. Swedish Journal of Agricultural Research, 18:129–133.

Jansman, A. J. M. 1993. Tannins in feedstuffs for simple-stomached animals, Nutr. Res. Rev. 6:209-236.

Jansman, A. J. M., and M. Longstaff. 1993. Nutritional effects of tannins and vicine/convicine in legume seeds. In Recent Advances of Research in Antinutritional Factors in Legume Seeds, van der Poel, A. F. B., J. Huisman, and H. S. Saini, Eds., Wageningen Pers, The Netherlands, 301-316.

Jensen, M. S., K. E. B. Knudsen, J. Inborr, K. Jakobsen. 1998. Effect of β-glucanase supplementation on pancreatic enzyme activity and nutrient digestibility in piglets fed diets based on hulled or hulless barley varieties. Anim. Feed Sci. Technol. 72: 329-345.

Ji, F., and S. W. Kim. 2004. Effects of carbohydrase supplement on lactation performance of primiparpous sows. Asian-Austral. J. Anim. Sci. 17:533-537.

Just, A., J. A. Fernandez, and H. Jorgensen. 1983. The net energy value of diets for growth in pigs in relation to the fermentative processes in the digestive tract and the site of absorption of the nutrients. Livest. Prod. Sci. 10:171-186.

Kass, M. L., P. J. Van Soest, and W. G. Pond. 1980. Utilization of dietary fiber from alfalfa by growing swine. II. Volatile fatty concentrations in and disappearance from the gastrointestinal tract. J. Anim. Sci. 50:192-197.

Kennelly, J. J., and F. X. Aherne. 1980a. The effect of fiber addition to diets formulated to contain different levels of energy and protein on growth and carcass quality of swine. Can. J. Anim. Sci. 60:385-393.

Kennelly, J. J., and F. X. Aherne. 1980b. The effect of fiber addition to diets formulated to contain different levels of energy and protein on growth and carcass quality of swine. Can. J. Anim. Sci. 60:385-393.

Kennelly, J. J., F. X. Aherne, and W. C. Sauer. 1978. The effects of levels of isolation, or varietal differences in high fiber hull fraction of low glucosinolate rapeseed meals on rat or pig performance. Can. J. Anim. Sci. 58:743-752.

Kim, S. W., D. L. Knabe, K. J. Hong, and R. A. Easter. 2003. Use of carbohydrases in corn-soybean meal-based nursery diets. J. Anim. Sci. 81:2496-2504.

Kim, S. W., J. H. Zhang, K. T. Soltwedel, and D. L. Knabe. 2006. Use of carbohydrates in corn-soybean meal-based grower-finisher pig diet. Anim. Res. 55:563-578.

King, R. H., and M. R. Taverner. 1975. Prediction of the digestible energy in pig diets from analyses of fibre contents. Anim. Prod. 21:275-284.

Le, P. D., A. J. A. Aarnink, N. W. M. Ogink, P. M. Becker, and M. W. A. Verstegen. 2005. Odour from animal production facilities: its relationship to diet. Nutr. Res. Rev. 18:3-30.

Lewis, A. J., and H. S. Bayley. 1995. Amino acid bioavailability. In: Bioavailability of Nutrients for Animals: Amino Acids, Minerals, and Vitamins. Ammerman, C. B., D. H. Baker, and A. J. Lewis, eds. San Diego, CA: Academic Press. Pp. 35-65.

Li, S., W. C. Sauer, S. X. Huang, and V. M. Gabert. 1996a. Effect of β-glucanase supplementation to hulless barley- or wheat-soybean meal diets on the digestibilities of energy, protein, β-glucans, and amino acids in young pigs. J. Anim. Sci. 74:1649-1656.

Li, S., N. C. Sauer, R. Mosenthin, and B. Kerr. 1996b. Effect of β-glucanase supplementation of cereal-based diets for starter pigs on the apparent digestibilities of dry matter, crude protein and energy. Anim. Feed Sci. Technol. 59:223-231.

Liener, I. E. 1994. Implications of antinutritional components in soybean foods, Crit. Rev. Food Sci. Nutr., 34:31.

Liu, Y. and S. K. Baidoo. 1997. Exogenous enzymes for pig diets: an overview. In: Enzymes in Poultry and Swine Nutrition. Ronald R. Marquardt and Zhengkang, Eds. http://www.idrc.ca/en/ev-30967-201-1-DO_TOPIC.html.

Low, A. G. 1985. The role of dietary fibre in digestion absorption and metabolism. Proc. 3rd Int. Seminar on Digestive Physiology in the Pig. Report No. 580. Copenhagen, Denmark: Beret. Statens. Husdyrbugsfors.

Marquardt, Ronald R. and Michael, Beford. 1997. Recommendations for future research on the use of enzymes in animal feeds. In: Enzymes in Poultry and Swine Nutrition. Ronald R. Marquardt and Zhengkang, Eds. http://www.idrc.ca/en/ev-30969-201-1-DO_TOPIC.html.

Mavromichalis, I., J. D. Hancock, B. W. Senne, T. L. Gugle, G. A. Kennedy, R. H. Hines, and C. L. Wyatt. 2000. Enzyme supplementation and particle size of wheat in diets for nursery and finishing pigs. J. Anim. Sci. 78:3086-3095.

Medel, P., F. Baucells, M. I. Gracia, C. de Blas, and G. G. Mateos. 2002. Processing of barley and enzyme supplementation in diets for young pigs. Anim. Feed Sci. Technol. 95:113-122.

Moeser, Adam J. and Theo A. T. G. van Kempen. 2002. Dietary fibre level and enzyme Inclusion affect nutrient digestibility and excreta characteristics in grower pigs. J. Sci. Food Agric. 82:1606-1613.

Moughan, Paul J., and V. Ravindran. 2000. Maximizing nutrient utilization:from enzymes to nutraceuticals. Pages: 61-78.

Noblet, J., and J. M. Perez. 1993. Prediction of digestibility of nutrients and energy values of pig diets from chemical analysis. J. Anim. Sci. 71:3389-3398.

NRC. 1998. Nutrient Requirements of Swine, 10th rev. ed., National Academy Press, Washington, D.C.

Nyachoti, C. M., Arntfield, S. D., Guenter, W., Cenkowski, S. and F. O. Opapeju. 2006. Effect of micronized pea and enzyme supplementation on nutrient utilization and manure output in growing pigs. J. Anim. Sci. 84:2150-2156.

Officer, D. I. 1995. Effect of multienzyme supplements on the growth performance of piglets during the pre- and post-weaning periods. Anim. Feed Sci. Technol. 56:55-65.

Omogbenigun, F. O., C. M. Nyachoti, and B. A. Slominski. 2004. Dietary supplementation with multienzyme preparations improves nutrient utilization and growth performance in weaned pigs. J. Anim. Sci. 82:1053-1061.

Pettey, L. A., S. D. Carter, B. W. Senne, and J. A. Shriver. 2002. Effects of β-mannanase addition to corn-soybean meal diets on growth performance, carcass traits, and nutrient digestibility of weanling and growing-finishing pigs. J. Anim. Sci. 80:1012-1019.

Pond, W. G., R. S. Lowrey, and J. H. Maner, 1962. Effect of crude fiber level on ration digestibility and performance in growing-finishing swine. J. Anim. Sci. 21:692-696.

Ritter, W. F. 1989. Odour Control of Livestock Wastes: State-of-the-Art in North America, J. Agric. Engng, Res. 42:51-62.

Sauber, T. E. and F. N. Owens. 2001. Cereal Grains and By-Products for Swine. In: Swine Nutrition, Second Ed. Austin J. Lewis and L. Lee Southern, Eds., U.S.A. Pp. 785-802.

Sheppy, C. 2001. The Current Feed Enzyme Market and Likely Trends. In: Enzymes in Farm Animal Nutrition. M. R. Bedford and G. G. Partridge. Eds. 1-10.

Southern, L. L. 1991. Digestible Amino Acids and Digestible Amino Acid Requirements for Swine. BioKyowa Technical Review-2. Chesterfield, MO: Nutri-Quest, Inc.

Stahly, T. S. 1984. Use of fats in diets for growing pigs. In: Fats in Animal Nutrition, J. Wiseman, ed. London: Butterworth. Pp. 313-331.

Sutton, A. L., K. B. Kephart, M. W. A. Verstegen, T. T. Canh, and P. J. Hobbs. 1999. Potential for reductions of odorous compounds in swine manure through diet modification. J. Anim. Sci. 77:430-439.

Tangendjaja, B., Z. B. Johnson, and P. R. Noland. 1988. Effect of cooking and addition of enzymes on feeding value of rice bran for swine. Nutrition Reports International, 37: 449–458.

Thacker, P. A., G. L. Campbell, and J. W. D. Grootwassink. 1988. The effect of beta-glucanase supplementation on the performance of pigs fed hulless barley. Nutr. Rep. Int. 38:91-99.

Thacker, P.A., G. L. Campbell, and J. W. D. GrootWassink. 1989. The effect of sodium bentonite on the performance of pigs fed barley-based diets supplemented with beta-glucanase. Nutr. Rep. Int. 40:613-620.

Thacker, P., G. Campbell, and J. Groot Wassink. 1991. The effect of enzyme supplementation on the nutritive value of rye-based diets for swine. Can. J. Anim. Sci. 71:489-496.

Thacker, P.A., G. L. Campbell, and J. W. D. Groot Wassink. 1992a. Effect of salinomycin and enzyme supplementation on nutrient digestibility and the performance of pigs fed barley- or rye-based diets. Can. J. Anim. Sci. 72:117–125.

Thacker, P. A., G. L. Campbell, J. Groot Wassink. 1992b. The effect of organic acids and enzyme supplementation of the performance of pigs fed barley-based diets. Can. J. Anim. Sci. 72:395-402.

van Heugten, E. 2001. Mycotoxins and Other Antinutritional Factors in Swine Feeds. In: Swine Nutrition, Second Ed. A. J. Lewis and L. L. Southern, Eds., U.S.A. Pp. 563-383.

Van Soest, P. J., and R. W. McQueen. 1973. The chemistry and estimation of fibre. Proc. Nutr. Soc. 32:123-130.

Williams, C. M., and S. S. Schiffman. 1995. Impact of feed and lagoon additives on animal odor. In: (J. B. Longenecker and J. W. Spears) New horizons in animal nutrition and health. Symposium, Research Triangle Park, NC.

Xavier, E. G. 2005. Production Economics and pig health: use of Allzyme Vegpro in feed formulation. In: Proceedings of Alltech's Twenty First Annual Symposium, Nutritional Biotechnology in the feed and food industries. T. P. Lyons and K. A. Jacques, Eds., Nottingham, U.K. Pp: 221-228.

Table 1. Enzyme efficacy: growth performance studies.

Enzymes	Level	Diet	Type of Pig	Length	ADG (kg)		AFI (kg)		GF		Reference
					0	+	0	+	0	+	
Cellulase	0.01%	Wheat-maize-fish-soya-rice bran	12 piglets, avg. initial B.W. 5.61kg	48 days	0.206	0.300[b]			0.478	0.538[b]	Suga et al. (1978)
	ABG sprayed on at 5 kg/tonne				0.211	0.234	0.282	0.259	0.769	0.885	
α-amylase, β-glucanase, and glucoamylase	ACP added at 5 kg/tonne				0.211	0.191	0.282	0.208[b]	0.769	0.909	
(ABG) and amylolytic, cellulolytic and	ABG and ACP supplemented at 5		48 pigs, avg. initial B.W. 3.9								Inborr and Ogle
proteolytic enzymes (ACP)	kg/tonne	barley-based	kg	7 weeks	0.211	0.203	0.282	0.231[b]	0.769	0.909	(1988)
		Maize-soya-ensiled cooked rice bran + cellulase			0.23	0.29	0.45	0.64[b]	0.510	0.444	
	7.9 ml/kg of cellulase to rice bran, 270	Maize-soya-ensiled cooked rice	48 pigs (avg. initial B.W.								Tangendjaja et al.
Cellulase, cellulase-amylase	mg/kg of cellulase-amylase to rice bran	bran + cellulase-amylase	6.15kg)	3 weeks	0.23	0.26	0.45	0.65[b]	0.510	0.400	(1988)
											Thacker et al.
β-glucanase	0.25% enzyme	barley-based	60 pigs (avg. initial B.W. 20kg)	12 weeks	0.74	0.76	2.32	2.35	3.190	0.322	(1988)
		barley-based diet + enzyme		individual pigs were slaugtered at	0.82	0.83	2.33	2.39	0.352	0.347	
		barley-based diet + enzyme +									Thacker et al.
β-glucanase	0.25% enzyme	3.0% sodium bentonite	96 pigs (avg. B.W. 25kg)	an avg. B.W. of	0.82	0.83	2.33	2.37	0.352	0.347	(1989)
				individual pigs were slaugtered at an avg. B.W. of							
		rye-based, mash form	90 pigs (avg initial B.W. 20 kg)	99 kg	0.73	0.76	2.53	2.38	3.480	3.12[b]	
				individual pigs were slaugtered at							
			48 pigs (avg. initial B.W. 25	an avg. B.W. of							Thacker et al.
Pentosanase	0.25%	rye-based, pelleted form	kg)	98 kg	0.76	0.78	2.02	2.05	2.640	2.640	(1991)
			Weaner pigs, average of 26								
Pentosanase (850 U xylanase activity g^{-1})		Rye-soya	days, Avg. Initial B.W. 10.7 kg	10 days	4.24	4.24	6.94	6.64	0.608	0.637	Bedford et al.
β-glucanase (1086 U g^{-1})	2 g of enzyme per kg of diet	Barley-soya	Avg. Initial B.W. 12.1 kg	10 days	5.24	6.14[b]	9.05	9.57	0.572	0.646	(1992)
		Barely-soya	72 pigs (avg. initial B.W. 22.5		0.84	0.86	2.14	2.22	2.53	2.59	
		Barley-soya + salinomycin	+/- 4.4kg)	75 days	0.87	0.84	2.18	2.16	2.53	2.56	
	750 units g^{-1} of beta-glucanse and 650	Rye-soya			0.74	0.71[b]	1.77	1.75	2.4	2.48	Thacker et al.
β-glucanase, pentosanse	units g^{-1} of pentosanase	Rye-soya + salinomycin	96 pigs (avg. B.W. 19.25kg)	88 days	0.72	0.75	1.71	1.81	2.38	2.43	(1992a)
		barley-based	48 pigs (avg. initial B.W.		0.83	0.84	2.56	2.47	0.324	0.329	
		barley-based + propionic acid	25.1kg)	77 days	0.86	0.79	2.47	2.35	0.346	0.333	
		barley-based	120 pigs (avg. initial B.W.		0.254	0.274	0.534	0.547	0.455	0.493	Thacker et al.
β-glucanase	0.25% enzyme	barley-based + fumaric acid	8.1kg)	5 weeks	0.275	0.275	0.538	0.522	0.503	0.524	(1992b)
		Barley-wheat-soya w/MF β-									
	1.0 g/kg^{-1}	glucanase			0.204	0.233	0.281	0.307	0.725	0.725	
		Barley-wheat-soya w/CL β-									
β-glucanase, xylanase, and α-amylase	0.95 g/kg^{-1}	glucanase	96 barrows, 21-24 days old	20 days	0.204	0.205	0.281	0.281	0.725	0.719	Inborr et al. (1993)
	0.25 g kg-1 bacterial protease, 0.25 g kg-1 lipase, 0.5 g kg-1 β-glucananse, 0.5 g kg-1 amylase, 0.5 g kg-1 cellulase, supplied by Alltech Pty Ltd		172 piglets avg. initial B.W. 6.5kg	29 days	0.286	0.289	0.357	0.331	0.649	0.714	
	1.0 g kg-1 of a mixture of β-glucanase, hemi-cellualse, pentosanase and cellulase, 0.333 g kg-1 amylase, 0.333 g kg-1 proteinase, 0.333 g kg-1 lipase supplied by Novo Nordisk Pty Ltd		218 piglets avg. initial B.W. 6.8kg	34 days	0.270	0.259	0.327	0.318	0.568	0.549	
	amylase, xylanase, β-glucanase and		253 piglets avg. initial B.W.								
Multienzyme preparations	pectinase supplied by Finfeed International	Wheat-fish-meat-tallow-soya-blood meal	6.7kg	35 days	0.313	0.293	0.346	.303[b]	0.641	0.671	Officer (1995)

[b] Values with different superscripts within a row are different (P<0.05).

Continued

Table 1. Continued

Enzymes	Level	Diet	Type of Pig	Length	ADG (kg)		AFI (kg)		GF		Reference
					0	+	0	+	0	+	
	.02% of diet	hulless barley-based diet + Endofeed enzyme			0.86	0.86	2.16	2.15	0.397	0.407	
	.07% of diet	hulless barley-based diet + Biofeed enzyme			0.86	0.83	2.16	2.06	0.397	0.408	
	.11% of diet	hulless barley-based diet + Biofeed Plus enzyme			0.86	0.85	2.16	2.15	0.397	0.403	
	.10% of diet	hulless barley-based diet + Porzyme SF enzyme			0.86	0.83	2.16	2.09	0.397	0.400	
β-glucanase	.16% of diet	hulless barley-based diet + Porzyme TP enzyme	138 pigs (avg. initial B.W. 30kg)	10 weeks	0.86	0.84	2.16	2.08	0.397	0.407	Baas and Thacker (1996)
	SBM sprayed with protease (1 μL/g of soybean meal)				0.1604	0.1042	0.3771	0.3402	0.0004	0.0002	
Protease	SBM incubated in protease (1 μL/g of soybean meal)	cornstarch-based with soybean meal	12 pigs (avg. initial B.W. 6.4 +/- .3kg)	9 days	0.1521	0.1479	0.342	0.3437	0.0004	0.0004	Caine et al. (1997)
cellulase, β-glucanase	0.1 g kg-1	hulless barley-based	48 pigs (avg. initial B.W. 9kg)	until approx. 60kg	0.721	0.782[b]			0.498	0.543	Baidoo et al. (1998)
		Hulled Barley (var. Arra)	40 pigs (avg. initial B.W.		0.197	0.209	0.349	0.346	0.575	0.613	
β-glucanase	0.25 g/kg feed	Hulless Barley (var. Condor)	9.4kg)	21 days	0.223	0.217	0.359	0.349	0.625	0.633	Jensen et al. (1998)
	0.08 units/kg feed, 400 mg/kg of α-galactosidase		54 gilts (avg. initial B.W. 43 kg) Grower	28 days	0.806	0.844	2.097	2.07	0.383	0.407[b]	Baucells et al.
α-1, 6 galactosidase		soybean meal	Finisher	28 days	0.678	0.785[b]	2.304	2.47[b]	0.292	0.317[b]	(2000)
		wheat-based			0.508	0.530	0.338	0.360	0.654	0.676	
		barley-based	240 piglets (avg. initial B.W.		0.518	0.531	0.329	0.350	0.625	0.654	
xylanase, amylase, pectinase, β-glucanase	1g/kg	sugar-beet pulp-based	8.17kg)	4 weeks	0.535	0.532	0.345	0.363	0.641	0.676	Gill et al. (2000)
		wheat-based, particle size 1,300 μm			0.41	0.43	0.55	0.54	0.75	0.80	
		wheat-based, particle size 600 μm			0.45	0.47	0.52	0.57	0.87	0.82	
		wheat-based, particle size 400 μm	180 pigs (avg. initial B.W. 5.7kg)	35 days	0.43	0.42	0.51	0.50	0.84	0.84	
		wheat-based, particle size 1,300 μm			0.88	0.87	3.11	3.02[b]	0.28	0.29	
		wheat-based, particle size 600 μm	160 pigs (avg. initial B.W. 67kg)	58 days	0.89	0.87	3.05	2.89[b]	0.29	0.3	
		wheat-based, particle size 600 μm			0.90	0.93	2.91	2.99	0.31	0.31	
		wheat-based, particle size 400	160 pigs (avg. initial B.W.								Mavromichalis et al.
Xylanase	.01% of the diet	μm	63kg)	61 days	0.91	0.92	2.84	2.84	0.32	0.32	(2000)
	600 mg/kg of enzyme complex				0.374	0.391	0.484	0.048	0.775	0.813	
3100 U of α-amylase/g, 275 U/g of β-glucanase, 275 U/g of β-glucanase	1200 mg/kg of enzyme complex	Barley diet	120 males (avg. initial B. W. 5.5 kg)	28 days	0.374	0.391	0.484	0.489	0.775	0.800	Medel et al. (2002)
		complex corn-soybean meal			0.383	0.387	0.620	0.602	0.618	0.646[b]	
		simple corn-soybean meal	156 pigs (avg. initial B.W. 6.27kg)	42 days	0.377	0.391	0.621	0.622	0.607	.628[b]	
		corn-soybean meal	117 pigs (avg. initial B.W. 13.62kg)	21 days	0.543	0.558	0.955	0.938	0.568	.595[b]	
β-mannanase	0.05% β-mannanase	corn-soybean meal	60 pigs (avg. initial B.W. 22.5kg)	pigs were slaughtered at an avg. block weight of 109 kg	0.842	.872[b]	2.500	2.480	0.337	.351[b]	Pettey et al. (2002)

[b] Values with different superscripts within a row are different (P<0.05).

Continued

Table 1. Continued

Enzymes	Level	Diet	Type of Pig	Length	ADG (kg)		AFI (kg)		GF		Reference
					0	+	0	+	0	+	
		corn-soybean meal supplemented with 0.1% CS			0.364	0.365	0.635	0.594	0.57	0.62[b]	
		corn-soybean meal supplemented with 0.2% CS	108 pigs (avg initial B.W. 6.29 +/- 0.08kg)	5 weeks	0.364	0.367	0.635	0.614	0.57	0.6	
Carbohydrases	7 units/g of α-1, 6-galactosidase, 22 units/g of β-1, 4-mannase, β-1, 4-mannosidase and trace amounts of other enzymes	corn-soybean meal supplemented with 0.1% CS	90 pigs (avg initial B.W. 8.0 +/- 0.1kg)	3 weeks	0.335	0.352	0.588	0.577	0.57	0.61[b]	Kim et al. (2003)
	5,500xylanase XU/kg				0.282	0.311	1.15	1.33	0.223	0.228	
	11,000xylanase XU/kg				0.282	0.331	1.15	1.24	0.223	0.260	
Xylanase	16,500xylanase XU/kg	wheat-based diet	30 pigs (21.4 +/- 1.8 kg avg. initial B.W.)	28 days	0.282	0.306	1.15	1.31	0.223	0.223	Barrera et al. (2004)
Xylanase	0.4g/kg (1000 units of xylanase per kg product)	wheat-based steeped for 1 hr	60 pigs (avg. initial B.W. 8.27kg)	21 days	0.279	0.321[b]	0.310	.359[b]	1.16	1.14	
		wheat-based steeped for 15 hr			0.340	0.324	0.364	0.388	1.07	1.21	Choct et al. (2004)
β-glucanase and β-xylanase	1 g/kg diet each enzyme	triticale, wheat bran, barley with low NSP	32 barrows, avg. initial B.W. 8.9kg	14 days	0.175	0.195					Hogberg et al. (2004)
		triticale, wheat bran, barley with high NSP			0.218	0.198					
Carbohydrase	7 units/g of α-1, 6-galactosidase, 22 units/g of β-1, 4-mannase, β-1, 4-mannosidase and trace amounts of other enzymes	Corn and SBM	20 Primiparous sows (avg. initial B.W. 164.25kg)	21 days	-0.610	-.100[b]	5.59	5.86			Ji and Kim (2004)
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, and mannanase			0.2236	0.2521[b]	0.4323	0.4354	0.52	0.58[b]	
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase and pectinase			0.2236	0.2628[b]	0.4323	0.4564	0.52	0.58[b]	
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, mannanase, and pectinase	24 pigs, avg. initial B.W. 7.0kg	4 weeks	0.2236	0.2491[b]	0.4323	0.4136	0.52	0.61[b]	
Multienzyme preparations	250 units of xylanase, 150 units of glucanase, 0.001% amylase, 0.0003% protease, 0.002% invertase, and 400 units of phytase activities per kg of diet	Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, mannanase, and pectinase	48 pigs, avg. initial B.W. 6.21kg	38 days	0.23107	0.25765[b]	0.41037	0.41035	0.56	0.63[b]	Omogbenigun et al. (2004)

[b] Values with different superscripts within a row are different (P<0.05).

Table 2. Enzyme efficacy: ileal digestibility studies

Enzymes	Level	Diet	Type of Pig	Length	CP		Ileal Lysine		Ileal Methionine		Ileal Threonine		Ileal Tryptophan		Reference
					0	+	0	+	0	+	0	+	0	+	
β-glucanase	β-glucanse (10,000 U/kg^{-1}) was supplemented to the diet at 1 g/kg^{-1}	Barley-soya	females (avg initial B.W. 30-50kg)	12 days	71.6	72.9									Graham et al. (1986)
β-glucanase-xylanase	diet supplemented with 2g/kg of enzyme containing 16 β-glucanase units/g and 1300 xylanase units/g	Barley-pollard-soya	2 gilts, 2 barrow (avg. initial B.W. 19-25kg)	14 days	64.5	70.1^{b}									Graham et al. (1988)
		Barley-soya mash		6-d adaption, 3-d feces collection, 2-d ileal collection	52.2	53.3									
β-glucanase	5 g of β-glucanase per kg of diet	Barley-soya pelleted	4 pigs (approx. initial B.W. 80kg)		54.9	52.3									Graham et al. (1989)
Pentosanase	2 g enzyme per kg of diet	Rye-soya,	24 pigs, avg. initial B.W. 10.7 kg	15 days, 5-d acclimation, 10-d experimental period	63.4	57.7									
β-glucanase	2 g enzyme per kg of diet	Barley-soya	24 pigs, avg. initial B.W. 12.1 kg	15 days, 5-d acclimation, 10-d experimental period	17.1	42.6									Bedford et al. (1992)
	1.0 g/kg-1	Barley-wheat-soya w/MF β-glucanase	24 barrows (avg ending B.W. 10-13kg)	20 days			0.768	0.784							
β-glucanase, xylanase, and α-amylase	0.95 g/kg^{-1}	Barley-wheat-soya w/CL β-glucanase					0.768	0.779							Inborr et al. (1993)
		Hulless Barley-Soybean Meal	6 pigs (avg. initial B.W. 7.3 +/- .8kg)	9 days	65.2	73.5^{b}	69.0	73.1			57.9	67.7^{b}			
β-glucanase	0.2% β-glucanase	Wheat-Soybean Meal	6 pigs (avg. initial B.W. 9.2 +/- 1.2kg)	9 days	68.8	75.9	74.1	79.4			64.2	71.5			Li et al. (1996a)
		cornstarch-based with untreated soybean meal			70.4	65.2	77.9	74.5	72.9	82.6	68.5	60.5			
Protease	1 μL/g of soybean meal	cornstarch-based with incubated soybean meal	12 pigs (avg. initial B.W. 6.4 +/- .3kg)	9 days	72.4	70.3	79.0	78.8	74.6	74.1	68.7	68.1			Caine et al. (1997)
		Lupin seed meal (species *L. luteus* cv Amulet)			83.3	85.3	79.1	84.1	86.6	90.9^{b}	79.0	82.8^{b}	78.2	81.6	
α-1, 6 galactosidase	(Alpha Gal - 500 1,500 galactosidase units (GALU) g^{-1}) at the level of 5 g/kg^{-1}	Lupin seed meal (species *L. angustifolius* cv Saturn)	12 barrows (avg initial B.W. 11 to 15 kg)	14 days	85.4	86.2	82.8	84.3	86.0	90.5^{b}	81.9	84.0^{b}	83.0	84.3	Gdala et al. (1997a)
		soybean meal + α-galactosidase			78.8	80.3									
	0.2 g kg^{-1} α-galactosidase, 1 g kg^{-1} protease, 0.19 g kg^{-1} xylanase, 0.18 g kg^{-1} β-glucanase, 0.06 g kg^{-1} α-amylase and a mixture of .028 g kg^{-1} α-galactosidase, 1.0 g kg^{-1} protease and .019 g kg^{-1} xylanase	soybean meal + protease			78.8	79.4									
		soybean meal + xylanase			78.8	80.7									
		soybean meal + β-glucanase			78.8	81.3									
		soybean meal, pea, rapeseed cake + mixture			77.4	77.5									
α-galactosidase, protease, xylanase, β-glucanase, α-amylase		soybean meal, pea, rapeseed cake + α-amylase	20 barrows (avg initial B.W. 9.3kg)	9 days	77.4	79.0									Gdala et al. (1997)
cellulase, β-glucanase	0.1 g kg^{-1}	hulless barley-based	8 pigs (avg. initial B.W. 14.3 kg)	10 days	57.6	61.8^{b}	54.6	61.7^{b}	65.4	75.0^{b}	52.4	61.0^{b}			Baidoo et al. (1998)
Carbohydrases	7 units/g of α-1, 6-galactosidase, 22 units/g of β-1, 4-mannase, β-1, 4-mannosidase and trace amounts of other enzymes	corn-soybean meal supplemented with 0.1% CS	10 gilts (avg initial B.W. 16.77 +/- 0.66kg)	5-d adjustment and 3-d collection			80.0	81.0^{b}	84.5	85.3	67.9	71.6^{b}	80.0	83.1^{b}	Kim et al. (2003)
Xylanase	5,500 xylanase XU/kg		8 barrows (20.5 +/- 1.2 kg avg. initial B.W.)	8-d adaptation and 4-d collection	78.5	81.7^{b}	56.3	59.9^{b}			50.2	51.4^{b}			Barrera et al. (2004)
	11,000 xylanase XU/kg				78.5	84.1^{b}	56.3	65.9^{b}			50.2	58.6^{b}			
	16,500 xylanase XU/kg	wheat-based			78.5	82.6^{b}	56.3	62.4^{b}			50.2	55.5^{b}			
		wheat-based + xylanase			83.6	85.2^{b}	87.0	88.3	89.1	90.8^{b}	76.4	77.5			
Xylanase and phospholipase	5,600 EXU/kg of xylanase, 1,000 IU/kg phospholipase	wheat-based + phospholipase	16 barrows (avg. initial B.W. 4.1kg)	12 days	83.6	84.8^{b}	87.0	87.5	89.1	89.8^{b}	76.4	76.7			Diebold et al. (2004)
		wheat-based + both enzymes			83.6	86.0^{b}	87.0	88.8	89.1	91.3^{b}	76.4	78.0			
β-glucanase and β-xylanase	1 g/kg diet each enzyme	triticale, wheat bran, barley with low NSP	32 barrows, avg. initial B.W. 8.9kg	14 days	0.64	0.65									Hogberg et al. (2004)
		triticale, wheat bran, barley with high NSP			0.67	0.66									
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, and mannanase			62.1	71.5^{b}									
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase and pectinase			62.1	71.4^{b}									
Multienzyme preparations	250 units of xylanase, 150 units of glucanase, 0.001% amylase, 0.0003% protease, 0.002% invertase, and 400 units of phytase activities per kg of diet	Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, mannanase, and pectinase	24 pigs, avg. initial B.W. 7.0kg	4 weeks	62.1	73.2^{b}									Omogbenigun et al. (2004)
Multienzyme blend of β-glucanase, phytase, protease, amylase, cellulase, and pectinase	500 units of β-glucanase, 300 units of phytase per kg plus a broad spectrum of other enzyme activities	barley-based diet with raw peas	8 barrows (21.5 +/- 1.2 kg of initial B.W)	5-d acclimation, 3-d fecal and urine collection, 2-d ileal collection	80.6	80.6	79.4	84.0^{b}			66.6	77.9^{b}			Nyachoti et al. (2006)
		barley-based diet with micronized peas			80.7	81.0	83.6	85.3			75.1	76.9			

[b] Values with different superscripts within a row are different (P<0.05).

Table 3. Enzyme efficacy: fecal digestibility studies

Enzymes	Level	Diet	Type of Pig	Length	Fecal DM		Fecal CP		Fecal GE		Reference
					0	+	0	+	0	+	
3100 U of α-amylase/g, 400 U/g of xylanase, 275 U/g of β-glucanase	600 mg/kg of enzyme complex				0.874	0.876	0.870	0.872	0.876	0.877	
	1200 mg/kg of enzyme complex	Barley diet	120 males (avg. initial B. W. 5.5 kg)	28 days	0.874	0.876	0.870	0.874	0.876	0.874	Medel et al. (2002)
Fiber degrading enzyme	2.0g/kg^{-1} of enzyme added to diet, 100 xylose unit g^{-1} of activity	Corn, soybean meal, soyhull diet	12 barrows (avg. initial B.W. 33kg)	28 days	81.6	83.5[b]	75.4	77.5	80.4	82.5[b]	Moeser and van Kempen (2002)
β-mannanase	0.05% β-mannanase	corn-soybean meal	12 barrows (avg. initial B.W. 93.0kg)	14 days, fecal collections on d 13 and d 14	91.7	92.1	80.8	80.7	85.9	86.5	Pettey et al. (2002)
Xylanase and phospholipase	5,600 EXU/kg of xylanase, 1,000 IU/kg phospholipase	wheat-based + xylanase	16 barrows (avg. initial B.W. 4.1kg)	13 days			90.9	90.8	87.7	87.5	
		wheat based + phospholipase					90.9	89.9[b]	87.7	86.6[b]	
		wheat-based + both enzymes					90.9	90.6	87.7	87.1	Diebold et al. (2004)
β-glucanase and β-xylanase	1 g/kg diet each enzyme	triticale, wheat bran, barley with low NSP	32 barrows, avg. initial B.W. 8.9kg	14 days	0.78	0.77					
		triticale, wheat bran, barley with high NSP			0.75	0.77					Hogberg et al. (2004)
Multienzyme preparations	250 units of xylanase, 150 units of glucanase, 0.001% amylase, 0.0003% protease, 0.002% invertase, and 400 units of phytase activities per kg of diet	Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, and mannanase	24 pigs, avg. initial B.W. 7.0kg	4 weeks	75.6	78.1[b]	67.1	71.2[b]	77.8	79.8[b]	Omogbenigun et al. (2004)
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase and pectinase			75.6	77.2[b]	67.1	71.6[b]	77.8	79.8[b]	
		Corn, wheat, barley, SBM, canola meal and peas + enzyme cocktail and cellulase, galactanase, mannanase, and pectinase			75.6	80[b]	67.1	74.2[b]	77.8	81.1[b]	

[b] Values with different superscripts within a row are different (P<0.05).

Chapter 2

Effect of enzyme supplementation on apparent ileal and fecal nutrient digestibilities of corn, soybean meal, and soybean hulls in growing pigs

Running head: Improving ingredient digestibility with enzymes

Effect of enzyme supplementation on apparent ileal and fecal nutrient digestibilities of corn, soybean meal, and soybean hulls in growing pigs [1, 2, 3]

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Abstract

Two experiments were conducted to determine the effects of adding an enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase on nutrient digestibility of growing pigs. In Exp. 1, 32 barrows (BW was 25.3 ± 1.8 kg) were grouped based on BW and alloted to 1 of 4 dietary treatments using a randomized complete block design with 8 weight blocks. The pigs were housed in individual pens (1.5 m x 0.9 m) in three temperature controlled rooms. The four dietary treatments were: 1) corn, 2) corn plus enzyme cocktail, 3) soybean meal, and 4) soybean meal plus enzyme cocktail. Diets contained both acid insoluble ash (Celite Corporation, Lompoc, CA) and chromium oxide as indigestible markers. The experimental period lasted for 11 d (excluding the 2-d dietary adaptation to the pens) with each experimental period consisting of a 7-d dietary adaptation period followed by a 3-d collection of feces. On d 10 and 11 of the experiment pigs were euthanized and samples from the distal ileum were collected. There were no differences in fecal digestibility with the addition of the enzyme when acid insoluble ash was used to determine digestibility. There were no differences in fecal digestibility between pigs fed corn and corn plus enzyme or between pigs fed soybean meal and soybean meal plusy enzyme when chromium oxide was used to determine digestibility. There were no differences in amino acid digestibility between pigs fed corn and corn plus enzyme or between pigs fed soybean meal and soybean meal plus enzyme. This study shows that enzyme supplementation does not improve nutrient digestibility of corn or soybean meal.

In Exp. 2, 20 pigs (11 barrows, 9 gilts) (BW was 29.9 ± 4.0 kg) were grouped based on BW and allotted to 1 of 4 dietary treatments using a randomized complete block design

with 5 weight blocks. The pigs were housed in individual pens (1.5 m x 0.9 m) in two temperature controlled rooms. The four dietary treatments were: 1) basal diet, 2) basal diet plus enzyme cocktail, 3) basal diet plus soy hulls, 4) basal diet plus soy hulls and enzyme cocktail. Diets contained acid insoluble ash (Celite Corporation, Lompoc, CA) as an indigestible marker. The experiment lasted 12 d with the experimental period consisting of an 8-d adaptation period followed by a 3-d collection of feces. On d 11 and 12 of the experiment, pigs were euthanized and samples from the distal ileum were collected from 16 of the 20 pigs. The addition of the enzyme to the basal diet lowered the fecal digestibility of CP ($P< 0.003$), Ca ($P<0.003$), NDF ($P<0.03$), P ($P<0.001$), Mg ($P<0.001$), Zn ($P<0.001$), Cu ($P<0.001$), Mn ($P<0.001$), Mb ($P<0.001$) in the basal diet. There were no differences in the amino acid digestibility with the addition of the enzyme to either the basal diet or the basal plus soy hulls diet.

Overall, no positive effects of adding enzyme to swine diets were observed.

Key Words: pigs, enzymes, ileum, digestibility, soy hulls

Introduction

Swine nutritionists are constantly searching for ways to optimize the utilization of feedstuffs. The use of dietary enzymes is one approach that can increase the release of nutrients from feed. One option would be to add a single enzyme to target one component of the diet. However, an enzyme cocktail with several different activities may be more efficient in increasing the availability of nutrients from feedstuffs.

Use of an enzyme cocktail to liberate the protein present in seed coats may increase the efficiency of protein digestion as well as reduce indigestible fiber. Most enzyme

cocktails to date focus on fiber degrading enzymes, perhaps by using an enzyme cocktail that also contains protein and starch degrading enzymes, increased nutrient digestibilites will result from creating more substrate for the enzyme to act upon. This effect may be greater in corn-soybean meal diets containing fiberous ingredients because they provide a greater amount of substrate for enzyme action.

Only a few studies (Caine et al., 1997; Tangendjaja et al., 1988; Li et al., 1996b; Baucells et al., 2000; Moeser and van Kempen, 2002; Pettey et al., 2002) have published data using exogenous enzymes in the typical corn-soy diet used in the U.S. Those that have been published have varying responses. Only a few studies (Kornegay, 1978; Kornegay, 1981; Mitaru et al., 1984; Moeser and van Kempen, 2002; Shriver et al., 2003; Stanogias et al., 1994) have evaluated the effect of adding soybean hulls to a typical corn-soybean meal diet and the efficacy of enzymes.

The objectives of the present study were to: 1) evaluate the effect of enzyme on the digestibility of corn and soybean meal, in order to assign a value of the enzyme to the individual ingredients; 2) determine the nutrient digestibility of a basal diet plus enzyme and a basal diet with 30% soy hulls with and without enzyme.

Materials and methods

The experimental protocol describing the management and animal care was reviewed and approved by the North Carolina State University Institutional Animal Care and Use Committee.

Experiment 1.

Animals and Experimental Design

Thirty-two barrows with an initial average body weight of 25.3 ± 1.2 kg were housed in individual pens (1.5 m x 0.9 m) in three temperature controlled rooms. Animals were allowed to adjust to the pens for a 2-d period. Following the acclimation to the pens, pigs were blocked by weight and assigned within block to one of four experimental diets according to a randomized complete block design with eight blocks.

The experiment lasted 11 days (excluding the 2-day dietary adaptation period to the pens). Feces was collected for 4 d after an adaptation period of 7 d. Acid insoluble ash included as 1.5% of the diet (Celite Corporation, Lompoc, CA) and a chromic oxide premix included at 0.5% of the diet were used as indigestible markers. On days 10 and 11 of the experiment, pigs were anaesthetized with a 5 mL intramuscular ketamine and xylazine combination injection (100 mg/mL) and then euthanized by lethal injection of 7 mL of sodium pentobarbital (390mg/mL) intravenous, according to approved procedures (FASS, 1999) approximately 2 h after feeding. Pigs were dissected and samples were collected from approximately 75 cm of the distal small intestine. The samples were then frozen at -20°C until further chemical analysis was conducted.

Diets and Feeding

Diets were formulated to contain either corn or soybean meal as the sole sources of energy and protein with or without enzyme (Table 1). Vitamins and minerals were added to meet nutrient requirements as suggested by NRC (1998). An enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase

(Allzyme Vegpro, Alltech Inc., Nicholasville, KY) was added at the inclusion rate of 0.10%. Pigs were fed experimental diets twice daily (0800 and 1700) in mash form at a feeding level equal to 90 g/kg body weight$^{0.75}$ of the group average body weight at the start of the period. Pigs were given *ad libitum* access to water via water nipples.

Experiment 2.

Animals and Experimental Design

Twenty grower pigs (11 barrows and 9 gilts) with an initial average body weight of 29.9 ± 4.1 kg were housed in individual pens (1.5 x 0.9 m) in two temperature controlled rooms. Pigs were blocked by weight and randomly assigned to one of four experimental diets according to a randomized complete block design with 5 blocks. The experiment lasted 12 d with the experimental period consisting of an 8-d dietary adaptation period followed by a 3-d collection of feces. Acid insoluble ash included at 1.5% of the diet (Celite Corporation, Lompoc, CA) was used as an indigestible marker. On d 12 of the experiment, 16 of the pigs (4 from each dietary treatment, 8 barrows and 8 gilts) were anaesthetized by a 1.5 mL intramuscular tiletamine and zolazepam combination (100 mg/mL) injection and then euthanized by lethal injection of 7 mL of sodium pentobarbital (390 mg/mL) intravenous according to approved procedures (FASS, 1999) approximately 2 h after feeding. Pigs were dissected and samples were collected from two and a half feet of the distal small intestine. The samples were then frozen at -20°C until further chemical analysis.

Diets and Feeding

The pigs were allotted to one of four dietary treatments (Table 2) consisting of: 1) a basal diet, 2) a basal diet plus enzyme, 3) a basal diet with 30% soy hulls, and 4) a basal diet

with 30% soy hulls plus enzyme. The basal diet was formulated based on available lysine and metabolizable energy, and they met all nutrient requirements as suggested by NRC (1998). An enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase (Allzyme Vegpro, Alltech Inc., Nicholasville, KY) was added at the inclusion rate of 0.05%. Pigs were fed experimental diets twice daily (0800 and 1700) in mash form at a feeding level equal to 90 g/kg body weight$^{0.75}$ of the group average body weight at the start of the period. Pigs were given *ad libitum* access to water via water nipples.

Collection Procedures and Chemical Analysis

During the collection periods, feces was collected twice daily (0800 and 1700) from each pen and placed in polyethylene bags and frozen at -20°C. Once the pigs were dissected, two and a half feet of the distal small intestine was removed, and the digesta was collected and placed in plastic Nalgene bottles and frozen at -20°C.

After collection, the fecal and ileal samples were freeze-dried (model LL3000, ATR, Inc., Laurel, MD). Dried samples were then ground through a 1-mm screen in a Retsch mill (model ZM 100, Haan, Germany) before chemical analysis. The DM content of the feed, fecal and ileal samples was determined by drying them to a constant weight in an oven at 60°C. Analysis of amino acid levels in the diets and ileal digesta were performed in accordance with AOAC (1997) procedures by the Experimental Station Chemical Laboratories, University of Missouri-Columbia, MO for both experiments. In experiment 1, acid insoluble ash was determined in the feed, fecal and ileal samples using the method described by Vogtmann (1975). Feed and fecal samples were analyzed in duplicate for GE content using an adiabatic bomb calorimeter (model C5000, IKA, Wilmington, NC).

The feed and fecal samples were also analyzed for Ca, P, and Cr using the dry ash method by the Analytical Services Laboratory, North Carolina State University, Raleigh, NC. The procedure for Ca, P, and Cr determination in feces and feed involved adding 20 mL of sodium carbonate (10% w/v) to 0.1 g of sample, placing them in a muffle furnace at 500°C to ash, followed by the addition of 8 mL of hydrochloric acid. Calcium, phosphorus, and chromium concentrations were determined using an Ion-Coupled Plasma Spectrophotometer (ICP; Perkin Elmer Model 2100DV ICP-Emission Spectrometer, PerkinElmer, Inc., Waltham, MA). In experiment 2, the fecal and feed samples were analyzed for DM, CP, Ca, ADF, NDF, P, Mg, K, Na, Zn, Cu, Mn and Mb, and the fecal, feed, and ileal samples were analyzed for acid insoluble ash in accordance with AOAC (1997) by the Dairy One Forage Laboratory, University of Cornell, Ithaca, NY.

Calculations and Statistical Analysis

For experiment 1 data were analyzed as a randomized complete block design using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). The model included diet, enzyme inclusion, and diet by enzyme interaction. Significant differences between the dietary treatments were determined following a significant F-test by using the least significant difference method. For experiment 2 data were analyzed as a 2 x 2 factorial randomized complete block design using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). The model included block, diet, enzyme inclusion, and diet by enzyme interaction.

Results and Discussion

There were no differences in fecal digestibility of corn or soybean meal with the addition of the enzyme when either acid insoluble ash or chromic oxide (Table 3) were used

as indigestible markers to determine digestibility. Pettey et al. (2002) found no differences in the apparent digestibility of energy, nitrogen, phosphorus, or dry matter in finishing barrows fed corn-soybean meal diets with or without β-mannanase supplementation. Likewise, Li et al. (1996b) found no effect on fecal digestibility with β-glucanase supplementation to a corn-soybean meal based diet. β-glucanase is a fiber degrading enzyme, as are three of the enzymes included in the enzyme cocktail used in this experiment, pentosanase, α-galactosidase, and cellulase. Medel et al. (2002) reported no differences in fecal digestibility of DM, OM, energy, and CP with the addition of an enzyme cocktail containing amylase, xylanase, and β-glucanase to a barley based diet, which contains more fiber than corn-soybean meal based diets and should have provided more substrate for the enzyme to break down. In contrast, Moeser and van Kempen (2002) reported an increase in DM and GE digestibility with the addition of a fiber-degrading enzyme cocktail added to a corn-soybean meal diet with 20% soybean hulls. In the present study, when acid insoluble ash was used as a marker, apparent fecal digestibility was significantly higher in soybean meal compared to corn for DM ($P=0.001$), GE ($P<0.001$), and P ($P=0.01$), but significantly lower for Ca ($P=0.01$). When chromic oxide was used as a marker, soybean meal had significantly lower digestibility for GE ($P<0.001$) and Ca ($P=0.01$) than corn.

There were no differences in ileal amino acid digestibility with the addition of the enzyme (Table 4). This agrees with Caine et al. (1997) who found that neither spraying nor incubating soybean meal with protease improved the apparent digestibilities of CP and amino acids in the newly weaned pig. The enzyme used in our diet was an enzyme cocktail and not only contained proteases but also starch and fiber degrading enzymes. Nonetheless, enzyme

supplementation still did not contribute to increasing the digestibility of fiber or increase amino acid digestibility in either corn or soybean meal. Corn had a higher ($P < 0.01$) digestibility of methionine, isoleucine, histidine, phenylalanine, threonine, leucine, valine, aspartic acid, serine, glutamine, alanine, cysteine, tyrosine and arginine than soybean meal. However, soybean meal had a higher ($P = 0.01$) digestibility for tryptophan and proline when compared to corn. In this trial results showed a low apparent ileal amino acid digestibility of corn and particularly soybean meal. Relative to NRC (1998), apparent ileal amino acid digestibility values in the present study are approximately 10% lower for corn and 33% for soybean meal. The reason for the lower amino acid digestibilites, particularly in pigs fed soybean meal are not known. The soybean meal used in this study had levels of trypsin inhibitor that were below the detection limit as determined by Carolina Analytical Services (NC Hwy 902, Bear Creek, NC). van Kempen et al. (2002, 2006) reported that apparent and standardized ileal digestibility of soybean meal samples from different regions and different processors were relatively similar, due to common, standardized processing conditions used in plants throughout the US.

In Experiment 2, the effects of enzyme supplementation to a basal corn-soybean meal based diet and a basal diet with 30% added soybean hulls was evaluated. Supplementation of diets with exogenous enzymes is thought to improve digestibility by causing cell-wall structure degradation, ultimately leading to increased nutrient availability and changes in the physical properties of non-starch polysaccharides (Liu, 1997). Non-starch polysaccharides will be more efficiently digested causing changes in the composition and content of bacteria in the small and large intestines (Liu, 1997).

It is not understood why the fecal DM digestibility did not decrease with addition of soy hulls, which disagrees with (Kornegay, 1978; Kornegay, 1981, Mitaru et al., 1984; and Dilger et al., 2004). The fecal digestibility of crude protein of the basal plus soy hull diet was lower ($P< 0.001$) than the basal diet alone. This agrees with Kornegay (1981) who found that the fecal digestibility of crude protein decreased as soybean hulls were substituted for the basal diet. It is to be expected that the addition of fiber to the diet would lower the apparent fecal digestibility of crude protein because it promotes a faster rate of passage as well as an increased level of endogenous nitrogen, which causes the lowering of the fecal digestibility of crude protein (Kornegay, 1978). Kornegay (1978) and Mitaru et al. (1984) also observed soy hulls to have a negative effect on the ileal digestibility of crude protein.

The digestibilities of Ca ($P<0.001$), ADF ($P<0.001$), NDF ($P=0.01$), P ($P<0.001$), Mg ($P<0.001$), K ($P<0.001$), Na ($P=0.005$), Zn ($P<0.001$), Cu ($P<0.001$), and Mb ($P<0.001$) were lower in the basal plus soy hull diet than the basal diet (Table 5). Digestibility is reduced much more for the basal diet when enzyme is added than the hull diet. The main reason for this is the relatively high digestibility of the basal diet. Our results agree with Partridge (1978) who reported that the addition of 6% cellulose to the diet decreased large intestinal absorption of Ca, P, Mg, K and Zn. Our results disagree with Stanogias et al. (1994) who found that the incorporation of soy hulls in the diet did not affect apparent absorption of Ca, P, or Mg. Partridge (1978) reported that addition of 6% cellulose decreased large intestinal absorption of Ca, P, Mg, K and Zn.

The addition of soy hulls to the basal diet reduced the apparent ileal digestibilities of amino acids ($P<0.001$), agreeing with Dilger et al. (2004). The addition of the enzyme to the

basal diet lowered the fecal digestibility of CP ($P< 0.003$), Ca ($P<0.003$), NDF ($P<0.03$), P ($P<0.001$), Mg ($P<0.001$), Zn ($P<0.001$), Cu ($P<0.001$), Mn ($P<0.001$), Mb ($P<0.001$). It is possible that interactions between endogenous enzymes, exogenous enzymes and feedstuffs could have been a reason for lowered digestibility of nutrients when the enzyme was added to the diet (Officer, 2000). Addition of the enzyme did not change the fecal digestibility of crude protein in the basal plus soy hull diet. Tangendjaja et al. (1988) saw no effects on fecal digestibility with the addition of cellulase and amylase or cellulase alone to a corn-soybean meal based diet with rice bran.

There were no differences in the ileal amino acid digestibility with the addition of the enzyme (Table 6). Gdala et al. (1997) found that enzyme supplementation of single enzymes, α-galactosidase, xylanase, β-glucanse, α-amylase, or protease, and a mixture of enzymes, α-galactosidase, xylanase and protease, to a soybean meal based diet with barley and wheat had no significant effect on the ileal digestibility or fecal digestibility of DM and CP of the diet.

It is not clear why the addition of this enzyme did not improve nutrient digestibility in pigs. Corn-soybean meal based diets contain relatively little dietary fiber which may have limited the amount of substrate available for action by the supplemental enzymes. However, when soybean hulls were added to the diet, no beneficial effects of enzymes were observed.

Conclusion

Addition of an enzyme containing protease, cellulase, pentosanase, α-galactosidase, and amylase same enzyme to corn-soybean meal based diets alone or with 30% added soy hulls did not improve the apparent ileal or fecal digestibilities of nutrients in growing pigs.

Literature Cited

AOAC. 1997. Official methods of analysis. 16th ed. Association of Official Analytical Chemists, Arlington, VA.

Baucells, F., J. F. Perez, J. Morales, and J. Gasa. 2000. Effect of α-galactosidase supplementation of cereal-soya-bean-pea diets on the productive performances, digestibility and lower gut fermentation in growing and finishing pigs. J. Anim. Sci. 71:157-164.

Caine, W. R., W. C. Sauer, S. Tamminga, M.W.A. Verstegen, and H. Schulze. 1997. Apparent ileal digestibilites of amino acids in newly weaned pigs fed diets with protease-treated soybean meal. J. Anim. Sci. 75:2962-2969.

Dilger, R. N., J. S. Sands, D. Ragland, and O. Adeola. 2004. Digestibility of nitrogen and amino acids in soybean meal with added soyhulls. J. Anim. Sci. 82:715-724.

FASS (Federation of Animal Science Societies). 1999. Guide for the care and use of agricultural animals in agricultural research and teaching, 1st revised edition. FASS, Savoy, IL.

Gdala, J., H. N. Johansen, K. E. Bach Knudsen, I. H. Knap, P. Wagner, and O. B. Jorgensen. 1997. The digestibility of carbohydrates, protein and fat in small and large intestine of piglets fed non-supplemented and enzyme supplemented diets. Anim. Feed Sci. Technol. 65:15-33.

Kornegay, E. T. 1978. Feeding value and digestibility of soybean hulls for swine. J. Anim. Sci. 47:1272-1280.

Kornegay, E. T. 1981. Soybean hull digestibility by sows and feeding value for growing-finishing swine. J. Anim. Sci. 53:138-145.

Li, S., N. C. Sauer, R. Mosenthin, and B. Kerr. 1996b. Effect of β-glucanase supplementation of cereal-based diets for starter pigs on the apparent digestibilities of dry matter, crude protein and energy. Anim. Feed Sci. Technol. 59:223-231.

Liu, Y. and S. K. Baidoo. 1997. Exogenous enzymes for pig diets: an overview. In: Enzymes in Poultry and Swine Nutrition. Ronald R. Marquardt and Zhengkang, Eds. http://www.idrc.ca/en/ev-30967-201-1-DO_TOPIC.html.

Medel, P., F. Baucells, M. I. Gracia, C. de Blas, and G. G. Mateos. 2002. Processing of barley and enzyme supplementation in diets for young pigs. Anim. Feed Sci. Technol. 95:113-122.

Mitaru, B. N., R. Blair, R. D. Reichert, and W. E. Roe. 1984. Dark and yellow rapeseed hulls, soybean hulls and purified fiber source: their effects on dry matter, energy, protein and amino acid digestibilities in cannulated pigs. J. Anim. Sci. 59:1510-1518.

Moeser, A. J. and T. A. T. G. van Kempen. 2002. Dietary fibre level and enzyme inclusion affect nutrient digestibility and excreta characteristics in grower pigs. J. Sci. Food Agric. 82:1606-1613.

NRC, 1998. Nutrient requirements of swine. 10th revised edition. National Academic Press, Washington, D.C.

Officer, D. I. 2000. Feed enzymes. In: J. P. F. D'Mello (ed.) Farm Animals Metabolism & Nutrition. CABI Publishing, New York, N. Y. p. 405-426.

Partridge, I. G. 1978. Studies on digestion and absorption in the intestines of growing pigs. 4. Effects of dietary cellulose and sodium levels on mineral absorption. Br. J. Nutr. 39:539.

Pettey, L. A., S. D. Carter, B. W. Senne, and J. A. Shriver. 2002. Effects of β-mannanase addition to corn-soybean meal diets on growth performance, carcass traits, and nutrient digestibility of weanling and growing-finishing pigs. J. Anim. Sci. 80:1012-1019.

Shriver, J. A., S. D. Carter, A. L. Sutton, B. T. Richert, B. W. Senne, and L. A. Pettey. 2003. Effets of adding fiber sources to reduced-crude protein, amino acid-supplemented diets on nitrogen excretion, growth performance, and carcass traits of finishing pigs. J. Anim. Sci. 81:492-502.

Stanogias, G., G. R. Pearce, T. Alifakiotis, J. Michaelidis, and B. Pappa-Michaelidis. 1994. Efects of dietary concentration and source of fiber on the apparent absorption of minerals by pigs. Anim. Feed. Sci. Technol. 47:287-295.

Tangendjaja, B., Z. B. Johnson, and P. R. Noland. 1988. Effect of cooking and addition of enzymes on feeding value of rice bran for swine. Nutrition Reports International. 37(6):449–458.

van Kempen, T. A. T. G., I. B. Kim, A. J. M. Jansman, M. W. A. Verstegen, J. D. Hancock, D. J. Lee, V. M. Gabert, D. M. Albin, G. C. Fahey, Jr., C. M. Grieshop, and D. Mahan. 2002. Regional and processor variation in the ileal digestible amino acid content of soybean meals measured in growing swine. J. Anim. Sci. 80:429–439.

van Kempen, T. A. T. G., E. van Heugten, A. J. Moeser, N. S. Muley, and V. J. H. Sewalt. 2006. Selecting soybean meal characteristics preferred for swine nutrition. J. Anim. Sci. 84:1387–1395

Vogtmann, H., P. Fritter, and A. L. Prabuck. 1975. A new method of determining metabolisability of energy and digestibility of fatty acids in broiler diets. Br. Poult. Sci. 67:641–646.

Table 1. Composition of the experimental diets for experiment 1 (as fed basis).

	Corn Control	Corn + Enzyme	Soybean Meal Control	Soybean Meal + Enzyme
Ingredient, %				
Corn	97.75	97.65	0.00	0.00
Soybean meal (48%)	0.00	0.00	97.75	97.65
Vitamins-Minerals[b]	0.25	0.25	0.25	0.25
Celite[c]	1.50	1.50	1.50	1.50
Chromic oxide premix[d]	0.50	0.50	0.50	0.50
Enzyme	0.00	0.10	0.00	0.10
Calculated Composition				
ME, Mcal/kg	3.34	3.34	3.30	3.30
Calcium, %	0.03	0.06	0.33	0.33
Phosphorus, %	0.27	0.27	0.67	0.67
Crude Protein, %	8.11	8.10	46.43	46.38
Lysine, %[e]	0.25	0.25	2.95	2.95
Methionine, %	0.17	0.17	0.65	0.65
Threonine, %	0.28	0.28	1.81	1.81
Tryptophan, %	0.06	0.06	0.64	0.63

[b] Supplied per kg of complete diet: 5,540 IU of vitamin A as retinyl acetate, 1,108 IU of vitamin D_3, 22 IU of vitamin E as dl-α-tocopherol acetate, 1.98 mg of vitamin K as menadione dimethylpyrimidinol bisulfite, 165 mg of choline as choline chloride, 22 mg of niacin, 17.6 mg of d-pantothenic acid as dl-calcium pantothenate, 4.4 mg of riboflavin, 1.1 mg of pyridoxine as pyridoxine•HCl, 0.55 mg thiamine as thiamine mononitrate, 0.022 mg of vitamin B_{12}, 0.33 mg of folic acid, 0.04 mg of d-biotin, 110 mg Zn as $ZnSO_4$, 110 mg Fe as $FeSO_4$, 22 mg Cu as $CuSO_4$, 55 mg Mn as MnO, 0.28 mg I as EDDI, and 0.30 mg Se as $NaSeO_3$.

[c] Used as an indigestible marker.

Table 1 (continued).

[d] Used as an indigestible marker, composed of chromic oxide and starch, 1 part chromic oxide and 2 parts starch.

Table 2. Composition of the experimental diets for experiment 2 (as fed basis).[a]

	Basal Diet
Ingredient, %	
Corn	65.50
Soybean meal (48%)	27.50
Limestone	1.00
Monocalcium Phosphate	1.25
Vitamins-Minerals[b]	0.25
Salt	0.50
Poultry Fat	2.00
Celite[c]	1.50
Chromic oxide premix[d]	0.50
Calculated Composition	
ME, Mcal/kg	3.33
Calcium, %	0.71
Phosphorus, %	0.64
Crude Protein, %	18.50
Lysine, %[e]	1.00
Methionine, %	0.30
Threonine, %	0.70
Tryptophan, %	0.22

[a] Experimental diets consisted of the basal diet, the basal with 0.05% enzyme, the basal with 30% added soybean hulls replacing an equal amount of basal diet, and the soybean hull containing diet with 0.05% enzyme.

[b] Supplied per kg of complete diet: 5,540 IU of vitamin A as retinyl acetate, 1,108 IU of vitamin D3, 22 IU of vitamin E as dl-α-tocopherol acetate, 1.98 mg of vitamin K as menadione dimethylpyrimidinol bisulfite, 165 mg of choline as choline chloride, 22 mg of niacin, 17.6 mg of d-pantothenic acid as dl-calcium pantothenate, 4.4 mg of riboflavin, 1.1 mg of pyridoxine as pyridoxine•HCl, 0.55 mg thiamine as thiamine mononitrate, 0.022 mg of vitamin B12, 0.33 mg

Table 2 (continued).

of folic acid, 0.04 mg of d-biotin, 110 mg Zn as ZnSO4, 110 mg Fe as FeSO4, 22 mg Cu as CuSO4, 55 mg Mn as MnO, 0.28 mg I as EDDI, and 0.30 mg Se as NaSeO3

[c] Used as an indigestible marker.

[d] Used as an indigestible marker, composed of chromic oxide and starch, 1 part chromic oxide and 2 parts starch.

[e] Amino acids are expressed on a calculated ileal apparent digestible basis.

Table 3. Effects of enzyme inclusion to either corn or soybean meal on fecal nutrient digestibility in growing pigs[a,b,c]

	Corn		SBM			P value		
Enzyme	-	+	-	+	SEM	Diet	Enz	D*E
Fecal digestibility (%) using acid insoluble ash as marker								
DM	84.0[ac]	83.9[a]	86.2[bc]	85.1[c]	0.4	0.001	0.17	0.27
GE	83.7[a]	83.6[a]	88.2[b]	86.9[b]	0.5	<0.001	0.21	0.24
P	33.7[a]	29.9[b]	40.8[c]	38.3[c]	2.9	0.01	0.29	0.82
Ca	38.7[a]	22.5[b]	14.0[b]	16.5[b]	5.7	0.01	0.23	0.12
Fecal digestibility (%) using chromium oxide as marker								
DM	88.6[a]	90.5[b]	91.6[c]	89.0[a]	0.4	0.06	0.29	<0.001
GE	88.4[a]	90.4[b]	92.8[c]	90.3[b]	0.4	<0.001	0.48	<0.001
P	52.8[a]	58.7[a]	64[ab]	53.9[b]	2.4	0.19	0.38	0.00
Ca	56.2[a]	54.1[a]	47.7[ac]	37.5[bc]	4.5	0.01	0.19	0.38

[a,b,c] Values with different superscripts within a row are different (P<0.05).

[d] Each value represents a mean of 32 barrows, 8 per treatment.

Table 4. Effects of enzyme inclusion to either corn or soybean meal on ileal amino acid digestibility in growing pigs[d]

	Corn		SBM			P value		
Enzyme	-	+	-	+	SEM	Diet	Enz	D*E
AA Digestibility %								
Lys	55.7[a]	53.0[a]	47.8[a]	41.2[b]	4.9	0.06	0.36	0.70
Met	80.8a	78.8a	66.4b	61.0b	3.7	<0.001	0.32	0.64
Trp	54.8[a]	56.9[ac]	69.3[b]	65.3[bc]	3.9	0.01	0.81	0.43
Ile	69.2[a]	68.8[ac]	60.5[a]	54.5[bc]	3.7	0.01	0.40	0.46
His	76.0[a]	73.7[a]	56.0[b]	52.5[b]	3.5	<0.001	0.42	0.86
Phe	77.6[a]	76.5[a]	60.5[b]	55.7[b]	3.2	<0.001	0.37	0.58
Thr	64.0[a]	59.3[ac]	48.1[b]	44.1[bc]	4.2	0.001	0.31	0.94
Leu	81.4[a]	80.3[a]	61.0[a]	55.8[b]	3.3	<0.001	0.36	0.54
Val	68.1[a]	67.6[a]	58.3b[c]	52.3[c]	3.8	0.003	0.40	0.48
Asp	68.2[a]	65.8[a]	48.0[b]	45.0[b]	3.6	<0.001	0.46	0.93
Ser	77.6[a]	70.6[a]	56.3[b]	53.6[b]	3.4	<0.001	0.16	0.53
Gln	79.9[a]	78.9[a]	47.7[b]	45.6[b]	3.4	<0.001	0.65	0.87
Pro	37.8	20.5	54.8	47.7	7.9	0.01	0.14	0.52
Gly	21.1[a]	18.3[a]	32.4[a]	27.1[a]	7.0	0.16	0.57	0.86
Ala	76.8[a]	73.7[a]	50.0[b]	45.7[b]	3.7	<0.001	0.33	0.89
Cys	76.8[a]	76.0[a]	35.9[b]	27.6[b]	4.2	<0.001	0.29	0.39
Tyr	79.2[a]	76.9[a]	61.8[b]	58.0[b]	3.3	<0.001	0.36	0.81
Arg	69.2[a]	68.7[a]	62.3[ab]	58.1[b]	3.2	0.01	0.48	0.58

[a,b,c] Values with different superscripts within a row are different (P<0.05).

[d] Each value represents a mean of 32 barrows, 8 per treatment.

Table 5. Effects of enzyme inclusion to either a basal corn-soybean meal diet or a basal corn-soybean meal diet plus soy hulls on fecal nutrient digestibility in growing pigs[e]

	Basal		Basal + Hull			P value		
Enzyme	-	+	-	+	SEM	Diet	Enz	D*E
Fecal digestibility (%) using acid insoluble ash as marker								
DM	92.3[a]	92.0[a]	93.3[b]	92.7[b]	0.4	0.030	0.26	0.62
CP	90.3[a]	86.0[bc]	72.6[b]	70.6[b]	0.8	<0.001	0.003	0.22
Ca	77.4[a]	66.1[b]	63.6[b]	61.4[b]	1.8	<0.001	0.003	0.03
ADF	80.3[a]	74.4[a]	54.4[b]	51.7[b]	5.1	<0.001	0.42	0.77
NDF	75.8[a]	65.3[b]	62.9[b]	59.4[b]	2.9	0.01	0.03	0.28
P	66.5[a]	50.9[bc]	50.1[bc]	44.7[b]	1.5	<0.001	<0.001	0.007
Mg	47.6[a]	26.1[bc]	28.7[bc]	19.9[b]	2.4	<0.001	<0.001	0.03
K	86.5[a]	80.4[b]	67.7[c]	68.1[c]	1.6	<0.001	0.09	0.07
Na	87.0[a]	80.0[ac]	73.6[bc]	67.9[b]	3.5	0.005	0.11	0.87
Zn	94.6[a]	56.5[b]	47.2[c]	40.6[d]	1.2	<0.001	<0.001	<0.001
Cu	44.2[a]	14.5[b]	23.8[c]	9.6[b]	1.8	<0.001	<0.001	<0.001
Mn	48.7[a]	16.0[b]	29.2[bc]	32.2[bc]	3.1	0.57	<0.001	<0.001
Mb	48.9[a]	21.7[b]	21.4[b]	18.4[b]	1.2	<0.001	<0.001	<0.001

[a,b,c,d] Values with different superscripts within a row are different (P<0.05).

[e] Each value represents a mean of 20 pigs, 5 per treatment.

Table 6. Effects of enzyme inclusion to either a basal corn-soybean meal diet or a basal corn -soybean meal diet plus soy hulls on ileal amino acid digestibility in growing pigs [c]

	Basal		Basal + Hull			P value		
Enzyme	-	+	-	+	SEM	Diet	Enz	D*E
AA Digestibility %								
Lys	86.3[a]	84.6[a]	66.1[b]	60.8[b]	4.1	<0.001	0.41	0.68
Met	89.1[a]	87.1[a]	72.0[b]	66.7[b]	3.5	<0.001	0.33	0.64
Trp	88.5[a]	89.8[a]	59.7[b]	52.2[b]	5.0	<0.001	0.56	0.40
Ile	87.5[a]	85.0[a]	67.0[b]	60.1[b]	3.8	<0.001	0.25	0.58
His	88.6[a]	86.9[a]	69.3[b]	64.6[b]	3.0	<0.001	0.32	0.64
Phe	87.8[a]	86.0[a]	70.8[b]	63.5[b]	3.3	<0.001	0.21	0.43
Thr	82.5[a]	80.2[a]	62.0[b]	54.0[b]	4.1	<0.001	0.25	0.53
Leu	87.9[a]	86.5[a]	70.0[b]	67.2[b]	2.9	<0.001	0.48	0.80
Val	85.3[a]	82.4[a]	64.1[b]	57.9[b]	3.7	<0.001	0.25	0.66
Asp	84.9[a]	82.7[a]	66.1[b]	57.5[b]	3.6	<0.001	0.17	0.40
Ser	86.3[a]	85.4[a]	68.3[b]	61.5[b]	2.6	<0.001	0.18	0.29
Gln	87.3[a]	85.8[a]	71.7[b]	63.8[b]	3.2	<0.001	0.17	0.35
Pro	84.4[a]	84.7[a]	65.8[b]	58.0[b]	3.4	<0.001	0.30	0.26
Gly	69.0[a]	65.3[a]	39.9[b]	28.3[b]	6.5	<0.001	0.27	0.56
Ala	84.7[a]	82.8[a]	65.0[b]	58.6[b]	3.6	<0.001	0.28	0.54
Cys	80.8[a]	78.6[a]	56.8[b]	45.4[b]	4.0	<0.001	0.12	0.28
Tyr	87.6[a]	85.6[a]	69.2[b]	63.2[b]	3.2	<0.001	0.24	0.54
Arg	91.2[a]	89.9[a]	75.7[b]	70.5[b]	3.3	<0.001	0.34	0.57

[a,b] Values with different superscripts within a row are different (P<0.05).

[c] Each value represents a mean of 20 pigs, 5 per treatment.

Chapter 3

Growth performance, carcass characteristics, nutrient digestibility, ammonia emission, and odor in growing-finishing pigs fed corn-soybeanmeal diets with or without enzyme

Running head: Enzymes in growing-finishing pig diets

Growth performance, carcass characteristics, nutrient digestibility, ammonia emission, and odor in growing-finishing pigs fed corn-soybean meal diets with or without enzyme[1,2,3]

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Abstract

This study was designed to determine the effects of enzyme inclusion on growth performance, carcass characteristics, nutrient digestibility, ammonia emission and odor. The enzyme used was an enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase. One hundred and seventeen grower pigs (72 barrows and 45 gilts); BW=35.1 ± 3.4 kg were allotted based on BW and gender to 1 of 3 dietary treatments using a randomized complete block design with 8 blocks. Pigs were housed 4 (3 pens) or 5 (21 pens) pigs per pen using a total of 24 pens. The three dietary treatments were: 1) positive control corn and soybean meal diet, 2) negative control corn and soybean meal diet with soy hulls, 3) negative control diet supplemented with enzyme. Growth performance was measured on days 28, 56, 70 and 89. Fecal samples were collected on days 52 and 53. Celite (Celite Corporation, Lompoc, CA) was used as an indigestible marker. Fifteen barrows (BW was 99.0 ± 7.2 kg), 5 from each treatment, were moved to individual metabolism crates that allowed for feces and urine to be collected quantitatively. The urine and feces were mixed to form a slurry, and this manure was used for determination of ammonia emission. Ultrasound measurements were taken at the end of the trial on d 89, excluding the 15 pigs on metabolism crates. There were no effects of the enzyme added to the corn/soybean meal/soy hull diet of pigs on growth performance, carcass characteristics, ammonia emission or odor. However, the addition of enzyme improved gross energy digestibility, the negative control diet plus enzyme was higher than that of the negative control diet (P=0.001). The gross energy digestibility of the negative control diet plus enzyme was not different from the positive

control diet meaning the enzyme improved the gross energy digestibility of the more fibrous diet bringing it up to that of the positive diet. The addition of enzyme also improved the fecal digestibilities of Ca ($P=0.01$), P ($P=0.001$), Mg ($P<0.001$), K ($P<0.001$), Fe ($P<0.001$), Zn ($P<0.001$), Cu ($P<0.001$), and Mn ($P=0.04$), which were higher for pigs fed the negative control plus enzyme diet than those fed the negative control diet. The digestibilities of Ca, Mg, K, Fe, and Mn were not different for pigs fed the positive control diet and pigs fed the negative control plus enzyme diet. The digestiblities of P ($P=0.001$), Zn ($P<0.001$), and Cu ($P<0.001$) were higher for pigs fed the negative control plus enzyme diet than those fed the positive diet. In conclusion, addition of enzymes to high fiber diets show promising results in increasing the digestibility of these feedstuffs. Even with an increase in average daily feed intake and gain to feed ratio, it may be cost effective to use less expensive, higher fiber ingredients supplemented with enzyme in the place of a certain percentage of more expensive, lower fiber ingredients such as corn.

Key Words: Enzyme; Swine; Growth; Carcass; Digestibility; Ammonia; Odor

Introduction

Due to increasing concerns of environmental implications due to animal production in the U.S., it is imperative to find an effective and practical approach for improving nutrient digestibility and, thereby, reducing waste production in pigs. Waste production and nutrient excretion is directly affected by the amount of dietary fiber present in a diet (Canh, 1998). If we can reduce indigestible fiber by the use of exogenous fiber degrading enzymes to improve the digestibility of nutrients we can have a positive effect on the environment. It would

further allow the swine industry to incorporate ingredients with high fiber content without impacting growth performance.

There have been many studies that have evaluated the use of exogenous enzymes in pig diets, including starch degrading enzymes in combination with fiber degrading enzymes with or without protein-degrading enzymes (Inborr and Ogle, 1988; Tangendjaja et al., 1988; Inborr et al., 1993; Gdala et al., 1997b; Mendel et al., 2002) and protein degrading enzymes alone (Caine et al., 1997). Studies with fiber degrading enzymes have included β-glucanase (Graham et al., 1986; Thacker et al., 1988; Thacker et al., 1989; Bedford et al., 1992; Thacker et al., 1992b; Baas and Thacker, 1996; Li et al., 1996a; Li et al., 1996b; Jensen et al., 1998; Moeser and van Kempen, 2002), or β-glucanase mixed with other fiber degrading enzymes (Graham et al., 1988; Thacker et al. 1992b; Baidoo et al., 1998; Gill et al., 2000; Hogberg et al., 2004), and very few with galactosidase (Gdala et al., 1997a; Baucells et al., 2000), xylanase (Mavromichalis et al., 2000; Barrera et al., 2004; Diebold et al., 2004), pentosanase (Thacker et al., 1991; Bedford et al., 1992), and β-mannanase (Pettey et al., 2002). Most of the studies have focused on the effects of these enzymes alone or in combination in barley, wheat or rye diets because of their high fiber content.

Only a few studies (Caine et al., 1997; Li et al., 1996b; Baucells et al., 2000; Moeser and van Kempen, 2002; Pettey et al., 2002) have published data using exogenous enzymes in typical corn-soybean meal diets used in the U.S. Caine et al. (1997) found that protease treatment of soybean meal had no effect on ileal digestibility of CP and amino acids in newly weaned pigs. Li et al. (1996b) reported no effect on the digestibility of DM, energy and crude protein in young pigs from β-glucanase

supplementation to corn-soybean meal diets. Baucells et al. (2000) supplemented cereal-soybean-pea diets with α-galactosidase, which improved the performance of growing and finishing pigs, and increased the apparent whole-tract digestibility of DM. In two of four experiments by Pettey et al. (2002), corn-soybean meal diets with added β-mannanase, had greater G:F than pigs fed diets with no added enzyme. In the third experiment, the addition of β-mannanase to the control diet significantly increased ADG compared with the control diet and the control diet with 2% soybean oil added. The addition of β-mannanase increased fat-free lean gain compared with that of pigs fed the control or soybean oil diets. The results of experiment four, a digestibility study, revealed no differences in the apparent digestibility of energy, nitrogen, phosphorus, or DM in pigs fed the control diet, the diet with 2% soybean oil, or the diet with the addition of 0.05% β-mannanase. Moeser and van Kempen (2002) found that inclusion of an enzyme cocktail to a high fiber corn-soybean meal diet with 20% soybean hulls, improved dry matter and energy digestibility by approximately 2 and 3% respectively.

Addition of fiber to the diet decreases ammonia emission by shifting N from urea in urine, to a more stable form of N in microbial protein in the hindgut (Canh et al., 1997). In addition, increased microbial activity with fiber supplementation causes a pH decline in the hindgut, which in turn will lower ammonia emission (Canh et al., 1998). Canh et al. (1998) observed that the addition of soybean hulls to the diet gave the greatest pH reduction and lowest ammonia emission when compared to coconut expeller and dried sugar beet pulp.

Attempts to reduce odor production and emission by altering diets has focused on reducing ammonia emission as well as other odorous compounds (Le et al., 2005). The

relationship between ammonia and odor is still not completely understood, however an excessive protein intake will increase both ammonia emission and odor emission (Le et al., 2005). Minimizing the amount of substrate available to microorganisms in the small intestine, thereby improving the digestibility of nutrients, may be important to reduce odor (van Heugten and van Kempen, 2001). If the use of exogenous enzymes in swine diets can improve the digestibility of nutrients, then we can hypothesize that their use would result in a reduction in odor emissions. In the present study, we hypothesized that by supplying an enzyme cocktail that included protein-degrading enzymes, both ammonia and odor production would be reduced, because proteins along with fermentable carbohydrates are the main precursors of odor formation (Le et al., 2005).

The objectives of this study were to evaluate the effects of supplementation of an enzyme cocktail to corn-soybean meal diets with or without soy hulls on growth performance, carcass characteristics, nutrient digestibility, ammonia emission and odor emission in growing-finishing pigs using.

Materials and Methods

The experimental protocol describing the management and animal care was reviewed and approved by the North Carolina State University Institutional Animal Care and Use Committee.

Animals and Experimental Design

One hundred and seventeen grower pigs (72 barrows and 45 gilts) of 35.1 ± 3.4 kg initial BW were used to evaluate the effects of enzyme (Allzyme® Vegpro, Alltech Inc., Nicholasville, KY) supplementation on growth performance, carcass characteristics, nutrient

digestibility, ammonia emission and odor . Pigs were weighed, blocked by sex and initial weight and allotted within block to one of three dietary treatments using a randomized complete block design with eight blocks. Pen groups were separated by sex, and litter mates were evenly distributed across pens within a block. Pigs were housed 4 (3 pens) or 5 (21 pens) pigs per pen using a total of 24 pens. There was a total of 15 pens for barrows, and 9 pens for gilts.

Diets and Feeding

Dietary treatments (Table 1) consisted of: 1) positive control corn and soybean meal diet, 2) negative control corn and soybean meal diet with 20% soy hulls, and 3) negative control diet supplemented with enzyme. We hypothesized that increasing the fiber in the corn-soybean meal diet, by including soy hulls, would give the enzyme more substrate to act upon, thus increasing the digestibility of fiber. Four different phases of diet were fed; 2 grower diets from 34-54 kg (phase I) and 54-73 kg (phase II), and 2 finisher diets from 73-91 kg (phase III) and 91-113 kg (phase IV). They were formulated with a constant lysine to metabolizable energy ratio, and they met all nutrient requirements as suggested by NRC (1998). Enzyme was added to the negative control corn and soybean meal diet with soy hulls at an inclusion rate of 0.05%. The enzyme used was an enzyme cocktail containing the activities of protease, cellulase, pentosanase, α-galactosidase, and amylase. Water via water nipples and feed in mash form were offered on an *ad libitum* basis.

Collection Procedures and Measurements

Growth performance was measured on d 28, 56, 70 and 89 of experiment. Fresh fecal grab samples were collected immediately at defecation from at least two pigs in each pen on

d 52 and 53 to determine fecal digestibility of GE, CP, ADF, NDF, Ca, P, Mg, K, Na, Fe, Zn, Cu and Mn. Fecal samples were combined by pen for all days collected and stored at -20°C until they were analyzed. Acid insoluble ash included at 1.5% (Celite Corporation, Lompoc, California) was used as an indigestible marker. Samples of each diet were collected at the time of manufacture for subsequent analysis. Ultrasound (Aloka 500, Ithaca, NY) measurements to determine backfat, loin eye area, percent fat-free lean and lean gain were taken when pigs were weighed at the end of the trial on d 89. Percent fat-free lean and lean gain were calculated according to NPPC (1991) equations.

On d 70 of the trial, 15 barrows (5 pigs per treatment), 1 barrow from each pen, were transferred to metabolism crates (1.5 m x 0.9 m) where they were fed the same diets they were originally assigned. The barrows that weighed closest to the mean weight of the pen they were housed were chosen. Pigs were fed experimental diets twice daily (0800 and 1700) in mash form at a feeding level equal to 90 g/kg body weight$^{0.75}$ of the average body weight of the block at the start of the period. They had *ad libitum* access to water *via* a water nipple for the adaptation period of six days. During the collection period, the pigs were offered as much water as they would drink twice a day in their feeders before feedings. Feeders were then cleaned out, dried and then pigs were fed their allotted feed. After a six day adaptation period to the crates, urine and feces were

collected separately for a period of five days. Feces were collected twice daily (0700 and 1900) onto wire screens fixed underneath the metabolism crates. Feces were weighed and immediately partitioned, one-half of the collection was pooled by pig and frozen at -20°C until further chemical analysis, and the other half was discarded.

Urine was voided on slope-shape stainless steel trays fixed under the metabolism cages and collected into plastic containers packed in ice to minimize any gaseous losses of nitrogen. Ice was replaced twice daily during the collection period. The quantity of urine was recorded, and one-half of the daily urinary excretion was stored, pooled by pig and stored at -20°C for further chemical analyses.

After the animal trial, frozen urine and feces from each pig were thawed and mixed and homogenized within respective animal (at the rates they were produced) using a hand mixer for 8 min to form a slurry. Half of this manure, considered fresh, was stored at -20°C for further chemical analyses, while the other half was aged for 21 d at room temperature in open 500 mL plastic containers and then stored at -20°C for further chemical analyses. The fresh manure and aged manure samples were used for determination of ammonia emission and odor offensiveness.

Chemical Analysis

The fecal samples collected on d 52 and 53 were freeze-dried (model LL3000, ATR, Inc., Laurel, MD). Freeze dried samples and feed samples were then ground through a 1-mm screen in a Retsch mill (model ZM 100, Haan, Germany) before chemical analysis. Feed and fecal samples were analyzed in duplicate for GE content using an adiabatic bomb calorimeter (model C5000, IKA, Wilmington, NC). Analysis of

dry matter, acid insoluble ash and CP, ADF, NDF, Ca, P, Mg, K, Na, Fe, Zn, Cu and Mn was performed on the fecal and diet samples in accordance with AOAC (1997) procedures by the Dairy One Forage Laboratory, University of Cornell, Ithaca, NY.

Ammonia emission of the fresh and aged manure samples were determined by placing 400 mL of the manure mixture in a rectangular (28 x 9.5 x 6 cm; length x width x height) container (Super Oval 1, Tupperware Co., Orlando, FL). Air was drawn through a flow meter (Cole Palmer, Vernon Hills, IL) at a rate of 1.0 L/min, through the container with manure, and then through a gas dispersion tube (Fisher, Pittsburgh, PA) placed in a 500-mL Erlenmeyer flask containing 400 mL of dilute sulfuric acid (0.1 N) to trap the ammonia released from the manure. This sulfuric acid solution was sampled (2.0 mL) at 12, 24, 36, 48, 72, and 96 h and analyzed for ammonia using the procedure of Willis et al. (1996).

The odor panel was conducted at West Texas A&M University. The panel was conducted without knowledge of sample identity for intensity and hedonic tone. The panelists (4) analyzed 1 sample at a time with no time limit, then stepped out of the room, drank a glass of water and were given 5 minutes before starting a new sample. The intensity standards used were prepared per ASTM E 544-99 with n-butanol at 5 concentrations for the 1-5 scale, in which 1 was the lowest odor intensity and 5 was the highest intensity. Standards were 250, 750, 2250, 6750, and 20250 ppm, respectively (Guo et al., 2001). The panelists smelled the sample and compared the standards for strength of odor assigning a number based on the standards. If the sample fell between two standards, a designation of 0.5 was used. The hedonic tone scale was -10 to +10,

with 0 being neutral. The panelists smelled each sample individually, then assigned his or her designation of degree for pleasantness or unpleasantness with 0 indicating the odor was neutral. Positive (+) designations indicated the odor was that degree of pleasant and negative

(-) designations indicated the odor was unpleasant, with +10 being the most pleasant and -10 being the most unpleasant.

Calculations and Statistical Analysis

Daily gain, feed intake, feed efficiency (gain/feed), ultrasound carcass composition and lean gain were calculated for each of the two grower (phases I and II), two finisher phases (phases III and IV), and overall. Data were analyzed as a randomized complete block design using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). The model included weight block, sex, treatment and sex by treatment interactions. Pig body weight at the end of the trial was used as a covariate for ultrasound carcass measurements. For odor analysis the model included mean intensity and mean hedonic tone and was evaluated for main effects for manure, treatment, and manure x treatment interaction. The ammonia analysis model included diet, time and diet by time interaction. Significant differences between the dietary treatments were determined following a significant F-test by using the least significant difference method.

Results

Health of Pigs

No treatment related health problems in pigs were observed in this study. Two pigs were removed from the study due to not being castrated, and 1 pig was removed due to a bad leg. Eight pigs were removed from the study due to prolapse, 2 from the positive control treatment, 2 from the negative control treatment, and 4 from the negative control treatment with enzyme. The corn used in the feed was tested by the North Carolina Department of Agriculture Feed Division (Blue Ridge Rd., Raleigh, NC) for zearalonone,

which was found not to be present. In the metabolism crates, one pig was euthanized due to a fatal injury.

Growth Performance and Carcass Characteristics

There were no differences in the growth performance of pigs with the addition of enzyme to the diet (Table 2). There were no differences in BW between the negative control diet and the negative control diet plus enzyme, between the negative control diet and the positive control diet, or between the positive control and the negative control diet plus enzyme. However, there was a tendency for pigs fed the positive control diet to weigh more than pigs fed the negative control diet in phase II (P=0.09) and phase III (P=0.07). There were no differences in ADG between the negative control diet and negative control diet plus enzyme, between the negative control diet and the positive control diet, or between the positive control and the negative control diet plus enzyme. There was, however, a tendency for pigs fed the positive control diet versus the negative control diet to gain more per day during phase II (P=0.07) as well as overall (P=0.12).

There were no differences in feed intake between the negative control diet and negative control diet plus enzyme. Pigs fed the negative control diet tended to consume more feed in phase II and IV (P=0.07) and consumed more feed in phase III (P=0.04) and overall (P=0.05) than pigs fed the positive control diet. Pigs fed the negative control diet plus enzyme consumed more feed in phase I (P=0.01), phase II (P=0.01), phase III (P=0.03), and overall (P=0.01) than pigs fed the positive control diet There were no differences in feed efficiency between the negative control diet and negative control diet plus enzyme. Similarily, pigs fed the positive control diet had an improved feed efficiency during phases I,

II, and all phases combined (P=0.01) than pigs fed the negative control diet. The pigs fed the positive control diet had an improved feed efficiency during phases I (P=0.002), II (P=0.01), and all phases combined (P=0.01) than pigs fed the negative control diet plus enzyme. There were no differences in metabolizable energy intake or gain/metabolizable energy intake between treatments (Table 3).

Enzyme supplementation did not have any effects on carcass characteristics measured when comparing pigs fed the negative control diet versus those fed the negative control diet plus enzyme (Table 4). Loin eye area was less in pigs fed the negative control diet versus the pigs fed the positive control diet (P=0.002) and loin eye area was less in pigs fed the negative control diet plus enzyme versus the pigs fed the positive control diet (P<0.001). There was a tendency for pigs fed the positive control diet to have more kg of lean gain per day (P=0.06) *versus* those fed the negative control diet. Pigs fed the positive control diet had more kg of lean gain per day (P=0.02) and more fat-free lean percent (P=0.03) versus those fed the negative control diet plus enzyme.

Apparent Total Tract Digestibility

Gross energy digestibility of the positive control diet was higher (P<0.001) than that of the negative control diet (Table 5). Gross energy digestibility of the negative control diet plus enzyme was higher than that of the negative control diet (P=0.001) but, there was no significant difference in the gross energy digestiblility between the positive control diet and the negative control plus enzyme diet. The enzyme had no effect on the fecal digestibility of crude protein (P=0.21). The crude protein digestibility of the positive control diet was significantly higher (P=0.001) than the negative control plus enzyme diet.

Pigs fed the positive control diet had a higher digestibility of acid detergent fiber (ADF) than those fed the negative control diet ($P<0.001$), and those fed the negative control diet plus enzyme had a higher digestibility of ADF than those fed the negative control diet ($P<0.001$). Pigs fed the negative control plus enzyme diet had a lower digestibility of ADF than those fed the positive control diet ($P=0.02$). Pigs fed the positive control diet ($P<0.001$) or the negative control diet plus enzyme ($P<0.001$) had higher digestibilities of neutral detergent fiber (NDF) than those fed the negative control diet. There was no significant difference between the digestibilites of NDF for pigs fed the positive control diet and those fed the negative control plus enzyme diet. The digestibilities of Ca ($P=0.04$), Mg ($P<0.001$), K ($P<0.001$), Fe ($P<0.001$), and Cu ($P=0.001$) were higher in the positive control diet than the negative control diet. The digestibilities of Ca ($P=0.01$), P ($P=0.001$), Mg ($P<0.001$), K ($P<0.001$), Fe ($P<0.001$), Zn ($P<0.001$), Cu ($P<0.001$), and Mn ($P=0.04$) were higher for pigs fed the negative control plus enzyme diet than those fed the negative control diet. The digestibilities of Ca, Mg, K, Fe, and Mn were not significantly different for pigs fed the positive control diet and pigs fed the negative control plus enzyme diet. The digestiblities of P ($P=0.001$), Zn ($P<0.001$), and Cu ($P<0.001$) were higher for pigs fed the negative control plus enzyme diet than those fed the positive control diet.

Ammonia Emission and Odor

There were no effects of enzyme supplementation on the emission of ammonia from either fresh or aged manure (Tables 6 and 7). There was also no difference in the amount of ammonia produced between the negative control diet and the positive control diet in either the fresh manure or the aged manure at 12, 24, 36, and 48 h. We did, however, observe a

reduction in the amount of ammonia produced for pigs fed the negative control diet compared to the positive control diet in the aged manure after 72 (P=0.004) and 96 h (P=0.003). The negative control plus enzyme diet had a lower ammonia emission from fresh manure at 36 h (P=0.04) compared to the positive diet. The negative control plus enzyme diet had lower ammonia emissions from aged manure at 48 h (P=0.04), 72 h (P=0.001), and 96 h (P=0.004) compared to the positive diet.

There were no significant effects of the enzyme on odor intensity or hedonic tone score (Table 8). There was a manure effect on odor intensity and hedonic tone, with the aged manure having a stronger odor intensity (P=0.01) and more unpleasant hedonic tone (P=0.03) than fresh manure.

Discussion

Pigs eat to meet their energy needs (NRC, 1998). Therefore, as energy density in the diet decreases, voluntary feed intake increases. Diets high in fiber have a lower energy density than diets lower in NSP (NRC, 1998). Fiber in the diet may depress growth rate and feed efficiency if feed intake is reduced due to increased bulk or reduced palatability (NRC, 1998). Energy intake was not a problem for pigs in this study because they were fed *ad libitum*, and as stated earlier, pigs consume feed to meet their energy needs unless prevented by bulk or palatability (Baird et al., 1975). Merkel et al. (1958) reported that pigs eating diets reduced in energy had an increased daily feed consumption. In our study pigs fed both of the diets containing soy hulls, the negative control diet and the negative control diet plus enzyme overall consumed more than pigs fed the positive control diet. Calculated metabolizable energy was the same between treatments. If the enzyme

made more energy available, then pigs fed the negative control diet plus enzyme would be expected to have consumed less than pigs fed the negative control diet, which we did not see observe. Kornegay (1978) reported that pigs fed 24% soy hulls were not able to maintain a growth rate equal to those fed 0, 6, or 12%, although feed intake increased with increasing levels of hulls. However, Kornegay (1981) reported that pigs fed 15% soy hulls were able to maintain a growth rate equal to those fed 0%. We did not see a significant reduction in ADG in the present study, but there was a tendency overall for pigs fed the negative control diet to weigh less than pigs fed the positive control diet, resulting in a numerical reduction in BW of 4 kg. Pigs fed the soy hull diet with the addition of enzyme were 2 kg heavier than those on the soyhull diet alone. The addition of the enzyme numerically increased ADG, but not completely equal with the pigs fed the positive diet. Adding fiber to the diet in the present study lowered feed efficiency agreeing with Kornegay (1981) for pigs fed 30% soy hulls. No differences across treatments in gain/metabolizable energy were observed in the present study. Adding enzyme to the high fiber, negative control diet did not improve ADG or feed efficiency of the diet, agreeing with (Thacker et al., 1988; and Thacker et al., 1989).

The only significant differences in carcass characteristics were that the pigs fed the positive diet had an increased loin eye area ($P<0.002$) *versus* the pigs fed the negative diet and tended to have a higher lean gain than the pigs fed the negative control diets. Shriver et al. (2003) observed no effects of adding soy hulls to the diet on carcass characteristics except in the lowering of back fat thickness. The addition of enzyme to the soy hull diet was unable to increase the loin eye area when compared to pigs fed the positive control, and also signifantly reduced the percentage of fat-free lean, as well as lean gain.

There were no positive significant effects of enzyme supplementation on carcass characteristics, which agree with previous work (Thacker et al., 1988; Thacker et al., 1989; and Mavromichalis et al., 2000).

There was an increase in the fecal digestibility of gross energy in the positive control diet ($P<0.001$) and negative control diet plus enzyme ($P=0.001$) *versus* the negative diet, showing the enzyme had a positive effect on gross energy digestibility, also seen by Thacker et al., (1988), Thacker et al., (1989), Li et al., (1996a), and Li et al., (1996b) when they added β-glucanase to a barley-based diet, also high in fiber. This can be explained by the replacement of more digestible feedstuffs, corn and soybean meal, with one of lower digestibility, soy hulls (Kornegay, 1978; Mitaru et al., 1984). Addition of the enzyme increased the energy digestibility of the negative diet similar to the positive control diet.

The fecal digestibility of crude protein was lower in the pigs consuming the negative control diet *versus* the positive control diet. Kornegay (1978) and Mitaru et al. (1984) also observed soy hulls to have a negative effect on the digestibility of crude protein. There was no improvement however, in crude protein digestibility between the negative control diet and the negative control diet plus enzyme, which agrees with previous work by Thacker et al. (1989).

Stanogias et al. (1994) found that incorporating soy hulls did not significantly affect apparent absorption of Ca, P or Mg, but the added NDF did reduce the apparent absorption of Na by 22 to 57% and of K by 7 to 70%. In contrast, Partridge (1978) reported that addition of 6% cellulose decreased fecal digestibility of Ca, P, Mg, K and Zn. Agreeing with Partridge (1978), the addition of 20% soy hulls to the negative diet in our study decreased the

digestibility of Ca, Mg, K, Fe, and Cu. The addition of the enzyme increased the digestibilities of Ca, P, Mg, K, Fe, Zn, Cu, and Mn similar to the positive control diet. This suggests that the enzyme was able to degrade fiber, thereby improving the digestibility of these nutrients. The enzyme increased the nutrient digestibility of P, Zn, and Cu of the negative diet, higher than those of the positive diet. Even without the incorporation of phytase, the enzyme blend added to the diet was able to increase P availability. It is clear that increasing the digestibility of fiber increases mineral digestibility which in turn can decrease excretion thus having a positive effect on the environment.

Ammonia emission is reduced when more fibrous feedstuffs are included in the diet (Canh et al. 1997). This agrees with the results of the present study that overall in both the fresh and aged manure the negative control diet released less ammonia than the positive control diet. Other studies have shown that increased fiber can reduce ammonia emission because fermentable carbohydrates can shift nitrogen from urine to feces in the form of bacterial protein (Canh et al., 1998; Sutton et al., 1999). The enzyme had no effect on ammonia emissions. Kim et al. (2006) also saw no difference in the average ammonia concentration between pigs fed a corn-soybean meal diet compared to pigs fed a corn-soybean diet plus 0.05% carbohydrase.

The aged manure was more intense and had a more unpleasant hedonic tone than the fresh manure. This was expected because fermentation produces more odorants. Addition of the enzyme to the diet had no effect on odor emissions of the fresh or aged manure. It was hypothesized that minimizing the amount of substrate available to microorganisms in the large intestine by improving the digestibility of nutrients could reduce odor (van Heugten and

van Kempen, 2001). However, in this study the enzyme did improve gross energy digestibility and increase the digestibilities of several minerals, therefore we did reduce the amount of substrate, but we did not see a decrease in odor intensity or hedonic tone. The addition of the enzyme did not increase CP digestibility, therefore undigested N going into the large intestine was not reduced. This may explain why we did not see a reduction in ammonia emissions with the addition of the enzyme. Otto et al. (2003) observed reduced ammonia emission with the reduction of dietary CP and providing crystalline amino acids to meet digestible amino acid requirements, but no reduction in odor offensiveness.

Overall, our data imply that dietary enzyme addition was able to effectively aid in the degradation of fiber, thus allowing for more efficient nutrient utilization. Inconsistent results on enzyme efficacies reported in the literature may be due to a combination of factors including level and source of the fiber, where and in what conditions feedstuffs are grown, how they are milled, enzyme substrate availability, and the age of the pig.

Implications

Dietary incorporation of an enzyme cocktail to a corn-soybean meal diet with soybean hulls may be an efficient way to utilize the addition of high fiber ingredients in a typical corn-soybean meal diet, while maintaining the same levels of growth performance, carcass characteristics, ammonia emission and odor. Our results suggest more work needs to be conducted to understand the interactions among the pig, endogenous and exogenous enzymes, and different components of feedstuffs in order to effectively and efficiently use enzymes as cost effective feed additives in swine diets. From a practical swine nutrition perspective, it would appear that supplementing enzymes to pig diets will be more beneficial

when using diets based on ingredients that are of lower quality and poorly digested. The application of enzyme supplementation in swine diets, once perfected, will not only economically benefit producers but also, and perhaps more importantly, have a positive impact on the environment.

Literature Cited

AOAC. 1997. Official methods of analysis. 16th ed. Association of Official Analytical Chemists, Arlington, VA.

Baird, D. M., H. C. McCampbell and J. R. Allison. 1975. Effect of levels of crude fiber,protein and bulk diets for finishing hogs. J. Anim. Sci. 41:1039-1047.

Barrera, M., M. Cervantes, W. C. Sauer, A. B. Araiza, N. Torrentera, and M. Cervantes.2004. Ileal amino acid digestibility and performance of growing pigs fed wheat-based diets supplemented with xylanase. J. Anim. Sci. 82:1997-2003.

Bass, T. C., and P. A. Thacker. 1996. Impact of gastric pH on dietary enzyme activity andsurvivability in swine fed β-glucanase supplemented diets. Can. J. Anim. Sci.76:245-252.

Baidoo, S. K., Y. G. Liu, and D. Yungblut. 1998. Effect of microbial enzyme supplementation on energy, amino acid digestibility and performance of pigs fed hulless barley based diets. Can. J. Anim. Sci. 78:625-631.

Baucells, F., J. F. Perez, J. Morales, and J. Gasa. 2000. Effect of α-galactosidase supplementation of cereal-soya-bean-pea diets on the productive performances, digestibility and lower gut fermentation in growing and finishing pigs. J. Anim. Sci. 71:157-164.

Bedford, M.R., J. F. Patience, H. L. Classen, and J. Inborr. 1992. The effect of dietary enzyme supplementation of rye- and barley-based diets on digestion and subsequent performance in weanling pigs. Can. J. Anim. Sci. 72: 97-105.

Caine, W. R., W. C. Sauer, S. Tamminga, M.W.A. Verstegen, and H. Schulze. 1997. Apparent ileal digestibilites of amino acids in newly weaned pigs fed diets with protease-treated soybean meal. J. Anim. Sci. 75:2962-2969.

Canh, T. T., M. W. A. Verstegen, A. J. A. Aarnink, J. W. Schrama. 1997. Influence of dietary factors on nitrogen partitioning and composition of urine and feces of fattening pigs. J. Anim. Sci. 75:700-706.

Canh, T. T., A. L. Sutton, A. J. A. Aarnink, M. W. A. Verstegen, J. W. Schrama, and G. C. M. Bakker. 1998. Dietary carbohydrates alter the fecal composition and pH and the ammonia emission from slurry of growing pigs. J. Anim. Sci. 76:1887-1895.

Diebold, G., R. Mosenthin, H.-P. Piepho, and W. C. Sauer. 2004. Effect of supplementation of xylanase and phospholipase to a wheat-based diet for weanling pigs on nutrient digestibility and concentrations of microbial metabolites in ileal digesta and feces. J. Anim. Sci. 82:2647-2656.

Gdala, J., A. J. M. Jansman, L. Buraczewska, J. Huisman and P. van Leeuwen. 1997a. The influence of α-galactosidase supplementation on the ileal digestibility of lupin seed carbohydrates and dietary protein in young pigs. Anim. Feed Sci. Technol. 67:115-125.

Gdala, J., H. N. Johansen, K. E. Bach Knudsen, I. H. Knap, P. Wagner, and O. B. Jorgensen. 1997b. The digestibility of carbohydrates, protein and fat in small and large intestine of piglets fed non-supplemented and enzyme supplemented diets. Anim. Feed Sci. Technol. 65:15-33.

Gill, B. P., J. Mellange, J. A. Rooke. 2000. Growth performance and apparent nutrient digestibility in weaned piglets offered wheat-, barley- or sugar-beet pulp-based diets supplemented with food enzymes. Anim. Sci. 70:107-118.

Graham, H., K. Hesselman, and P. Aman. 1986. Influences of β-glucanase supplementation of a barley-based diet in the pig gastrointestinal tract. Nutr. Rep. Int. 34:1089-1096.

Graham, H., W. Lowgren, D. Pettersson, and P. Aman. 1988. Effect of enzyme supplementation on digestion of a barley/pollard-based pig diet. Nutr. Rep. Int. 38:1073-1079.

Guo, H., L. D. Jacobson, D. R. Schmidt, and R. E. Nicolai. 2001. Calibrating Inpuff-2 model by resident-panelists for long-distance odor dispersion from animal production sites. Appl. Eng. Agric. 17: 859–868.

Hogberg, A., J.E. Lindberg. 2004. Influence of cereal non-starch polysaccharides and enzyme supplementation on digestion site and gut environment in weaned piglets. Anim. Feed Sci. Technol. 116:113-128.

Inborr, J., M. Schmitz, and F. Ahrene. 1993. Effect of adding fiber and starch degrading enzymes to a barley/wheat based diet on performance and nutrient digestibility in different segments of the small intestine of early weaned pigs. Anim. Feed Sci. Technol. 44:113-127.

Inborr, J., and R. B. Ogle. 1988. Effect of enzyme treatment of piglet feeds on performance and post-weaning diarrhoea. Swedish Journal of Agricultural Research. 18:129–133.

Jensen, M. S., K. E. B. Knudsen, J. Inborr, K. Jakobsen. 1998. Effect of β-glucanase supplementation on pancreatic enzyme activity and nutrient digestibility in piglets fed diets based on hulled or hulless barley varieties. Anim. Feed Sci. Technol. 72: 329-345.

Kim, S. W., J. H. Zhang, K. T. Soltwedel, and D. L. Knabe. 2006. Use of carbohydrates in corn-soybean meal-based grower-finisher pig diet. Anim. Res. 55:563-578.

Kornegay, E. T. 1978. Feeding value and digestibility of soybean hulls for swine. J. Anim. Sci. 47:1272-1280.

Kornegay, E. T. 1981. Soybean hull digestibility by sows and feeding value for growing-finishing swine. J. Anim. Sci. 53:138-145.

Le, P. D., A. J. A. Aarnink, N. W. M. Ogink, P. M. Becker, and M. W. A. Verstegen. 2005. Odour from animal production facilities: its relationship to diet. Nutr. Res. Rev. 18:3-30.

Li, S., W. C. Sauer, S. X. Huang, and V. M. Gabert. 1996a. Effect of β-glucanase supplementation to hulless barley- or wheat-soybean meal diets on the digestibilities of energy, protein, β-glucans, and amino acids in young pigs. J. Anim. Sci. 74:1649-1656.

Li, S., N. C. Sauer, R. Mosenthin, and B. Kerr. 1996b. Effect of β-glucanase supplementation of cereal-based diets for starter pigs on the apparent digestibilities of dry matter, crude protein and energy. Anim. Feed Sci. Technol. 59:223-231.

Mavromichalis, I., J. D. Hancock, B. W. Senne, T. L. Gugle, G. A. Kennedy, R. H. Hines, and C. L. Wyatt. 2000. Enzyme supplementation and particle size of wheat in diets for nursery and finishing pigs. J. Anim. Sci. 78:3086-3095.

Medel, P., F. Baucells, M. I. Gracia, C. de Blas, and G. G. Mateos. 2002. Processing of barley and enzyme supplementation in diets for young pigs. Anim. Feed Sci. Technol. 95:113-122.

Merkel, R. A., R. W. Bray, R. H. Grummer, P. H. Phillips, and G. Bohnstedt. 1958. The influence of limited feeding, using high fiber rations, upon growth and carcass characteristics in swine. I. Effects upon feedlot performance. J. Anim. Sci. 17:3-12.

Moeser, A. J. and T. A. T. G. van Kempen. 2002. Dietary fibre level and enzyme inclusion affect nutrient digestibility and excreta characteristics in grower pigs. J. Sci. Food Agric. 82:1606-1613.

Mitaru, B. N., R. Blair, R. D. Reichert, and W. E. Roe. 1984. Dark and yellow rapeseed hulls, soybean hulls and purified fiber source: their effects on dry matter, energy, protein and amino acid digestibilities in cannulated pigs. J. Anim. Sci. 59:1510-1518.

NPPC. 1991. Procedures to evaluate market hogs. Third Ed. National Pork Producers Council, Des Moines, IA.

NRC, 1998. Nutrient requirements of swine. 10th revised edition. National Academic Press, Washington, D.C.

Otto, E. R., M. Yokoyama, S. Hengemuehle, R. D. von Bermuth, T. van Kempen, and N. L. Trottier. 2003. Ammonia, volatile fatty acids, phenolics and odor offensiveness in manure from growing pigs fed diets reduced in protein concentration. J. Anim. Sci. 81:1754-1763.

Partridge, I. G. 1978. Studies on digestion and absorption in the intestines of growing pigs. 4. Effects of dietary cellulose and sodium levels on mineral absorption. Br. J. Nutr. 39:539.

Pettey, L. A., S. D. Carter, B. W. Senne, and J. A. Shriver. 2002. Effects of β-mannanase addition to corn-soybean meal diets on growth performance, carcass traits, and nutrient digestibility of weanling and growing-finishing pigs. J. Anim. Sci. 80:1012-1019.

Shriver, J. A., S. D. Carter, A. L. Sutton, B. T. Richert, B. W. Senne, and L. A. Pettey. 2003. Effets of adding fiber sources to reduced-crude protein, amino acid-supplemented diets on nitrogen excretion, growth performance, and carcass traits of finishing pigs. J. Anim. Sci. 81:492-502.

Stanogias, G., G. R. Pearce, T. Alifakiotis, J. Michaelidis, and B. Pappa-Michaelidis. 1994. Efects of dietary concentration and source of fiber on the apparent absorption of minerals by pigs. Anim. Feed. Sci. Technol. 47:287-295.

Sutton, A. L., K. B. Kephart, M. W. A Verstegen, T. T. Canh, and P. J. Hobbs. 1999. Potential for reduction of odorous compounds in swine manure through diet modification. J. Anim. Sci. 77:430-439.

Tangendjaja, B., Z. B. Johnson, and P. R. Noland. 1988. Effect of cooking and addition of enzymes on feeding value of rice bran for swine. Nutrition Reports International. 37: 449–458.

Thacker, P. A., G. L. Campbell, and J. W. D. Grootwassink. 1988. The effect of beta-glucanase supplementation on the performance of pigs fed hulless barley. Nutr. Rep. Int. 38:91-99.

Thacker, P.A., G. L. Campbell, and J. W. D. GrootWassink. 1989. The effect of sodium bentonite on the performance of pigs fed barley-based diets supplemented with beta-glucanase. Nutr. Rep. Int. 40:613-620.

Thacker, P., G. Campbell, and J. GrootWassink. 1991. The effect of enzyme supplementation on the nutritive value of rye-based diets for swine. Can. J. Anim.Sci. 71:489-496.

Thacker, P. A., G. L. Campbell, and J. GrootWassink. 1992b. The effect of organic acids and enzyme supplementation of the performance of pigs fed barley-based diets. Can. J. Anim. Sci. 72:395-402.

van Heugten, E. and T. A. T. G. van Kempen. 2001. The Science of Odor. In: Swine News. Published by North Carolina Cooperative Extension Service. Oct. Vol. 24. No. 9.

Willis, R., M. Montgomery, and P. Allen. 1996. Improved method for manual, colorimetric determination of total Kjeldahl nitrogen using salicylate. J. Agric. Food Chem. 44:1804–1807.

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

	Grower 1[a]		Grower 2		Finisher 1		Finisher 2	
	Control	Soyhull	Control	Soyhull	Control	Soyhull	Control	Soyhull
Ingredient, %								
Corn	72.73	58.83	74.88	60.54	78.29	63.66	81.68	66.76
Soybean hulls	0.00	20.00	0.00	20.00	0.00	20.00	0.00	20.00
Soybean meal (48%)	23.60	17.55	20.10	14.50	16.80	11.50	13.50	8.50
Limestone	1.03	0.78	1.00	0.74	0.87	0.61	0.86	0.60
Monocalcium Phosphate	0.89	1.09	0.77	0.97	0.79	0.98	0.71	0.89
Vitamins-Minerals[b]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Salt	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Poultry Fat	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Celite[c]	0.00	0.00	1.50	1.50	1.50	1.50	1.50	1.50
Calculated Composition								
ME, kcal/kg	3367	3040	3322	2995	3327	3001	3314	2988
Calcium, %	0.650	0.653	0.607	0.607	0.550	0.550	0.522	0.521
Phosphorus, %	0.553	0.553	0.510	0.511	0.501	0.501	0.470	0.469
Crude Protein, %	17.25	15.42	15.76	14.11	14.48	12.95	13.15	11.74
Lysine, %[d]	0.902	0.811	0.802	0.723	0.711	0.641	0.619	0.557
Methionine, %	0.28	0.24	0.26	0.22	0.25	0.21	0.23	0.19
Threonine, %	0.65	0.75	0.59	0.70	0.54	0.66	0.49	0.61
Tryptophan, %	0.20	0.16	0.18	0.14	0.16	0.13	0.14	0.11

[a] Basal diet was made and enzyme was added to all soyhull diets at an inclusion rate of 0.05%.

[b]Supplied per kg of complete diet: 5,540 IU of vitamin A as retinyl acetate, 1,108 IU of vitamin D3, 22 IU of vitamin E as dl-α-tocopherol acetate, 1.98 mg of vitamin K as menadione dimethylpyrimidinol bisulfite, 165 mg of choline as choline chloride, 22 mg of niacin, 17.6 mg of d-pantothenic acid as dl-calcium pantothenate, 4.4 mg of riboflavin, 1.1 mg of pyridoxine as pyridoxine•HCl, 0.55 mg thiamine as thiamine mononitrate, 0.022 mg of vitamin B12, 0.33 mg of folic acid, 0.04 mg of d-biotin, 110 mg Zn as

Table 1 (continued).

ZnSO4, 110 mg Fe as FeSO4, 22 mg Cu as CuSO4, 55 mg Mn as MnO, 0.28 mg I as EDDI, and 0.30 mg Se as NaSeO3.

[c] Used as indigestible marker (Celite Corporation, Lompoc, CA).

[d] Amino acids are expressed on an ileal apparent digestible basis.

Table 2. Effects of enzyme inclusion to corn-soybean meal diets on growth performance of grower-finisher pigs

					P value[a]		
Variable	Positive	Negative	Negative+Enzyme	SEM	P vs. N	N vs. N+E	P vs. N+E
Body weight, kg							
Day 0[b]	33.93	33.74	33.77	0.33	0.68	0.95	0.72
Day 28[c]	54.85	53.67	54.08	0.77	0.28	0.70	0.48
Day 56[c]	81.97	78.66	79.94	1.32	0.09	0.49	0.28
Day 70[d]	95.46	92.07	93.70	1.28	0.07	0.36	0.33
Day 89	114.82	110.86	112.06	1.94	0.15	0.65	0.31
Average daily gain, kg/d							
Day 0 to 28[b]	0.75	0.71	0.73	0.02	0.17	0.54	0.43
Day 29 to 56[c]	0.97	0.89	0.92	0.03	0.07	0.44	0.27
Day 57 to 70[c]	0.96	0.96	0.98	0.05	0.93	0.70	0.76
Day 71 to 89[d]	0.99	0.99	0.95	0.05	0.97	0.63	0.61
Total	0.91	0.87	0.88	0.02	0.12	0.60	0.27
Average daily feed intake, kg/d							
Day 0 to 28[b]	1.64	1.74	1.85	0.05	0.17	0.16	0.01
Day 29 to 56[c]	3.10	3.30	3.44	0.07	0.07	0.18	0.01
Day 57 to 70[c]	3.30	3.61	3.64	0.10	0.04	0.85	0.03
Day 71 to 89[d]	3.38	3.69	3.69	0.12	0.07	1.00	0.08
Total	2.62	2.82	2.90	0.07	0.05	0.37	0.01
Gain/Feed							
Day 0 to 28[b]	0.46	0.41	0.40	0.01	0.01	0.45	0.002
Day 29 to 56[c]	0.31	0.27	0.27	0.01	0.01	0.85	0.01
Day 57 to 70[c]	0.29	0.27	0.27	0.01	0.17	0.91	0.21
Day 71 to 89[d]	0.29	0.27	0.26	0.01	0.26	0.58	0.10
Total	0.35	0.31	0.30	0.01	0.01	0.74	0.01

[a] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

Table 2 (continued).

[b] Each value represents a mean of 24 pens with 4 pigs per pen in 3 pens and 5 pigs per pen in 21 pens.

[c] Each value represents a mean of 24 pens with 3 pigs per pen in 1 pen, 4 pigs per pen in 9 pens, and 5 pigs per pen in 15 pens.

[d] Each value represents a mean of 24 pens with 2 pigs per pen in 3 pens, 3 pigs per pen in 6 pen, 4 pigs per pen in 8 pens, and 5 pigs per pen in 7 pens.

Table 3. Effects of enzyme inclusion to corn-soybean meal diets on energy of grower-finisher pigs[a]

					P value[b]		
Variable	Positive	Negative	Negative+Enzyme	SEM	P vs. N	N vs. N+E	P vs. N+E
Metabolizable Energy (Mcal/d)							
Day 28	5.54	5.23	5.60	0.17	0.30	0.19	0.77
Day 56	10.32	9.87	10.30	0.23	0.17	0.19	0.95
Day 70	10.99	10.83	10.91	0.32	0.72	0.85	0.86
Day 89	11.19	11.01	11.01	0.37	0.73	1.00	0.72
Total	8.73	8.45	8.71	0.21	0.34	0.39	0.93
Gain/Metabolizable Energy (kg/Mcal)							
Day 28	0.136	0.135	0.130	0.004	0.74	0.42	0.27
Day 56	0.094	0.091	0.090	0.003	0.47	0.85	0.37
Day 70	0.088	0.089	0.090	0.004	0.86	0.91	0.77
Day 89	0.089	0.090	0.087	0.005	0.78	0.57	0.77
Total	0.104	0.103	0.101	0.003	0.71	0.73	0.47

[a] Each value represents a mean of 24 pens with 4 pigs per pen in 3 pens and 5 pigs per pen in 21 pens.

[b] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

Table 4. Effects of enzyme inclusion to corn-soybean meal diets on ultrasound carcass characteristics of grower-finisher pigs[a,b,c]

					P value[d]		
Variable	Positive	Negative	Negative+Enzyme	SEM	P vs. N	N vs. N+E	P vs. N+E
Backfat, cm	1.89	1.79	1.90	0.09	0.41	0.37	0.94
Loin eye, cm^2	44.25	41.11	40.31	0.62	0.002	0.35	<0.001
Fat-free lean, %	51.76	51.22	50.49	0.43	0.38	0.23	0.03
Lean gain, g/d	353.15	332.77	328.53	7.19	0.06	0.67	0.02

[a] Each value represents a mean of 24 pens with2 pigs per pen in 3 pens, 3 pigs per pen in 6 pens, and 4 pigs per pen in 8 pens, and 5 pigs per pen in 7 pens.

[b] All measurements taken on day 89.

[c] Pig final BW was used as a covariate in the data analysis.

[d] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

Table 5. Effect of enzyme inclusion to corn-soybean meal diets on fecal digestibility of grower-finisher pigs[1,2]

	Positive	Negative	Negative + Enzyme				
Fecal digestibility (%)					P value[b]		
				SEM	P vs. N	N vs. N+E	P vs. N+E
GE	84.9[a]	79.3[b]	83.1[a]	0.7	<0.001	0.001	0.07
CP	79.8[a]	75.2[b]	76.6[b]	0.8	0.001	0.21	0.001
ADF	87.0[a]	21.9[b]	67.6[a]	5.4	<0.001	<0.001	0.02
NDF	79.3[a]	31.4[b]	72.1[a]	2.8	<0.001	<0.001	0.08
Ca	58.9[a]	53.5[b]	61.2[a]	1.7	0.04	0.01	0.33
P	51.9[a]	51.7[a]	59.7[b]	1.4	0.93	0.001	0.001
Mg	46.7[a]	33.5[b]	48.6[a]	2.1	<0.001	<0.001	0.50
K	79.2[a]	72.8[b]	78.6[a]	1.0	<0.001	<0.001	0.64
Na	78.7	70.2	73.9	3.0	0.06	0.39	0.26
Fe	54.8[a]	23.3[a]	56.5[a]	0.9	<0.001	<0.001	0.18
Zn	25.2[a]	28.5[a]	37.2[b]	1.2	0.06	<0.001	<0.001
Cu	22.8[a]	34.5[b]	50.3[c]	2.1	0.001	<0.001	<0.001
Mn	30.6[a]	26.8[a]	43.2[b]	5.3	0.60	0.04	0.11

[1] At least 2 samples from 2 pigs were collected from each of the 24 pens.

[2] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

[a,b,c] Values with different superscripts differ by P<0.05.

Table 6. Effect of enzyme inclusion to corn-soybean diets on ammonia emissions of 400 ml fresh manure slurry[a]

				P value[b]		
Variable	Positive	Negative	Negative+Enzyme	P vs. N	N vs. N+E	P vs. N+E
Hours						
12	6.54	8.05	5.60	0.82	0.72	0.90
24	19.91	20.77	16.38	0.90	0.52	0.62
36	42.64	32.77	27.78	0.15	0.46	0.04
48	46.91	35.87	34.14	0.11	0.80	0.08
72	52.86	48.28	42.30	0.50	0.38	0.15
96	39.55	34.73	38.3	0.48	0.60	0.86

[a] Each observation is a mean of 4 or 5 replications, with a pooled SEM of 4.7.

[b] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

Table 7. Effect of enzyme inclusion to corn-soybean diets on ammonia emissions of 400 ml aged manure slurry[a]

				P value[b]		
Variable	Positive	Negative	Negative+Enzyme	P vs. N	N vs. N+E	P vs. N+E
Hours						
12	7.15	3.60	3.44	0.36	0.97	0.34
24	13.36	6.61	6.30	0.08	0.93	0.07
36	15.15	9.04	8.41	0.12	0.88	0.08
48	19.36	12.14	11.13	0.06	0.78	0.04
72	32.05	20.61	18.80	0.004	0.61	0.001
96	31.11	19.20	19.61	0.003	0.91	0.004

[a] Each observation is a mean of 4 or 5 replications, with a pooled SEM of 2.7.

[b] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

Table 8. Effects of enzyme inclusion to corn-soybean meal on manure odor intensity and hedonic tone score.

	Fresh Manure[a]			Aged Manure[b]				P value		
Diet[c]	P	N	N + E	P	N	N + E	SEM	manure	treatment	manure x treatment
Intensity[d]	2.89	2.85	2.70	3.31	3.00	3.35	0.10	0.01	0.65	0.37
Hedonic Tone[e]	-4.88	-4.80	-4.85	-6.00	-4.90	-5.60	0.20	0.03	0.22	0.31

[a] Manure samples mixed by individual feces and urine collections.

[b] Manure samples after anaerobic aging at room temperature for 21 days.

[c] P = positive control diet, N = negative control diet, and N+E = negative control diet plus enzyme.

[d] Odor intensity was evaluated by comparing the odor intensity of the headspace air to the odor intensities of a series of concentrations of n-butanol.

[e] Hedonic tone, degree of pleasantness or unpleasantness according to a -10 to +10 hedonic tone scale.